<Original Article>

# Relationship between team performance levels and serum SH (sulfhydryl group) changes brought about by intensive athletic training summer camp

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**Summary** Serum SH (sulfhydryl group) exists as either bound SH (B-SH), which is bound to various stress substances, or free SH (F-SH). Total SH (T-SH) is the sum of the two forms of SH. We previously reported that aerobic exercise increases serum F-SH, while anaerobic exercise decreases serum F-SH and increases serum B-SH.

Athletes are subjected to exercise stress brought about by daily training, and it can be assumed that the motor performance of athletes is correlated with their ability to relieve exercise stress. Among various sports, rugby football involves both aerobic and anaerobic exercises, and the relationship between athletes' performance levels and the changes in serum SH caused by exercise was investigated in athletes attending summer training camp, in which athletes train intensely before the start of the season.

Before training camp, the F-SH levels was the highest in teams A, followed in order by team B and C. The F-SH levels was high for teams with high performance levels, and the lower the performance level, and the B-SH level was the lowest in Team A, followed in order by team B and C. After training camp, changes in F-SH and B-SH levels differed among the teams, suggesting that training exercises affected F-SH.

Key words: T-SH, F-SH, B-SH, Physical stress

## 1. Introduction

Training improves the motor performance of athletes, but it is well known that alternating between

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Received for Publication June 25, 2009 Accepted for Publication September 15, 2009 mance, but also harm athletes' health.

Exercise stress is a generic term for various factors that are harmful to health. In general, exercise stress is caused by exercising, and it is conjugated and buffered in the body's organs while resting or exercising. For example, lactic acid (LA) generated in muscles is broken down by lactate dehydrogenase (LD)<sup>1), 2)</sup>. Active oxygen generated by intense breathing exercise can be easily decomposed by superoxide dismutase (SOD) in the serum<sup>3)-5)</sup>. However, when intense exercise stress generates too much active oxygen to be broken down and buffered, exercise stress causes psychosomatic damage and then pathologic states. Research is currently being conducted in order to determine the relationship between rest and intense exercise that are essential to athletes, but decisions regarding the amounts of rest and intense exercise are being made based mostly on the experiences of individual managers and coaches.

Serum SH (sulfhydryl group) exists as free-SH (F-SH), which is not bound to any substance and accounts for about 75% of all forms of SH in the body, and bound-SH (B-SH), which is bound to substances such as cysteine or glutathione. The sum of the two forms of SH is total-SH (T-SH). The conversion between B-SH and F-SH is a reversible reaction that is routinely observed, and the ratio of these compounds changes with age or disease. We developed an improved method that not only can measure the F-SH concentration rapidly, conveniently and accurately, but also can measure the B-SH concentration using an ammonium oxalate treatment<sup>6)</sup>. The results of clinical application showed that hemodialysis increased F-SH levels<sup>6</sup>, and in the field of exercise physiology, long-distance running increased F-SH levels7). An animal study using mice showed that aerobic exercise increased the F-SH level, while anaerobic exercise decreased the F-SH level and increased the B-SH level<sup>8)</sup>, suggesting that F-SH conjugates and buffers various forms of stress and is then converted into B-SH.

Rugby football involves both aerobic and anaerobic exercises<sup>9)</sup>. The present study investigated the effects of exercise stress on biochemical test findings and serum SH in members of three university rugby teams with different performance levels during preseason intense summer training camp.

### 2. Materials and methods

The subjects were members of the following three university rugby football teams in the Tokai Student Rugby Football League (Tokai Student League): University A (average age,  $19.5 \pm 1.3$  years; n=21); University B (average age,  $18.9 \pm 0.8$  years; n=13); and University C (average age,  $20.8 \pm 2.0$  years; n=13). These volunteers participated in a summer training camp in Sugadaira, Nagano Prefecture. Team A belonged to the Tokai Student League A; Team B is one of the top teams in the Tokai Student League B; and Team C is one of the bottom teams in the Tokai Student League C. Team A had the highest team ranking, followed in order by Teams B and C. While it was highly possible that the performance level of individual athletes varied within each team, the ranking in the Tokai Student League was an indicator of overall team performance level. During the training camp, the number of training days, the amount of exercise, exercise methods and lifestyle habits were not standardized.

Blood samples were collected before and after the training camp. Prior to training camp, blood samples were collected before the first day of training held at each university, and blood samples were collected after the end of training on the last day of training camp. Written informed consent was obtained from all volunteers, and our study was in compliance with the rule for human experimentation at our institution.

With regard to biochemical tests, total protein (TP), serum albumin (ALB), total bilirubin (TB), direct bilirubin (DB), AST (GOT), ALT (GPT), ALP, LD, cholinesterase (CHE),  $\gamma$ -GT, inorganic phosphorus (IP), CK, ZTT and TTT levels were measured using an automatic analyzer (HITACHI 7600). Serum LA level was measured using Lactate Pro (ARKRAY).

Serum SH levels were measured using the modified thiocholine method<sup>6)</sup> that we developed. For comparison (Control group), the serum SH levels

|                     | in the ave      | erage before a     | und after | training ca | ump (%). P va    | lues indicate      | signific | ant differe | nces before ai     | nd after trainin   | g camp.  |         |
|---------------------|-----------------|--------------------|-----------|-------------|------------------|--------------------|----------|-------------|--------------------|--------------------|----------|---------|
|                     |                 | Team A             |           |             |                  | Team B             |          |             |                    | Team C             |          |         |
|                     | before          | after              | rate (%)  | Р           | before           | after              | rate (%) | Р           | before             | after              | rate (%) | Р       |
| LA (mmol/L)         | $1.42 \pm 0.53$ | $1.15 \pm 0.29$    | 88.7      | P<0.05      | $1.36 \pm 0.24$  | $1.19 \pm 0.18$    | 88.7     | P<0.01      | $1.49 \pm 0.37$    | $1.05 \pm 0.20$    | 73.3     | P<0.001 |
| TP (g/dL)           | $7.28 \pm 0.29$ | $7.35 \pm 0.19$    | 101.1     | n.s.        | $7.43 \pm 0.33$  | $7.37 \pm 0.30$    | 99.4     | n.s.        | $7.53 \pm 0.31$    | $7.23 \pm 0.24$    | 96.1     | P<0.001 |
| ALB (g/dL)          | $4.64 \pm 0.18$ | $4.70 \pm 0.21$    | 101.3     | n.s.        | $4.70 \pm 0.25$  | $4.66 \pm 0.24$    | 99.3     | n.s.        | $4.88\pm0.19$      | $4.68 \pm 0.19$    | 95.9     | P<0.005 |
| TB (mg/dL)          | $0.83 \pm 0.29$ | $0.86 \pm 0.24$    | 112.1     | n.s.        | $0.80 \pm 0.27$  | $0.67 \pm 0.24$    | 2.68     | n.s.        | $0.99 \pm 0.23$    | $1.23 \pm 0.25$    | 125.3    | P<0.001 |
| DB (mg/dL)          | $0.17 \pm 0.03$ | $0.19 \pm 0.03$    | 109.2     | n.s.        | $0.20 \pm 0.00$  | $0.23 \pm 0.23$    | 115.4    | n.s.        | $0.27 \pm 0.04$    | $0.33\pm0.04$      | 121.6    | P<0.001 |
| AST (IU/L)          | $30.8 \pm 10.6$ | $33.4\pm16.5$      | 112.3     | n.s.        | $19.1 \pm 5.1$   | $33.2 \pm 9.1$     | 176.7    | P<0.001     | $28.2 \pm 8.8$     | $132.4 \pm 77.5$   | 474.5    | P<0.001 |
| ALT (IU/L)          | $19.8 \pm 9.1$  | $24.4 \pm 9.2$     | 123.2     | P<0.01      | $20.5 \pm 11.2$  | $33.7 \pm 16.6$    | 173.9    | P<0.001     | $22.7 \pm 13.6$    | $59.1 \pm 31.4$    | 313.6    | P<0.001 |
| ALP (IU/L)          | $300.0\pm82.5$  | $345.0 \pm 90.7$   | 115.6     | P<0.001     | $284.0\pm71.5$   | $278.5 \pm 67.6$   | 6.7      | n.s.        | $248.5 \pm 67.1$   | $238.8\pm69.2$     | 96.1     | n.s.    |
| LD (IUL)            | $228.7\pm50.2$  | $298.8 \pm 101.1$  | 129.6     | P<0.001     | $167.1 \pm 21.0$ | $234.3 \pm 73.5$   | 142.4    | P<0.01      | 254.8±42.5         | $623.6\pm 208.1$   | 246.0    | P<0.001 |
| CHE (IU/L)          | 5725.6±789.7    | $5472.1 \pm 736.8$ | 95.8      | P<0.005     | $5461.8\pm855.0$ | $5279.9 \pm 954.6$ | 96.9     | n.s.        | $4931.8 \pm 688.8$ | $4589.1 \pm 599.6$ | 93.3     | P<0.001 |
| $\gamma$ -GT (IU/L) | $24.0 \pm 8.1$  | $21.6\!\pm\!9.5$   | 90.3      | n.s.        | $22.4 \pm 10.4$  | $28.6 \pm 12.1$    | 132.1    | n.s.        | $19.8\pm9.8$       | $17.5 \pm 5.8$     | 92.2     | n.s.    |
| IP (mg/dL)          | $3.64 \pm 0.64$ | $3.51\pm0.38$      | 99.4      | n.s.        | $3.31 \pm 0.32$  | $3.92 \pm 0.39$    | 118.6    | P<0.001     | $2.40 \pm 0.30$    | $2.69 \pm 0.27$    | 114.1    | P<0.05  |
| CK (IU/L)           | $30.1 \pm 11.2$ | $49.0 \pm 31.0$    | 164.6     | P<0.005     | $24.0 \pm 10.7$  | $219.8 \pm 146.1$  | 974.5    | P<0.001     | $554.5 \pm 365.4$  | $7831.5\pm 5934.0$ | 1624.4   | P<0.001 |
| ZTT (U)             | $7.16\pm 2.07$  | 8.17±2.45          | 114.5     | P<0.001     | $7.71 \pm 2.04$  | $10.98 \pm 2.85$   | 143.9    | P<0.001     | $5.84 \pm 1.98$    | $5.37 \pm 1.90$    | 91.6     | P<0.05  |

Results of serum biochemical tests values for each team before and after training camp. All values are means  $\pm$  SD. The rate of change **Fable 1** 

were measured in male students in the same age group (average age,  $19.5 \pm 1.2$  years; n=67).

Paired *t*-test was used to examine the significance of differences before and after training camp, and multiple comparison test was performed to assess the significance of differences among the teams. The level of significance was set at p<0.05.

## 3. Results

### 1. Biochemical tests

The LA levels decreased significantly after training camp in all three teams. The CHE level tended to decrease in Team A and significantly decreased in Team C. The levels of TP and ALB did not markedly change in Teams A and B, but significantly decreased in Team C. The levels of TB and DB did not markedly change in Teams A and B, but significantly increased in Team C. Levels of AST, ALT, LD and CK either tended to increase or significantly increased for all teams. In addition, the lower the team rank, the greater the rate of increase. The ZTT and TTT levels of each team either tended to increase or significantly increased, but their dynamics lacked consistency (Table 1).

# 2. Serum SH

# (1) Changes in T-SH levels

Before the training camp, the T-SH level of Team A was the highest, followed in order by Teams B and C. In other words, the lower the team rank, the lower the T-SH level. Significant differences were seen between Teams A and B (p<0.001) and between Teams A and C (p<0.001), but not between Teams B and C. When compared with the Control group, the T-SH level was significantly higher in Team A (p<0.001), but was significantly lower in Teams B and C (p<0.05 each) (Fig. 1-1).

After the training camp, significant differences in T-SH level were seen between Teams A and B (p<0.01), between Teams A and C (p<0.001) and between Teams B and C (p<0.05). When compared with the Control group, the T-SH level was significantly higher in Team A (p<0.05), but was significantly lower in Team C (p<0.001) (Fig. 1-2).

P<0.001

66.1

 $1.89 \pm 1.50$ 

 $2.77 \pm 1.83$ 

n.s.

106.2

 $3.10 \pm 1.99$ 

 $2.95 \pm 1.40$ 

P<0.001

64.0

 $1.40 \pm 0.77$ 

 $2.35 \pm 1.23$ 

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Fig. 1-1 Levels of T-SH before training camp. Levels of T-SH for Team A were the highest, followed in order by Teams B and C.



\*:P<0.05, \*\*:P<0.01, \*\*\*\*:P<0.001

Fig. 1-2 Levels of T-SH after training camp. Like before training camp, levels of T-SH for Team A were the highest, followed in order by Teams B and C.



Fig. 1-3 Levels of T-SH for each team before and after training camp



Fig. 2-1 Levels of F-SH before training camp. Levels of F-SH for Team A were higher than those for the other two teams.



Fig. 2-2 Levels of F-SH after training camp. Levels of F-SH for Team C were lower than those for the other two teams.



Fig. 2-3 Levels of F-SH before and after training camp. Levels of F-SH for Team B increased after training camp.

Before and after the training camp, no significant differences were confirmed in any team (Fig. 1-3). (2) Changes in F-SH levels

Before training camp, the F-SH level was the highest in Team A, followed in order by Teams B and C. In other words, the lower the team rank will have a the lower the F-SH level. Significant differences in F-SH level were seen between Teams A and B (p<0.001) and between Teams A and C (p<0.001), but not between Teams B and C. When compared with the Control group, the F-SH level of Team A was significantly higher (p<0.001) (Fig. 2-1).

After training camp, significant differences in F-SH level were seen between Teams A and C (p<0.001) and between Teams B and C (p<0.05). When compared with the Control group, the F-SH level of

Team A was significantly higher (Fig. 2-2).

Comparing the F-SH levels before and after training camp, there were significant differences in Teams A and B (both p<0.05) (Fig. 2-3).

(3) Changes in B-SH level

Before training camp, the B-SH level was the lowest in Team A, followed in order by Teams B and C. In other words, the lower the team rank, will have a the higher the B-SH level. Significant differences in B-SH level were seen between Teams A and B (p<0.05) and between Teams A and C (p<0.01), but not between Teams B and C. When compared with the Control group, the B-SH level of Team A was significantly lower (p<0.01) (Fig. 3-1).

After training camp, the B-SH level of Team B was the lowest, followed in order by Teams A and C, but no significant differences were seen among the groups. When compared with the Control group, the B-SH levels of Teams A and B were significantly lower (both p<0.005) (Fig. 3-2). Before and after the training camp, a significant difference was seen only in Team B (p<0.05) (Fig. 3-3).

(4) Ratio of F-SH to T-SH

Before the training camp, the ratio of F-SH to T-SH was the highest in Team A, followed in order by Teams B and C. Significant differences in the ratio of F-SH to T-SH were seen between Teams A and B (p<0.05) and between Teams A and C (p<0.005). When compared with the Control group, a significant difference was only seen with Team A (p<0.001).

After the training camp, the ratio of F-SH to T-SH in Team A was slightly lower, but was still significantly higher when compared with the Control group (p<0.05). In Team B, the ratio of F-SH to T-SH increased to a level comparable to that in Team A, and a significant difference was confirmed between the Control group and Team B (p<0.01). After the training camp, the ratio of F-SH to T-SH in Team C increased slightly, but no significant difference was seen between Team C and the Control group (Fig. 4).

#### 4. Discussion

1. Changes in laboratory test findings due to exercise LA is believed to be a fatigue-related substance



Fig. 3-1 Levels of B-SH before training camp. Levels of B-SH for Team A were lower than those for the other two teams.



Fig. 3-2 Levels of B-SH after training camp



Fig. 3-3 Levels of B-SH before and after training camp. Levels of B-SH for Team B decreased markedly.



Fig. 4 Ratios of F-SH to T-SH before and after training camp for each team.

that accumulates in muscles after excess exercise. However, in recent years, it has been thought that LA is produced when fast muscle fibers break down glucose, and LA is used as the source of energy for mitochondria in heart muscle cells and slow muscle fibers while exercising<sup>10)-13)</sup>. The LA level significantly decreased after training camp in all teams, and the results suggest that LA was consumed as a source of energy via the above-mentioned metabolic pathway.

After the training camp, the levels of AST, ALT, LD and CK either tended to increase or significantly increased in all teams. It is well known that these levels increase after exercise<sup>14), 15)</sup>. The increases in these enzyme levels may be due to concentration by perspiration, but even in Team B, in which the TB level tended to decrease after training camp, the levels of these enzymes increased, thus suggesting that exercise stress damages the liver. This observation is supported by the finding that the CHE level tended to decrease or significantly decreased in all teams. The reasons for this are that exercise stress induced hepatopathy, thus impairing CHE synthesis, destroying hepatocytes and increasing the levels of liver-derived enzymes in the serum. The finding of an inverse relationship between team ranking and the rate of increase suggests that the motor performance of athletes in high-rank teams was high on average and was less likely to be impacted by the amount of exercise during training camp.

The marked fluctuations in ZTT and TTT were due to the marked fluctuations in the electrolyte balance as a result of sweating caused by vigorous exercise in hot weather and water consumption. This result suggests the importance of replenishing the water and mineral supply during exercise.

## 2. Changes in serum SH due to exercise

Before and after training camp, the T-SH levels of Team A were the highest, followed in order by Teams B and C. Hence, the lower the team rank, the lower the T-SH level. This suggests that T-SH resists or buffers exercise stress.

Before and after training camp, the F-SH levels of Team A were the highest, followed in order by Teams B and C. Hence, the lower the team rank, the lower the F-SH level. This suggests that the higher the F-SH level, the higher the team rank. T-SH is the sum of F-SH and B-SH, and F-SH is antagonistic to B-SH, thus suggesting that F-SH is the main substance that conjugates and buffers exercise stress.

Before training camp, the F-SH level of Team A was higher than that of the Control group, and the normal exercises performed by Team A consisted mostly of aerobic exercise and the increased F-SH level in this group was also due to aerobic exercise. On the other hand, the F-SH level of Teams B and C was lower than that of the Control group, suggesting that F-SH conjugated and buffered exercise stress that was elevated by routine exercise.

After training camp, the F-SH level of Team A tended to decrease; this suggests that the exercises during training camp consisted mostly of anaerobic exercise. Even after training camp, the F-SH level of Team A was significantly higher than that of the Control group, suggesting that the team had excess ability to conjugate and buffer exercise stress. The F-SH level of Team B increased significantly after training camp this suggests that exercise during training camp mostly consisted of aerobic exercise and that Team B was better able to conjugate and buffer exercise stress. The F-SH level of Team C tended to increase after training camp, suggesting that exercise during training camp mostly consisted of mild aerobic exercise. However, the F-SH level of Team C was comparable to that of the Control group, suggesting that the team did not have excess ability to conjugate and buffer exercise stress.

## 3. Significance of serum SH due to exercise

In the present study, the ranking of rugby teams was correlated with the F-SH level and was inversely proportional to the B-SH level. The amount of daily exercise varies among teams, and players who can endure training make the team. Motor performance naturally improves in teams that train and exercise, but athletes who lack the ability to relieve exercise stress become fatigued and do not make the team, thus increasing the motor performance of the entire team. Inevitably, the levels of F-SH, which resists exercise stress and conjugates and buffers exercise stress, are high, while the levels of B-SH are low; in other words, the higher the ratio of F-SH to T-SH, the higher the motor performance of the entire team.

To date, it has generally been thought that good outcomes indicate high motor performance in individual athletes. However, the outcome of games is based on athletic competition and also represents the results of training of athletes and maintenance of physical condition. By measuring blood components after exercise, it is possible to estimate performance based on increases and decreases in laboratory test findings, but the results of blood component analyses only reflect the changes brought about by exercise, and it is not possible to predict training performance or maintenance of physical condition based on precompetition test results. The levels of F-SH and B-SH and the ratio of F-SH to T-SH that were measured before competition may serve as indicators for the motor performance and physical condition maintenance of teams.

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