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Original Article

A Study for Prevention of Chronic Fatigue. Part 2. Effects of Strenuous Physical Exercise Performed in a Training Camp on Serum Enzyme Activity Levels and Subjective Fatigue.

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Abstract

The principal objective of this paper is to develop a simple and rapid method of estimating levels of fatigue so that chronic fatigue can be prevented. Long-distance runners belonging to a successful corporate team (Group A; 25 males) and representative runners at the prefectural level (Group B; 14 males) participated in this study. We examined the effects of strenuous physical exercise on serum enzyme activity and the fatigue level felt by the runners (subjective fatigue).

The following parameters were measured on two consecutive mornings during a training period: physical characteristics, serum-biochemistry using the dry-chemistry method, and subjective fatigue determined using the questionnaire regarding subjective symptoms authorized by the Japan Association of Industrial Health and the Profile of Mood State (POMS). Group A was divided into A-Senior (17 males; highest performance level) and A-Freshman (8 males) subgroups according to the length of employment within the corporation (one year or more and less than one year, respectively).

The levels of serum aspartate aminotransferase (AST) and creatine kinase (CK) were significantly lower in the A-Senior group than the other groups and this group displayed the "iceberg" POMS profile at both examinations. Some significant correlations between the elements of POMS and serum enzyme activity levels were observed among all three groups during both examinations. The decline of serum CK levels tended to accompany a decrease in "Fatigue" according to POMS among 9 of 17 members of the A-Senior group. This tendency between the A-Senior and the A-Freshman groups statistically differed according to the χ-square analysis.

Our results suggest that the effects of physical stress on serum enzyme activity levels and subjective fatigue are affected by performance levels. Physical fatigue seemed to be reflected by serum CK levels. Monitoring subjective fatigue while measuring serum enzyme activity levels using the dry-chemistry method immediately provides clinical value to players and coaches on site, and should therefore help to prevent a shift from "overreaching" to "overtraining".

Key words: Male Adults, Overtraining, Serum enzyme activity, Subjective fatigue, Dry-chemistry method

INTRODUCTION

Chronic fatigue has been defined as a decreased capacity to perform physical and mental work, overwhelming sustained exhaustion, a lack of energy, tiredness and a combination of these.[1] Overtraining due to exercise stress is considered to be one type of chronic fatigue syndrome[2] and the amount of research into this condition is increasing.[3] However, there are no diagnostic or warning tests that indicate overtraining,[4,5] as there are obvious ethical limitations associated with intentionally overtraining athletes.[6] In athletic training, workloads are gradually increased, thereby exceeding that previously applied. Improvements in an athlete's ability are achieved through

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adaptation to overload stimuli applied during the training programme.[7] When the overload is too great and recovery and adaptation do not occur, the athlete overreaches or overtrains.[8] Falsetti[9] stated that short term overtraining is overreaching. The symptoms of overreaching can be reversed with supercompensation by a longer than normal regeneration period and overreaching is used to improve performance levels.[10] Such systems have been applied by following training plans determined by experience.

Many investigators have reported the effects of training on blood properties among athletes.[11-14] However, such examinations are time-consuming and expensive. Monitoring subjective fatigue levels using a questionnaire is a more convenient and inexpensive means of estimating fatigue. However, questionnaires[15,16] applied through many types of field surveys,[17,18] have shown that personal estimates of physical fatigue are affected by mental stress.[19] Therefore psychological monitoring of overtraining may be useful if supported by physiological measurements.[20]

The principal objective of this paper is to develop methods to prevent chronic fatigue. Long-distance runners belonging to a successful corporate team and representative runners at the prefectural level participated in this study. The effects of strenuous physical exercise performed during a training camp upon levels of serum enzyme activities and upon the amount of fatigue felt by the runners (subjective fatigue) were studied.

MATERIALS AND METHODS

1. Athletes and protocols

Long-distance runners belonging to a successful corporate team (Group A; 25 males) and representative runners at the prefectural level (Group B; 14 males) participated in a one week training camp during September 1994. The purpose of the study was explained to the participants before the survey, which then proceeded with their informed consent. Group A was divided into A-Senior (17 males) and A-Freshman (8 males) groups, according to the length of employment in the company (one year or more and less than one year, respectively).

Examinations were conducted on two consecutive mornings after the day when 3 times of 5,000 meter time-trial, which were most strenuous physical exercise performed in the training camp, were carried out.

2. Physical characteristics

Athletes were physically examined twice. Age and height were recorded once during the first examination. Body weight, body fat weight, percentage of body fat, lean body mass, and body water were measured by the bioelectrical impedance using a Tanita body fat analyzer (TBF-102, Tanita Co.) in the athlete mode.[21]

3. Blood sampling and serum-biochemistry

Blood was sampled twice at similar times (between 6:00 to 6:30 am) in one room at the athletes' accommodation. Peripheral venous blood samples (1 ml) were drawn by antecubital venepuncture with the athletes in a resting position. Serum was separated using an exclusive micro-amount centrifuge (CF-9510, Kyoto Daiichi Kagaku Co.). Serum samples were immediately measured by the dry-chemistry method[22] using a Spotchem system (SP-4410, Kyoto Daiichi Kagaku Co.) as described.[23] Serological tests for aspartate aminotransferase (AST), lactate dehydrogenase (LD), and creatine kinase (CK) were simultaneously performed using a reagent strip (Heart-2, Kyoto Daiichi Kagaku Co.).

4. Monitoring subjective fatigue.

We monitored subjective fatigue by using the subjective symptoms of fatigue (SSF) questionnaire authorized by the Japan Association of Industrial Health[15] and the Profile of Mood State (POMS) questionnaire.[16,17] These questionnaires were clearly explained before the athletes received and completed them in the room described above.

The scores for the three elements in SSF, "drowsiness and dullness" (SSF-I), "difficulty in concentration" (SSF-II), and "projection of physical disintegration" (SSF-III), were counted, with 10 as the maximum score for each element.[16] Scores for the six elements in POMS were counted and converted into a T-score.[16,17] The total mood state (TMS) of POMS was computed by adding the five negative mood states (tension, depression, anger, fatigue, and confusion) and subtracting one positive mood state (vigor).

5. Relationship between serum CK levels and "fatigue" in POMS.

To clarify the relationship between serum CK levels and "fatigue" in POMS, changes in serum CK levels and T-scores of POMS over time were calculated as follows: the value difference between the second and the first examinations was divided by the corresponding value of the first examination. Results obtained for each athlete were plotted on a scatter plot with changes in serum CK levels as the horizontal axis and "fatigue" in POMS as the vertical axis. The scatter plot was divided into four sections, (A), (B), (C), and (D), by two axes. When there was no change in serum CK level or "fatigue" in POMS over time, these results were plotted on the vertical or the horizontal axis, respectively.

Section (A) represents increasing CK levels and increasing "fatigue" in POMS and section (C) represents decreasing CK levels and decreasing "fatigue" in POMS at the second examination. Section (B) represents increasing CK levels and decreasing "fatigue" in POMS and section (D) represents decreasing CK levels and increasing "fatigue" in POMS at the second examination. Sectionized as on the each axis, the vertical or the horizontal axis, represents no change in CK levels or "fatigue" in POMS over time, respectively.

6. Statistical methods

Statistical analysis was performed on a Macintosh IIsi using Stat View software. Differences between the first and the second examinations were compared using Student's paired t-test. Three groups were compared using the one factor ANOVA test. The regression analysis of subjective fatigue was examined using Spearman's rank correlation. The χ-square analysis revealed differences between groups. The level of significance was $P < 0.05$.

RESULTS

1. Characteristics of athletes and performance levels

Results of the physical examinations are summarized in Table 1. Body weight, the body fat weight (Body Fat), percentage of fat (% Fat), lean body mass (LBM), and total body water (Body Water) of the athletes did not significantly change

Table 1 Physical characteristics of participants during this study.

		Group A								Group B			
		A-Senior (n=17)				A-Freshman (n=8)				(n=14)			
		First Exam.		Second Exam.		First Exam.		Second Exam.		First Exam.		Second Exam.	
Items		Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Age	(year)	25.9	4.6	-	-	19.1	1.2!	-	-	24.4	5.2	-	-
Height	(cm)	167.8	4.8	-	-	173.8	8.4#	-	-	174.0	5.9#	-	-
Body Weight	(Kg)	54.7	3.2	54.7	3.6	58.9	6.6*	58.8	6.0*	58.7	3.1#	58.7	3.1#
Body Fat	(Kg)	3.4	0.9	3.5	1.0	3.8	1.1	3.8	1.0	3.9	0.8	4.0	0.8
% Fat	(%)	6.3	1.3	6.2	1.4	6.3	1.4	6.3	1.3	6.7	1.1	6.7	1.1
LBM	(Kg)	51.3	2.4	51.3	2.7	55.1	5.6*	55.0	5.1*	54.8	2.4#	54.8	2.2#
Body Water	(Kg)	37.5	1.8	37.5	1.8	40.3	4.1*	40.3	4.1*	40.1	1.8#	40.1	1.8#

Examinations were conducted on two consecutive mornings after the day when 3 times of 5,000 meter time-trial, which were most strenuous physical exercise performed in the training camp, were carried out. First Exam. and Second Exam. in this Table indicate the examinations conducted in the first and second morning after the day time-trials were performed, respectively. For further details see Materials and Methods in the text.
LBM, lean body mass.
Statistically significant differences between A-Senior and the other groups are represented as: *, $P < 0.05$; #, $P < 0.01$; !, $P < 0.001$.

Table 2. Performance levels of participants.

	Best records of 10,000 meter running		
Groups	Mean	SD	P value
A-Senior	28' 50" 86	0' 30" 51	
A-Freshman	30' 14" 59	1' 11" 97	$P < 0.001$
B	31' 36" 62	0' 45" 59	$P < 0.001$

Statistical significance of differences between A-Senior and the other groups is shown.

throughout the study. As shown in Table 1, the A-Freshman group was significantly younger than the other two groups and the A-Senior group was smaller, weighed less, had lower LBM, and less Body Water than the other two groups. However, Body Fat and % Fat did not significantly differ among the three groups.

Performance levels of each group is shown in Table 2 indicated by personal best results of running 10,000 m. The A-Senior group had the best performance levels. The mean value of the personal best of the A-Freshman group was significantly delayed by about 1 min 24 sec from that of the A-Senior group. The mean value of the Group B differed from that of the A-Freshman group by a similar range.

2. Serum enzyme activity levels

Serum enzyme activity levels in each group are summarized in Table 3. At the first examination, the AST level of the A-Senior group was higher than the reference level (38 IU/L).[24,25] The A-Senior and B groups significantly differed at both test times ($p < 0.001$ and $p < 0.01$ at the first and second examinations, respectively). Serum AST levels significantly decreased in all groups at the second examination and that of the A-senior group became lower than the reference level. The serum LD levels of the A-Freshman and B groups at the first examination were higher than the reference level (460 IU/L).[24,25] In contrast to AST levels, there was no significant difference among the LD levels of the three groups at both examinations. Serum CK levels of all three groups at the first examination were higher than the reference level (195 IU/L) [24,25] and CK levels significantly decreased at the second examination ($p < 0.001$ for all groups) but remained considerably above the reference level. Serum CK levels of the A-Freshman and B groups were significantly higher than the A-Senior group at both examinations.

We also examined the change of serum enzyme activity levels over time. Levels at the first and second examinations were measured and the difference were divided by the corresponding value at the first examination. Results are summarized in Table 3. There was no significant difference among the three groups. CK levels considerably decreased and LD levels only slightly varied.

3. Correlations among serum enzyme activity levels.

Serum enzyme activity levels significantly correlated in the Group B at the first examination ($r=0.913$, $p < 0.001$; $r=0.838$, $p < 0.001$ and $r=0.930$, $p < 0.001$ between AST and LD, CK and LD, and AST and CK, respectively) and AST and CK levels also significantly correlated in the A-Senior and A-Freshman groups ($r=0.815$, $p < 0.001$ and $r=0.870$, $p < 0.01$, respectively). AST and LD were significantly correlated in the A-Freshman group ($r=0.732$, $p < 0.05$).

At the second examination, AST and CK levels significantly correlated in the A-Senior, A-Freshman, and B groups ($r=0.697$, $p < 0.01$; $r=0.939$, $p < 0.001$ and $r=0.942$, $p < 0.001$, respectively). AST and LD levels significantly correlated in the A-Freshman and B groups ($r=0.774$, $p < 0.05$ and $r=0.907$, $p < 0.001$, respectively) and CK and LD levels significantly correlated in the A-Senior and B groups ($r=0.547$, $p < 0.05$ and $r=0.888$, $p < 0.001$, respectively).

4. Subjective fatigue.

Results of the subjective symptoms of fatigue are summarized in Table 4. At the first examination, the score for element I of the subjective symptom of fatigue (SSF-I), "drowsiness and dullness", in the A-Senior group was lower than that of the other groups and the A-Senior and B groups significantly differed ($p < 0.05$). At the second examination, the SSF-I level of the Group B significantly decreased ($p < 0.05$) with no significant differences from the A-Senior group. As shown in Table 4, SSF-II and SSF-III did not significantly differ among all three groups and did not significantly decrease at the second examination.

Mean levels of the six elements in POMS are also summarized in Table 4. The A-Senior group displayed the classical "iceberg" profile with "vigor" (Vig) as the apex in both examinations. In contrast to the A-Senior group, the A-Freshman group displayed the "iceberg" profile with "fatigue" (Fat) as the apex. In the Group B, "vigor" and "fatigue" in

Table 3 Serum enzyme activity levels in each group during this study.

	Group A						Group B		
	A-Senior (n=17)			A-Freshman (n=8)			(n=14)		
	First Exam.	Second Exam.	Change(%)	First Exam.	Second Exam.	Change(%)	First Exam.	Second Exam.	Change(%)
Items	Mean (SD)	Mean (SD)	Mean (SD)	Mean (SD)	Mean (SD)	Mean (SD)	Mean (SD)	Mean (SD)	Mean (SD)
AST (IU/L)	41.4 (11.7)	34.6 (9.5)[a]	-15.1 (13.7)	62.1 (17.6)	51.2 (18.7)[a]	-19.3 (9.3)	73.6 (36.4)[!]	60.9 (30.3)[#,b]	-17.7 (12.7)
LD (IU/L)	457.7 (122.5)	429.6 (97.6)	- 1.0 (24.5)	515.9 (94.2)	490.7 (76.5)	- 4.3 (10.8)	518.7 (142.0)	484.0 (130.2)[b]	- 6.7 (5.5)
CK (IU/L)	734.9 (328.5)	538.6 (255.8)[a]	-26.5 (9.9)	1315.4 (510.3)[#]	970.9 (460.7)[*,a]	-28.3 (10.3)	1349.0 (549.4)[!]	1010.4 (513.9)[#,a]	-28.5 (11.0)

First Exam. and Second Exam. in this Table indicate the examinations conducted in the first and second morning after the day time-trials were performed, respectively. For further details see Table 1 and Materials and Methods in the text.
Statistically significant differences between A-Senior and the other groups are evaluated by the one factor ANOVA test and represented as:
*, P < 0.05; #, P < 0.01; !, P < 0.001.
Statistically significant differences between First and Second Exams are evaluated by the paired t-test and represented as: a, P < 0.001; b, P < 0.01.
Changes (%) calculated as follows:
Changes (%) = ([values of Second Exam - values of First Exam] / values of First Exam) x 100
No statistically significant differences between A-Senior and the other groups in Changes (%).

Table 4 Subjective fatigue levels in each group during this study.

	Group A						Group B		
	A-Senior (n=17)			A-Freshman (n=8)			(n=14)		
	First Exam.	Second Exam.	Change (%)	First Exam.	Second Exam.	Change (%)	First Exam.	Second Exam.	Change (%)
Items	Mean (SD)	Mean (SD)	Mean (SD)	Mean (SD)	Mean (SD)	Mean (SD)	Mean (SD)	Mean (SD)	Mean (SD)
Subjective symptoms of fatigue (score) :									
SSF-I	1.89 (1.88)	1.88 (2.42)	-	3.38 (3.02)	2.88 (2.85)	-	3.93 (2.56)*	2.21 (2.61)[c]	-
SSF-II	0.06 (0.24)	0.53 (1.18)	-	0.75 (1.49)	0.38 (1.06)	-	1.14 (1.88)	0.57 (0.85)	-
SSF-III	0.78 (1.06)	0.59 (1.00)	-	0.75 (1.17)	1.13 (1.25)	-	1.64 (1.34)	1.21 (1.37)	-
POMS (T-score) :									
Tension	37.8 (4.1)	37.9 (5.0)	0.6(11.7)	37.4 (4.2)	36.7 (2.4)	- 1.0 (10.8)	42.6 (6.9)	39.5 (5.4)[c]	- 6.3(10.8)
Depression	35.9 (0.3)	35.9 (0.4)	0.1 (1.1)	36.0 (0.4)	35.8 (0.3)	- 0.3 (0.5)	36.2 (0.5)*	35.9 (0.3)	- 0.6 (1.2)
Anger	39.4 (4.6)	40.4 (6.2)	2.3(11.1)	38.5 (2.4)	38.5 (2.4)	0.1 (5.1)	43.2 (7.0)*	39.1 (3.2)[c]	- 8.1(10.8)[#]
Vigor	50.4(12.9)	50.4(10.1)	3.0(19.5)	41.8 (6.7)	42.8(11.0)	1.2(12.1)	50.3 (7.1)	48.1 (8.2)	- 4.4 (9.1)
Fatigue	43.7 (5.9)	42.9 (5.7)	- 1.9 (9.1)	48.3 (7.1)	47.2 (8.4)	- 1.9(13.2)	50.8 (6.7)[#]	47.8 (6.8)	- 5.6(10.1)
Confusion	38.3 (4.4)	39.1 (5.2)	2.0 (9.4)	40.0 (5.0)	39.5 (3.1)	- 0.6 (9.7)	42.8 (4.9)	40.2 (4.5)[c]	- 5.0 (6.6)*
TMS	111.6 (22.8)	113.2 (23.5)	0.6 (7.9)	121.8 (20.2)	117.6 (13.9)	- 1.2 (5.0)	132.8 (24.5)*	119.9 (16.3)	- 3.7 (5.8)

First Exam. and Second Exam. in this Table indicate the examinations conducted in the first and second morning after the day time-trials were performed, respectively. For further details see Table 1 and Materials and Methods in the text.
Statistically significant differences between A-Senior and the other groups are evaluated by the one factor ANOVA test and represented as: *, P < 0.05; #, P < 0.01.
Statistically significant differences between First and Second Exams are evaluated by the paired t-test and represented as: c, P < 0.05.
Changes (%) calculated as follows:
Changes (%) = ([values of Second Exam - values of First Exam] / values of First Exam) x 100

POMS were the apices and mean levels of "depression," "anger," and "fatigue" were significantly higher than those of the A-Senior group at the first examination (Table 4). The profiles of the groups did not change at the second examination and there was no significant difference among the three groups.

Table 4 shows a significant difference between the total mood state (TMS) of the A-Senior group and that of the Group B at the first examination ($p < 0.05$). However, the TMS levels of the A-Senior and A-Freshman groups remained constant at the second examination, whereas that of the Group B decreased. The groups did not significantly differ at the second examination.

To clarify the POMS elements over time, changes were calculated as described and are summarized in Table 4. The elements of "anger" and "confusion" among the Group B decreased to a great extent and there were significant differences from the A-Senior group.

5. Correlations among the elements of subjective fatigue.

Correlations among the elements of subjective fatigue were examined by regression analysis using Spearman's rank correlation. Except for "vigor" in POMS, SSF-I significantly correlated with the elements in POMS in the A-Senior and B groups at the first examination. There were only two significant correlations in the A-freshman group between SSF and POMS (r_s=0.945, $p < 0.001$ and r_s=0.783, $p < 0.05$ between SSF-I and "fatigue" in POMS and SSF-I and TMD, respectively). "Vigor" and the other POMS elements did not significantly correlate among all groups.

At the second examination, SSF-I significantly correlated with the elements in POMS, except for "vigor" and "confusion", in the A-Senior group, whereas SSF-I in the Group B significantly correlated only with "confusion" in POMS (r_s=0.572, $p < 0.05$). SSF-I and "fatigue" significantly correlated in the A-freshman group (r_s=0.805, $p < 0.05$).

6. Correlations between serum enzyme activity levels and subjective fatigue.

We applied regression analysis using Spearman's rank correlation to examine the correlation between serum enzyme activity and subjective fatigue. The results are summarized in Table 5. Serum enzyme activity levels and the elements of subjective symptoms of fatigue (SSF) at both examinations were not significantly correlated. Serum enzyme activity levels of the A-freshman group positively correlated with "vigor" in POMS but negatively with TMS (AST and CK), "tension," (LD) and "depression" (CK) at the first examination (Table 5). Serum CK

Table 5 Correlation between serum enzyme activity levels and subjective fatigue.

			Subjective symptoms of fatigue			POMS						
Items	Groups (n)	Examination	SSF-1	SSF-2	SSF-3	Tension	Depression	Anger	Vigor	Fatigue	Confusion	TMS
AST	A-Senior (n=17)	First	0.022	-0.305	0.055	-0.126	-0.122	0.098	0.413	0.011	-0.031	-0.137
		Second	0.356	0.292	0.109	0.082	-0.091	0.090	0.009	0.217	-0.087	0.133
	A-Freshman (n=8)	First	-0.458	-0.514	-0.273	-0.446	-0.687	-0.424	0.988'	-0.419	-0.627	-0.810*
		Second	0.244	-0.415	-0.006	-0.552	-0.426	-0.295	0.909'	0.271	-0.364	-0.551
	B (n=14)	First	0.026	-0.066	0.281	-0.155	-0.373	-0.227	-0.156	0.206	0.104	0.048
		Second	0.093	-0.130	0.281	0.360	0.197	-0.183	-0.300	0.378	0.072	0.516
LD	A-Senior (n=17)	First	-0.099	-0.397	0.062	0.011	-0.020	-0.085	0.229	-0.182	-0.024	-0.205
		Second	0.479	0.107	0.330	0.360	0.298	0.498*	0.386	0.328	-0.092	0.036
	A-Freshman (n=8)	First	-0.217	-0.187	0.218	-0.759*	-0.374	-0.509	0.868'	-0.252	-0.615	-0.667
		Second	0.388	-0.412	0.063	-0.422	-0.135	0.220	0.554	0.287	-0.110	-0.238
	B (n=14)	First	0.118	-0.024	0.314	-0.193	-0.442	-0.212	-0.248	0.248	0.040	0.108
		Second	-0.034	-0.228	0.214	0.066	0.048	-0.298	-0.481	0.337	-0.104	0.398
CK	A-Senior (n=17)	First	0.118	-0.257	0.048	0.139	0.005	0.275	0.565*	0.168	-0.055	-0.092
		Second	0.421	0.279	0.067	0.148	0.385	0.446	0.215	0.522*	0.120	0.136
	A-Freshman (n=8)	First	-0.590	-0.514	-0.355	-0.458	-0.819*	-0.533	0.892'	-0.599	-0.518	-0.857'
		Second	-0.061	-0.412	-0.013	-0.554	-0.577	-0.415	0.928'	0.156	-0.405	-0.595
	B (n=14)	First	-0.067	0.012	0.231	-0.180	-0.397	-0.228	-0.221	0.073	0.013	0.020
		Second	0.156	-0.098	0.262	0.301	0.133	-0.170	-0.393	0.374	0.032	0.569*

Statistically significant correlations between serum enzyme activity levels and subjective fatigue are evaluated by Spearman's rank correlation. Numbers shown are rank correlation coefficients and statistical significance is represented as: ns, not significant; *, $P < 0.05$; #, $P < 0.01$; !, $P < 0.001$.

Table 6 Distribution of changes in Fatigue in POMS and serum CK levels.

Groups	Section (C)	On the line	Section (D)
A-Senior (n)	9	5	3
Fatigue	45.0 ± 3.9	37.0 ± 0 #	46.3 ± 8.7
CK	563 ± 257	383 ± 164	725 ± 300
A-Freshman (n)	3	0	5
Fatigue	41.0 ± 3.5		50.9 ± 8.5
CK	686 ± 296		1142 ± 480
B (n)	8	2	4
Fatigue	46.0 ± 6.7	49.7 ± 14.1	50.3 ± 3.4
CK	903 ± 482	486 ± 203	1487 ± 269 *

Section (C) represents decreasing CK levels and decreasing "fatigue" in POMS at the second examination. Section (D) represents decreasing CK levels and increasing "fatigue" in POMS at the second examination. "On the line" represents no change in T-score of "fatigue" in POMS over time plotted on the horizontal axis.

For further details of Section (C), (D), and "On the line" see Materials and Methods in the text.

Fatigue (POMS) and serum CK levels of each category in the 3 groups are means and standard deviations at the second examination. The second examination indicate the examination conducted in the second morning after the day time-trials were performed. For further details see Table 1 and Materials and Methods in the text.

Statistically significant differences between Section (C) and the other groups are represented as: *, $P < 0.05$; #, $P < 0.01$.

levels of the A-Senior group correlated with "vigor" in POMS at the first examination. At the second examination, serum AST and CK levels of the A-freshman group positively correlated with "vigor" in POMS. LD levels and "anger" were significantly correlated, as were CK levels and "fatigue" in the A-Senior group. CK levels and TMS were significantly correlated at the second examination in the Group B.

7. Relationship between serum CK levels and "fatigue" in POMS.

To clarify the relationship between serum CK levels and "fatigue" in POMS, T-scores of "fatigue" over time were calculated for each athlete and plotted in a scatter plot representing changes in serum CK level as the horizontal axis and "fatigue" in POMS as the vertical axis. Table 6 shows that athletes are scattered in sections (C), (D), and on the horizontal axis. To reveal different tendencies, the athletes in each group were divided into the following categories: "Section (C)," "On the line" and "Section (D)." Results are summarized in Table 6 with "fatigue" in POMS and CK levels at the second examination. Tendencies did not significantly differ among the three groups. However, the χ-square analysis revealed a statistical difference between the A-Senior and A-Freshman groups ($p < 0.05$).

In Table 6, the five athletes in the A-Senior group categorized as "On the line" showed significant lower levels of "fatigue" in POMS ($p < 0.01$) than those in the other two categories. The CK level in "Section (D)" of the Group B was significantly higher ($p < 0.05$) than that of the other categories.

DISCUSSION

We did not study serum enzyme activity levels and subjective fatigue before or immediately after starting the training camp because these values did not represent resting conditions. Endurance athletes, especially well-trained long-distance runners run almost every day. Furthermore, the resting or initial values of athletes/patients complaining of fatigue are essentially impossible to measure. Training fatigue is the normal result of several days of heavy training associated with an overload training stimulus [10]. However, one critical problem is that diagnostic or warning tests for the overtraining syndrome have not yet been established[4,5]. Therefore, it is important for the prevention of overtraining, which is one type of chronic fatigue, to categorize athletes feeling fatigue into high-risk and adaptable groups by objective means.

Serum enzyme activity levels are affected and increased by physical stress[26,27]. Sympathetic overtraining may affect mainly speed and power athletes while parasympathetic overtraining seems to affect mostly endurance athletes[5]. Lehmann et al.[5]

reported in agreement with other investigators[8], that changes in blood-chemistry such as muscle enzymes, total protein, and immunoglobulins are probably not obligatory in parasympathetic overtraining syndromes and are therefore presumably of less diagnostic importance. However, Lehmann et al.[5] found that CK levels are elevated later in the experiment (day 24), when athletes are expected to exhibit the overtrained condition. We reported that norepinephrine, testosterone and the ratio of testosterone to cortisol may be the most useful indices of blood properties to determine the early stage of overtraining[28]. However, these measurements are too expensive for daily monitoring. Flynn et al[29]. reported that changes in total testosterone, free testosterone, and CK were concomitant with decreased performance and increased TMS. Therefore, they concluded that CK may be an effective marker for monitoring overtraining in endurance athletes.

Table 3 shows that the serum AST and CK levels were initially higher than the reference values and that they significantly decreased at the second examination. These findings correlated well with the report of Cannon et al.[30], which shows that the plasma CK concentration was significantly increased in the morning following exercise. CK levels between the A-Senior and A-Freshman groups and between the A-Senior and B groups at both examinations significantly differed. These findings may agree with the report of Koutedakis et al.[31], which shows that Olympic rowers demonstrated higher resting AST ($p < 0.05$) and CK ($p < 0.001$) levels than untrained persons. One explanation for our results may be deferences in the adaptability. In other words, stress is reduced to some extent in the high fitness group such as the A-Senior group probably because of more frequent exposure to exercise[32]. Another explanation for the lower levels of serum enzyme activity found in the A-Senior group may be the repeated bout effect[33], namely, that changes caused by exercise are diminished by repetition of the same exercise.

Serum AST and CK levels were significantly decreased at the second examination. However, as summarized in Table 3, serum enzyme activity levels (Change (%)) did not statistically differ among the groups. These results contradicted a report[31] indicating that Olympic rowers showed higher CK clearance rates (55%) from the blood compared with untrained counterparts (44%), suggesting an adaptive response to the specialized training undertaken by elite rowers. As shown in Table 2, the A-Senior group including one Olympic marathon runner showed highest performance level but even the lesser groups had the same performance level as the highly trained distance runners (personal best 10-km time, 31 min 4 sec) described by Verde et al.[34]. These results may reflect the adaptive response of athletes to specialized training. Another cause of this discrepancy may be the blood sampling time. Koutedakis et al.[31] examined athletes one hour after ergometer exercise, whereas we examined our athletes one and two days after strenuous physical exercise.

Measuring the effects of exercise on blood properties is useful for obtaining objective indices of physical fatigue but it is also expensive and time-consuming. Therefore, monitoring subjective fatigue levels using a questionnaire is applied in field surveys as a more convenient and inexpensive means of estimating fatigue[15-18]. POMS is to date, the best single marker of disturbed function, indicating increased fatigue and decreased vigor[34]. It is also rapidly completed and widely accepted in sports psychology[35]. Exercise and sport science can considerably benefit from additional research in psychophysiology[36]. The A-Senior group displayed the classical "iceberg" profile in both examinations and as Morgan[37] noted, the athletes had significantly above average vigor scores as measured by POMS. The elements in the questionnaire for subjective fatigue significantly correlated, in agreement with the report by Hooper et al.[38]. Table 4 shows that the subjective fatigue of the Group B compared with that of the A-Senior group was identified by the SSF-I, namely "drowsiness and dullness" and by the mean levels of three elements and TMS in POMS at the first examination. However, the three groups did not significantly differ at the second examination. Thus, identifying differences in fatigue levels among the three groups by psychological monitoring at the second examination seemed to be difficult.

In contrast to psychological monitoring, Table 3 shows statistical significances in CK levels at both examinations. To clarify the relationship between the serum CK levels and "fatigue" in POMS over time, we calculated changes and summarized them in Table 6. The A-Senior and A-Freshman groups seemed to dominantly pool in sections (C) and (D), respectively. Table 6 shows CK levels and "fatigue" in POMS at the second examination in each category of the three groups. Athletes in the Group B categorized into "Section (D)" showed higher CK levels than other two categories. This finding suggests that the athletes categorized into "Section (D)" can not sufficiently recover within one day. Overtraining appears to be caused by too much high intensity training and/or too little regeneration (recovery) time [10]. The athletes in "Section (D)" are more likely to become overtrained.

The χ-square analysis revealed a statistical difference between the A-Senior and A-Freshman groups (Table 6). This result suggests that the athletes in the A-Freshman group do not sufficiently regenerate compared with the A-Senior group. On the other hand, Table 5 shows that the AST and CK levels of the A-Freshman group positively correlated with the one positive mood state, "vigor," in POMS at the second examination. These correlations in the A-freshman group were contrary to those of the other groups. According to this result, the athletes in the A-Freshman group had tended to feel more "vigor" at higher AST and CK levels; that is, insufficiently regenerated. Dissociation of physical and psychological fatigue has been reported[39,40]. These results suggest that physical and psychological fatigue are dissociated in the A-Freshman group.

Exercise is a form of physiological stress, and the degree of stress encountered is dependent on the intensity and duration of the exercise[32]. Furthermore, exercise is not a single homogeneous stressor, rather it is a multi-stressor combination that varies in different situations[41]. Physiological mechanisms may mediate some of the mood changes associated with overtraining, and that psychological monitoring of overtraining can be useful if supported by physiological measurements[20]. Long-term athletic training has followed established training plans learned by experience to prevent overtraining. It is important to regulate training plans according to an objective estimation of physical fatigue, not only to prevent a shift from over-reaching to overtraining, but also to improve performance levels.

Further studies with large number of individuals are needed to determine how age, workload, duration of exercise and physical fitness affect the magnitude of physical fatigue. In conclusion, serum enzyme activity levels are thought to be more important indices, even in well-trained endurance athletes, for reflecting physical fatigue in relatively short periods of training

such as micro-cycles[39]. Monitoring subjective fatigue concurrently with measuring serum enzyme activity levels using the dry-chemistry method, immediately provides clinical value to players and coaches on site. These procedures should be useful for preventing shift from "overreaching" to "overtraining".

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