Europ. J. appl. Physiol. 37, 243–254 (1977)

European Journal of
Applied
Physiology
and Occupational Physiology

Modifications of Serum Glycoproteins the Days Following a Prolonged Physical Exercise and the Influence of Physical Training*

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Summary. Eight male subjects (mean age 24.1 ± 2.6 years) performed at intervals of 2 weeks successively a 3 h and two 2 h runs of different running speed. The days following the running there were moderate elevations of C-reactive protein, haptoglobin, alpha-1-acid glycoprotein, coeruloplasmin, transferrin, alpha-1-antitrypsin and plasminogen. There were small or no changes of albumin, alpha-2-macroglobulin and hemopexin. The elevations of the "acute phase reactants" were examined in three male subjects following a 2 h run before and after an endurance training period of 9 weeks. This demonstrated a decreased acute phase response after training as illustrated by the changes of C-reactive protein, haptoglobin and alpha-1-acid glycoprotein in spite of higher posttraining running speeds. Well-trained athletes have elevated levels of the serum protease inhibitors alpha-1-antitrypsin, alpha-2-macroglobulin and C_1-inhibitor. These antiproteolytic glycoproteins might limit exercise-induced inflammatory reactions.

Key words: Glycoproteins – Acute phase reaction – Inflammation – Physical exercise – Physical training.

Introduction

After a 12 weeks' lasting training period we found in elderly people a significant increase of the rest serum levels of plasminogen, coeruloplasmin, transferrin and alpha-1-antitrypsin. Plasma volume was also increased. There was no change of the alpha-1-acid glycoproteins, haptoglobin and hemopexin levels [43, 44]. Haralambie (1976) found after a 5 weeks' training period in altitude in young well-trained people a significant increase of alpha-1-acid glycoprotein, coeruloplasmin, transferrin, haptoglobin, hemopexin, alpha-2-HS-glycoprotein, alpha-1-antitrypsin, alpha-2-macroglobulin and beta-1A-globulin. Changes in plasma volumes were not reported.

* This research was supported by the "Bundesinstitut für Sportwissenschaften (Köln-Lövenich)"

With the present study we wanted to investigate the modifications of serum glycoproteins during the days following a prolonged physical exercise. The observed changes of C-reactive protein, alpha-1-acid glycoprotein, coeruloplasmin, transferrin, haptoglobin, hemopexin, alpha-1-antitrypsin and alpha-2-macroglobulin although moderate resembled in many aspects to what is seen during a delayed inflammatory response.

Material and Methods

Eight healthy male subjects between 21 and 29 years (mean 24.1) performed at intervals of 2 weeks succesively a 3 h and two 2 h runs. Blood samples were collected from a cubital vein before, immediately after, on the next day and after 2, 4 and 7 days. Three other male subjects ran for 2 h before and after an endurance training period of 9 weeks which consisted of a 10 km run four times a week. Blood samples were also collected before and after the run, on the next day and after 3 and 7 days. Blood samples were also taken at rest from 50 healthy subjects between 18 and 25 years old (mean 22.4 $\pm$ 2.9), from 26 very well trained German national top-swimmers aged between 15 and 23 years (mean 19.2 $\pm$ 2.5) and from 18 international professional bike riders aged between 23 and 34 years (mean 26.8 $\pm$ 3.0). The blood was centrifugated and the serum refrigerated at minus 25° C until analysis. Quantification of the individual serum glycoproteins was performed by standard radial immunodiffusion [46] on plates containing monospecific antiserum obtained from Behring-Werke, Marburg/Lahn. All samples were run against three provided standards for each glycoprotein. The C-reactive protein (CRP) gel plate was treated with tannin. The plate was rinsed for 24 h with phosphate-buffered saline, pH 7.2 (0.85% NaCl, 0.86% $NA_2HPO_4 \cdot 2H_2O$, 0.24% KH_2PO_4). After rinsing the gel surface was covered with a 4% aqueous solution of tannin for 30 min and finally rinsed for 1 h with distillated water. We calculated the mean values ($\bar{x}$), the standard-deviation (s) and the standard error ($s/\sqrt{n}$). The level of significance was proved by the t-test for independent or correlated observations.

Results

The days following a 3 h and two 2 h runnings there was an increase of the acute-phase globulins. Figure 1 represents the elevations of C-reactive protein (CRP) in the eight subjects after the three runnings. The individual running speed for the first 2 h test was about 90% of the threshold speed, determined on a threadmill, at which there is a passage from a predominantly aerobic energy contribution to a predominantly anaerobic contribution. This aerobic-anaerobic threshold has been determined experimentally to be situated around 4 mmol/l serum lactic acid concentration [45]. The individual running speed of the 3 h and the second 2 h test was about 75% of the aerobic-anaerobic threshold speed (Table 1).

Figures 2 and 3 represent the modifications of the different glycoproteins and of albumin after a total of 24 runnings by 8 subjects in the three running-tests performed at 14 days interval. The changes are expressed as percentages of the values found before the tests. The most pronounced elevations were observed for CRP (Fig. 1) and haptoglobin (Fig. 2) with peak values respectively after 1 day and 2–4 days. After a week both proteins almost had disappeared from the blood. Some of the subjects (sport-students) showed low levels of CRP at rest before the runnings. This may have resulted from low intensity sport practice the days preceeding the tests. In the same individual the elevations of CRP showed a very similar configuration after

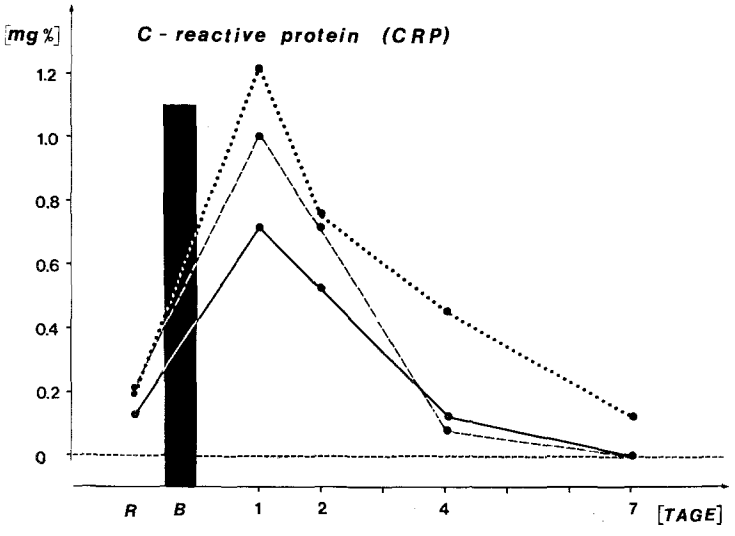

Fig. 1. Changes of serum CRP in 8 males following a fast 2 h run (– – –), a slower 3 h run (· · · ·), and a second 2 h run (——) of nearly the same running speed as the 3 h run. One day (Tag) after the runs there were significant differences in the CRP-concentrations

Table 1. The mean running speed of each participant in the three runs in meter per second. The percentage values are applied to the individual pure aerobic performance capacity (= 100%), measured by the running intensity, who lead to a serum lactiol acid of 4 mmol/l

Participants	Aerobic-anaerobic threshold [4 mmol/l LA]	Fast running		Slower runnings			
		First 2 h		Second 2 h		3 h	
	m/s	m/s	%	m/s	%	m/s	%
M.	3.06	2.52	82	2.08	68	2.05	67
H.	3.54	3.33	94	2.80	79	2.78	78
B.	3.68	3.28	89	2.78	75	2.78	75
P.	3.68	3.22	87	2.79	75	2.78	75
K.	3.86	3.47	90	2.83	73	2.82	73
L.	3.88	3.65	94	3.11	80	3.15	81
B. P.	3.90	3.68	94	2.82	72	2.81	72
S.	4.19	3.98	95	3.52	84	3.79	90

the 3 h and the two 2 h runnings, only the intensity of the response differed significantly. As is shown in Figure 1 the running speed affected the elevation of CRP as shown by the larger increase after the fast 2 h compared with the slower 2 h run. The duration of the runnings influenced very markedly the CRP response as seen by the higher and more prolonged elevations after the 3 h run compared with the 2 h test even performed at higher speed (see first 2 h run, Fig. 1).

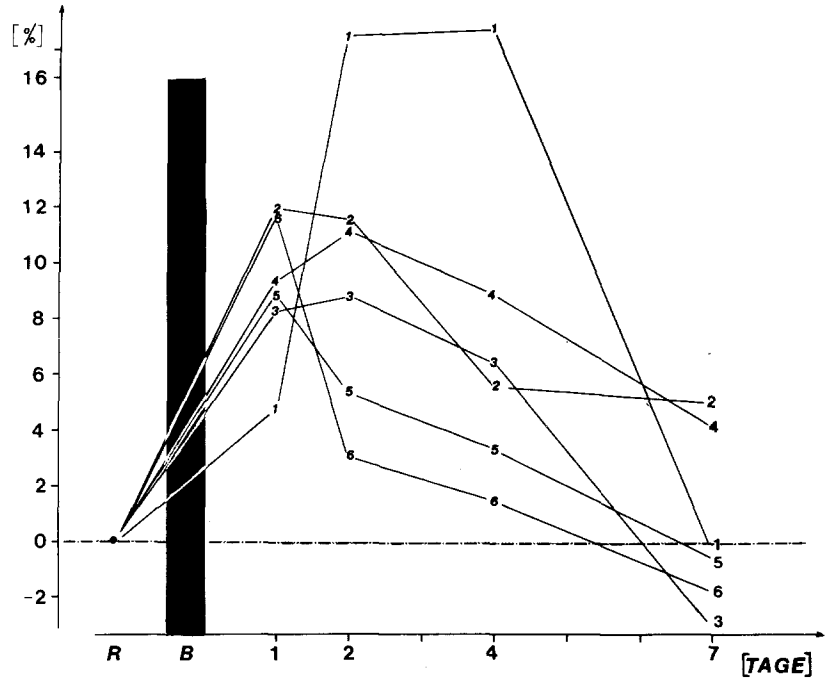

Fig. 2. Changes of serum haptoglobin (1), transferrin (2), coeruloplasmin (3), alpha-1-acid glycoprotein (4), alpha-1-antitrypsin (5), and plasminogen (6) in the days (Tage) after a long distance running (B) of 2 or 3 h in per cent of the resting values (R) before the run ($n = 24$). In the first days after running the increase of these glycoproteins were significant

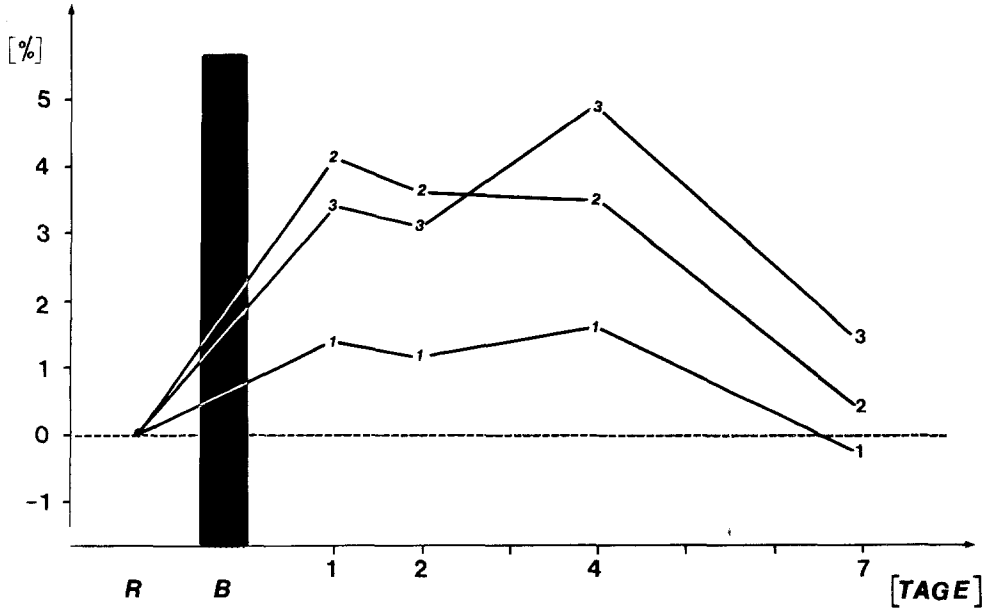

Fig. 3. Changes in serum albumin (1) ($p < 0.05$), alpha-2-macroglobulin (2) ($p < 0.05$) and hemopexin (3) in the days (Tage) following 2 or 3 h runnings (B) in per cent of the serum concentration before starting (R)

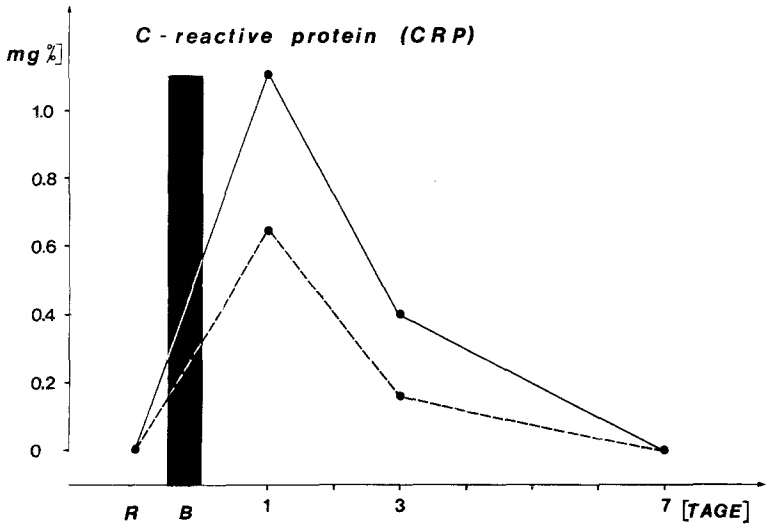

Fig. 4. Changes of serum CRP in three subjects following a 2 h run (B) before (—) and after (– – –) a 9 weeks' training period. During the post-training runs the distances covered were respectively 10, 12 and 20% larger as before training

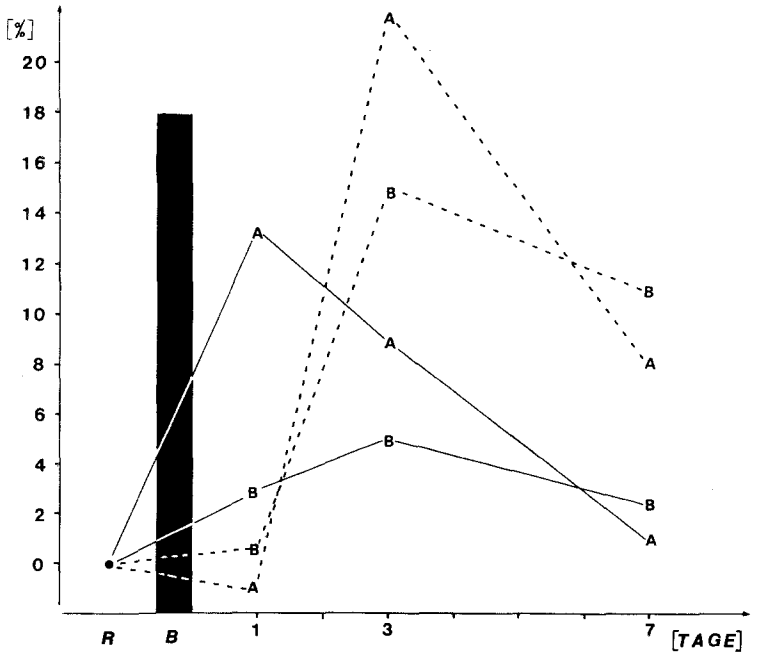

Fig. 5. Changes of alpha-1-acid glycoprotein (—) and haptoglobin (– – –) in three males in the days (Tage) following 2 h exhaustive runs (B) before (A) and after (B) a 9 weeks' endurance training period

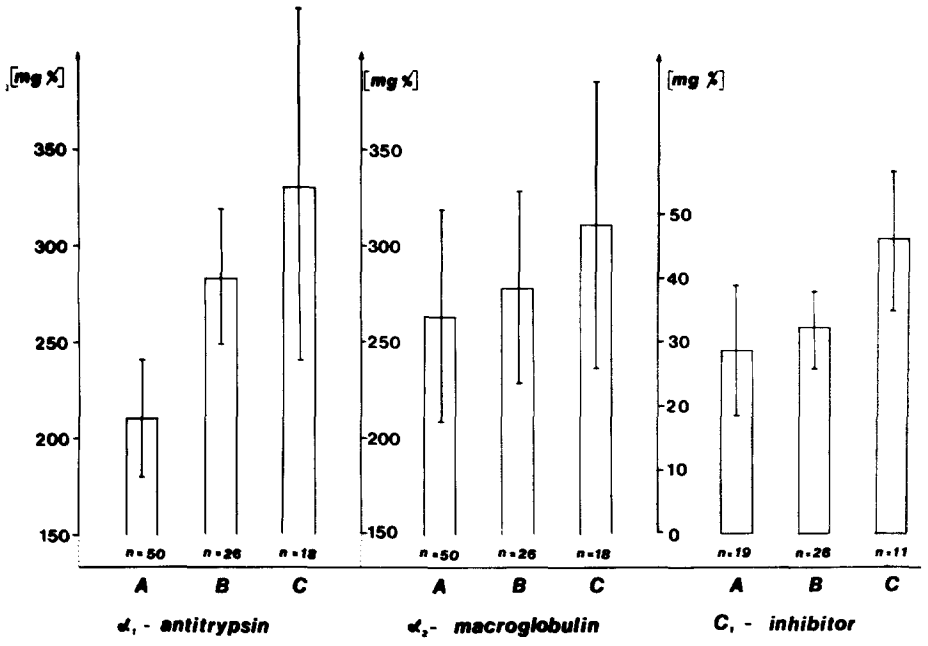

Fig. 6. The serum levels (at rest) of alpha-1-antitrypsin, alpha-2-macroglobulin, and C_1-Inhibitor in untrained healthy males (A), German national top swimmers (B), and international professional bike riders (C). The top athletes have significant higher values for these proteinase-inhibitors: alpha-1-antitrypsin: A–B < 0.01, A–C < 0.001; alpha-2-macroglobulin: A–B N.S., A–C < 0.05; C_1-inhibitor: A–B N.S., A–C < 0.01

Moderate increase were observed for haptoglobin, α_1-acid glycoprotein, coeruloplasmin, transferrin, α_1-antitrypsin, and plasminogen with peak values after 1 or 2 days (Fig. 2). Small to absence of increases were observed for albumin, α_2-macroglobulin and hemopexin (Fig. 3). None of the studied proteins demonstrated a decrease the days following the runnings.

After an endurance training period of 9 weeks, the elevations of CRP (Fig. 4) haptoglobin and α_1-acid glycoprotein (Fig. 5) following a 2 h run were much lower, although the distances covered during the posttraining running test by the 3 subjects were respectively 10, 12 and 20% larger as before training. In top-swimmers and before all in professional bike riders, which train extremely hard, the serum levels of the protease inhibitors alpha-1-antitrypsin, alpha-2-macroglobulin and C_1-inhibitor were higher than in 50 healthy subjects of the same age group taken as control (Fig. 6).

Discussion

In the presence of an aseptic inflammation as well as in infection there are changes in certain serum glycoproteins known collectively as "acute phase reactants". Among these are C-reactive protein (CRP), α-1-acid glycoprotein, coeruloplasmin, haptoglobin, transferrin, α-1-antitrypsin, plasminogen [71, 72]. The modification of the serum glycoproteins have been studied after surgical trauma [11, 14, 67, 71, 72],

bone fracture [32], myocardial infarction [34, 62], acute infections [26]. The C-reactive protein [3], α-1-acid glycoprotein [57], haptoglobin [47], α-1-antitrypsin [2], transferrin [59] are of hepatic origin. Studies from Cockerell et al. (1973), Kampschmidt et al. (1973), and Pekarek et al. (1974) suggested that changes in the α-1- and α-2-globulins during inflammation and infection may be mediated by a leucocyte endogenous mediator (L.E.M.) released from phagocytizing polymorphonuclear leucocytes which stimulates the protein synthesis by the liver.

Adequate amounts of "acute phase reactants" during and after severe exercise are available intravascularly by two successive mechanisms. An initial consumption of plasminogen, haptoglobin, α-1-antitrypsin, hemopexin, and α-2-macroglobulin during and immediately following a severe exercise could be partly or completely compensated by an afflux of these proteins from the intersitium as a result of an enhanced lymphatic return [74]. Then in the following hours and days the new synthesis of "acute phase reactants" assures that extra amounts can be disposed of if physical exercise is repeated. In this way the organism might adapt to a higher level of exercise-stress exposure.

Recent findings concerning the functions of the examined glycoproteins may help to clarify the significance of their modifications after exercise. CRP is a trace constituent of normal blood which increases very rapidly and as much as 1000 fold during the acute phase of an inflammatory reaction [14]. Recent studies show that CRP is remarkably similar functionally to the immunoglobulins and that it may play a role in nonspecific resistance to infection as well as in modulation of tissue injury and repair. CRP possesses the ability to initiate reactions of precipitation [21], agglutination [25] promotion of phagocytosis [38] and complement consumption [36, 52, 60]. It is further suggested that CRP plays a role in modulating T-cell function during inflammation [48].

Besides its biochemical interest, CRP may be of value in the appreciation of the amount of physical work performed by an individual or by a small group of subjects. As is shown in Figure 1 the increase of CRP in the same 8 subjects after a 3 h running was greater and of longer duration than after the 2 h running even performed at higher speed.

Recently several new biochemical functions have been attributed to α-1-acid glycoprotein. Van Oss et al. (1974) ascribed to α-1-acid glycoprotein phagocytosis inhibiting properties. Onda et al. (1975) have proposed that every cell excretes mitosis inhibiting proteins. The intracellular concentrations of this proteins would control the process of cell division. Following this mechanism α-1-acid glycoprotein would play a primary role in the regulation of the liver cell proliferation. Franzblau et al. (1976) have shown that in vitro α-1-acid glycoprotein is capable of influencing the formation of collagen fibers. Although α-acid glycoprotein demonstrates an acute phase reaction after prolonged exercise, there was no change of the rest values after a 12 week lasting training period [42]. Well trained athletes have similar values as non-athletes (unpublished results).

The primary function of coeruloplasmin may be to promote the oxidation of a variety of plasma components among which are ascorbic acid, catecholamines and ferrous iron [50, 51]. However only the oxidation of ferrous iron is likely to be important in vivo. To leave the iron storage compound ferritin from the liver, iron has to be reduced to the ferrous state. Coeruloplasmin is required for the reoxida-

tion to ferric iron to permit its complexation with transferrin [24, 51, 56]. Several observations such as the findings of Evans et al. (1973) which show that hemoglobin and coeruloplasmin levels parallel one another during copper deprivation and repletion in the growing rat, speak for a relationship of copper metabolism and the iron availability. Coeruloplasmin plays further a role in the transport and metabolism of copper [19], but it is not considered to be a copper transport protein in the manner of transferrin [58].

In contrast to what is observed in many other inflammatory conditions [26, 62, 67, 72] transferrin increase the day following prolonged running. This increase may affect iron metabolism and transport and contribute to prevent the excretion in urin of the iron liberated during exercise. Of interest may be the role of transferrin in the nonspecific immunological defenses. A large amount of studies now implicate iron and transferrin in the mechanism of resistance of the mammalian organism to a variety of bacterial infections [7, 37, 68, 69, 70]. Following severe exercise serum iron increases [1]. This may be particularly pronounced in running when some hemolysis occurs [33]. A greater susceptibility to bacterial and fungal pathogens may be the consequence of this hyperferremia [8]. Rised transferrin levels the days following a severe exercise and after training [42] as well as low serum iron levels in well-trained athletes [16] may be useful adaptations of the antimicrobial defense mechanisms to the exercise-induced serum-iron elevations. A role of transferrin in the metabolism of the lymphocytes has been proposed [10, 54, 55].

Prolonged running induces a certain degree of hemolysis [15, 33] and is followed by a fall of serum haptoglobins (Fig. 5) and hemopexin [43], most probably as a consequence of the rapid disappearance from the blood of haptoglobin complexed with hemoglobin and hemopexin complexed with heme molecules. Bullen et al. (1968) have shown that the presence of hemoglobin can stimulate the growth of E. coli in vivo. The pronounced rise of haptoglobin and hemopexin the days following a severe exercise may be of value for a rapid and complete elimination of hemoglobin if the organism has to endure a repeated running-hemolysis.

Plasminogen is activated by a variety of blood and tissue activators to become plasmin which has a broad proteolytic activity intervening in fibrinolysis, coagulation, kinin liberation, complement activation and platelet aggregation and release [63]. Rised levels of the plasmin inhibitors α-1-antitrypsin and α-2-macroglobulin [13] during the post-exercise days and after training [43] may bring about a reduction of the exercise induced fibrinolysis [20]. The effects of physical exercise on the deposition and removal of fibrin in the arterial wall are unknown. Some authors [4, 61] consider that the balance between the breakdown of fibrinogen and its conversion to fibrin within the arterial intima are key factors in atherogenesis.

The widespread tissue distribution of plasminogen activators support the concept that fibrinolysis has an extravascular as well as an intravascular role. New data suggest that plasminogen activation may be involved in the normal process of tissue growth [27, 65].

The physiological importance of the protease inhibitors α-1-antitrypsin [31], C_1 inhibitor, and α-2-macroglobulin is that they intervene in the control of proteolytic-regulated processes [30] which may be activated during exercise. Among these are fibrinolysis and blood clotting [20, 39], kinin liberation [43], complement activation [17], collagen and elastin digestion by polymorphonuclear neutrophil proteinases.

Proteolytic processes are also implicated in division of normal [27] and transformed cells [5].

α-1-antitrypsin is considered as an acute phase globulin whereas α-2-macroglobulin is not [72]. The days following prolonged exercise however there was a small elevation (Fig. 2) and α-2-macroglobulin is markedly elevated in athletes as compared to non athletes (Fig. 6). These elevations may intervene in a posttraining reajustment of degradation and biosynthesis of collagen, elastin and proteoglycans, which are of major importance in the control of growth, morphogenesis and repair of the connective tissue structures. Hauss et al. (1968) reported that in rats after a training period there is a reduction of the post-exercise enhanced turn over of heart glycosaminglycans.

Several authors have shown that α-2-macroglobulin is involved in the development and function of lymphocytes [9, 22, 64]. α-2-macroglobulin could promote lymphocyte reactivity and/or influence the antigen recognition by immunocompetent cells. An elevation of this protein in the post-exercise days and after training might be a factor of rised unspecific immunological defense.

It is thought that the acute phase response of serum proteins in inflammation, which follows various types of aggressions, is protective in nature. In this regard the control role of the proteolytic enzymes and their inhibitors should be considered. There is a rapidly growing mass of evidence which indicates that lysosomal enzymes released from polymorphonuclear leucocytes under influence of specific and nonspecific stimuli cause the acute inflammatory reactions with irreversible tissue damage. Immediately after and in the days following a prolonged exercise there is a liberation into the blood of cellular enzymes [41] suggesting the occurrence of tissue destruction. The complement system is activated [17] possibly by proteolytic enzymes. Physical training may realize a new balance between the inflammatory mediators and their inhibitors. Well-trained athletes show significantly elevated levels of three major protease inhibitors: α-1-antitrypsin, α-2-macroglobulin, and C_1-inhibitor (Fig. 6). For three subjects the acute phase reaction as illustrated by the elevations of CRP (Fig. 4), haptoglobin and α-1-acid glycoprotein following a 2 h run was much less pronounced after a 9 weeks endurance training. This lower response appeared although the posttraining distances covered by the three subjects were respectively 10, 12 and 20% larger as before training. One can speculate that higher levels of the serum protease inhibitors assign a limitation to the proteolytic activation during exercise and restrain by unknown mechanism the induction of the synthesis by the liver of the acute phase reactants.

It has been suggested by some authors that chronic inflammation has an immunological basis and could be the result of the inability of the body to get rid of the products of acute inflammation [73]. A weakened inflammatory response could prevent the release of autoantigens from the injured tissues.

The progressive increase of the examined glycoproteins and more particularly of the protease inhibitors after prolonged exercise and training may provide a biochemical basis for the necessity to adapt the intensity of the physical stress exposure to the degree of physical training. In this way detrimental effects on inflammatory defense mechanisms could be avoided.

Further studies might show that the acute phase response which follows exercise as well as many other aggressions [26, 32, 62, 71, 72] is part of a general response of the organism to stressful conditions.

References

1. Ahlborg, B., Brohult, J.: Immediate and delayed metabolic reactions in well-trained subjects after prolonged physical exercise. Acta med. scand. **182,** 41–54 (1967)
2. Asofsky, R., Thorbecke, G. J.: Sites of formation of immune globulins and of a component of C'_3. 2. Products of immunelectropheretically identified serum proteins by human and monkey tissues in vitro. J. exp. Med. **114,** 471–488 (1961)
3. Asofsky, R., Thorbeck, G. J., Hochwald, G. M., Williams, C. A.: Formation in vitro of plasma proteins by tissues from normal and infected animals. Fed. Proc. **22,** 264–269 (1963)
4. Astrup, T.: New developments in pathogenesis of arteriosclerosis. Circulation **48,** 239–243 (1973)
5. Blumberg, P. M., Robbins, P. W.: Effect of proteases on activation of resting chick embryo fibroblast and on cell surface proteins. Cell. **6,** 137–147 (1975)
6. Bullen, J. J., Leigh, L. C., Rogers, H. J.: The effect of iron compounds on the virulence of Escherichia coli for guinea pigs. Immunology **15,** 581–588 (1968)
7. Bullen, J. J., Rogers, H. J., Griffiths, E.: Bacterial iron metabolism in infection and immunity. In: Microbial iron metabolism (J. B. Nerlands, ed.), pp. 517–548. New York-London: Academic Press 1974
8. Bullen, J. J.: Iron and infection. Brit. J. Hann. **28,** 139 (1974)
9. Calder, E. A., Urbaniak, S. J., Irvine, W. J., James, K.: The effect of anti-alpha-2-macroglobulin on k-cell cytolisis and T- and B-cell rosette formation. Clin. exp. Immunol. **22,** 112–120 (1975)
10. Chester, J. K.: The role of zine ions in the transformation of lymphocytes by phytohaemagglutinin. Biochem. J. **130,** 133–139 (1972)
11. Clarke, H. G. M., Freeman, T., Pryse-Phillips, W.: Serum protein changes after injury. Clin. Sci. **40,** 337–344 (1971)
12. Cockerell, G. L.: Changes in plasma protein bound carbohydrates and glycoprotein patterns during infection, inflammation and starvation. Proc. Soc. exp. Biol. (N.Y.) **142,** 1072–1076 (1973)
13. Crawford, G. P., Ogston, D.: The influence of alpha-1-antitrypsin on plasmin, urokinase and Hageman factor cofactor. Biochim. biophys. Acta (Amst.) **354,** 107–113 (1974)
14. Crockson, R. A., Payne, C. J., Ratcliff, A. P., Soothill, J. F.: Time sequence of acute-phase reactive proteins following surgical trauma. Clin. chim. Acta **14,** 435–441 (1966)
15. Davidson, R. J. L.: Exertional haemoglobinuria: a report on three cases with studies on the hemolytiy mechanism. J. clin. Path. **17,** 536–540 (1964)
16. De Wyn, J. F., Dejongst, J. L., Mosterd, W., Willebrand, D.: Hemoglobin, packed cell volume, serum iron and iron-binding capacity of selected athletes during training. Nutr. Metab. **13,** 129–139 (1971)
17. Dufaux, B., Liesen, H.: Modifications of complement and of its components after a prolonged physical exercise and the influence of physical training (in preparation)
18. Evans, J. L., Abraham, P. A.: Anemia, iron storage and coeruloplasmin in copper nutrition in the growing rat. J. Nutr. **103,** 196–201 (1973)
19. Evans, J. L., Brar, B. S.: Tissue cytochrome oxidase activity and copper, iron, and pantothenic-acid nutrition in growing rat. J. Nutr. **104,** 1285–1291 (1974)
20. Ferguson, E. W., Guest, M. M.: Exercise physical conditioning, blood coagulation and fibrinolysis. Thrombos. Diathes. haemorrh. (Stuttg.) **31,** 69–71 (1974)
21. Fletcher, T. C., Baldo, B. A.: Immediate hypersensitivity responses in flat fish. Science **185,** 360–361 (1974)
22. Ford, W. H., Caspary, E. A., Shenton, B.: Purification and properties of a lymphocyte inhibition factor from human serum. Clin. exp. Immunol. **15,** 169–179 (1973)
23. Franzblau, C., Schmid, K., Faris, B., Beldekas, J., Garvin, P., Kagan, H. M., Baum, B. J.: The interaction of collagen with α-1-acid glycoprotein. Biochim. biophys. Acta (Amst.) **427,** 302–314 (1976)
24. Frieden, E.: Coeruloplasmin, a link between copper and iron metabolism. Nutr. Rev. **28,** 87–91 (1970)
25. Gal, K., Miltenyi, M.: Hemagglutination test for the demonstration of CRP. Acta microbiol. Acad. Sci. hung. **3,** 41–45 (1955)

26. Ganrot, K.: Plasma protein pattern in acute infections diseases. Scand. J. clin. Lab. Invest. **34,** 75–81 (1974)
27. Goldberg, A. R., Lazarowitz, S. G.: Plasminogen activators of normal and transformal cells. In: Proteinase inhibitors. Proc. of the 2nd Int. Res. Conf. (H. Fritz, H. Tschesche, L. J. Greene, E. Truscheit, eds.), pp. 631–648. Berlin-Heidelberg-New York: Springer 1974
28. Haralambie, G., Keul, J., Theumert, F: Protein-, Eisen- und Kupferveränderungen im Serum bei Schwimmern vor und nach Höhentraining. Europ. J. appl. Physiol. **35,** 21–31 (1976)
29. Hauss, W. H., Junge-Hülsing, G., Gerlach, U.: Die unspezifische Mesenchymreaktion, pp. 141–142. Stuttgart: Thieme 1968
30. Heimburger, N.: Biochemistry of proteinase inhibitors from human plasma. A review of recent development. In: Proteinase inhibitors. Proc. of the 2nd Int. Res. Conf. (H. Fritz, H. Tschesche, L. J. Greene, E. Truscheit, eds.), pp. 14–22. Berlin-Heidelberg-New York: Springer 1974
31. Hercz, A.: The inhibition of proteinases by human α_1-antitrypsin. Europ. J. Biochem. **49,** 287–292 (1974)
32. Herzberg, M., Oberman, Z., Weissman, S. L., Herold, H. Z.: Dynamic changes of different serum glycoproteins after bone fractures. Clin. Chem. **13,** 1065–1070 (1967)
33. Hornbostel, H., Huengers, R., Montz, R., Osburg, K.: Neuere Erkenntnisse bei der Marschhämoglobinurie. Dtsch med. Wschr. **95,** 458–461 (1970)
34. Johansson, B. G., Kindmark, C. O., Trell, E. Y., Wollheim, F. A.: Sequential changes of plasma proteins after myocardial infarction. Scand. J. clin. Lab. Invest. **29,** 17–22 (1972)
35. Kampschmidt, R. F., Upchurch, H. F., Eddington, C. L., Pulliam, L. A.: Multiple biological activities of a partially purified leucocyte endogenous mediator. Amer. J. Physiol. **224,** 530–533 (1973)
36. Kaplan, M. H., Volanakis, J. E.: Interaction of C-reactive protein complexes with the complement system. I. Consumption of human complement associated with reaction of C-reactive protein with pneumonical C-polysaccharide and with the choline phosphatides, lecithin and sphingomyelin. J. Immunol. **112,** 2135–2147 (1974)
37. Kaplan, S. S., Quie, P. G., Basford, R. E.: Effect of iron on leucocyte function: inactivation of H_2O_2 by iron. Infect. Immun. **12,** 303–308 (1975)
38. Kindmark, C. O.: Stimulating effect of C-reactive protein on phagocytosis of various species of pathogenic bacteria. Clin. exp. Immunol. **8,** 941–948 (1971)
39. Korsau-Bengsten, K., Conradso, T. B.: Influence of 2 different beta-adrenergic blocking agents on increase in fibrinolytic activity and factor VIII activity during submaximal and maximal exercise. Scand. J. Haemat. **13,** 377–384 (1974)
40. Liesen, H., Hollmann, W.: Arbeitsuntersuchungen über die Wirkung des Proteinasen-Inhibitors Trasylol auf kardio-pulmonale Stoffwechelparameter. Med. Welt **22,** 1381–1385 (1971)
41. Liesen, H., Michel, D., Hollmann, W.: Aktivitätsveränderungen von Serumenzymen bei jüngeren und älteren Athleten durch einen Marathonlauf. Sportwissenschaft **4,** 323–335 (1973)
42. Liesen, H., Dufaux, B., Weber, K., Lohmann, W., Fischer, W., Hollmann, W.: Das Verhalten von Serumimmunglobulinen bei Ausdauertraining und extremen Ausdauerbelastungen. Sportarzt u. Sportmedizin **6,** 119–123 (1976)
43. Liesen, H., Dufaux, B., Fischer, W., Hollmann, W.: Effects of endurance training on changing in serum glycoproteins and immunoglobulins in 55–70 years aged man. Proc. Int. Congress Physical Activity Sciences, Quebec 1976 (in press)
44. Liesen, H., Dufaux, B., Hollmann, W.: Ausdauertraining und natürliche Proteinaseninhibitoren: Der Einfluß auf die Plasmavolumenregulation in Ruhe und bei dosierter körperlicher Belastung. Sportarzt u. Sportmedizin (im Druck)
45. Mader, A., Liesen, H., Heck, H., Philippi, H., Rost, R., Schürch, P., Hollmann, W.: Zur Beurteilung der sportartspezifischen Ausdauerleistungsfähigkeit im Labor. Sportarzt u. Sportmedizin **4,** 80–88 (1976); **5,** 109–112 (1976)
46. Mancini, G., Carbonara, A. O., Heremans, J. F.: Immunochemical quantitation of antigens by single radial immunodiffusion. In: Immunochemistry, Vol. 2 (D. H. Campbell, ed.), pp. 235. New York: Pergamon Press 1965
47. Merrill, D. A., Kirkpatr, C. H., Wilson, W. E. C., Riley, C. M.: Change in serum haptoglobin type following human liver transplantation. Proc. Soc. exp. Biol. (N.Y.) **116,** 748 (1964)

48. Mortensen, R. F., Osmand, A. P., Gewurz, H.: Effects of C-reactive protein on the lymphoid system. J. exp. Med. **141,** 821–839 (1975)
49. Onda, H., Yoshikawa, J.: Alpha-1-acid glycoprotein as a hepatocyte-specific mitosis-inhibiting protein in regenerating rat liver. Gann **66,** 227–235 (1975)
50. Osaki, S., McDermott, I. A., Frieden, E.: Proof for the ascorbate oxidase activity of coeruloplasmin. J. biol. Chem. **239,** 3570–3575 (1964)
51. Osaki, S., Johnson, D. A., Frieden, E.: The possible significance of coeruloplasmin in normal human serum. J. biol. Chem. **241,** 2746–2751 (1966)
52. Osmand, A. P., Mortensen, R. F., Siegel, J., Gewurz, H.: Interaction of C-reactive protein with complement system. 3. Complement dependant passive hemolysis initiated by CRP. J. exp. Med. **142,** 1065–1077 (1975)
53. Pekarek, R., Wannemacher, R., Powanda, M. et al.: Further evidence that leucocyte endogenous mediator (LEM) is not endotoxin. Life Sci. **14,** 1765–1776 (1974)
54. Phillips, I. L., Azari, P.: Zinc transferrin-enhancement of nucleidacid synthesis in phytohemagglutinin-stimulated human lymphocytes. Cell Immunol. **10,** 31–37 (1974)
55. Phillips, J. L., Azari, P.: Effect of iron transferrin on nucleic acid synthesis in phytohemagglutinin-stimulated human lymphocytes. Cell Immunol. **15,** 94–99 (1975)
56. Ragan, H. A., Nacht, S., Lee, G. R., Bishop, C., Cartwright, G.: Effect of coeruloplasmin on plasma iron in copper-deficient swine. Amer. J. Physiol. **217,** 1320–1323 (1969)
57. Sarcione, E. J.: Synthesis of alpha-1-acid glycoprotein by isolated perfused rat liver. Arch. Biochem. **100,** 516–519 (1963)
58. Scheinberg, I. H.: Coeruloplasmin, a review. In: The biochemistry of copper (I. Peisach, Ph. Aisen, W. E. Blumberg, eds.), pp. 513–524. New York: Academic Press 1966
59. Schultze, H. E., Heremans, J. F.: Molecular biology of human proteins, Vol. 1, p. 354. Amsterdam: Elsevier 1966
60. Siegel, J., Rent, R., Gewurz, H.: Interactions of C-reactive protein with complement system. 1. Protamine induced consumption of complement in acute phase sera. J. exp. Med. **140,** 631–647 (1974)
61. Smith, E. B., Slater, R. S., Hunter, J. A.: Quantitative studies on fibrinogen and low density lypoprotein in human aortic intima. Atherosclerosis **18,** 479–487 (1973)
62. Snyder, S., Durham, B. C., Iskandrian, A. S., Coodley, E. L., Linhart, I. W.: Serum lipids and glycoproteins in acute myiocardial infarktion. Amer. Heart J. **90,** 582–586 (1975)
63. Stormosken, H.: Relation of fibrinolytic to other biological systems. Thrombos. Diathes. haemorrh. (Stuttg.) **34,** 378–385 (1975)
64. Tunstall, A. M., James, K.: Preliminary studies on the synthesis of alpha-2-macroglobulin by human lymphocytes in vitro. Clin. exp. Immunol. **17,** 697–701 (1974)
65. Unkeless, J. C., Gordon, S., Reich, E.: Secretion of plasminogen activator by stimulated macrophages. J. exp. Med. **139,** 834–850 (1974)
66. Van Oss, C. J., Gillman, C. F., Bronson, P. M., Border, J. R.: Phagocytosis – inhibiting properties of human serum alpha-1-acid glycoprotein. Immun. commun. **3,** 321–328 (1974)
67. Wandall, J. H.: Concentrations of serum proteins during and immediately after surgical trauma. Acta chir. scand. **140,** 171–179 (1974)
68. Weinberg, E. D.: Iron and susceptibility to infections disease. Science **184,** 952–956 (1974)
69. Weinberg, E. D.: Metal starvation of pathogens by hosts. Bio. Science **25,** 314–318 (1975)
70. Weinberg, E. D.: Nutritional immunity. Host's attempt to withhold iron from microbial invaders. J. Amer. med. Ass. **231,** 39–41 (1975)
71. Werner, M., Odenthal, D.: Serum protein changes after gastrectomy as a model of acute phase reaction. J. Lab. clin. Med. **70,** 302–310 (1967)
72. Werner, M.: Serum protein changes during the acute phase reaction. Clin. chim. Acta **25,** 299–305 (1969)
73. Willoughby, D. A., Ryan, G. B.: Evidence for a possible endogenous antigen in chronic inflammation. J. Path. **101,** 233–239 (1970)
74. Yoffey, I. M., Courtice, F. C.: Lymphatics, lymph and the lymphomyeloid complex, p. 208. London-New York: Academic Press 1970

Received November 2, 1976/Accepted July 7, 1977