THE ROLE OF TRYPTOPHAN IN FATIGUE IN DIFFERENT CONDITIONS OF STRESS

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ABSTRACT

Tryptophan is the precursor for the neurotransmitter 5-hydroxytryptamine (5-HT), which is involved in fatigue and sleep. It is present in bound and free form in the blood, where the concentration is controlled by albumin binding to tryptophan. An increase in plasma free tryptophan leads to an increased rate of entry of tryptophan into the brain. This should lead to a higher level of 5-HT which may cause central fatigue. Central fatigue is implicated in clinical conditions such as chronic fatigue syndrome and post-operative fatigue. Increased plasma free tryptophan leads to an increase in the plasma concentration ratio of free tryptophan to the branched chain amino acids (BCAA) which compete with tryptophan for entry into the brain across the blood-brain barrier.

The plasma concentrations of these amino acids were measured in chronic fatigue syndrome patients (CFS) before and after exercise (Castell *et al.*, 1998), and in patients undergoing major surgery (Yamamoto *et al.*, 1997). In the CFS patients, the pre-exercise concentration of plasma free tryptophan was higher than in controls ($p < 0.05$) but did not change during or after exercise. This might indicate an abnormally high level of brain 5-HT in CFS patients leading to persistent fatigue. In the control group, plasma free tryptophan was increased after maximal exercise ($p < 0.001$), returning towards baseline levels 60 min later. The apparent failure of the CFS patients to change the plasma free tryptophan concentration or the free tryptophan/BCAA ratio during exercise may indicate increased sensitivity of brain 5-HT receptors, as has been demonstrated in other studies (Cleare *et al.*, 1995).

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In post-operative recovery after major surgery plasma free tryptophan concentrations were markedly increased compared with baseline levels; the plasma free tryptophan/BCAA concentration ratio was also increased after surgery. Plasma albumin concentrations were decreased after surgery: this may account for the increase in plasma free tryptophan levels.

Provision of BCAA has improved mental performance in athletes after endurance exercise (Blomstrand *et al.*, 1995, 1997). It is suggested that BCAA supplementation may help to counteract the effects of an increase in plasma free tryptophan, and may thus improve the status of patients during or after some clinically stressful conditions.

1. THE ROLE OF TRYPTOPHAN IN CENTRAL FATIGUE

Tryptophan is predominantly taken up and metabolised by the liver. However, some tryptophan is taken up by the brain where it is converted to the neurotransmitter 5-hydroxytryptamine (5-HT) which can influence behaviour such as sleep, mood and fatigue (Newsholme and Leech, 1983).

Tryptophan is unique among amino acids in that it is bound to albumin in the blood. Plasma fatty acids also bind to albumin: this decreases the affinity for tryptophan. Thus, an increase in plasma free fatty acids can lead to an increased plasma free tryptophan level. An increased level of brain tryptophan can increase the rate of formation, and the level of 5-HT in some areas of the brain (Blomstrand *et al.*, 1989).

A high 5-HT level could result in an increased amount of this neurotransmitter being released into the synaptic cleft during neuronal firing, leading to a greater postsynaptic stimulation in some 5-HT neurones, some of which may be involved in fatigue.

The branched chain amino acids (leucine, isoleucine and valine) compete with tryptophan for entry into the brain across the blood-brain barrier (Oldendorf and Szabo, 1976; for review see Fernstrom, 1990). It is considered that the plasma concentration of free tryptophan, in competition with branched chain amino acids (BCAA), governs the rate of entry of tryptophan into the brain, the level of tryptophan in the brain and hence that of 5-HT (Blomstrand *et al.*, 1988; Fernstrom, 1990). An increase in the level of brain 5-HT would lead to fatigue.

2. CENTRAL FATIGUE AND EXHAUSTIVE EXERCISE

In middle distance exercise, depending upon the fitness of the individual and the duration/distance of the event, fatigue may occur because of either the depletion of glycogen or the accumulation of protons in the muscle (Newsholme *et al.*, 1994). If glycogen levels fall too early in an event, fatty acid mobilisation may cause fatigue before the finish. Increased mobilisation of fatty acids from adipose tissue is probably the result of sympathetic stimulation. In the early stages of prolonged exercise such as a marathon, the plasma concentration of fatty acids may be only slightly increased, because the rates of fatty acid uptake and oxidation by the active muscle are increased (Winder, 1996). Eventually, however, there is insufficient glycogen to provide the energy required for the whole event and thus fatty acids must be mobilised.

It is also possible that, in unfit subjects, the plasma fatty acid concentration and, therefore, the free tryptophan level increases markedly in exercise because fatty acid mobilisation may not be precisely regulated in relation to demand and control of oxidation within the muscle.

Some of the experimental findings, which provide evidence to support the hypothesis, are set out below.

(i) The plasma concentration ratio of free tryptophan to BCAA is increased in humans after prolonged, exhaustive exercise; in the rat, the brain levels of tryptophan and 5-HT are increased after exercise (Blomstrand *et al.*, 1989, 1991; Chaouloff *et al.*, 1986).

(ii) Administration of a 5-HT agonist impaired running performance whereas a 5-HT antagonist improved running performance in rats (Bailey *et al.*, 1992).

(iii) In humans, the administration of a 5-HT re-uptake blocker (paroxetine) decreased exercise time to exhaustion during standardised exercise (Wilson and Maughan, 1992).

(iv) Prolactin secretion from the hypothalamus is controlled, in part, by 5-HT-neurones, and 5-HT stimulates the rate of secretion. A correlation between plasma prolactin and free tryptophan during exercise suggests that an increased plasma concentration of free tryptophan can influence the 5-HT level in the hypothalamus (Fischer *et al.*, 1991).

(v) An increase in 5-HT levels in the hypothalamus due to, *e.g.* fenfluramine, caused a much smaller increase in plasma prolactin in well-trained endurance athletes, compared with controls. This could be caused by down-regulation of 5-HT receptors as a result of chronic elevation of the 5-HT level in the hypothalamus (Jakeman *et al.*, 1994).

(vi) In rats with genetically controlled lack of albumin, the intrasynaptosomal concentration of tryptophan, 5-hydroxytryptophan and 5-HT was markedly increased in the striatum after running to fatigue (Yamamoto *et al.*, 1997).

In the past few years, supplementary feeding with BCAA in athletes has produced some results supporting the hypothesis and some which show no effect. Some of these studies are included in a brief comparison in Table 1.

Mittleman *et al.* (1998) observed a positive effect of BCAA supplementation on performance in moderate exercise during heat stress in men and women. Blomstrand *et al.* (1997) monitored perceived effort and mental fatigue in seven endurance cyclists with and without BCAA supplementation. When they received BCAA the subjects perceived that less effort was required to sustain a specific level of exercise, compared to that experienced when receiving a placebo.

The data on athletes in Table 1 indicate that, the higher the dose of BCAA, the more likely it is that plasma ammonia levels will be elevated. This suggests that lower doses are more likely to be beneficial. In several studies BCAA were administered before exercise. It may be that administration during exercise was the reason that Blomstrand *et al.* (1997) and that Mittleman *et al.* (1998) failed to observe a change in plasma ammonia. Whether a bolus dose is given or whether separate doses are given during exercise could be important for the release of ammonia from muscle.

In rats, Calders *et al.* (1997) observed an increase in the time to fatigue, as well as an increase in plasma ammonia, in fasting rats injected with 30 mg of BCAA five minutes before exercise, compared to those injected with placebo.

In genetically analbuminemic rats, sustained running to the point of fatigue caused an increase in the concentration of tryptophan and in the turnover of 5-HT in the striatal synaptosomes. The reason proposed for this is an increase in plasma free tryptophan which may be due to the lack of albumin. The rate-limiting enzyme, tryptophan hydroxylase, catalyzes the conversion of tryptophan to 5-hydroxytryptophan, the immediate pre-

Table 1. A comparison of studies on branched chain amino acid supplementation in humans during endurance exercise. Peak p[BCAA] denotes the highest plasma concentration of branched chain amino acids observed in each study. N/M denotes not measured.

No. of subj.	Exercise	Amount of BCAA ingested	Peak p[BCAA]	Effect on NH_3	Effect on Performance		VO_{2max} %	Ref
					Mental	Physical		
13	Cycling	12.8 g	1,250 μM	None	None	Improved*	40	1
10	Cycling	23.4 g	2,400 μM	Rise	N/M	None[h]	70–75	2
	Cycling	7.8 g	950 μM	Rise	N/M	None[h]	70–75	2
193[m]	Marathon[d]	16 g	1,250 μM	N/M	Improved	Improved[e]	N/M	3
5	Cycling	6.3 g/L[b]	1,000 μM	N/M	N/M	None	75	4
10	Cycling	16 g/day[j]	N/M	N/M	N/M	Improved	72.7	5
7	Cycling	30 g	3,000 μM	Rise	N/M	None		7
52	30 km run[d]	5.3 g[c]	650 μM	N/M	Improved	N/M	N/M	9
7	Cycling	6–9 g	1,050 μM	None	Improved	Improved	70	10
9	Cycling[k]	18 g[a]	1,026 μM	Rise	N/M	None	63.1	11
10	Cycling	21 g	ca.1,250 μM	Rise	Improved	None	N/M	12

Exceptional conditions:

*Heat stress; [d]Field study; [e]In subset of slower runners; [h]High day-to-day intra-individual variation in time to fatigue; [j]14-day study; [k]100-km trials; [m]Not all subjects gave blood samples.

Additions to BCAA:

[a]5% carbohydrate; [b]6% carbohydrate; [c]7% carbohydrate.

References: [1]Mittleman *et al.* (1998); [2]van Hall *et al.* (1995); [3]Blomstrand *et al.* (1991); [4]Blomstrand *et al.* (1995); [5]Hefler *et al.* (1995); [7]Wagenmakers (1992); [9]Hassmen *et al.* (1994); [10]Blomstrand *et al.* (1997); [11]Madsen *et al.* (1996); [12]Struder *et al.* (1998).

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cursor of 5-HT. The marked increase in 5-hydroxytryptophan observed in the analbuminemic rats after exercise, might be explained by increased activity of tryptophan hydroxylase due to increased level of the substrate tryptophan in the neurones. This enzyme is not saturated in the mammalian brain (Pardridge, 1977), and thus 5-HT production in the brain may be sensitive to dietary tryptophan (Fernstrom and Wurtman, 1974) via a change in the concentration of plasma tryptophan (Blomstrand *et al.*, 1988). This may result in an increase in the neuronal level of 5-HT, a subsequent increase in the rate of 5-HT neuronal firing, and consequently to central fatigue.

Injection of BCAA has both increased the time to fatigue of exercising rats and prevented the normal increase in brain tryptophan level caused by exhaustive exercise (T. Yamamoto, personal communication).

3. CHRONIC FATIGUE SYNDROME

Central fatigue may be an important factor in chronic fatigue syndrome, since muscle function in this condition is normal (Bigland-Ritchie *et al.*, 1995).

Patients with chronic fatigue syndrome (CFS), compared with sedentary controls were studied before and after exercise (for full details see Castell *et al.*, 1998). Exercise was undertaken on a bicycle ergometer, with 30 W increments every 3 min until the subjects stopped of their own volition. Subjects were monitored during exercise with (1) a

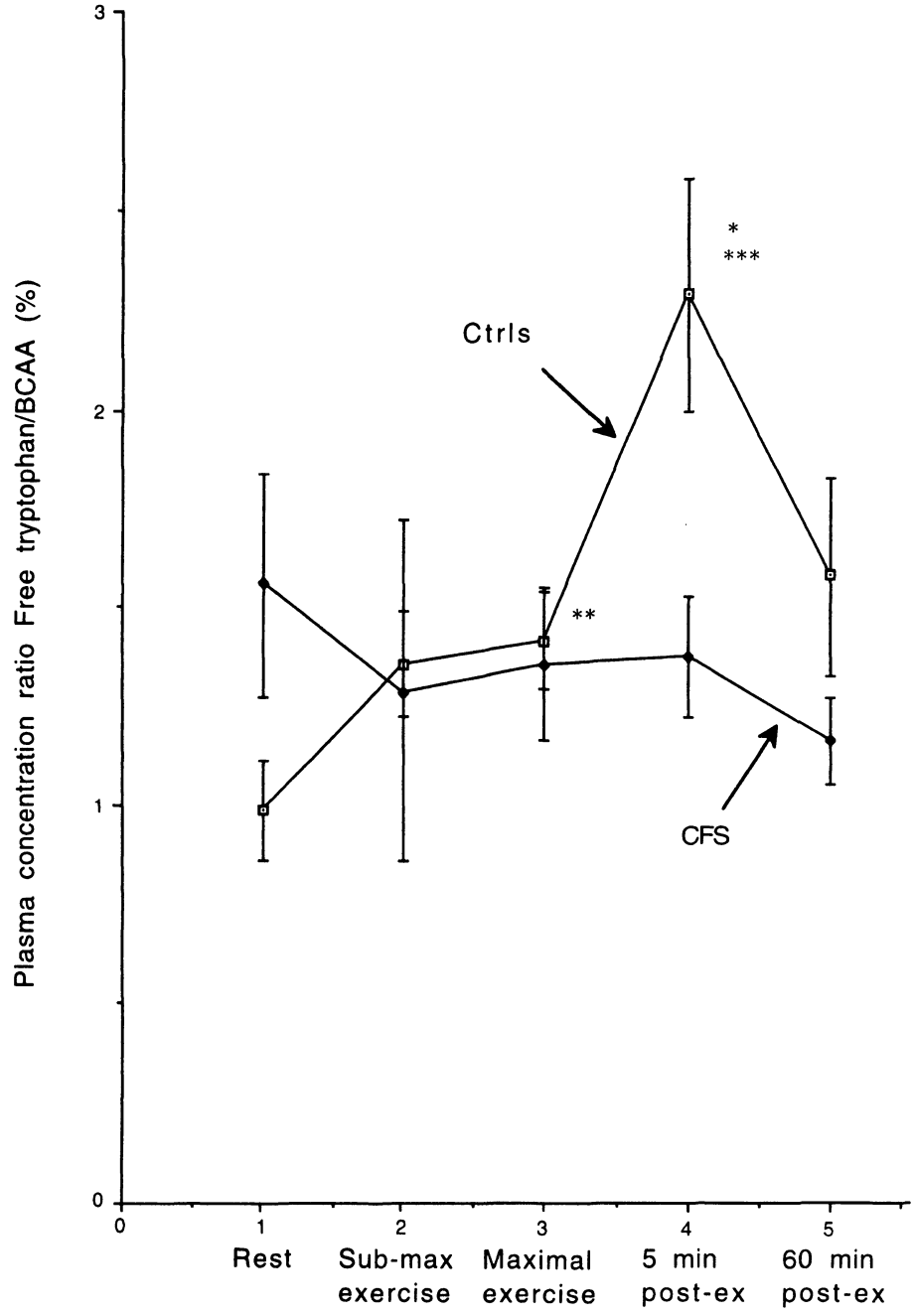

Figure 1. Plasma concentration ratio of free tryptophan/BCAA in chronic fatigue syndrome patients, compared with age-matched controls, before and after exercise. Statistical significance is indicated by *$p < 0.02$, between CFS and controls; **$p < 0.05$; ***$p < 0.001$, compared with baseline values.

continuous heart rate monitor; (2) the Borg scale for perceived exertion. Blood samples were taken at rest, and after sub-maximal and maximal exercise, and assayed for free and total tryptophan and branched chain amino acids.

The total amount of work done at maximal exercise reflected the patients' inability to reach maximal exercise, despite their perception (according to the Borg scale) of a significantly greater exertion than the controls.

In the CFS patients, the pre-exercise concentration of plasma free tryptophan was higher than in controls ($p < 0.05$) but did not change during or after exercise. This might indicate an abnormally high level of brain 5-HT in CFS patients leading to persistent fatigue. In the control group, plasma free tryptophan increased at maximal exercise ($p < 0.02$), peaking at 5 min post-ex ($p < 0.001$), returning to baseline levels at 60 min.

The resting plasma concentration ratio of free tryptophan/BCAA appeared 31% higher (only close to significance, $p < 0.1$) in the CFS patients than the controls and remained at similar levels during and after exercise (Fig. 1). The apparent failure of these patients to change the free tryptophan/BCAA ratio during exercise may indicate an increased sensitivity of brain 5-HT receptors, such as has been demonstrated in other studies (Cleare *et al.*, 1995).

4. POST-OPERATIVE FATIGUE

A study was undertaken to investigate whether or not the plasma concentration ratio of free tryptophan/BCAA and the affinity of plasma tryptophan binding to albumin were altered post-operatively in humans (for details see Yamamoto *et al.*, 1997).

Samples were taken before and after major surgery from elderly patients undergoing restorative surgery and from coronary artery bypass graft (CABG) patients. In both the elderly and the CABG patients plasma free tryptophan concentrations were increased after surgery, compared with baseline levels; the plasma free tryptophan/BCAA concentration ratio was also increased significantly after surgery . Plasma albumin concentrations were decreased significantly after surgery in both the elderly and the CABG patients.

Plasma free tryptophan concentrations were markedly increased after surgery in the elderly patients, compared with baseline levels pre-surgery and two weeks after surgery. The plasma concentration ratio of free tryptophan/BCAA after surgery was markedly increased compared with those at two weeks after surgery. A decrease in plasma albumin after surgery was maintained for two weeks.

In the elderly patients, there was a decrease in the affinity of albumin binding of tryptophan after surgery, a decrease in plasma albumin concentrations , and an increase in plasma free tryptophan concentrations. It should also be taken into consideration that binding is relatively low in old age (Koch-Weser and Sellers, 1976).

In the CABG patients, plasma free tryptophan levels were markedly increased after surgery, compared with baseline levels. The plasma concentration ratio of free tryptophan/BCAA was increased after surgery and, similar to the plasma free tryptophan concentration, returned nearly to baseline levels 5 days later. Plasma albumin concentrations were decreased significantly after surgery, returning to baseline concentrations 5 days later. A smaller increase in plasma free tryptophan concentration was observed in these patients than in the elderly patients. One factor which may have affected the plasma concentrations of amino acids in the CABG study is the plasma dilution which occurs during cardiopulmonary bypass.

5. DISCUSSION

The post-operative fatigue study reported the first evidence that there is a significant increase in the plasma concentration ratio of free tryptophan/BCAA after major surgery. This might be an important factor in post-operative recovery in humans. Despite the unique nature of the coronary artery bypass graft operation, the results from the different groups of surgical patients were very similar. Thus, it seems likely that post-operative fatigue occurs after major surgery which may be related to an increase in plasma free tryptophan via an increase in 5-HT.

Beneficial effects of BCAA supplementation have been seen in aspects of both mental and physical fatigue in exhaustive exercise. Most of these feeding studies in athletes have not investigated effects on mental fatigue. However, the mental exertion necessary to maintain a given power output is an integral feature of central fatigue.

It is suggested that dietary provision of BCAA might help to overcome some problems of post-operative recovery which may be due to 5-HT-induced fatigue at certain times after surgery, and that this supplementation might also be useful in other clinical situations which involve fatigue, such as chronic fatigue syndrome or overtraining.

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