Simultaneous Alterations of Circulating Mg Subfractions and Liver Related Parameters during Stress in Human Probands

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Zusammenfassung

Streß führt angeblich [1, 2] zu Mg-Verlust aus dem Herzen und der Leber, was zu einer Veränderung der zirkulierenden Mg-Fraktionen führen sollte. Sollte die Leber tatsächlich wesentlich am veränderten Mg-Verhalten beteiligt sein, könnte das leicht auch zu veränderten Harnstoff-, Cholesterin-, Blutzucker- und Cholinesterase-Werten führen. Um diese Vermutung näher zu untersuchen, hatten 12 Pilotenanwärter des Österreichischen Bundesheeres eine Serie von psychischen und physischen Streßsituationen vor und nach einem Trainingsflug zu bewältigen. Unsere Versuchsanordnung beinhaltete somit Streßsituationen unterschiedlicher Art, Dauer und Intensität. Vor dem Flug wurden insgesamt 6, nach dem Flug 5 Blutproben (jeweils nach den Kurzzeitstreßsituationen) abgenommen. Aus diesen Blutproben bestimmten wir jeweils totales, ionisiertes und daraus berechnetes gebundenes Magnesium wie auch die oben genannten Parameter. Die nach dem Flug abgenommenen Blutproben zeigten, im Vergleich zu den Blutkonzentrationen vor dem Flug, einen deutlichen Anstieg des ionisierten Mg (unter physischem und psychischem Streß: p = 0,001) mit einem gleichzeitigen Abfall des gebundenen Mg (unter psychischem Streß: p = 0,001, unter physischem Streß: p = 0,006). Nach dem Flug waren aber auch die Werte von Cholinesterase (p = 0,046), Cholesterol (p = 0,008) und Harnstoff (p = 0,023) erniedrigt. Diese Resultate zeigen eine deutliche streßbedingte Veränderung der zirkulierenden Mg-Fraktionen und eine gleichzeitige Veränderung der oben genannten Leberparameter in eine Richtung, die keinen Anhaltspunkt für eine erhöhte Leberaktivität bietet.

Summary

Allegedly [1, 2] stress leads to Mg loss from heart and liver, which should result in changed levels of circulating Mg and – if the liver did really participate – perhaps also in altered levels of urea, cholesterol, blood glucose and cholinesterase activity. This should be investigated by application of a series of ergometric and psychological stresses to 12 young officer pilot trainees of the Austrian Federal Army before and after an aerobatic flight. Thus we provided stressful situations of various duration, type and intensity. Their effect was checked by 6 blood samples after short-term stresses before the flight and 5 samples afterwards, whereby total, ionized and calculated bound magnesium values were determined in addition to the liver parameters mentioned above. Blood sampled in stressful situations after the aerobatics, showed significantly increased levels of ionized magnesium (under physical and psychological stress: p = 0.001) with a simultaneous decrease in the calculated bound fraction (under psychological stress: p = 0.001, under physical stress: p = 0.006). At the same time the levels of cholesterol (p = 0.008) and urea (p = 0.023) were lowered as was the activity of cholinesterase (p = 0.046). There are distinct stress induced alterations in the circulating subfractions of magnesium, which are accompanied by significant alterations of liver parameters in a direction, which can well be interpreted as an indication of restricted liver function.

Résumé

Le stress soi-disant mène aux pertes de Mg [1, 2] dans le coeur et le foie, ce qui devrait rèsulter dans un changement du taux de Mg circulant. Si en effet le foie en fait partie, cela pourrait facilement mener à des taux altérés dans l'urine, dans le cholestérol, le sucre sanguin et dans la cholinestérase. Pour vérifier ce soupçon, 12 aspirants de pilotes de l'armée fédérale d'Autriche devaient endurer des situations de stress psychiques et physiques avant et après un vole d'entraînement. Cette étude comprenait des situations de stress diverses, en ce qui concernait la manière, la durée et l'intensité. En somme, 6 échantillons du sang furent prélevé avant le vol, tandis que 5 échantillons du sang furent prelevé après le vol (chaque fois après des situations de stress de courte durée), dont nous avons déterminé le Mg total, le Mg ionisé et le Mg lié qui se calculait du Mg total et ionisé, en addition des taux susmentionnés. Les échantillons du sang prelevé après le vol montraient – en comparaison des taux avant le vol, une augmentation signifiante du Mg ionisé (sous des conditions de stress physique et psychique: p = 0,001), avec une diminution simultanée du Mg lié (sous stress psychique: p = 0,001, sous stress physique: p = 0,006). Pourtant, après le vol, les taux de cholinestérase (p = 0,046), du cholestérol (p = 0,008) et de l'urée (p = 0,023) étaient diminués. Ces résultats montrent clairement un changement des fractions de Mg circulants au stress, ainsi qu'un changement simultané des taux du foie dans un sens qui ne peut pas être interprété comme activité du foie augmentée.

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Introduction

It has been shown in animal experiments [1, 2], partly in vivo and in vitro, that an increase in catecholamine levels leads to depletion of Mg from soft tissue organs, mainly heart and liver, whereby the time course is still unclear. We tried to follow up those results in human probands by subjecting them to defined stresses. Before and after those stresses we took blood samples and measured ionized and total magnesium and calculated bound Mg. At the same time we estimated blood glucose levels along with typical liver parameters like urea and cholesterol concentrations as well as cholinesterase activity.

The aim of the study was to investigate, what kind of alterations in circulating Mg and its subfractions would take place and – since diminishment of liver magnesium in such situations is postulated – what kind of simultaneous alterations of liver parameters could be measured.

Material and Methods

We used a standard system of cumulating stress situations, consisting of a series of alternating physical and psychological stresses out of a relaxed standard situation, and called them "internal standardization" (fig. 1). On another day we repeated these series of short stresses in the same way but not before a 45 min intensive mixed physical and psychical stress was absolved, namely the very first combat like aerobatics in the training of the young pilots.

30 min before the experiment started, the young pilot trainees got their antecubital vein canulated. Blood was drawn immediately before the experiment and after each step of the experiment:

1st step: 7 min cycle ergometry
2nd step: break of 30 min
3rd step: 45 min of psychological stress, consisting of a visual discrimination test, a signal detection test, and a short visual memory test
4th step: 7 min cycle ergometry
5th step: break of 30 min

The same procedure was performed on the second day, but only after 45 min of combat like aerobatics were performed.

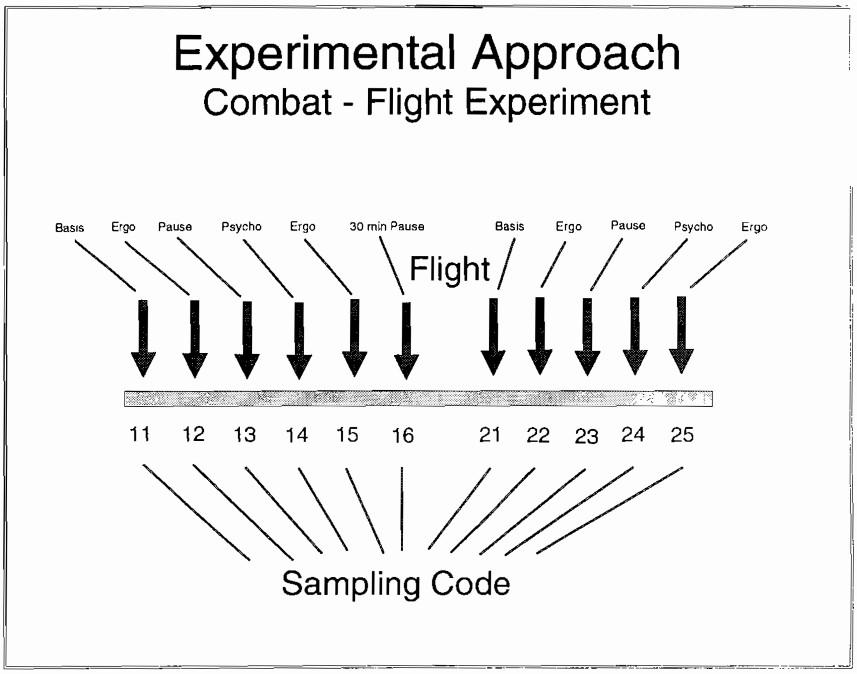

Fig. 1: Experimental approach: 11: 1st day, basal level, 12: 1st day, level after 7 min cycle ergometry, 13: 1st day, level after 30 min rest, 14: 1st day, level after 45 min psychological test, 15: 1st day, level after 7 min cycle ergometry, 16: 1st day, level after 30 min rest, 21: 2nd day, basal level, 22: 2nd day, level after 7 min cycle ergometry, 23: 2nd day, level after 30 min rest, 24: 2nd day, level after 45 min psychological test, 25: 2nd day, level after 7 min cycle ergometry.

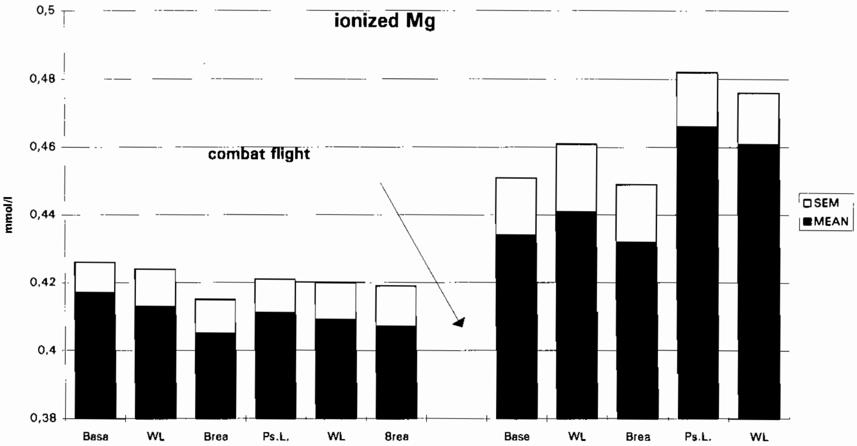

Fig. 2: Ionized Mg: x: Base: basal level, WL: 7 min workload, Brea: 30 min break, Ps.L.: a psychological test of 45 min, y: ionized Mg in mmol/l plasma.

From those blood samples we measured:

1. total Mg concentrations (Kodak dry chemical method),
2. ionized Mg concentrations (with the AVL 9884 magnesium electrode magnesium analyzer, AVL, Graz),
3. by subtraction of the ionized from the total fraction [3], a bound fraction was characterized, consisting of a small complexometric bound [4] and a bigger protein bound fraction,
4. glucose, cholesterol, urea, as well as cholinesterase activity in blood.

Results

During the first day, when just a series of short standard stresses **without previous aerobatics** was applied, there was nearly no alteration of ionized Mg subsequent to those standard stresses (fig. 2).

The bound fraction on the other hand slowly increased along with the number of the short stresses in such a clear way that it figured prominently in shaping the curve of total Mg (fig. 3 and 4).

It is important to notice, that in the very first blood sample, taken as the basal level, the concentration of ionized Mg was about 0.03 mmols higher than that of the calculated bound fraction.

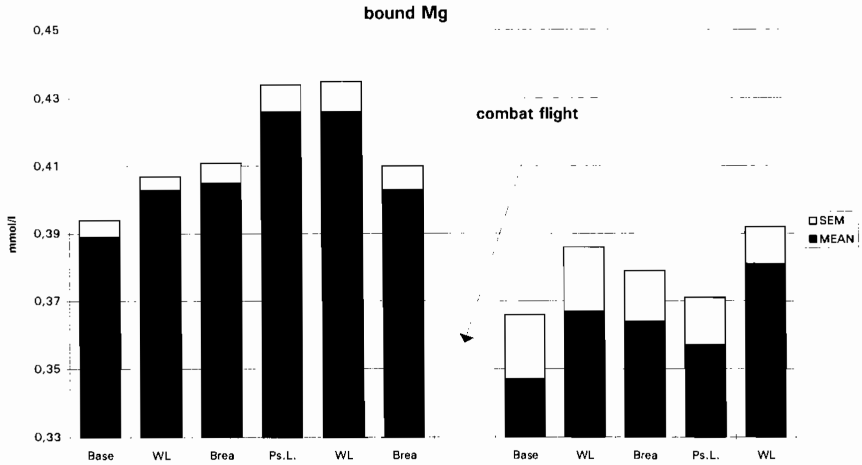

Fig. 3: Bound Mg: x: Base: basal level, WL: 7 min workload, Brea: 30 min break, Ps.L.: a psychological test of 45 min, y: bound Mg in mmol/l plasma.

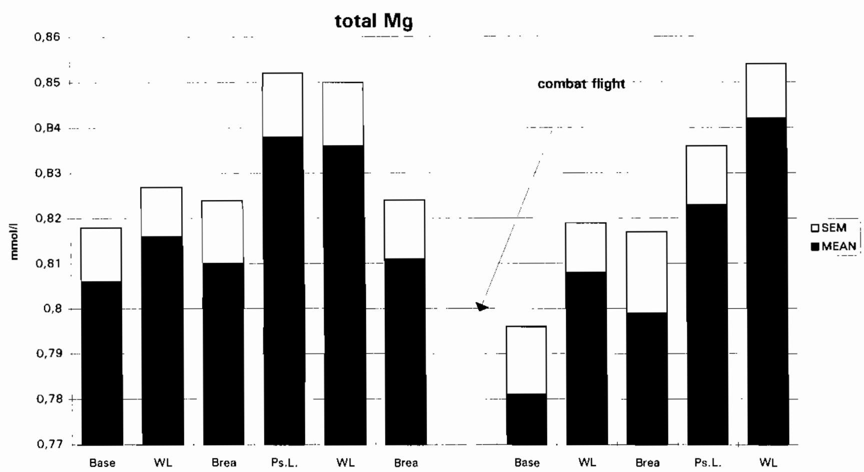

Fig. 4: Total Mg: x: Base: basal level, WL: 7 min workload, Brea: 30 min break, Ps.L.: a psychological test of 45 min, y: total Mg in mmol/l plasma.

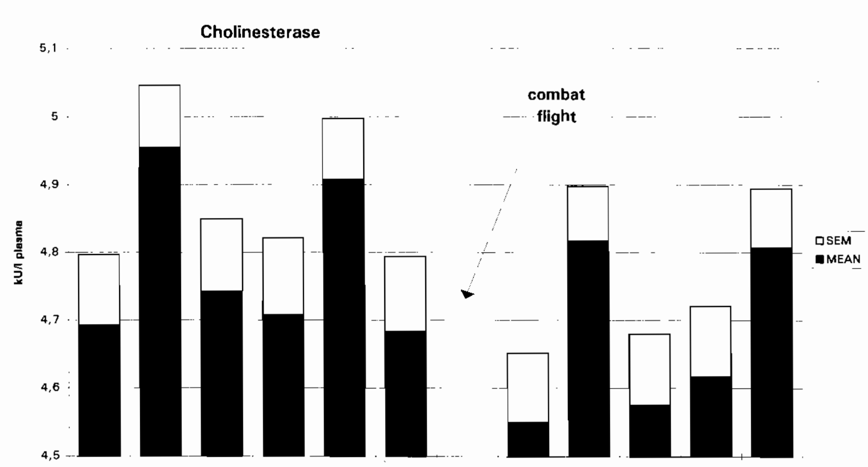

Fig. 5: Cholinesterase: x: Base: basal level, WL: 7 min workload, Brea: 30 min break, Ps.L.: a psychological test of 45 min, y: Cholinesterase in kU/l plasma.

The second basal level, however, taken at the day of aerobatic flight, half an hour after its cessation, showed a lower total Mg when compared to the first basal level. Interestingly, the calculated bound Mg subfraction was also significantly lower than at the first day (under physical stress: $p = 0.006$, under psychological stress: $p = 0.001$), while the ionized fraction was significantly higher (under physiological as well as under physical stress: $p = 0.001$).

The difference between the levels of ionized and bound fraction thus increased after stress from 0.03 mmols to 0.09 mmols.

After aerobatics however, bound Mg levels showed only slight alterations along with the physical stresses, while the ionized Mg increased steadily and markedly, a behaviour, which decisively influenced the behaviour of the total Mg.

A longer uninterrupted stress of about 45 min led therefore to a fall of total Mg in blood, mostly brought about by the unproportional diminishment of the bound fraction. The difference between the levels of ionized and bound Mg fraction was higher when compared with the first day.

At the same time the liver parameters, like urea ($p = 0.023$), cholesterol ($p = 0.008$) and cholinesterase activity ($p = 0.046$) were in the average clearly lower during the series of stresses after the aerobatics than they had been on the day before the flight (fig. 5).

In the course of the stress series, however, those parameters behaved individually differently.

While cholinesterase activity and cholesterol concentration did show clear increases immediately after physical exercise, urea values just steadily increased along the general time course from a very low level immediately after the flight to an end concentration similiar or undistinguishable from those won during the stress series before the flight (fig. 6 and 7).

While aerobatic stress lowered liver related substrate concentrations and enzyme activities, blood glucose levels were differently affected (fig. 8).

The blood glucose level after aerobatics was undistinguishable from the

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basal level on the first day without aerobatics.
The reaction of glucose levels on the first day before aerobatics only consisted in a slight decrease of glucose after the 1st 7 min of physical workload. Although the basal levels after aerobatics had been the same as on the day without aerobatics, the blood glucose reaction to the physical workload after aerobatics was at least threefold higher than that seen before aerobatics (p = 0,027).

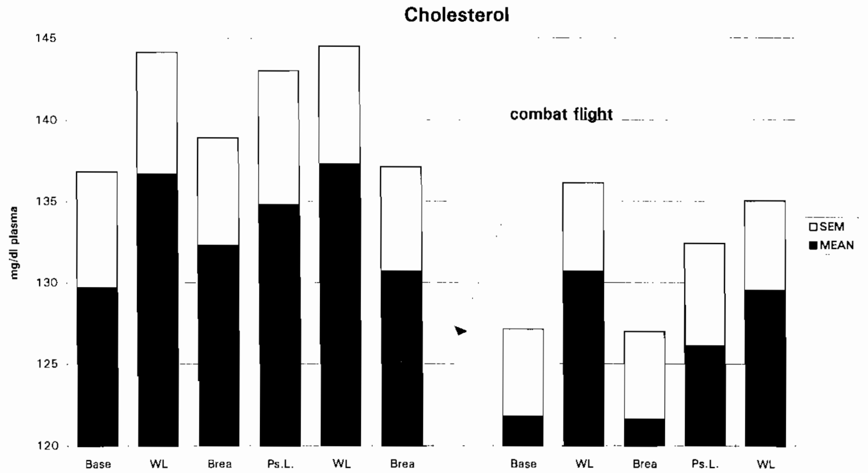

Fig. 6: Cholesterol: x: Base: basal level, WL: 7 min workload, Brea: 30 min break, Ps.L.: a psychological test of 45 min, y: Cholesterol in mg/dl plasma.

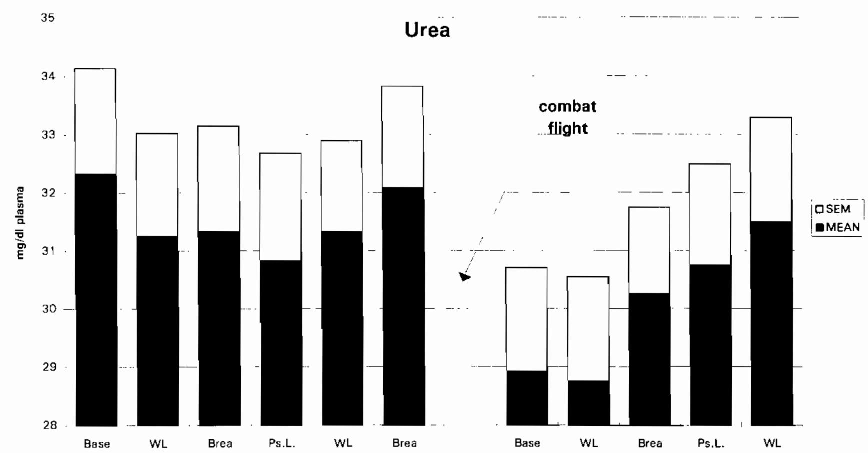

Fig. 7: Urea: x: Base: basal level, WL: 7 min workload, Brea: 30 min break, Ps.L.: a psychological test of 45 min, y: Urea in mg/dl plasma.

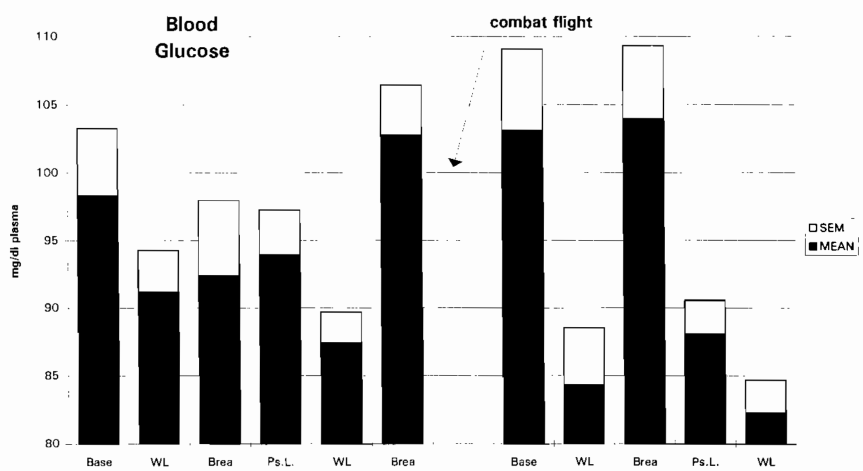

Fig. 8: Blood Glucose: x: Base: basal level, WL: 7 min workload, Brea: 30 min break, Ps.L.: a psychological test of 45 min, y: Blood glucose in mg/dl plasma.

Discussion

The most immediate reaction of Mg subfractions in blood to a series of short stresses seems to be their drifting apart, at the second day even accompanied with an unproportional lowering of the bound fraction and an increase of the ionized Mg. Thus a short time stress or even a series of such stresses may be diagnostically characterized by this drifting apart of the values of the bound and ionized Mg fractions.
Moreover, stress of longer duration lowers the bound Mg in such a way that a clearly recognizable decrease of the total Mg results.
There are then measurable reactions of the circulating magnesium to different stresses. They seem to be quicker as hitherto suspected (already after minutes of stress) and they seem to afflict the circulating magnesium subfractions in a much more clearcut way than total magnesium levels. During all those alterations the threefold increased lag between the bound and free fraction of Mg was steadily kept up. If one follows the reasoning of the authors postulating Mg depletion from the liver under stress [5, 6, 7], according to our results also a simultaneous Mg loss from circulation should occur, whereby, of course, both – extravascular or extracorporal loss – is possible. Our results show further, that Mg depletion from the liver does not seem to be a process which goes hand in hand with increased liver activity.
Therefore stress-induced Mg depletion of the liver does not seem to be an active process, but on the contrary, the stress induced Mg loss from the liver seems to reduce liver activities, not in a pathological, but nevertheless in a recognizable way. This can be deduced from the generally lower activities of cholinesterase, from the lower cholesterol levels and from the significant lowering of the urea levels after aerobatics.

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The remarkable increased fluctuations of glucose concentrations, provoked by physical stress after the aerobatics also fit into the picture in so far, as smoothly running glucose turnover during stress induced increased demand can only be provided by plentiful of Mg supply. This means that aerobatic stress did not manifest itself in changing the basal level of blood glucose, but it had striking influence upon the reagibility of glucose levels to short physical stress. Since in our stress experiments Mg content of the liver per definition should be not at all plentiful, buffer capacity of glucose supply seems to be diminished, which leads to these unusual high fluctuations of blood glucose levels under physical stress.

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