The stress-induced disruption of hippocampal LTP is associated to a deficient CaMKII signaling pathway

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Summary

Experiences of stress are known to disrupt rapidly the long-term potentiation (LTP) at excitatory synapses in the hippocampus. To understand which synaptic mechanisms are long-lastingly altered we employed somatic whole-cell recording of the evoked $\alpha$-amino-3-hydroxy-5-methyl-4-isoxazole propionate receptor (AMPA-R) responses in hippocampal slices of rats exposed to social defeat on two consecutive days followed by three weeks of post-stress delay. We demonstrate that in stressed rats the recurrent CA3 synapses are enhanced in baseline transmission. However, activity-dependent regulation of the AMPA-R currents was selectively impaired in LTP induction ($98 \pm 8\%$) together with a normal expression of long-term depression (LTD) and intact de-potentiation. Control animals showed larger responses in synaptic strength upon LTP tetanization. Consistent with the physiological characteristic of the CA3 recurrent network, stress impaired the LTP-induced transition from unitary excitatory-postsynaptic currents (EPSCs) to AMPA-R mediated network bursting. We aimed to test the role of Ca$^{2+}$-calmodulin-dependent kinase II (CaMKII) mediated phosphorylation to establish LTP by functionally activating the enzyme using intracellular postsynaptic perfusion of Ca$^{2+}$-calmodulin (Ca$^{2+}$-CaM) and monitoring EPSCs. In control animals the treatment with Ca$^{2+}$-CaM (4:1) resulted in a progressive, kinase-dependent up-regulation of EPSC peak amplitudes (~190% from baseline). In slices from stressed animals, however, Ca$^{2+}$-CaM treatment was significantly less effective (90%). We conclude that a brief stress episode induces long-lasting modifications in activity-dependent adjustments of synaptic strength by selectively disrupting LTP processes. This might be regulated by a decreased Ca$^{2+}$-CaM sensitivity of AMPA receptors or differences in the CaMKII signaling in general. These data suggest an essential role for postsynaptic kinases in regulating the excitatory network activity in the CA3 area even long after a time-limited stressful episode.
Introduction

Synaptic long-term potentiation (LTP) and long-term depression (LTD) are thought to be crucial mechanisms for the information storage in the hippocampus (Bliss and Lømo, 1973; Martin et al., 2000). Particularly well documented is that short or long episodes of stress disrupt LTP at excitatory synapses of CA3-CA1 Schaffer-collaterals (SC) (Foy et al., 1989; Garcia et al., 1997; Xu et al., 1997; Baker and Kim, 2002; Alfarez et al., 2003) which is implicated in the diminished learning capacity in spatial memory tasks (reviewed by Kim and Diamond, 2002). Far less understood, however, is which particular molecular mechanisms are involved in the stress-LTP interactions. Identified signaling factors include: activation of the glucocorticoid receptor (GR) (Xu et al., 1998; Alfarez et al., 2003), increased cytokine leukine-1β concentrations (Murray and Lynch, 1998), N-methyl-D-aspartate (NMDA) receptor activation (Xu et al., 1997; Kamphuis et al., 2003), or alterations in corticotropin-releasing factor (Blank et al., 2003). These factors, however, still provide little information how downstream at the glutamate receptor complex specifically the LTP activity-dependent mechanisms are corrupted.

Very recent hypotheses on stress-induced suppression of LTP implicate a critical role for the multifunctional enzyme Ca²⁺-calmodulin-dependent kinase II (CaMKII) (Gerges et al., 2003). The α-subunit of CaMKII is highly distributed in cell bodies and dendrites of hippocampal pyramidal neurons and constitutes the main protein of the postsynaptic density. The enzyme is involved in essential processes such as release probability or intracellular protein regulation (Lisman et al., 2002; Hinds et al., 2003). Very recent studies showed that CaMKII protein levels are decreased in the CA1 area following four weeks of chronic psychosocial stress (Gerges et al., 2003) and that acute immobilization reduces the amount of autophosphorylated Ca²⁺/calmodulin-dependent protein kinase II (Blank et al., 2003).

Here, we aimed to investigate functional properties of the excitatory signaling of the commissural-associational (C/A) synapses three weeks after a brief stress, which leads to a complete abolishment of LTP (Kole et al., submitted). We postulated a role of a deficiency in CaMKII activation in establishing LTP at the CA3 recurrent synapses. Particularly the α-amino-3-hydroxy-5-methyl-4-isoxazole propionate (AMPA) ion channels contain cytosolic intracellular phosphorylation sites that consensus undergo activation and deactivation according to rapid turnover of available CaMKII and phosphatase levels, that enhances AMPA channel conductance (Yakel et al., 1995; Lee et al., 2000). We tested the functional activation of CaMKII by use of physiological whole-cell perfusion of Ca²⁺-calmodulin (Wang and Kelly, 1995).
Materials and Methods

Animals and animal experiments
Male Wistar rats (Harlan-Winkelmann, Borchen, Germany) were housed in pairs with food and water ad libitum. Animals were offered acclimatization for two weeks before onset of the experiment and kept under reversed light/dark conditions (lights on 2100-0900 h), room temperature of 21 ± 1°C. Resident rats (wild-type strain, Haren, University of Groningen, The Netherlands) were housed in pairs of one male and one sterilized female in large cages in a separate room. All manipulations were performed during the active period of the rats between 13.00 and 15.00 (during the dark phase) under additional dim red light (< 1 Lux). Animal experiments were conducted in accordance with the European Council Directive of November 24, 1986 (86/609/ECC) and approved by the Government of Lower Saxony, Germany. The minimum number of animals required to obtain consistent data was employed.

Stress procedure
Wistar rats (n = 15) were exposed to a psychosocial type of stress (social defeat), using a modified resident–intruder paradigm as described previously (Buwalda et al., 1999; 2001). In brief, the female wild-type rat was removed from the cage and the experimental male rat (Wistar) introduced into the resident’s cage. In typically less than a minute, the resident male wild-type rat attacked the Wistar, followed by submissive behaviors of the Wistar intruder. For the remaining hour, the intruder was enclosed in a small wire-mesh within the resident’s cage. As such, animals were protected from direct physical contact but remained in olfactory, visual and auditory contact with the resident male rat. Afterwards the intruder animal was housed singly, for 21 days until the electrophysiological experiments. Control Wistar animals (n = 13) were exposed for the same duration (1 h) to an open field and subsequently housed singly. Control and stressed animals were housed in the same room excluding non-specific environmental effects on CA3 cells (Lee and Kesner, 2002).

Slice preparation
At the 22nd day, 2 hr after lights-off (1100 h) animals were weighed, deeply anesthetized (mixture of 50 ketamine, 10 xylazine and 0.1 atropine mg/mL, i.p. injection) and tail blood collected in EDTA-containing tubes. Subsequently, animals were transcardially perfused with an ice-cold carbogenated (95% O2/5% CO2) sucrose-based artificial-cerebrospinal fluid (ACSF) solution (Aghajanian and Rasmussen, 1989) for 2 min followed by decapitation. Sucrose-ACSF contained (in mmol/L) 206 Sucrose, 1 MgCl2, 2.5 KCl, 2 MgSO4, 1.25 Na2H2PO4, 26 NaHCO3, 14 d-Glucose, 1 Kynurenic acid, 1.5 CaCl2, 1 L (+)-Ascorbic acid. The brain was rapidly removed and transverse 400 µm hippocampal slices were cut with a vibroslicer (Vibracut 2, FTB, Bensheim, Germany) in the sucrose-ACSF. Slices were stored in normal containing chamber at 33°C, for 1 h and afterwards stored at room temperature. ACSF for slice storing consisted of 125 NaCl, 2.5 KCl, 1.25 Na2HPO4, 2 MgSO4, 26 NaHCO3, 1.5 CaCl2, 1 L (+)-Ascorbic acid, 14 D(+)-Glucose, ~300 mOsm. (all chemicals from Merck, Darmstadt, Germany). Adrenals were removed and weighed with an analytical balance.
Corticosteroid assay

The EDTA-blood (serum) was analyzed for the unbound corticosterone with a corticosterone RIA kit protocol (Diagnostic System Laboratories Inc., Webster, TX, USA) as recommended by the manufacture.

Patch-clamp recording

Pyramidal shaped somata within CA3b were recorded using patch-clamp pipettes with 3-5 MΩ resistance and an intracellular patch solution composed of (in mmol/L) 130 KMeSO4, 20 KCl, 10 HEPES, 0.2 EGTA, 2 ATP-Mg2, 0.3 Tris-GTP, 10 phosphocreatine set at 280 mOsm and pH 7.2 with KOH. Somatic whole-cell recordings in either current- and voltage-clamp configuration were performed with an Axopatch 200B amplifier (Axon Instruments, Foster City, CA, USA), data collected by and ITC-16 and PULSE software (v. 8.54, HEKA, Lambrecht, Germany). Immediate after whole-cell configuration the initial access resistance measured typically 10 MΩ and was during the course of recording not allowed to exceed 16 MΩ. Therefore, the bridge voltage (current-clamp) or series resistance (voltage-clamp) were continuously monitored by applying small negative steps of -100 pA 150 ms preceding the EPSP or -10 mV, 50 ms preceding the EPSC (see Fig. 2). Tungsten electrodes (0.1 MΩ) were used to stimulate the C/A fibers in stratum radiatum 200 - 300 µm from stratum pyramidale, 100 µm lateral to prevent direct triggering of the cell. Basal stimulation and recording frequency was always 0.05 Hz. In LTP experiments evoked AMPA-R EPSCs were recorded at basal frequency for 8 min or longer and then responses potentiated by applying a mild low-frequency paradigm of 3 Hz for 3 min. All data were collected at 32 ± 2°C.

For LTP induction α-aminobutyric acid (GABA)A mediated activity was suppressed by bath application of 10 µM (-)-bicuculline methobromide and 50 µM Picrotoxin (Tocris, Bristol, UK). Extracellular Ca2+ was elevated to 2.5 mM. LTP was induced in current-clamp mode using 100 pulses at 100 Hz, 3 trains at 0.05 HZ, delivered at C/A-CA3 synaptic pathways. In addition a 200-400 pA depolarization pulse of 1 sec assured comparable number of spikes (control: 9.7 ± 0.5, stress: 11.2 ± 1.2, P > 0.8). Recordings were afterwards continued in voltage-clamp configuration. The first population of EPSCs, before tetanization, were defined as baseline (100%) and the following successive EPSC amplitudes normalized to this baseline period.

To perfuse the cell, patch pipettes (3-5 MΩ) were loaded with Ca2+ and calmodulin (Sigma-Aldrich, Steinheim, Germany) according to established protocols (Wang and Kelly, 1995, 2001). The stock solution was prepared with distilled water (100 times the final concentration) and diluted with the standard pipette solution before use. Ca2+-CaM consisted of Ca2+ and CaM at a molar ratio of 4:1 (Wang and Kelly, 1995, 2001). Whole-cell perfusion was performed by filling the tip with the patch solution alone, and then back-fill the pipette with the Ca2+ -containing solution. The pipette solution contained the standard pipette solution but without phosphocreatine to rule out non-specific phosphorylation actions. The first population of EPSCs (0-2 min) was defined as baseline of 100%. While the EPSCs with this internal solution alone exhibited a run-down of the amplitudes (~15% in 30 min) the amplitudes were corrected for the time-dependent shift in amplitude reduction using a linear correction factor. The highly specific CaMKII blocker KN-93 (Sigma-Aldrich, Steinheim,
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Germany) was applied at 10 µM in the bath solution, 15 min before start of the recording.

Data analysis

Data were explored by One-way ANOVA or paired/unpaired student’s t-test with the SPSS software (SPSS Inc., Chicago, Illinois). Significance level was set to \( P < 0.05 \). Graphical processing done with Origin v6.1 (Microcal Software, Northampton, USA). Data in the tables and graphs represent means ± SEM

Results

Long-lasting effects of stress on adrenocortical system

Male rats subjected to stress possessed significantly increased adrenal to body weight ratio (stress: 13.5 ± 0.4, \( n = 16 \), vs. control: 10.5 ± 0.5, \( n = 12 \), \( F_{1, 26} = 23.50, P < 0.001 \)). However, both absolute body weight and the body weight change during the experimental period were not affected (\( P > 0.5 \)). The serum corticosterone levels, sampled at 2 h after onset of the dark phase, i.e. shortly after the rising phase of the diurnal corticosterone peak were on average slightly lower (control 225.0 ± 31 ng/mL, \( n = 13 \), vs. stress: 175.5 ± 29 ng/mL, \( n = 13 \)) but did not reach significant difference (\( F_{1, 24} = 1.34, P < 0.26 \)). Therefore, and in accordance to previous studies the social defeat experience three weeks earlier mildly but persistently affect HPA-axis activity (Buwalda et al., 1999).

Stress affects baseline excitatory synaptic transmission

Somatic whole-cell recordings were obtained from a total of 84 CA3 pyramidal cells. There were no differences in input resistance \( R_N \) (stress: 87.0 ± 4.6 MΩ, \( n = 42 \), control: 92.9 ± 3.6, \( n = 35 \)). The glutamate-receptor mediated synaptic transmission was examined at the C/A synapses by evoking composite EPSCs at ~300 µm from the soma in the stratum radiatum (Fig 1. in Chapter 5). As reported previously (Kole et al., 2002) EPSCs could be completely blocked by CNQX (20 µM 97.3 ± 0.03 %, \( n = 17 \)) indicating ample specificity for AMPA-receptor mediated currents. Upon inspection of the EPSC onset latency we observed that stress decreased the duration by ~100 µs (1.40 ± 0.06, \( n = 41 \) ms vs. control: 1.53 ± 0.04 ms, \( n = 35 \), \( P < 0.05 \)). In the same line of a facilitated EPSC onset, also the input-output curve of the dual-component EPSCs showed a significant facilitation in the slope (Fig 1A-B). No difference was found in EPSC decay time (~12 ms for both groups, not shown).
Stress-induced block of LTP

Fig. 1. AMPA-receptor mediated synaptic transmission long-term after stress. A. Stress increased the evoked stimulus-dependent EPSCs. Amplitudes were significant increased at stimulus intensities of 8 and 10 µA ($P < 0.01$). The slope of the linear part of the curve showed coefficient of $164.5 \pm 7.4$, $n = 9$ and in control $66.0 \pm 4.6$, $n = 6$ ($P < 0.01$). B. Examples of the AMPA currents elicited from -80 mV at various stimulus intensities. Currents could be fully blocked by the AMPA-R antagonist CNQX (20 µM). Data are shown as mean ± SEM.

Stress selective impairs LTP

After adjustment of the EPSC response to peak amplitudes of ~250 pA, long-lasting recordings were performed (80 min) to monitor activity-dependent plasticity in a sequel of three phases. First, high frequency tetanus (100 Hz) was applied in current-clamp mode to induce LTP. Switching back in voltage-clamp we observed that in some cells the EPSC developed into an oscillation pattern (Fig. 2A-C). Such a recruitment of multiple synapses is consistent with previous reports (Bains et al., 1999, Staley et al., 2000). Consistent with the inability to induce LTP in stress, no oscillation was observed in slices from stressed animals. In control cells without oscillatory bursting the onset the peak EPSC amplitudes at 20-30 min interval reached 150% increase (see Fig. 3A-C). No increase was detected in slices from stressed animals. There were no changes in the decay kinetics.

After 30 min we applied LTD using 15 min 1 Hz stimulation. Both in control recordings and those from stressed animals the EPSCs significantly decreased in peak amplitudes. In slices from stressed animals the LTD induction reduced EPSC amplitudes compared to baseline to 70% of EPSC peak amplitudes. To evaluate whether the previous activity might influence the possibility to establish LTP we tested after LTD establishment whether synapses could be ‘de-de-potentiated’ (Lee et al., 2000). In control animals this produced a remarkable increase in EPSC amplitude of 190% ($P < 0.001$) and also EPSC amplitudes from stressed animals responded by increasing the EPSCs towards their baseline amplitudes. This shows that only the initial early LTP is disrupted long after stress.
Fig. 2. LTP-induction at C/A synapses is followed by an oscillatory pattern of two distinct firing modes. A. After a 100-Hz tetanus at the C/A axons, the compound EPSCs grew steadily and finally resulted in an oscillation (4/15 cells). These were characterized by mono-synaptic currents alternated with large unclamped inward currents, existing of multiple summating EPSCs (arrows). B. During such recurrent polysynaptic burst response, action potentials were summed on a large envelope EPSP. C. Polysynaptic bursts are observed in an oscillatory pattern, within the delta frequency bandwidth (0.3 ± 0.02 Hz, n = 4).
Stress-induced block of LTP

Deficient response of AMPA-Rs to Ca^{2+}-calmodulin

In view of the existing models on LTP in naïve synapses pointing to a critical role of selective protein phosphorylation (Lee et al., 2000) and the recently suggested effects of stress at CaMKII (Gerges et al., 2003) we set out to physiologically test the activation of this protein by intracellular perfusion of its agonist (Wang and Kelly, 1995; 2001). We tested the role of CaMKII at CA3 collateral synapses by perfusing control slices with the specific membrane-permeable CaMKII antagonist KN-93 (1 µmol/L, Sumi et al., 1991). There was a significant block in the early phase (14 min, \( P < 0.001 \)) but not the late phase of LTP (\( > 15 \) min, \( P > 0.5 \)). Perhaps activates the high-frequency stimulation mossy fiber LTP transsynaptically, which does not require CaMKII (Ito et al., 1991; Salin et al., 1996). Alternatively, other proteins (e.g. PKC) might underlie late mechanisms of LTP at the C/A synapses.

We reasoned that when the CaMKII cascade is changed after stressful experience the activation of CaMKII might be impaired. Therefore, we directly monitored EPSCs during cytosolic whole-cell perfusion with Ca^{2+}-CaM (Wang and Kelly, 1995, 2001). In control cells the application of Ca^{2+}-CaM led to a time-dependent increase of the EPSC peak amplitudes (Fig. 4A-C). After two minutes the amplitudes started to increase progressively, reaching \(~190\%\) at 20 min. The upregulation in control animals was CaMKII-dependent since application of KN-93 (1 µmol/L) 15 min before recording significantly suppressed the effects of Ca^{2+}-CaM (15 min, 102 ± 2%, \( n = 4 \)). The CA3 C/A synapses of stressed animals were significantly less susceptible for
stimulation with Ca\textsuperscript{2+}-CaM and did not respond by an increase in the AMPA currents. At 20 min, only \sim 90\% of the baseline EPSC amplitude was reached (\( P < 0.001 \), Fig 4A-D).

**Fig. 4.** Differential AMPA receptor responses to Ca\textsuperscript{2+}-CaM application. A-B. Ca\textsuperscript{2+}-calmodulin was added to the pipette solution in a stochastic ratio of 40 \( \mu \)M:10 \( \mu \)M, similarly in CA3 pyramidal cells from control and stressed animals. C. Ca\textsuperscript{2+}-CaM produced a slow developing increase in the compound EPSCs, reaching peak amplitudes of 193 \pm 16 \% from baseline at 20 min (\( n = 8 \), \( P < 0.01 \)). Recordings from stressed animals showed that at 15 min only 103 \pm 17\% of baseline EPSC amplitude was reached (unpaired t-test, compared to control, \( P < 0.03 \)) and at 20 min even resulted in decreased amplitudes of 86 \pm 17\% (compared to control, \( P < 0.001 \)). D. The distribution of EPSC amplitudes shows a slightly bi-directional effect of Ca\textsuperscript{2+}-CaM at AMPA responses, as a depression of EPSCs in stressed animals, and increase in control animals.

**Discussion**

The present investigation demonstrates that a short-lasting stress exposure produces long-lasting opposite effects on AMPA-R-mediated C/A synaptic properties. It increases the basal synaptic strength but blocks the probability for induction of LTP. Consistent with this, the LTP-dependent bursting could also not be elicited. Seeking for a possible mechanism that prevents this experience-dependent blockade in synaptic plasticity we obtained functional evidence that a reduced response of AMPA-Rs towards Ca\textsuperscript{2+}-calmodulin might be involved.
**Increased excitatory baseline transmission**

Recently, Karst and Joëls (2003) showed that following three weeks of repetitive stress the AMPA-R responses in hippocampal dentate granule cells were elevated, but only when external high corticosterone levels were applied. In the present study, we observed similar changes but investigated in animals in the onset of their activity phase, i.e., when corticosterone is endogenously elevated. The corticosterone levels appeared not to be changed by the stress exposure, which is consistent with previous studies (Buwalda et al., 1999; 2001). How the AMPA-R-mediated currents at the CA3 C/A synapses (Fig. 1) are increased is not clear. The conductance change might be caused by an elevation in mRNA for the AMPA channel subtype GluR1 after stress (Schwendt and Jezova, 2002) although no information is available on channel expression long after stress episodes. In addition to the increased current amplitudes, the facilitation of the EPSC onset latency might depend on the reduced apical dendritic geometry of the CA3 pyramidal neurons (Kole et al., submitted). Irrespective of the precise source of increased AMPA currents it is clear that even a short stressor leads to long-lasting modification of basal excitatory signaling at the CA3 pyramidal cell. Despite this facilitation, the activity-dependent regulation of the AMPA channels appeared to be blocked.

The currently best-supported scenario for LTP induction involves phosphorylation of AMPA receptors by CaMKII (Lisman et al., 2002). During NMDA-mediated Ca\(^{2+}\) influx at naïve synapses the enzyme CaMKII is phosphorylated, and subsequently undergoes autophosphorylation at its Thr\(^{286}\) site (Barria et al., 1997, Lee et al., 2000). This biochemical signal leads to enhanced conductance of GluR1 subunit of the AMPA receptor by the phosphorylation of its Ser\(^{831}\) site (Lledo et al., 1995; Lisman et al., 2002) and there is evidence that this shares mechanisms with the tetanization-induced LTP. We show here that, identical to the CA1 SC synapses, the C/A synapses also utilizes CaMKII signaling, since we could block both the Ca\(^{2+}\)-calmodulin activation of AMPA EPSCs and the induction of the early phase of LTP by KN-93, a known antagonist for the autophosphorylation of the α- and β-CaMKII sites as well as the calmodulin binding at the protein (Sumi et al., 1991). While the present experiments demonstrate that there is a disruption in functional activation of AMPA-Rs by the CaMKII-dependent mechanisms after stress (Fig. 4), other forms of activity-dependent plasticity were still intact. Both LTD and de-depotentiation differ also greatly from LTP in their biochemical correlate by inducing dephosphorylation of the AMPA-R sites via protein phosphatases or PKA, thus act independently from the CaMKII enzyme (Lee et al., 2000).

**Implications**

The ability of CA3 C/A synapses to induce polysynaptic bursting (Fig. 2) together with its topographical wiring, render the CA3 recurrent collaterals strongly involved in feed-forward excitation. This facilitates bi-directional modification of synaptic strength that depends on the synchronized activity between network-connected populations of neurons (Zalutsky and Nicoll, 1990; Debanne et al., 1998; Bains et al., 1999; Staley et al., 2000; Nakazawa et al., 2002). Network transitions from quiescence to fully active occur when recurrent synapses are sufficiently synchronized to initiate action potential bursts, which on its turn lead to opening of NMDA receptors (Bains et
al., 1999, Staley et al., 2000). The stress-impaired blockade of LTP would thus predict a long-lasting disruption of CA3-dependent mnemonic tasks that rely on the changes in synaptic strength. CA3 C/A synapses are recognized to be involved in associative or fast learning about single events (Rolls, 1996; Nakazawa et al., 2002; 2003).

Whereas some types of acute stress impair spatial and recognition memory and hippocampal LTP transiently (Baker and Kim, 2002; Garcia et al., 1997) the exposure to social defeat, followed by single housing, can produce long-lasting effects on social recognition memory or anxiety-related behaviors (Koolhaas et al., 1997; von Frijtag et al., 2000). This might be compared to the effects of traumatic stressful events that, via elevated epinephrine and glucocorticoids, impair the recall or acquisition of non-emotional memories (spatial and contextual information) but contrastingly lead to the consolidation of memories with emotional content, consistently shown in laboratory animals and humans (Buchanan and Lovallo, 2001; Roozendaal, 2002). This raises the question: are C/A synapses after stress merely saturated by a history of learning-induced LTP? Kim et al. (1996) proposed that baseline changes in hippocampal excitatory transmission after stress shift the modification range of synaptic plasticity towards LTD. Our empirical observations partially support this hypothesis. Although basal excitatory transmission was potentiated, the dynamic range is nevertheless suppressed compared to control animals. This does not occur during learning of motor tasks, where LTP is also lower but the synaptic modification range is maintained (Rioulte-Pedotti et al., 2000).

Where is the CaMKII enzyme altered? Currently, we can only speculate. Recent studies report a reduced availability of total or phosphorylated fraction of the CaMKII after stress (Gerges et al., 2003; Blank et al., 2003). Since we recorded synaptic responses three weeks after stress, it cannot be excluded that the enzyme is changed in another way then being reduced. The multifunctional enzyme CaMKII is involved in the transitions of branch dynamics of dendrites and axon during maturation of the brain (Cline, 1999; Lisman et al., 2002).

Since we have recently shown that CA3 pyramidal dendrites exhibit extraordinary reorganization following three weeks after two social defeats (Kole et al., submitted) it is tempting to propose that a reduction in postsynaptic CaMKII might be involved in the observed increase in branch dynamics, allowing both branch addition as well as elimination. These processes require the simultaneous reduction of CaMKII and microtubuli-associated proteins (Wu and Cline, 1998; Vaillant et al., 2002). In this view, changes in CaMKII activity after stress might act as a switch between synaptic Hebbian plasticity and structural plasticity, of which the latter provides a larger capacity for storage of information (Poirazi and Mel, 2001).