

# Syrian hamster neuroplasticity mechanisms fail as temperature declines to 15 °C, but histaminergic neuromodulation persists

Jock S. Hamilton<sup>1</sup> · Sat M. Chau<sup>1</sup> · Kevin J. Malins<sup>1</sup> · Giancarlo G. Ibanez<sup>1</sup> · John M. Horowitz<sup>1</sup> · Barbara A. Horowitz<sup>1</sup> 

Received: 28 August 2016 / Revised: 27 December 2016 / Accepted: 26 February 2017 / Published online: 9 April 2017  
© Springer-Verlag Berlin Heidelberg 2017

**Abstract** Previous research suggests that hippocampal neurons in mammalian hibernators shift their major function from memory formation at euthermic brain temperatures ( $T_b = \sim 37^\circ\text{C}$ ) to modulation of hibernation bout duration as  $T_b$  decreases. This role of hippocampal neurons during torpor is based in part on in vivo studies showing that histamine (HA) infused into ground squirrel hippocampi lengthened torpor bouts by  $\sim 50\%$ . However, it was unclear if HA acted *directly* on hippocampal neurons or on downstream brain regions via HA spillover into lateral ventricles. To clarify this, we used hippocampal slices to determine if HA would modulate pyramidal neurons at low levels of synaptic activity (as occurs in torpor). We tested the hypotheses that although LTP (a neuroplasticity mechanism) could not be generated at low temperatures, HA (via H2 receptors) would increase population spike amplitudes (PSAs) of Syrian hamster CA1 pyramidal neurons at low stimulation voltages and low temperatures. PSAs were recorded following Schaffer collateral stimulation from subthreshold levels to a maximum response plateau. We found that tetanus evoked LTP at 35 °C but not 15 °C; and at temperatures from 30 to 15 °C, HA significantly enhanced PSA at near threshold levels in slices from non-hibernating hamsters housed in “summer-like” or “winter-like”

conditions and from hibernating hamsters. Cimetidine (H2 antagonist) blocked HA-mediated PSA increases in 8 of 8 slices; pyrilamine (H1 antagonist) had no effect in 7 of 8 slices. These results support our hypotheses and show that HA can directly enhance pyramidal neuron excitability via H2 receptors and thus may prolong torpor bouts.

**Keywords** Hibernation · Torpor · Histamine · CA1 pyramidal cells · Ascending arousal system

## Abbreviations

|         |  |
|---------|--|
| AAS     | Ascending arousal system   |
| aCSF    | Artificial cerebral spinal fluid   |
| CaMKII  | Calmodulin-dependent protein kinase II   |
| EEG     | Electroencephalogram   |
| fEPSP   | Field excitatory post-synaptic potential   |
| HA      | Histamine  |
| HH      | Hamster in hibernation (torpor) at $6 \pm 2^\circ\text{C}$ and short photoperiod (8:16 LD) |
| HS-aCSF | High-sucrose artificial cerebral spinal fluid  |
| LTD     | Long-term depotentiation   |
| LTP     | Long-term potentiation   |
| NMDA    | N-methyl-D-aspartate   |
| OGD     | Oxygen glucose deprivation   |
| PS      | Population spike   |
| PSA     | Population spike amplitude   |
| SH      | Hamster housed at $22 \pm 2^\circ\text{C}$ and long photoperiod (14:10 LD)                 |
| $T_b$   | Brain and core temperature   |
| $T_s$   | Temperature of a hippocampal slice in the recording chamber                                |
| WH      | Non-hibernating hamster housed at $6 \pm 2^\circ\text{C}$ and short photoperiod (8:16 LD)  |

Communicated by F. Breukelen.

This manuscript is part of the special issue Hibernation—Guest Editors: Frank van Breukelen and Jenifer C. Utz.

✉ Barbara A. Horowitz  
bahorwitz@ucdavis.edu

<sup>1</sup> Department of Neurobiology, Physiology, and Behavior, College of Biological Sciences, University of California at Davis, One Shields Ave, Davis, CA 95616, USA

## Introduction

Torpor is characterized by a dramatic reduction in systemic metabolism (Carey et al. 2003) and a profound decrease in neural activity throughout the central nervous system (Kilduff et al. 1982, 1990). While the neocortex is essentially silenced in torpor, deeper subcortical regions continue to function, albeit their electrical activity is greatly reduced (Heller 1979; Mihailovic 1972; Zimmer and Milson 2001). In EEG studies, two subcortical brain regions, the brainstem reticular formation and the hippocampus, were identified as contributing to the neural control of hibernation (Mihailovic 1972; South et al. 1972). Recordings in the brainstem reticular formation indicated that neural activity in this region was inhibited when the animal was in hibernation. Based on this finding, the role of the brainstem reticular formation in hibernating species was extended to include not only regulation of sleep and waking [as in non-hibernating species (Moruzzi and Magoun 1949)], but also regulation of hibernation bouts. In addition, EEG recordings identified the hippocampus as a major region sending signals to inhibit the brainstem reticular formation (Mihailovic 1972; South et al. 1972). Citing additional microinjection studies, two reviews (Beckman and Stanton 1982; Heller 1979) include hippocampal signaling to the brainstem as a core element in their diagrams of brain regions controlling hibernation.

The finding that histamine (HA) receptors are present in greater density in hippocampi of hibernating than non-hibernating golden-mantled ground squirrels (*Spermophilus lateralis*) (Sallmen et al. 2003a) suggested that HA modulates hippocampal activity over hibernation bouts. [Histaminergic neurons have their cell bodies in the tuberomammillary nucleus (located within the posterior hypothalamus) and project not only to the hippocampus, but to numerous other areas including the cortex, thalamus, and posterior pituitary where they broadly modulate brain activity (Diewald et al. 1997; Haas and Panula 2003).] Supporting the possibility that HA takes on an additional modulatory role in hibernating species is the increase in bout duration following bilateral infusion of HA into the hippocampi of hibernating ground squirrels (Sallmen et al. 2003b).

In their *in vivo* study showing that HA prolonged bout duration, Sallmen et al. (2003b) proposed that it was the hippocampal CA1 pyramidal neurons that sent the signals inhibiting brainstem nuclei and lengthening the duration of torpor. This same class of neurons has been extensively studied in non-hibernating mammals because at brain temperatures of ~35 °C they exhibit cellular neuroplasticity mechanisms supporting memory formation. In fact, CA1 pyramidal cells in non-hibernating mammals have become exemplars for detailed study of cellular pathways associated

with long-term potentiation (LTP) and long-term depotentiation (LTD) (Herring and Nicoll 2016; Mayford et al. 2012). Brain slice studies using Syrian hamsters, a facultative hibernating mammalian species, have shown that Syrian hamster CA1 pyramidal neurons (at slice temperatures between 30 and ~20 °C) also have cellular pathways supporting a variety of plasticity mechanisms including LTP, depotentiation, and LTD (Bronson et al. 2006; Arant et al. 2011; Krelstein et al. 1990), and thus have the same cellular plasticity mechanisms proposed to form memories as do non-hibernating mammalian species.

Combining HA and neural plasticity studies on hippocampal neurons led to the unifying proposal that CA1 pyramidal neurons in Syrian hamsters have cellular properties supporting a shift in hippocampal function from memory formation to modulation of hibernation bout duration as brain temperature declined (Arant et al. 2011). Experiments in the present study are designed to further test this proposal. Our first objective was to determine how LTP studies are related to recent behavioral and synaptic data on memory formation in Syrian hamsters (Bullmann et al. 2016). Precisely how these recent studies fit with those focused on neuroplasticity mechanisms in hippocampal neurons is limited by incomplete LTP data on hibernating species adapted to environments simulating seasonal variations. To address this limit, we determined the range of temperatures over which LTP could be generated in slices from non-hibernating hamsters housed in a summer-like environment (SH slices), slices from hamsters housed in a winter-like environment but have not entered torpor (WH slices), and slices from hamsters housed in a winter-like environment that are in torpor (HH slices). A second objective was to determine if at 15 °C, pyramidal neurons continue to generate action potentials (a necessary property if they play a role in extending bout duration).

Our third objective was to test the proposal that HA *directly* modulates CA1 pyramidal neural excitability as temperature decreases and neural activity is reduced, and if so, to determine if the histamine effect is mediated via H1 or H2 receptors. There appears to be only one study showing that HA enhanced hippocampal pyramidal cell neural excitability in any hibernating species (i.e., increased the ability of the neuron to receive and respond to excitatory synaptic signals and generate action potentials). This *in vitro* study (Nikmanesh et al. 1996) utilized relatively strong stimulation and temperatures above 22 °C. Notably, a species difference between rats and Turkish hamsters was identified—i.e., HA increased pyramidal cell population spike amplitude (PSA) only in the hamsters (Nikmanesh et al. 1996). However, the limited data that hamster pyramidal cells are *directly* modulated by HA at low temperatures and low stimulation intensities leave open an alternative interpretation of results obtained in the *in vivo* hamster

study (Sallmen et al. 2003b). That is, because insertion of the infusion cannula into the hamster hippocampus required passage through the lateral ventricle, some HA could have leaked back into these ventricles and been swept along with the ventricular fluid to caudal brain regions. Such potential spillover raises the possibility that the observed increase in bout length resulted from excitation of targets downstream of the hippocampus with HA having minimal, if any, effect on hippocampal neuron excitability.

The present study deals with the objectives discussed above by testing the following hypotheses: (1) although LTP can be generated at 35 °C, it cannot be generated in CA1 pyramidal neurons at low slice temperatures (e.g., at ~15 °C) in any of the three hamster groups (SH, WH, HH); (2) at low temperatures, CA1 pyramidal cells in all three hamster groups can still generate action potentials even if they cannot generate LTP; (3) HA modulation of CA1 pyramidal neurons significantly increases CA1 pyramidal cell PSAs near threshold levels of stimulation at 30, 20, and 15 °C in all three hamster groups, and it does so via H2 receptors.

## Materials and methods

All experimental protocols used in this study were approved by the UC Davis Institutional Animal Care and Use Committee in compliance with the Animal Welfare Act and in accordance with Public Health Service Policy on Humane Care and Use of Laboratory Animals.

### Animals and housing

Syrian hamsters (*Mesocricetus auratus*) from our colony were bred from animals selected over generations to more readily hibernate when exposed to cold and short photoperiod (the breeders were originally from the colony of Dr. John R. Willis). One group of adults ( $9.1 \pm 0.4$  months) from our colony was housed at  $22 \pm 2$  °C in long photoperiod, 14:10 light–dark (LD), to simulate summer-like conditions, a group referred to as SH. Under these summer-like conditions, the hamsters did not enter torpor and had core body temperatures that averaged  $37.2 \pm 0.2$  °C at time of killing. A subgroup of colony hamsters was transferred to a hibernaculum (8:16 LD; ambient temperature of  $6 \pm 2$  °C). Those that did not enter hibernation within their first 3 weeks in the hibernaculum (and had a core temperature of ~37 °C) were deemed acclimated to the winter-like environment, a group denoted WH. These hamsters had lower ratios of gonadal weight/body mass than did those in the SH group ( $P < 0.05$ ), consistent with cold acclimation and preparation for entering hibernation. The HH group was comprised of hamsters that were killed while in torpor,

having entered torpor at least 24 h prior to killing (determined by the lack of movement of a small piece of folded tape placed on top of their back). These hamsters continued to remain tucked in a ball position when removed from their cage and had low core and brain temperatures at time of death (within ~2 °C of the hibernaculum temperature). There were 26 hamsters in the SH group, 11 hamsters in the WH group, and 16 hamsters in the HH group.

### Brain slice preparation and perfusion

Within 30 s following killing of each hamster, their brain was removed and placed into chilled (4 °C) high-sucrose, low-NaCl artificial cerebral spinal fluid (HS-aCSF), which was oxygenated with 95% O<sub>2</sub>/5% CO<sub>2</sub> and contained (mM final concentration): 62 NaCl, 2.5 KCl, 2 CaCl<sub>2</sub>, 26 NaH<sub>2</sub>CO<sub>3</sub>, 1.5 NaH<sub>2</sub>PO<sub>4</sub>, 2 MgSO<sub>4</sub>, 124 sucrose, 5 dextrose. [Lowering brain tissue to ~4 °C before and during sectioning minimizes hypoxia-induced damage in hibernating species as it does in non-hibernating species (Pakhotin et al. 1990).] Hippocampi were sectioned into 400 μm slices using a McIlwain tissue chopper, and slices were immediately placed in HS-aCSF at  $30 \pm 1$  °C and incubated for 30 min. At the end of the incubation period, HS-aCSF was replaced by standard aCSF containing (mM final concentration): 124 NaCl, 2.5 KCl, 2.0 CaCl<sub>2</sub>, 26 NaHCO<sub>3</sub>, 1.5 NaH<sub>2</sub>PO<sub>4</sub>, 1.5 MgSO<sub>4</sub>, and 5 dextrose and gassed with 95% O<sub>2</sub>/5% CO<sub>2</sub> at  $30 \pm 1$  °C. Each slice was incubated in this standard aCSF for at least 30 min before transfer to the recording chamber [The warm incubation temperature of 30 °C for a minimum of 60 min was chosen because under these conditions, population spike and unit activity in slices from both hibernating and non-hibernating ground squirrels have been shown to have recovered from the trauma of slicing (Pakhotin et al. 1990).]

Hippocampal slices were transferred to a recording chamber (2 ml volume; 2 ml/min flow rate of standard aCSF). By adjusting the temperature of a circulating water bath in thermal contact with the recording chamber, the temperature of the recording chamber was set to a specific value (either 35, 30, 25, 20, or 15 °C) and then held within  $\pm 0.5$  °C of the selected temperature while a slice was in the recording chamber and evoked responses were obtained. Over this range of temperatures, data acquired using the in vitro slice preparation in hibernating species approximate in vivo conditions (Igelmund 1995). As  $T_S$  increases above 35 °C and cells increase their metabolic rates, hypoxia becomes a factor that affects slice viability because oxygen reaches neurons near the center of the slice only via diffusion. Therefore, hippocampal slice protocols commonly set recording chamber temperature no higher than 35 °C (Lein et al. 2011) and interpret data in LTP experiments to be applicable to the euthermic ( $T_b =$

~37°C) animal. With  $T_S$  below 35°C and with an adequate flow of oxygenated solution and a chamber design promoting laminar flow, neurons maintain physiologically realistic patterns of network activity for many hours (Hajos et al. 2009).

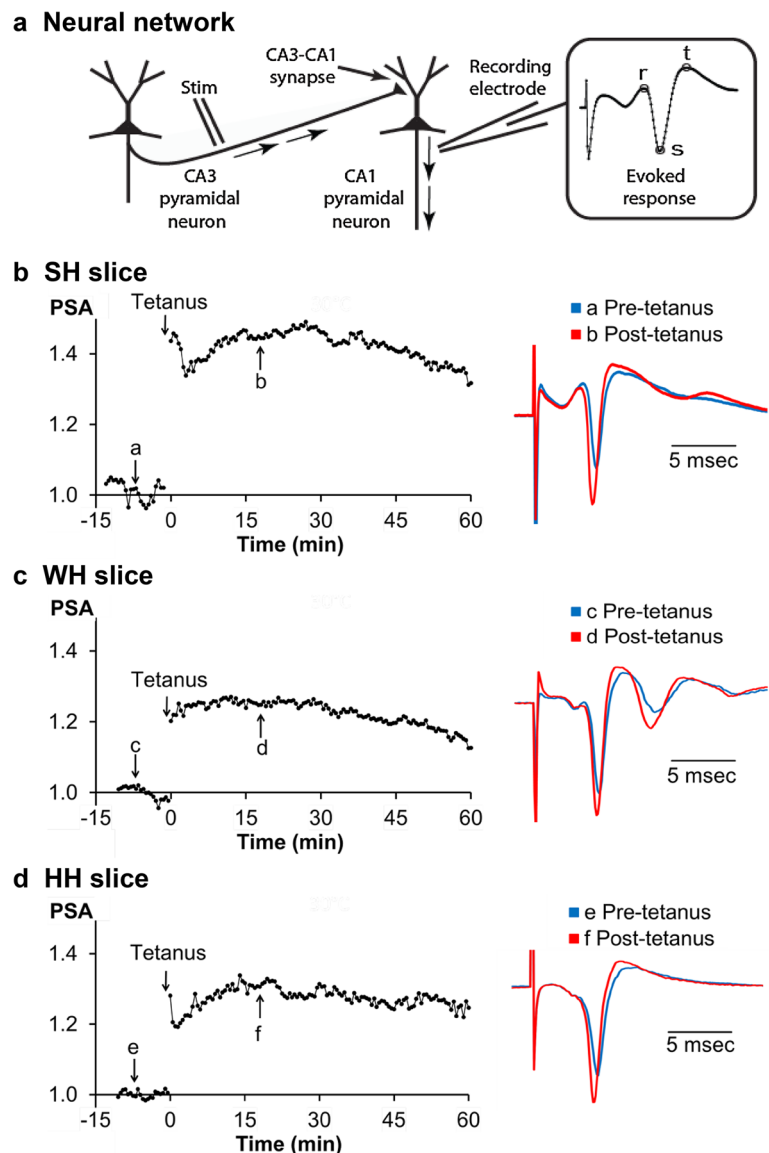
## Electrophysiology

Hippocampal slices were transferred to a recording chamber, and stimulating and recording electrodes were placed on the slice to excite fibers to CA1 pyramidal neurons and record their evoked responses (diagrammed in Fig. 1a). Specifically, bipolar tungsten electrodes were inserted into the stratum radiatum layer of the CA3 region to stimulate the Schaffer collateral fiber pathway (Fig. 1a). Electrical stimulation (100  $\mu$ sec duration pulses delivered at 1 Hz) evoked responses that were recorded with extracellular

glass pipettes filled with 3 M NaCl (resistance 2–20 megohms) placed in the stratum pyramidale of the CA1 region.

A population spike (PS) in the evoked response was identified as a large negative potential (e.g., labeled “s” in Fig. 1a) between two positive local maxima potentials (e.g., “r” and “t” in Fig. 1a). The PS, a field potential, reflected the summed response of action potentials from a population of CA1 pyramidal cells synchronously excited by single-shock Schaffer collateral stimulation (Andersen et al. 1971). PSA was measured as the difference between the potential at site “s” and the average of the two maxima voltages at “r” and “t” [ $PSA = \frac{1}{2}(r+t) - s$ ]. For each slice, threshold and maximal PSA values were determined after the evoked response had stabilized (PSA variation less than 10% over a 10 min interval). The greatest voltage that produced no detectable PSA in the averaged evoked response to ten successive shocks was taken as being just below

**Fig. 1** PS enhancement is sustained following tetanus at 35°C in CA1 pyramidal neurons. **a** Electrode configuration for exciting CA1 pyramidal cells and recording their evoked responses. A PS is shown on the evoked response trace as a downward “spike” labeled “r s t”. **b** PSA enhancement in a slice from a hamster housed in a summer-like environment (an SH slice). The evoked response (prior to tetanus) at the arrow labeled **a** on the plot of PSA versus time is shown on the insert on the right (*blue curve*). A second evoked response (following tetanus) at arrow **b** is also shown on the insert on the right (*red curve*). **c** PSA enhancement in a slice from a non-hibernating hamster housed in a winter-like environment (a WH slice). The evoked responses at arrows **c** and **d** are plotted on the insert on the right (*blue and red curves* respectively). **d** PSA enhancement in a slice from a hibernating hamster housed in a winter-like environment (a HH slice). The evoked responses at arrows **e** and **f** are plotted on the right as *blue and red curves*, respectively. The PSAs recorded over the last 10 min of the control period were averaged and normalized to 1.0. Thus, following tetanus (at arrow **f**), the PSA had increased to ~1.3, an increase of 30% above control



threshold. In response to a stepwise increase in stimulus intensity, PSA increased steadily from threshold to a maximal response ( $V_{max}$ ). Following determination of threshold (VT) and  $V_{max}$ , various protocols were used in LTP and HA experiments as described below.

### Protocol for LTP generation

As in previous slice studies on hibernating species (Krelstein et al. 1990; Pakhotin et al. 1990), our criterion for concluding that neurons in SH, WH, or HH slices at a fixed temperature could generate LTP was that a tetanizing protocol lasting <2 min rapidly evoked an increase in PSA that was sustained for at least 30 min. Throughout the slice experiment (a pre-tetanus control period, a tetanizing stimulus, and a post-tetanus period), PSs were recorded in response to shocks delivered at half-max stimulus intensity (i.e., half the voltage that elicited the maximal PS). In the 15-min control period, responses to single shock stimulation at a rate of 1 shock per second were recorded. This was followed by a tetanizing stimulus involving five pulse trains. Each train consisted of 100 shocks to Schaffer collaterals over a 1-s interval (i.e., at a rate of 100 shocks per second). Each individual shock had a duration of 100  $\mu$ sec and a half-max voltage stimulus intensity. The time between pulse trains was 15 s. Following this tetanizing stimulus, PSAs were recorded in response to single shock stimulation at the rate of one shock per second for at least 30 min. Our criterion for LTP generation required the following sequence of events: PSAs rapidly increased to a higher amplitude within a few minutes following the high-frequency trains, they stabilized at a higher value, and they remained significantly above the baseline response for at least 30 min (often recordings were extended to 60 min as in Fig. 1b).

### Protocol for HA input–output curves (plots of PSA versus stimulus intensity)

Input–Output (IO) curves were constructed during three successive time periods: (a) just before the end of the initial 10-min control period (with the slice in standard aCSF); (b) 5 min into the 10-min treatment period with HA-aCSF (10  $\mu$ M HA added to the standard aCSF); (c) at the end of a 30-min washout period (with the slice in standard aCSF). Each IO curve was constructed as stimulus intensity was increased stepwise from subthreshold to near maximum levels. That is, following each step increase in stimulus intensity to a higher level, PSA was calculated as the average of 10 responses evoked by Schaffer collateral stimulation (delivered at a rate of one pulse per second) with stimulus intensity held constant at the higher level. [Testing the effects of histamine by adding HA to the bath solution and

measuring CA1 pyramidal neuron responses was necessary because the diffuse distribution of histaminergic fibers in the hippocampus (Diewald et al. 1997) from neurons with their cell bodies in the tuberomamillary nuclei do not form a discrete bundle in the hippocampus.]

### Evaluation of histaminergic receptor type and drugs

Histamine obtained from Sigma (Sigma-Aldrich Corporation, St. Louis, MO) was added to the solution (10  $\mu$ M final concentration) being delivered to the slice chamber for the 15-min treatment period. The HA receptor blockade experiments utilized separate groups of slices. PSAs at approximately 50–80% of saturation were used to analyze the effect of 20  $\mu$ M pyrilamine, an H1 receptor antagonist, and 20  $\mu$ M cimetidine, an H2 antagonist (both obtained from Sigma–Aldrich) on neural circuit excitation mediated by 10  $\mu$ M HA. The use of these H1 and H2 antagonists in hippocampal slices is reviewed in Panula et al. (2015).

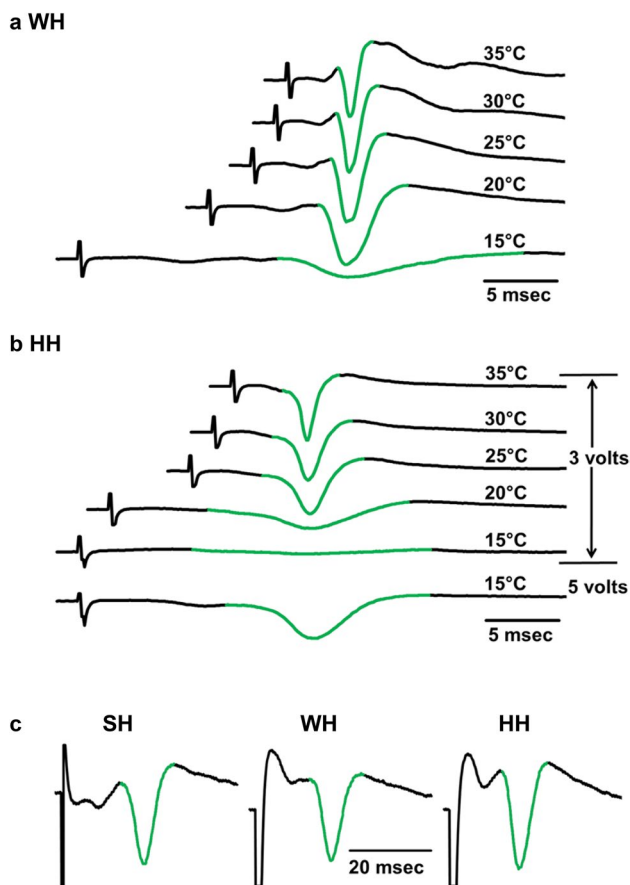
### Data analysis

At each IO curve voltage setting, we used a paired, two-tailed Student's *t* test to compare responses obtained during the control and the HA perfusion periods. For antagonist experiments, an unpaired Student's *t* test was used. Significance was set at  $P < 0.05$  in both analyses.

## Results

### LTP generation (hypotheses 1 and 2)

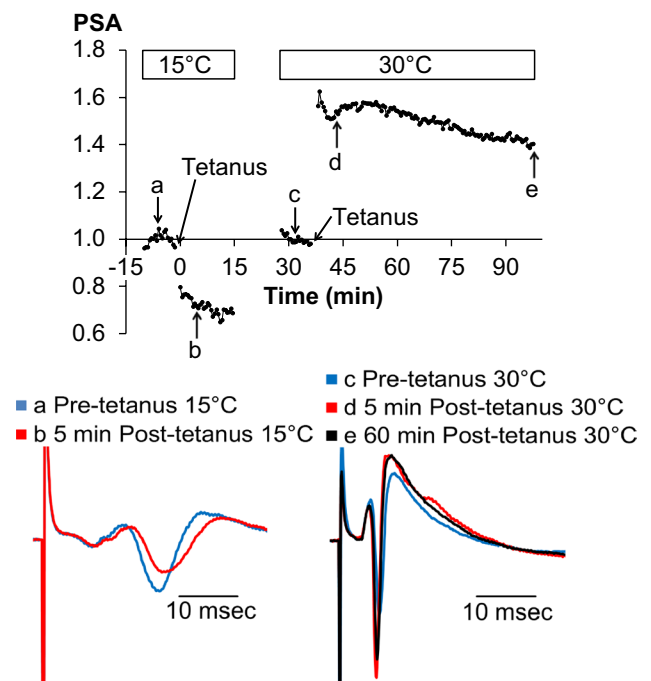
Several “gaps” in the description of the temperature dependence of LTP generation in CA1 pyramidal cells were addressed in this study. The first involved extension of previous research showing that LTP could be generated in the temperature range of  $T_s = \sim 22^\circ\text{C}$  to  $T_s = 30^\circ\text{C}$  (see discussion). As illustrated in Fig. 1, we determined that LTP could, in fact, be generated at  $T_s = 35^\circ\text{C}$  in slices from all three animal groups (SH, WH, HH). These increases in PSA lasted at least 30 min, and in slices where recording was continued for an additional 30 min, they were maintained over the entire hour. We also evaluated PS amplitude and width as  $T_s$  declined to  $15^\circ\text{C}$ . As shown in Fig. 2, during such a decline, PSA significantly decreased. In addition, the width of PSs increased, indicating a loss in synchronized activation and firing of the population of pyramidal cells contributing to the PS—i.e., signals activating LTP were degraded at  $15^\circ\text{C}$ . Additionally, although LTP could not be elicited in slices from any of the three groups at  $15^\circ\text{C}$  (i.e., the amplitude of an evoked PS spike was not increased following tetanus), a PS could be generated in all



**Fig. 2** Declining slice temperature ( $T_s$ ) induced an increase in population spike (PS) duration (a loss of synchronization) and a fall in PSA. The population spike is the *green portion* of the evoked response. **a** PS versus  $T_s$  in a WH slice; **b** PS versus  $T_s$  in a HH slice; **c** PSs evoked at 15°C in all three groups (SH, WH, HH)

groups (Fig. 2c). Moreover, while LTP could not be generated at 15°C, it could be generated when the same slice was rewarmed to 30°C (Fig. 3) in five of five slices. That is, although the percent increase in PSA following tetanus varied from slice to slice (depending on a variety of factors including electrode placement), our data on SH, WH, and HH groups extend the upper end of the range of temperatures where LTP can be readily generated to 35°C and further show that LTP cannot be generated at 15°C, thereby supporting hypothesis 1.

A common finding in all groups (SH, WH, and HH) was that at 15°C the amplitude of a PS was markedly smaller than that at 35°C (in response to the same stimulus intensity) (Fig. 2a). In some cases (when the constant stimulus voltage was just above threshold at 35°C), the amplitude of the PS at 15°C appeared to be negligible compared to the amplitude at 35°C (Fig. 2b). However, even in these cases, a PS could be observed at 15°C by simply increasing the stimulus intensity (e.g., from 3 to 5 volts in Fig. 2b). The width of PSs also increased as temperature was lowered,



**Fig. 3** An HH slice demonstrating the dependence of LTP generation on  $T_s$ . Following tetanus at  $T_s = 15^\circ\text{C}$ , PSA was not enhanced, but it was (in the same slice) when  $T_s$  was raised to 30°C. In fact, as seen at *arrow a* and *b*, at 15°C, PSA actually decreased in this slice after tetanus. However, at 30°C, PSA increased shortly following tetanus (*arrow d*), and this increase was thereafter sustained (*arrow e*), the hallmark of LTP

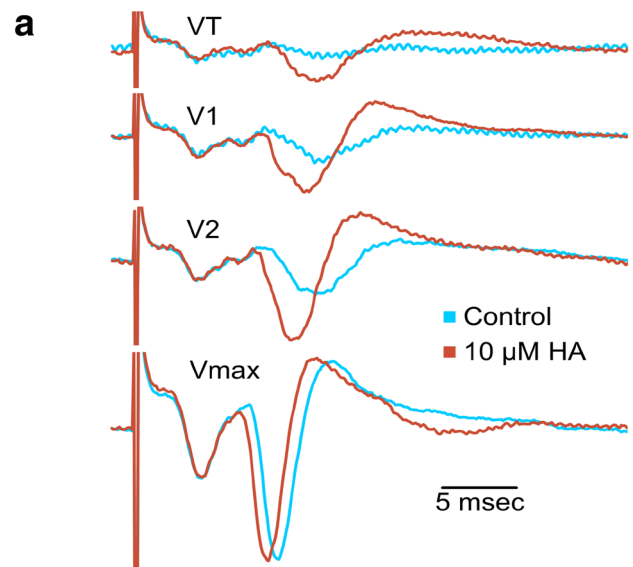
but as seen in Fig. 2c, rescaling the time axis shows that the PSs retained their “spike-like” appearance. The consistent finding that CA1 pyramidal cells in all three groups (SH, WH, HH) could still generate action potentials at 15°C (Fig. 2c) supports hypothesis 2.

While single shock stimulation evoked PSs at 15°C, tetanus failed to enhance PS amplitude. However, although LTP could not be evoked in a slice at 15°C, it could be evoked when slice temperature was raised to 30°C (Fig. 3).

### Histamine (HA) modulation of PSA (hypothesis 3)

Experiments involving HA addition to the oxygenated aCSF flowing through the recording chamber were designed to determine if HA modulated PSA as stimulus intensity was varied from threshold to maximum values (hypothesis 3). As an illustrative example, Fig. 4a, b show that with  $T_s$  fixed at 20°C throughout the experiment, adding 10  $\mu\text{M}$  HA to the standard aCSF increased PSAs of CA1 hippocampal pyramidal cells in an HH slice at all stimulus intensities except those close to maximum ( $V_{\text{max}}$ ). This is illustrated in Fig. 4a, where a threshold stimulus delivered during the control period (denoted in the traces labeled VT) was insufficient to evoke an appreciable

**Fig. 4** Modulation of CA1 pyramidal neuron activity by 10  $\mu\text{M}$  HA in a HH slice at 20°C. **a** Enhancement of evoked responses of CA1 pyramidal cells by HA. **b** IO curves constructed from PSAs calculated during control (blue), HA treatment (red), and recovery (purple) periods for the slice in (a). PSAs are normalized and expressed as a fraction of the control period response to a maximum stimulus. Voltages VT, V1, V2, and Vmax on IO plots in (b) correspond to their like-named traces in (a). **c** Group data for HA modulation of PSA in 15 HH slices. With recording chamber temperature set at 20°C throughout the slice experiment, 10  $\mu\text{M}$  HA was added to the aCSF. The pair of bars shows averaged PSA amplitude for the control period (blue bar) and the HA effect on PSA amplitude (red bar). VT, V1, and V2 correspond to their like-named sites in (b). Significant differences ( $P < 0.05$ ) are indicated by an asterisk (\*). PSAs recorded in the control (blue curve) at Vmax were normalized to 1.0



PS, but with addition of 10  $\mu\text{M}$  HA during the treatment period, a stimulus with the same intensity was able to do so. As stimulus intensity increased from threshold VT to the next two higher steps (V1 and V2), the response in the presence of HA was larger than that in its absence. Moreover, latency between shock artifact and the negative peak of the PS decreased with HA addition (Fig. 4a), a finding consistent with increased pyramidal cell excitability. At Vmax, the HA-induced PSA enhancement was much less pronounced (Fig. 4a).

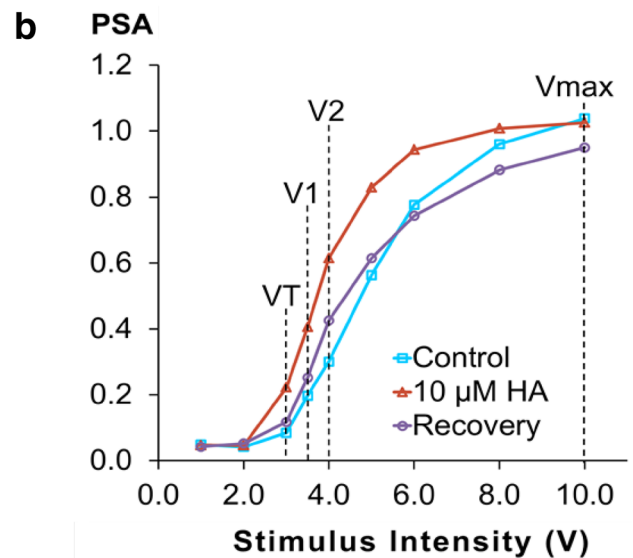
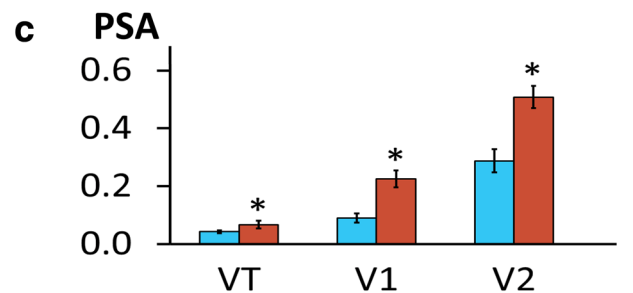


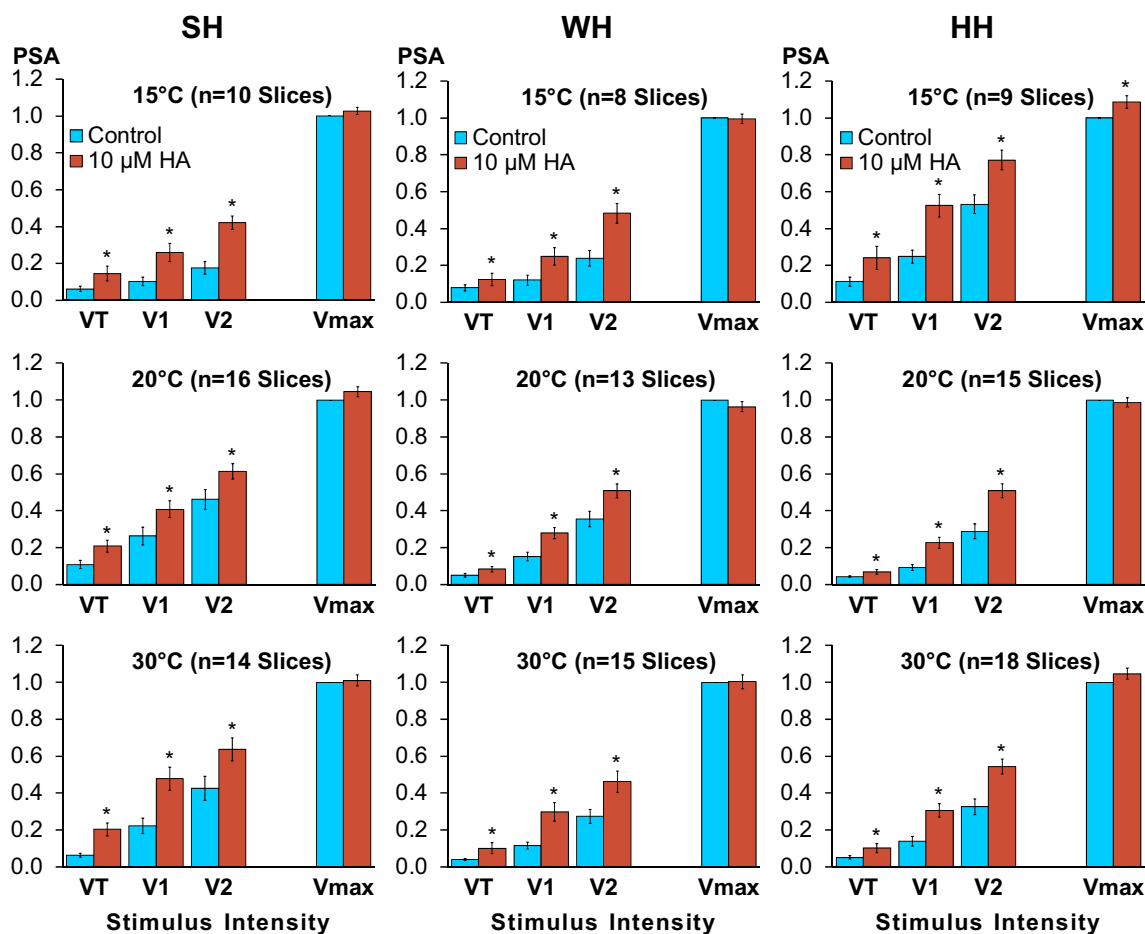
Figure 4b depicts IO curves of PSA versus stimulus intensity at all step increases (the points on the IO plots labeled VT, V1, V2, and Vmax correspond to the evoked responses with the same labels in Fig. 4a). These plots show the HA-induced enhancement at low stimulus intensities (VT, V1, and V2), with the magnitude of this enhancement tapering off as stimulus intensities increased to Vmax. As illustrated in Fig. 4b, during the recovery period (when the slice was perfused for 30 min with standard aCSF), the HA-induced enhancement was reversed.



The results in Fig. 4a, b for a single slice were mirrored by the HH group data (Fig. 4c) in  $n = 15$  slices. That is, the HA-induced PSA enhancement was significant at threshold and low stimulus intensities at 20°C (Fig. 4c). The pair of bars at V1 shows that HA more than doubled the responsiveness of CA1 pyramidal neurons to afferent signals over the Schaffer collaterals. This doubling occurred for a stimulus intensity only one-tenth of Vmax. The bars at VT, V1 and V2 in Fig. 4c display a pattern that serves as an exemplar of enhanced excitability for a range of stimulus intensities near threshold.

The pattern of HA enhancement of PSA in Fig. 4c for HH slices at 20°C was similar to groups of slices in SH, WH, and HH at 15, 20, and 30°C (Fig. 5). Results are consistent with the persistence of HA effects as the hamster enters torpor. While not shown in Fig. 5, an additional finding seen in comparing PSA at the end of recovery across all groups (SH, WH, and HH) and all temperatures was that the excitatory effect of HA

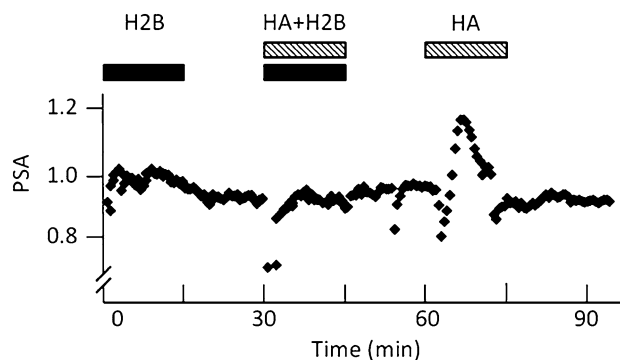
on PSA after submaximal stimulation at 15 and 20°C often (but not always) failed to completely return to the control value at the end of the 30-min recovery period while it did return at 30°C. That is, the effect of HA was often longer lasting at low temperatures. The results for slices from WH and HH were essentially the same



**Fig. 5** Data for HA modulation of PSA in all groups (SH, WH, HH) at a low range of stimulus intensities (VT, V1, V2) and at maximal stimulation. PSAs recorded in the control (blue bar) at Vmax were normalized to 1.0

as the results for SH slices—namely that HA enhanced PSAs near threshold stimulation (Fig. 5). Notably, the enhancement of PSA induced by HA did not decline with decreasing temperature and was as robust at 15 °C as at warmer temperatures. Moreover, significant HA enhancement of PSA was seen in all groups at all temperatures at the low stimulus intensities (VT, V1, and V2), an enhancement that gradually declined as stimulus intensity was increased, resulting in no HA-induced increase at maximal stimulus.

That the HA-mediated PSA excitation involves H2 receptors is supported by the data showing that 20 μM cimetidine (an H2 receptor inhibitor) reversibly blocked the increase in PSA evoked by 10 μM HA (8 of 8 slices at 30 °C; example in Fig. 6). In contrast, pyrilamine, an H1 blocker, reduced the magnitude of HA-mediated PSA excitation in only 1 of 8 slices at 30 °C.



**Fig. 6** H2 receptor antagonism of HA modulation. Addition of 20 μM cimetidine (an H2 antagonist) to the aCSF is indicated by black bars labeled H2B, while addition of 10 μM HA is indicated by cross-hatched bars labeled HA. Cimetidine prevented HA-induced PS enhancement. This inhibition was reversed after a 15-min washout of the cimetidine with aCSF, as demonstrated by subsequent PSA enhancement in HA-aCSF (bar at right). Records are from a single slice at submaximal levels of stimulation at 30 °C



**Discussion**

Our data on CA1 pyramidal neurons are consistent with our hypotheses and demonstrate that in all three groups of hamsters, LTP can be generated at a slice temperature of 35 °C, but not at 15 °C. In contrast, HA significantly increased the excitability of these neurons at 15 °C as well as at higher temperatures. This HA-induced increase was abolished by the H2 antagonist cimetidine but not the H1 antagonist pyrilamine. Since both 35 and 15 °C are well within the physiological range of body temperatures of Syrian hamsters measured by telemetry as they naturally enter and arouse from hibernation (Horwitz et al. 2013), delineation of cellular events in slice preparations provides insight into functional changes of CA1 pyramidal neurons as the hamsters transition between euthermic and torpor states.

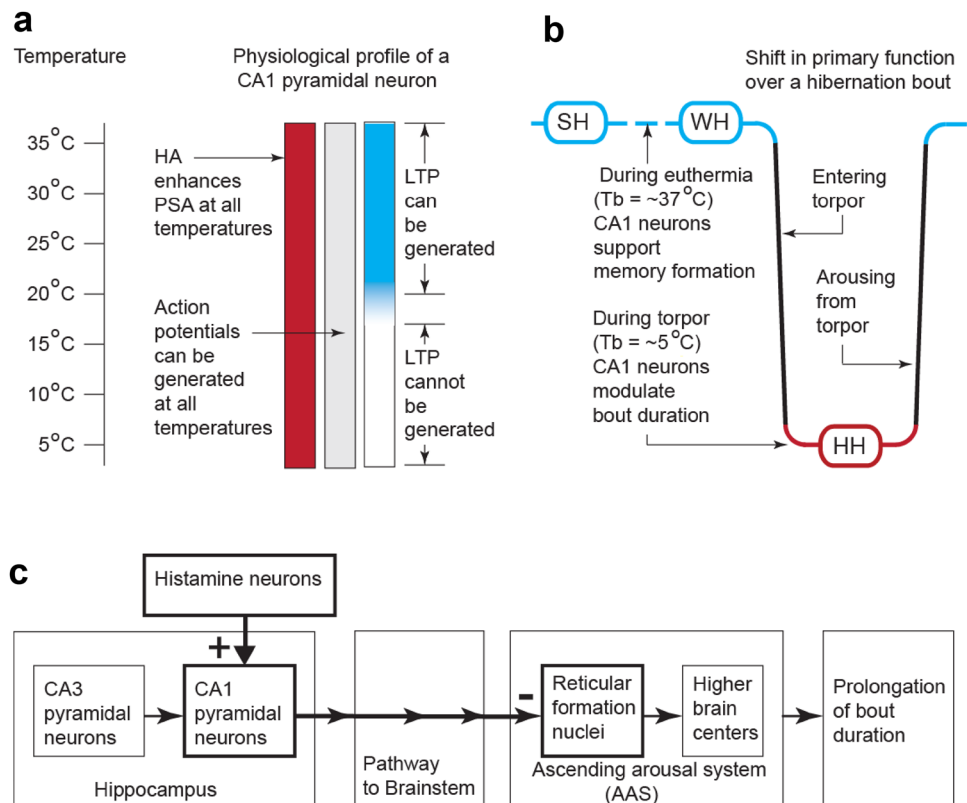
**LTP can be generated in all three hamster groups (SH, WH, HH) from 35 to 23 °C**

Hippocampal LTP generation is arguably a basic cellular mechanism for memory formation (Herring and Nicoll 2016), and as described below, delineating the physiological profile of the CA1 pyramidal neurons at brain temperatures encountered during hibernation (Fig. 7a) provides evidence for the temperature dependence of memory formation as well as additional roles the hippocampus may

take on in hibernating species. Specifically, our results showing that at  $T_s = 35\text{ °C}$ , tetanus evoked a sustained increase in PSA (Fig. 1) indicate that LTP could be generated in hippocampal slices from SH, WH, and HH groups and imply that at euthermic body temperatures, Syrian hamsters are able to form new memories via mechanisms like those used by non-hibernating mammalian species. Moreover, the ability of slices from SH, WH, and HH groups to generate sustained increases in PSA at 35 °C, but not at 15 °C, supports the concept that brain temperature per se is a major factor in LTP generation in CA1 pyramidal cells. These findings are in full alignment with the broader assertion (Carey et al. 2003) that during their active phase (with  $T_b \approx 37\text{ °C}$ ), hibernating mammalian species display physiological profiles that are essentially identical to those of non-hibernating mammalian species.

The data in Fig. 1 showing that at 35 °C LTP can be generated in all groups fill one gap in LTP slice studies and are consistent with previous PS studies (generally at temperatures below 30 °C) designed to test the assertion that several hibernating species have a cellular pathway that can support LTP generation. That is, LTP in Syrian hamster CA1 pyramidal cells has been previously shown to be generated at temperatures ranging from ~30 to ~22 °C (Bronson et al. 2006; Krelstein and Horowitz 1990; Krelstein et al. 1990) and in hippocampal slices from the Turkish hamster at 22 °C (*Mesocricetus banditi*) (Spangenberg et al. 1995).

**Fig. 7** Functional shift from memory formation to modulation of bout duration during torpor. **a** The physiological profile displays the temperature dependence of several features of CA1 pyramidal neurons that culminate in a shift in function with temperature—i.e., the neuron can generate LTP at warm but not cool/cold temperatures and can respond to HA at all temperatures tested. **b** Shift in function of the CA1 pyramidal neuron in SH, WH, and HH. **c** Histamine enhancement of CA1 pyramidal cell excitability leading to inhibition of the AAS and prolongation of torpor



Field potential recordings (fEPSPs) in these hamster studies further showed that LTP induced by Schaffer collateral stimulation was localized to the CA3–CA1 synapse. LTP data from Syrian and Turkish hamster hippocampal slices are comparable, with only slight differences that likely reflect species differences and/or somewhat different tetanizing protocols. LTP has also been shown to be generated at ~31 °C in hippocampal slices from the hibernating Yakutian ground squirrel (*Citellus undulates*) (Pakhotin et al. 1990). Yakutian ground squirrels are obligatory (seasonal) hibernators (i.e., they commonly hibernate only in winter) while the Turkish and Syrian hamsters are facultative hibernators (able to hibernate any time of year when exposed to a cold environment and short light–dark cycle). Although both types of hibernators naturally enter and arouse from torpor, obligatory species appear to more strongly link their neural networks associated with entering hibernation to their internal time-keeping networks and maintain robust circannual cycles of seasonal weight gain, breeding, and hibernation (Carey et al. 2003). Despite this difference, both facultative and obligatory hibernating species and non-hibernating mammalian species appear to have a similar intracellular hippocampal pathway for generating LTP.

#### **At 15 °C hamster pyramidal neurons can still generate action potentials, but can no longer generate LTP**

The precise threshold value where tetanizing stimuli fail to evoke LTP depends on the specific tetanizing protocol and the ionic composition of the solution in the recording chamber. Previous experiments (Arant et al. 2011) suggested that this threshold is ~18 °C for hamsters in the SH group. Although pyramidal cells lose their ability to generate LTP as temperature falls to 15 °C, they can still respond to afferent signals over Schaffer collaterals and generate action potentials (Fig. 2). This is consistent with the hippocampus in hibernating species taking on a “new” major function as temperature further declines—i.e., prolongation of hibernation bouts via inhibition of the AAS. Pyramidal neurons fulfill multiple functions dependent on temperature and seasonal adaptations, and this inhibitory role of the hippocampus has drawn particular attention in studies on the neural control of hibernation (Beckman and Stanton 1982; Heller 1979; Mihailovic 1972; South et al. 1972).

The failure to generate LTP in slice preparations at 15 °C may reflect several factors. One such factor is illustrated in Fig. 2—i.e., as temperature declined, with the stimulus amplitude held constant, the amplitude of the evoked PS decreased and its width increased. The marked increase in width of the PS at lower temperatures reflects a loss of synchronized excitation of pyramidal cells. Low body temperature generally increases signal dispersion under both in vivo and in vitro conditions as shown by the response

to a click at successive nuclei along the ground squirrel auditory pathway as it aroused from hibernation (Hamill et al. 1989). Because induction of LTP requires near-simultaneous coincidence of adequate postsynaptic depolarization and glutamate binding to NMDA receptors (Nicoll and Malenka 1999), a tetanizing stimulus (multiple single shocks) plausibly evokes “weakened” depolarization and hence gating signals for LTP generation at low temperatures (Krelstein and Horowitz 1990). An additional factor may be that the slowing of ATP production at low temperatures impairs recovery of ion homeostasis following a tetanizing stimulus.

In vivo hippocampal conditions during torpor are even less favorable to LTP induction because “natural” memory formation via LTP and LTD requires synchronized firing of pyramidal neurons (Axmacher et al. 2006). In torpor, synchronization (theta and gamma EEG activity) is lost as EEG activity is severely reduced at low brain temperatures (Chatfield and Lyman 1954; Mihailovic 1972). Spontaneous excitatory post-synaptic currents in single hamster brainstem neurons are also significantly reduced as temperature decreases (Sekizawa et al. 2013). In fact, neural activity throughout the brain is attenuated in torpor, as shown by <sup>14</sup>C-2-oxyglucose uptake studies (Kilduff et al. 1990, 1982). While one can impose synchronized excitation of Schaffer collaterals in in vitro slice preparations simply by applying a tetanizing stimulus, under in vivo conditions of deep hibernation hippocampal activity is muted and, in the absence of adequate postsynaptic depolarization, it appears unlikely that LTP is generated.

The recent finding that spine regression occurs during torpor in Syrian hamsters (Bullmann et al. 2016) extends previous studies on CA1 pyramidal cell synaptic/dendritic structure in obligatory hibernating species to facultative hibernators. This work is a significant advance in characterizing mnemonic mechanisms in Syrian hamsters as it is the first to show that hamsters housed in summer-like conditions can master a behavioral task, subsequently complete a series of hibernation bouts, and then, with minor retraining, perform the task well in summer-like conditions despite tau protein structural changes that occurred during torpor (and were reversed during arousal). LTP is a cellular mechanism that likely contributed to hamsters’ mastery of the maze and retraining as our data show that LTP can be generated at 35 °C (Fig. 1). Consistent with this assertion is the finding that the absence of tetanus-evoked LTP generation at 15 °C in HH slices was reversed upon warming the slice to 30 °C (Fig. 3), suggesting that, to the extent that reversal of spine regression is required to support LTP generation, such reversal occurred during the period encompassing slice preparation, incubation, and recording. Therefore, despite massive remodeling of dendrites and spine reduction in torpor (Bullmann et al. 2016), at near euthermic

temperatures, LTP mechanisms that support memory formation are in place in HH slices as in SH and WH slices (Figs. 1, 3).

### Histamine, acting via H2 receptors on CA1 pyramidal cells, enhances responses evoked by low-intensity stimulation

Because classical antihistamines have a well-known sedative action (Haas and Panula 2003), and HA has an excitatory effect on neurons, it may appear counterintuitive that HA prolongs rather than shortens hibernation bouts. In fact, hippocampal H1 and H2 receptors in hibernating hamsters are greater in number than those in non-hibernating hamsters, consistent with an excitatory effect of HA on the hippocampus during hibernation (Sallmen et al. 2003a). This *localized excitatory* modulation of CA1 pyramidal hippocampal neurons (the “+” sign in Fig. 7c) has been proposed (Sallmen et al. 2003a, b) to enhance pyramidal cell firing over a neural pathway from the hippocampus to brainstem nuclei that results in inhibition of the AAS and prolongs hibernation bouts (the “-” sign in Fig. 7c). Thus, an *excitatory* HA modulation of pyramidal neurons would be consistent with an *inhibitory* signal to the AAS. Our HA data showing that HA directly enhanced hippocampal excitability at 30, 20, and 15 °C support this sequence of events. Moreover, the use of hippocampal slice preparations ensured that the effects of HA on excitability (the PSAs) could be directly attributed to modulation of hippocampal activity (CA1 pyramidal cells) without the caveat that HA may have diffused into other brain regions.

The HA data in the present study show that HA application at low levels of afferent stimulation [“mimicking” brain activity during entrance into deep torpor (Heller 1979; Kilduff et al. 1990, 1982)] acts via H2 receptors to significantly increase CA1 pyramidal cell responses. Additionally, the longer lasting effects of HA modulation at 20 °C versus those at 30 °C are consistent with enhancement of the effectiveness of HA modulation at low brain temperatures.

Our results support the assertion (Sallmen et al. 2003b) that continuous low levels of HA infused into the hibernating ground squirrel hippocampus (3 fmol/h) directly excited CA1 pyramidal neurons as a step on a neural pathway to the brainstem that prolongs a bout of torpor. Our results also extend previous studies on Turkish hamsters (Nikmanesh et al. 1996) in which HA enhanced PSAs evoked by Schaffer collateral stimulation at 50–80% of their maximal amplitude, by showing that PSA enhancement was clearly evident at low, near-threshold stimulus intensities (Fig. 5), simulating the low level of neural activity throughout the central nervous system occurring in torpor (Kilduff et al. 1982).

The general proposal that a hibernation bout is lengthened by a hippocampal signal that inhibits brainstem arousal circuits implies that hippocampal neurons are well protected and functional at low temperatures. Indeed, hippocampi in hibernating mammalian species are significantly more tolerant to oxygen deprivation (Drew et al. 2004; Lewis et al. 2012) and to oxygen glucose deprivation (Dave et al. 2012; Mikhailova et al. 2016) than are hippocampi from non-hibernating mammalian species. As a result, a brief period of hypoxia and/or hypoglycemia may not significantly impair the hibernator’s ability to modulate its bout duration. The physiological profile of a pyramidal cell as diagrammed in Fig. 7a does not include these protective adaptations, but is rather a basic model involving a single neuromodulator (HA), action potential generation, and a single neuroplasticity mechanism (LTP) to describe core functional properties in CA1 pyramidal cells. The profile could be expanded by, for example, adding neuroprotective mechanisms providing tolerance to oxygen glucose deprivation (OGD) in hippocampi from hibernating hamsters (Mikhailova et al. 2016).

### Arousal from hibernation and the ascending arousal system

The in vivo experiments where HA was infused into the hippocampus via minipumps (Sallmen et al. 2003b) as an intervention to alter hippocampal neural activity is one of the few hibernation studies relating the hippocampus to the AAS that is not correlative in nature [see earlier reviews (Beckman and Stanton 1982; Heller 1979)]. The Sallmen et al. study and the reviews all postulate signal transmission during hibernation over a path from the hippocampus to the AAS, a system mediating waking, sleep, and coma in non-hibernating mammals first described by Moruzzi and Magoun (1949). In a recent reassessment of brainstem nuclei forming the anatomical structure of the AAS, cell-specific lesions of the brainstem parabrachial–precoeruleus complex (Fuller et al. 2011) produced the marked behavioral changes seen in the original experiments (Moruzzi and Magoun 1949). Future hibernation studies are needed to fully delineate the pathway from the hippocampus to these brainstem AAS nuclei. To date, evidence is consistent with the view that the AAS is inhibited in torpor (Harris and Milsom 2000), and one brain region that contributes to this inhibition is the hippocampus (Sallmen et al. 2003b).

### Summary

This study is the first to provide graphical data showing that LTP can be generated at 35 °C in hippocampal slices from SH, WH, and HH groups (Fig. 1), consistent with a role for the hippocampus in memory formation in

euthermic SH and WH groups and in animals that have aroused from hibernation. These findings extend previous work on SH and HH hippocampal slices over a temperature range from 18 to 30 °C (Arant et al. 2011). Taken together, the temperature dependence of LTP generation in Syrian hamsters indicates that LTP can be generated from euthermia to ~18 °C, suggesting that the role of the hippocampus in memory formation is limited to this range of brain temperatures. We also show for the first time that although LTP can no longer be generated at 15 °C, PSs can still be evoked (Fig. 2c) and HA can directly enhance CA1 pyramidal cell excitability (i.e., increased PSAs, Fig. 5) via H2 receptors (Fig. 6). These data support the view that at low brain temperatures, Syrian hamster hippocampal neurons can take on a new role—i.e., generation of inhibitory signals that suppress the AAS and prolong a hibernation bout. Overall, our data are consistent with a major temperature-dependent shift in the functional profile and behavioral role of Syrian hamster CA1 pyramidal neurons as they transition from euthermia to hibernation.

**Acknowledgements** Funds for this study were provided to B.A. Horowitz by the University of California, Davis. Additionally, Kevin Malins and Giancarlo Ibanez were each recipients of a Provost Undergraduate Fellowship, which supports students doing research under the guidance of UC Davis faculty.

#### Compliance with ethical standards

This article does not contain any studies with human participants performed by any of the authors. It does, however, contain studies involving animals for which all procedures were in accordance with the ethical standards of the institution at which the studies were conducted. Additionally, the authors had no conflicts of interest.

## References

- Andersen P, Bliss TV, Skrede KK (1971) Unit analysis of hippocampal population spikes. *Exp Brain Res* 13:208–221
- Arant RJ, Goo MS, Gill PD, Nguyen Y, Watson KD, Hamilton JS, Horowitz JM, Horowitz BA (2011) Decreasing temperature shifts hippocampal function from memory formation to modulation of hibernation bout duration in Syrian hamsters. *Am J Physiol-Reg I*(301):R438–R447
- Axmacher N, Mormann F, Fernandez G, Elger CE, Fell J (2006) Memory formation by neuronal synchronization. *Brain Res Rev* 52(1):170–182
- Beckman AL, Stanton TL (1982) Properties of the CNS during the state of hibernation. In: Beckman A (ed) *Neural Basis of Behavior*. Spectrum Publications, Inc, Jamaica
- Bronson NW, Piro JB, Hamilton JS, Horowitz JM, Horowitz BA (2006) Temperature modifies potentiation but not depotentiation in bidirectional hippocampal plasticity of Syrian hamsters (*Mesocricetus auratus*). *Brain Res* 1098:61–70
- Bullmann T, Seeger G, Stieler J, Hanics J, Reimann K, Kretschmann TP, Hilbrich I, Holzer M, Alpar A, Arendt T (2016) Tau phosphorylation-associated spine regression does not impair hippocampal-dependent memory in hibernating golden hamsters. *Hippocampus* 26:301–318
- Carey HV, Andrews MT, Martin SL (2003) Mammalian hibernation: cellular and molecular responses to depressed metabolism and low temperature. *Physiol Rev* 83:1153–1181
- Chatfield PO, Lyman CP (1954) Subcortical electrical activity in the golden hamster during arousal from hibernation. *Electroen Clin Neuro* 6:403–408
- Dave KR, Christian SL, Perez-Pinzon MA, Drew KL (2012) Neuroprotection: lessons from hibernators. *Comp Biochem Phys B* 162:1–9
- Diewald L, Heimrich B, Busselberg D, Watanabe T, Haas HL (1997) Histaminergic system in co-cultures of hippocampus and posterior hypothalamus: a morphological and electrophysiological study in the rat. *Eur J Neurosci* 9:2406–2413
- Drew KL, Harris MB, LaManna JC, Smith MA, Zhu XW, Ma YL (2004) Hypoxia tolerance in mammalian heterotherms. *J Exp Biol* 207:3155–3162
- Fuller PM, Sherman D, Pedersen NP, Saper CB, Lu J (2011) Reassessment of the structural basis of the ascending arousal system. *J Comp Neurol* 519:933–956
- Haas H, Panula P (2003) The role of histamine and the tuberomammillary nucleus in the nervous system. *Nat Rev Neurosci* 4:121–130
- Hajos N, Ellender TJ, Zemankovics R, Mann EO, Exley R, Cragg SJ, Freund TF, Paulsen O (2009) Maintaining network activity in submerged hippocampal slices: importance of oxygen supply. *Eur J Neurosci* 29(2):319–327
- Hamill NJ, McGinn MD, Horowitz JM (1989) Auditory brainstem responses in ground squirrels arousing from hibernation. *J Comp Biochem Phys B* 159:167–172
- Harris MB, Milsom WK (2000) Is hibernation facilitated by an inhibition of arousal? In: Heldmaier G, Klingenspor M (eds) *Life in the Cold*. Springer-Verlag, Berlin, pp 241–250
- Heller HC (1979) Hibernation: neural aspects. *Annu Rev Physiol* 41:305–321
- Herring BE, Nicoll RA (2016) Long-Term Potentiation: From CaMKII to AMPA Receptor Trafficking. *Annu Rev Physiol* 78:351–365
- Horowitz BA, Chau SM, Hamilton JS, Song C, Gorgone J, Saenz M, Horowitz JM, Chen CY (2013) Temporal relationships of blood pressure, heart rate, baroreflex function, and body temperature change over a hibernation bout in Syrian hamsters. *Am J Physiol-Reg I*(305):R759–R768
- Igelmund P (1995) Modulation of synaptic transmission at low temperatures by hibernation-related changes in ionic microenvironment in hippocampal slices of golden hamsters. *Cryobiology* 32:334–343
- Kilduff TS, Sharp FR, Heller HC (1982) [<sup>14</sup>C]2-deoxyglucose uptake in ground squirrel brain during hibernation. *J Neurosci* 2:143–157
- Kilduff TS, Miller JD, Radeke CM, Sharp FR, Heller HC (1990) <sup>14</sup>C-2-deoxyglucose uptake in the ground squirrel brain during entrance to and arousal from hibernation. *J Neurosci* 10:2463–2475
- Krelstein MS, Horowitz JM (1990) Tetanus during a high extracellular calcium pulse overrides the block of long-term potentiation seen at 20 degrees C in the hamster hippocampal slice. *Brain Res* 536:105–113
- Krelstein MS, Thomas MP, Horowitz JM (1990) Thermal effects on long-term potentiation in the hamster hippocampus. *Brain Res* 520:115–122
- Lein PJ, Barnhart CD, Pessah IN (2011) Acute hippocampal slice preparation and hippocampal slice cultures. *Method. Mol Biol* 758:115–134
- Lewis CJ, Becker JJ, Manis AD, Hamilton JS, Horowitz JM, Horowitz BA (2012) Neuroprotection supports signal processing in the hippocampus of Syrian hamsters, a facultative hibernator. *Neurosci Lett* 520:20–25

- Mayford M, Siegelbaum SA, Kandel ER (2012) Synapses and memory storage. *Cold Spring Harb Perspect Biol* 4:a005751
- Mihailovic LT (1972) Cortical and subcortical electrical activity in hibernation and hypothermia. In: South FE, Hannon J, Willis JR, Pengelley ET, Alpert NR (eds) *Hibernation and Hypothermia, Perspectives and Challenges*. Elsevier, Amsterdam, pp 487–532
- Mikhailova A, Mack J, Vitagliano N, Hamilton JS, Horowitz JM, Horwitz BA (2016) Recovery of Syrian hamster hippocampal signaling following its depression during oxygen-glucose deprivation is enhanced by cold temperatures and by hibernation. *Neurosci Lett* 621:98–103
- Moruzzi G, Magoun HW (1949) Brain stem reticular formation and activation of the EEG. *Electroen Clin Neuro* 1:455–473
- Nicol RA, Malenka RC (1999) Expression mechanisms underlying NMDA receptor-dependent long-term potentiation. *Ann NY Acad Sci* 868:515–525
- Nikmanesh FG, Spangenberg H, Igelmund P (1996) Histamine enhances synaptic transmission in hippocampal slices from hibernating and warm-acclimated Turkish hamsters. *Neurosci Lett* 210:119–120
- Pakhotin PI, Belousov AB, Otmakhov NA (1990) Functional stability of the brain slices of ground squirrels, *Citellus undulatus*, kept in conditions of prolonged deep periodic hypothermia: electrophysiological criteria. *Neuroscience* 38:591–598
- Panula P, Chazot PL, Cowart M, Gutzmer R, Leurs R, Liu WL, Stark H, Thurmond RL, Haas HL (2015) International Union of Basic and Clinical Pharmacology. XCVIII. Histamine Receptors. *Pharmacol Rev* 67(3):601–655
- Sallmen T, Lozada AF, Anichtchik OV, Beckman AL, Leurs R, Panula P (2003a) Changes in hippocampal histamine receptors across the hibernation cycle in ground squirrels. *Hippocampus* 13:745–754
- Sallmen T, Lozada AF, Beckman AL, Panula P (2003b) Intrahippocampal histamine delays arousal from hibernation. *Brain Res* 966:317–320
- Sekizawa S, Horwitz BA, Horowitz JM, Chen CY (2013) Protection of signal processing at low temperature in baroreceptive neurons in the nucleus tractus solitarius of Syrian hamsters, a hibernating species. *Am J Physiol-Reg I*(305):R1153–R1162
- South FE, Heath JE, Leucke RH, Mihailovic LT, Myers RE, Panuska JA, Williams BA, Hartner WC, Jacobs HK (1972) Status of the role of the central nervous system and thermoregulation during hibernation and hypothermia. In: South FE, Hannon JP, Willis JR, Pengelley ET, Alpert NR (eds) *Hibernation and hypothermia, perspectives and challenges*. Elsevier, Amsterdam, pp 629–633
- Spangenberg H, Nikmanesh FG, Igelmund P (1995) Long-term potentiation at low temperature is stronger in hippocampal slices from hibernating Turkish hamsters compared to warm-acclimated hamsters and rats. *Neurosci Lett* 194:127–129
- Zimmer MB, Milsom WK (2001) Effects of changing ambient temperature on metabolic, heart, and ventilation rates during steady state hibernation in golden-mantled ground squirrels (*Spermophilus lateralis*). *Physiol Biochem Zool* 74:714–723