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Transition from the locked in to the completely locked-in state: A physiological analysis

A. Ramos Murguialday [a,e,*], J. Hill [c], M. Bensch [d], S. Martens [c], S. Halder [a], F. Nijboer [e], B. Schoelkopf [c], N. Birbaumer [a,f], A. Gharabaghi [b]

[a] Institute of Medical Psychology and Behavioral Neurobiology, MEG Center, University of Tübingen, Tübingen, Germany
[b] Functional and Restorative Neurosurgery Unit, Department of Neurosurgery, and Neuroprosthetic Research Group, Werner Reichardt Centre for Integrative Neuroscience, University of Tübingen, Tübingen, Germany
[c] Empirical Inference Dept., Max Planck Institute for Biological Cybernetics, Tübingen, Germany
[d] Computer Engineering Dept., Eberhard-Karls-University Tübingen, Germany
[e] Fatronik-Tecnalia Germany, Tübingen, Germany
[f] Ospedale San Camillo, Istituto di Ricovero e Cura a Carattere Scientifico, Venezia-Lido, Italy

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HIGHLIGHTS

- This represents the first documentation of transition of a patient with ALS from the Locked In State the to Completely Locked In State, and the first EMG documentation of loss of all muscle activities, including sphincter function, but with retained cognition as measured with ERPs.
- In this patient, any stimulation, communication or learning using visual and tactile stimuli was lost. Visual BCI was useless.
- The findings suggest ALS as a multisystem disorder, even affecting afferent sensory pathways.

ABSTRACT

Objective: To clarify the physiological and behavioral boundaries between locked-in (LIS) and the completely locked-in state (CLIS) (no voluntary eye movements, no communication possible) through electrophysiological data and to secure brain–computer-interface (BCI) communication.
Methods: Electromyography from facial muscles, external anal sphincter (EAS), electrooculography and electrocorticographic data during different psychophysiological tests were acquired to define electrophysiological differences in an amyotrophic lateral sclerosis (ALS) patient with an intracranially implanted grid of 112 electrodes for nine months while the patient passed from the LIS to the CLIS.
Results: At the very end of the LIS there was no facial muscle activity, nor external anal sphincter but eye control. Eye movements were slow and lasted for short periods only. During CLIS event related brain potentials (ERP) to passive limb movements and auditory stimuli were recorded, vibrotactile stimulation of different body parts resulted in no ERP response.
Conclusions: The results presented contradict the commonly accepted assumption that the EAS is the last remaining muscle under voluntary control and demonstrate complete loss of eye movements in CLIS. The eye muscle was shown to be the last muscle group under voluntary control. The findings suggest ALS as a multisystem disorder, even affecting afferent sensory pathways.
Significance: Auditory and proprioceptive brain–computer-interface (BCI) systems are the only remaining communication channels in CLIS.

1. Introduction

Currently there is a lack of physiological measures to define the transition from the LIS to the CLIS. Furthermore, no standardized

* Corresponding author at: Institute of Medical Psychology and Behavioral Neurobiology, MEG Center, University of Tübingen, Tübingen, Germany. Tel.: +49 70712987712; fax: +49 7071295956.
E-mail address: aramos@fatronik.com (A.R. Murguialday).

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scale exists for the late stages of ALS. Once the zero value in the ALS functional rating scale (ALS-FRS) is obtained classification of the disease stage in ALS is complicated and this scale does not differentiate between CLIS and LIS. However, there is a fundamental difference between the two: communication is still possible within LIS but up to now impossible in the CLIS (Kübler and Birbaumer, 2008; Hinterberger et al., 2005a).

In 1966, Plum and Posner defined the "locked-in" syndrome as the clinical syndrome due to bilateral lesions of the corticospinal and corticobulbar tracts in the ventral portion of the pons with preservation of the tegmentum, describing a neurological condition of quadriplegia, anarthria and a paralysis of all facial muscles except the vertical eye movements. (Plum and Posner, 1966). Consciousness is thought to be fully preserved and can be demonstrated through voluntary blinking. However, LIS is not a homogenous neurological entity but has numerous variations (Bauer et al., 1979). Kübler and Birbaumer define LIS as a state of almost complete paralysis with voluntary eye movement control, eye blinks or twitches of the lip (Kübler and Birbaumer, 2008). The complete LIS (CLIS) is defined as a condition in which all motor control is lost.

Bauer and coworkers further differentiate these states: (1) the *classical LIS,* which refers to total paralysis except for eye movements and blinking, combined with preserved consciousness, (2) *incomplete LIS*, which refers to a state in which other voluntary motions are present (e.g. movement of thumb) and (3) *total LIS*, which consists of a total paralysis including paralysis of eye-muscles combined with preserved consciousness (Bauer et al., 1979).

Bauer and coworkers suggest that voluntary blinking could be a behavioral tool with which a differential diagnosis between the classical LIS and the coma and prolonged coma-like states can be made (Bauer et al., 1979). Furthermore, Bauer suggests that EEG could serve as an electrophysiological tool to differentiate CLIS from coma.

Several other progressive, systemic or traumatic neurological diseases may result in a LIS and CLIS such as ALS, Guillain–Barré, end-stage Parkinson disease, multiple sclerosis, traumatic brain injury and others with different etiological and neuropathological features. Thus, the enormous variation of LIS and CLIS asks for a physiologically based scale to quantify the degree of the "locked-in" state. ALS seems to be a particularly useful model for this scale because of the frequent change from LIS to CLIS within one patient.

Markland differentiated LIS from Coma by the presence of EEG-reactivity and "alertness" in LIS (Markand, 1976). However, Kotchoubey et al. have shown that EEG-reactivity (event related synchronization ERS, and event related desynchronization, ERD) is sometimes present in Vegetative State (VS) (Kotchoubey et al., 2003).

Eye movement paralysis, sensory dysfunctions and vesicorectal disorders (Yuki et al., 1995) are frequent complications after long respiratory support and paralysis. Therefore, their differentiation power is low as some groups reported patients with ventilation for many years with some patients showing incomplete and variable presence of the above mentioned signs (Hayashi et al., 1991; Okamoto et al., 1993; Yoshida et al., 1992).

The only remaining possibility to retain communication in the CLIS depends on neuroprosthetic devices, particularly brain–computer-interfaces (BCIs). Successful application of visual or autonomic signals or sniffing in the CLIS was only reported once in a single case using recordings of pH from mouth saliva (Wilhelm et al., 2006). BCIs depend on differentiable neural signals (i.e. to encode a "yes" and "no" signal). Therefore, a precise characterization and prediction of the CLIS seems mandatory (Kübler and Birbaumer, 2008). Peripheral autonomic psychophysiological measures could also be used for communication analogous to the use of skin conductance responses (SCR), heart rate (HR) and respiration in lie detection. Patients may signal "yes" or "no" by changing one of those response systems activities. In the paralyzed artificially ventilated ALS patient HR-variability might be severely reduced by the paced artificial respiration and lack of muscle activity necessary for HR-increases. Voluntary operant regulation of HR-decrease is extremely difficult to learn and uses different psychophysiological mechanisms (Cuthbert et al., 1981) than control of HR-increase. Therefore, HR-control, sniffing or breathing control is not possible in advanced ALS. The situation for SCR in ALS is not clear and no data exist. Skin alterations due to the disease and as a consequence of extended bed rest are frequent (Masur et al., 1995) and may prevent voluntary control. In the patient reported here SCR measured from the palm of the hand was virtually absent, probably correlated with the unresponsiveness of parts in the somatosensory system. Furthermore, late stage ALS and CLIS exhibit sympathetic and parasympathetic signs (Pinelli et al., 1995; Beck et al., 2002), decreased heart rate variation (Pisano et al., 1995), alterations of the excretory function of the salivary glands (Giess et al., 2000), and disturbance of the gastrointestinal tract [Toepfer et al., 1997,1999). These findings suggest parasympathetic abnormalities. Studies using 123IMIBG-SPECT have shown deficient sympathetic cardiac innervations (Druschky et al., 1999) and electrophysiological data demonstrated alterations of the sympathetic skin responses in ALS, indicating a degeneration of sympathetic nerve fibers (Masur et al., 1995).

Postmortem histology of ALS patient's tissue showed neuronal degeneration and loss of Onuf's nucleus in the ventral horns of the spinal cord, explaining alterations in bowel and bladder innervations (Pullen and Martin, 1995; Carvalho et al., 1995).

These findings challenge the previously accepted hypothesis about the non involvement of the striatic pelvic floor sphincter muscles and the survival of the Onuf nucleus motor neurons in CLIS. However the same group already reported an increase in neuromuscular jitter, fibrillation potentials and fiber density in the external anal sphincter (EAS) (de Carvalho et al., 1995).

Postganglionic sympathetic dysfunction affecting epidermal and dermal structures have been reported (Masur et al., 1995; Dettmers et al., 1993). Furthermore, some groups reported involvement of the peripheral sensory and autonomic nervous system (Bradley et al., 1983; Dyck et al., 1975; Steiner et al., 1984).

Recently, de Carvalho et al. (2008) concluded, that electrophysiological evidence for chronic neurogenic change plays an important role in ALS.

While new technologies are helping to understand the physiology of the LIS and CLIS and to communicate with these patients, a data based physiological measurement is needed to evaluate the degree of motor degeneration and physiological changes involved in the above mentioned states.

To investigate the transition between LIS and CLIS, we performed six different physiological tests in an ALS patient who went through the above mentioned transition to gain insight into the associated physiological changes and to propose the most appropriate approach for a communication system in CLIS, supported by neurophysiological data.

2. Methods

2.1. Patient

The patient was a 40 year old end-stage ALS patient. He was diagnosed with ALS in 1997, artificially ventilated since 2000 and entered the CLIS in March 2008. The last successful communication session was through vertical eye movement and was recorded on the 16th of March 2008. The last previous communication mode consisted of a mouth-twitch, therefore we performed electromyography (EMG) experiments in November 2007 to determine if the muscle contractions could be elicited and controlled by the patient

but could not produce any response. External sphincter control measurements were performed and from the 4th of February 2008 onwards were negative. There was, however although rare, weak eye movement control. Vision was severely compromised by necrosis of the cornea due to insufficient fluid availability caused by paralysis and apparent lack of adequate nursing. The patient underwent an epidural electrocorticographic (ECoG) 112 electrode grid and two 5 and 11 electrode strips implantation over the pre-motor, motor and somatosensory areas in December 2007 in order to guarantee BCI-based communication. Furthermore he suffered from diabetes type II, pneumonia, MRSA colonization, bedsores, and chronic constipation. There were no upper motor neuron degeneration signs (such as spasticity, hyperreflexia/increased tendon reflexes of the lower extremity etc.) indicating no severe affection of the corticospinal tract. We performed three efferent control tests at different days, recording eye, facial and sphincter muscles activity. Three selected brain activity assessment tests are reported. Vibrotactile, auditory and proprioceptive stimuli were used to investigate the integrity and functionality of the different afferent pathways. Motor imagery was used to study self modulated brain activity during LIS and CLIS. The experimental protocol was approved by the ethics committee of the University of Tübingen, Medical Faculty. Informed consent was obtained from the patient and given before entering CLIS and was confirmed several days before the implantation following the protocol described in Section 3 of Haselager et al. (2009). The patient gave consent by signaling "yes" or "no" with vertical eye movements 2 months before he became completely locked in. The sessions with his consent were videotaped and are available on request with the usual legal prerequisites; the following questions were asked repetitively to increase reliability among other personal questions:

Do you wish to continue the experiments with brain wave communication?
Do you want to continue brain communication and life support even if your eye movements stop in the future?
Do you want to receive electrodes implanted in your brain? (after extensive information by the neurosurgeon).

These questions were repeated 2 days before the operation, but due to the prominent paralysis no significant response pattern emerged. According to German law, informed consent in patients without communication capacity has to be given by the legal representative. In this case a social worker of the responsible hospital was declared legal representative and gave written informed consent to the neurosurgical and experimental procedure.

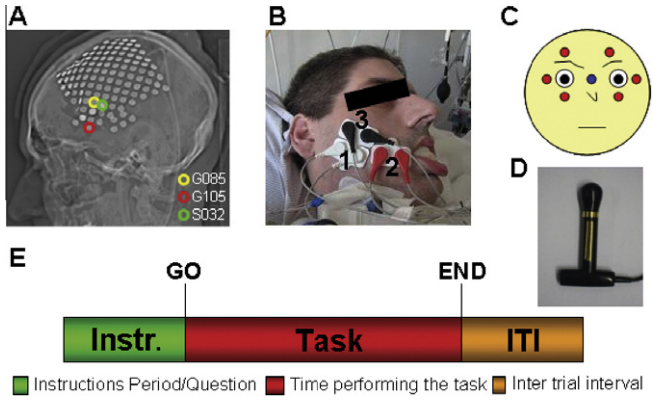

Fig. 1. (A) X-ray of patient with the epidurally implanted electrodes. (B) EMG electrodes distribution on the face muscles. (C) EOG electrodes location. (D) External anal sphincter electrode. (E) Timing diagram.

2.2. Experiment A: facial muscle control

2.2.1. Study design

Questions with known "yes–no" answers were presented to the patient. After each question the patient had 4 s to produce muscle activity around the mouth region in order to answer the questions "yes" and then a Stop cue was presented with an inter-trial interval of 4 s (Fig. 1E). The patient was asked to do nothing to respond to "No" question. We performed 3 sessions of 5 runs each. One run implies 12 repetitions of each answer class.

2.2.2. Data acquisition

Three bipolar Ag/AgCl electrodes from Myotronics-Noromed were used for EMG data acquisition and placed on the *risorius muscle* and two on the *zygomaticus major muscle*. The reference electrode was placed over the *olecranon* and the ground electrode was placed on the *clavicle*. (See Fig. 1B).

Data were acquired using a BrainAmp 32-channel amplifier from Brainproducts GmbH, Munich Germany. Sampling rate was 2500 Hz.

2.2.3. Signal processing

Data were filtered between 10 and 500 Hz, rectified and segmented in preselected time windows using the end of the "yes" or "no" question as trigger for the different movement classes. All the segments corresponding to one movement class were concatenated one after the other. Then we separated the EMG data in 200 ms windows with 25 ms overlap. Four different features that have been extensively used in muscle activity classification (Tenore et al., 2009) were calculated (mean absolute value, variance Willison amplitude and waveform length). The waveform length (WL) feature resulted in higher accuracy for discriminating between classes. The WL of the signal provides both information of the signal amplitude and its frequency expressed within the WL amplitude. It is obtained by the summation of the absolute values given by the difference in amplitude from a point in time (j) with a previous predefined time point ($j-1$) within a time window.

$$\mathrm{WL} = \sum_{j=1}^{N} \mid X_j - X_{j-1} \mid$$

Being X_j the jth point of N points time window of raw EMG.

2.3. Experiment B: external anal sphincter control

2.3.1. Data acquisition

It has been demonstrated (Lopez et al., 1999) that there is a strong correlation between surface electrodes applied to the perineal skin and concentric needle electrodes in the diagnosis of anal sphincter reaction. We used one single bipolar non-invasive anal sensor from Medicheck, Vossbuch, Germany for EAS activity data acquisition (see Fig. 1D).

2.3.2. Study design

Following the protocol used in the facial muscle control experiment, but the patient was asked to contract the sphincter for YES and relax for NO. During the contraction periods the subject was verbally instructed to maintain contraction and at certain point try to produce a peak in the EMG, to perform a short and intense EAS contraction. The signal was processed as in experiment A.

2.4. Experiment C: eye movement control

2.4.1. Data acquisition

Six monopolar Ag–AgCl sintered electrodes from EASYCAP GmbH, Herrsching-Breitbrunn, Germany were placed following

standard physiological landmarks around the eyes for electrooculography (EOG) acquisition. The reference electrode was placed on the patient's nose (see Fig. 1C). Sampling rate was 2500 Hz.

2.4.2. Study design

As in Experiment A, a "yes–no" protocol was selected, using eye movements in a predefined direction (vertical or horizontal) for YES and doing nothing for NO with inter-trial-intervals of 30 s and trial duration of 30 s.

2.5. Experiment D, E and F: electrocorticogram (ECoG)

2.5.1. Data acquisition/instruments

Intracranial brain activity was acquired using a BrainAmp 64-channel amplifier from Brainproducts GmbH, Munich Germany. Sampling rate was set at 500 Hz. An epidurally implanted 112 electrode custom made grid from Ad-Tech Medical Instrument Corporation, Wisconsin, USA was used for electrocorticogram (ECoG) data acquisition (see Fig. 1A) with electrodes S032 and G085 the ground and reference respectively (see Fig. 1A).

Vibrotactile stimuli were presented using the Quaerosys stimulator, Schotten, Germany. The envelope of the signal was 23–29 Hz with a carrier frequency of 200 Hz and duration of touch of 2000 ms.

2.5.2. Study design

In the *passive movement experiment D* we tried to measure patient's ability to perform motor imagery and his proprioceptive afferent pathways integrity. An auditory stimulus was presented requiring a right foot (dorsoplantar flexion and extension) or right hand movement imagery (hand open and close) in three different conditions: motor imagery task without passive movement, motor imagery task with passive movement and passive movement without motor imagery. For the motor imagery condition the patient was instructed to imagine "kinesthetically" the movement as if he was doing it actively. In the conditions with passive movements the hand and foot were extended and flexed passively every 2 s. Three seconds after the instruction period indicating hand or foot movement, a GO cue was presented, followed by a 25 s task period ending with an END auditory stimulus. If passive movement was requested, the patient limbs were passively moved for the 25 s varying between flexion and extension at a 0.5 Hz frequency. There was an inter-trial randomized rest time of 6.5–7 s between the END auditory cue and the beginning of the next instructions period (see Fig. 1E). We performed 3 sessions with 9 runs each. One run implies 15 repetitions of each movement.

In the *auditory oddball experiment E*, 245 standard tones (low pitch) and 45 deviant tones (high pitch) of 100 ms duration were presented at 70 dB via canal phones with an ITI of 850 ms. The patient was instructed to attend to rare, deviant tones.

In the *vibrotactile experiment F* vibrotactile stimulation was applied at three different locations. Right hand index finger tip, right foot big toe tip and the lip were used for stimulator placement. Three different auditory cues representing the three anatomical locations of the vibrotactile stimulators were presented to the subject before the actual vibrotactile stimulation. After the cues were presented, the vibration was applied for 12 s with an inter-trial interval of 10 s. The patient was instructed to focus his attention towards the stimulated body part.

2.5.3. Signal processing

In the *passive movements experiment D*, and the *vibrotactile experiments F,* data were band-pass filtered between 0.5 and 150 Hz. The data were transformed into the frequency domain generating 2 Hz frequency bins using the Power Spectral Density (PSD) estimate (Welch's method) with a time window of 500 ms and no overlap. A center-surround local spatial filtering approach, in which a radial difference-of-Gaussians function was used to weight the electrodes at each spatial location, was applied to the ECoG data. After discarding non-functioning bad channels, the remaining negative weights and positive weights were separately scaled to sum 1, so that the original reference cancelled out. In off-line cross-validation experiments, this procedure was found to improve signal-to-noise ratio slightly while ensuring that for each resulting signal, the contribution from electrodes more than one row or column distant was very small. The 25 s trials were divided into five segments of 5 s windows with no overlap. We calculated the area under the curve (AUC) scores for the comparison between the brain activity in the different conditions and a 3 s window of the inter-trial-interval used as rest. Hand versus foot brain activity was compared in each condition.

For the *auditory oddball experiment E* a standard P300 time domain analysis was done averaging over all trials (Polich et al., 1997). One of the electrodes on the grid was used as reference for the off-line analysis (G105 shown on Fig. 1A). We assumed that the distance from this electrode to the auditory cortex enabled a stable reference not influenced by auditory stimuli. The data were downsampled to 100 Hz and FIR-bandpass filtered (order: 500) between 0.5 and 9 Hz and cut into 1100 ms segments from 100 ms before the stimulus presentation to 1000 ms afterwards.

3. Results

Two months before the surgical intervention the patient could still communicate with a twitch of the mouth. This response disappeared slowly but some reflexive movements of his lips could still be observed weeks before the surgery.

The possibility of communication through the mouth-twitch was tested every day presenting questions with known "yes–no" answers to the patient and open questions with unknown answers, always with negative results after that date. Furthermore EMG activity of the previously active muscles was acquired in order to test if any remaining muscle activity was present, without success. No activity was found using any of the four calculated EMG features. Fig. 2 presents a comparison between a healthy subject and the ALS patient.

We used a feedforward multilayer perceptrons (MLPs) with varying numbers of hidden layer neurons (empirically chosen to be between 0.5 and 4 times the dimension of the input space) for classifying and testing the facial EMG data of the patient. A classification accuracy of 51% (chance level) was obtained (more information regarding classification process is presented in Tenore et al., 2009).

After the mouth-twitch based communication pathway ceased, the eyes appear to be the next suitable option. The patient could communicate with eye movements at two occasions before the surgery and two occasions after the surgery. This communication pathway was tested every day several times and the EOG was recorded. Fig. 3 depicts how the patient was able to move his eyes to answer questions with known answer for a successful session. The red triangles indicate YES answer questions and the black indicate NO answer questions.

Although the patient suffered fatigue, some days in which the patient could communicate for longer time were recorded until March 2008. In the beginning of February 2008 two EAS experiments were performed, with a week of separation between them. The possibility of remaining anal sphincter control was investigated with negative results. In Fig. 4A and B the raw and transformed EAS data of a healthy person is presented. Red and blue crosses indicate the start and ending of a verbally triggered contraction respectively. A clear difference between rest and EAS con-

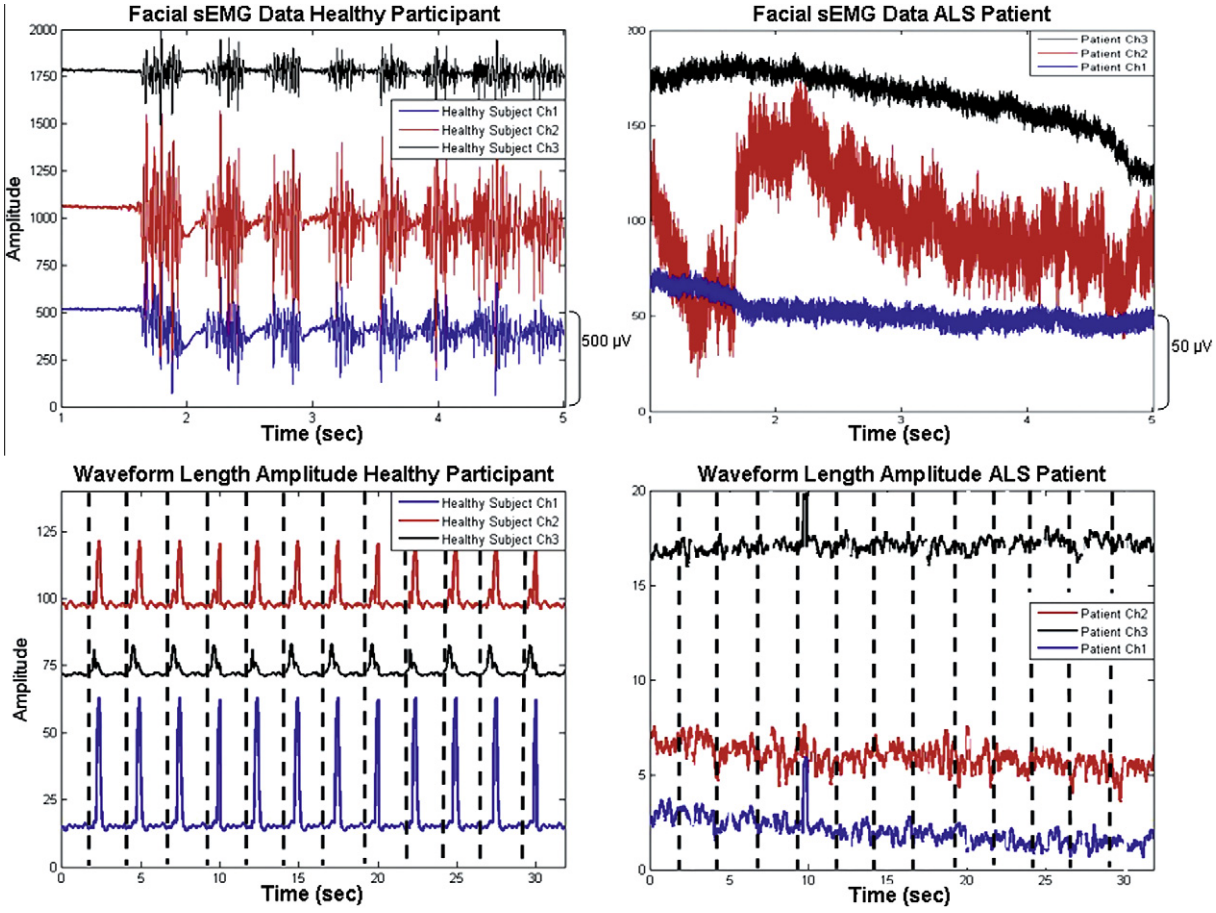

Fig. 2. Facial EMG: on the top representative facial EMG raw activity of the ALS patient in LIS (top right) and a healthy subject (top left) of three different bipolar EMG electrodes (see Fig. 1B) during a "yes" mouth-twitch. The bottom plot depicts the waveform length of concatenated trials of the ALS patient (bottom right) and of a control healthy participant (bottom left). The vertical black dashed lines indicate the start of each known YES answer questions in which EMG activity was expected. Please note different amplitude scaling between patient and control.

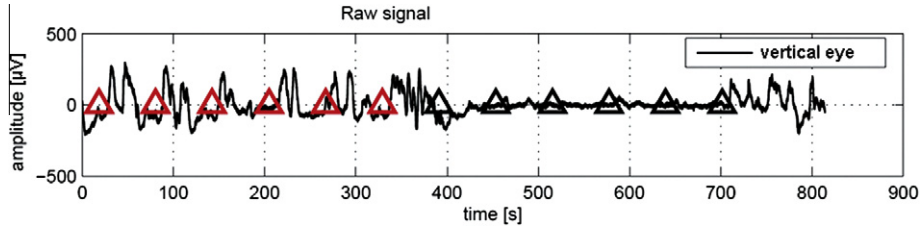

Fig. 3. *EOG activity* of the ALS patient during LIS. The red triangles were known "YES" answer questions and the black ones known "NO" answer questions. It is important to note that the EOG was clearly correlated with the presented cues at the beginning of the session.

traction is shown. In Fig. 4C and D, concatenated raw and transformed data from the ALS patient for YES (expected EAS activity) and NO (no expected EAS activity) answers is presented. In contrary to the healthy person there is no EAS activity. Corticospinal tract lesions were not visible in MR images from spinal cord excluding the motor neuron degeneration as the most likely cause of the sphincter pathology.

The last communication with the patient took place on the 16th of March 2008. After this date the patient was considered to be in the CLIS. During CLIS we performed somatosensory stimulation in order to test the non visual afferent pathways to the patient's brain. In locked-in ALS patients vision is compromised because of lack of adequate eye lubrication and moisture. Many different attempts for BCI communication not reported here were tried during CLIS, all without significant results. During the vibrotactile stimulation (Experiment C) no correlated activity was found in any of the implanted electrodes during any of the stimulations performed on the three different body parts (Index finger, toe and lip). This result is in complete contrast with similar procedures that elicited clear cortical responses in healthy and epilepsy samples (Diesch et al., 2001; Ray et al., 2008; Hansson and Brismar, 1999).

Nevertheless, while passive movements were performed on patient's foot and hand, activation of the somatosensory cortex was detected with and without simultaneous motor imagery task. The statistical analysis was performed, following (Agarwal et al., 2005) on the area under the curve (AUC) values, comparing hand

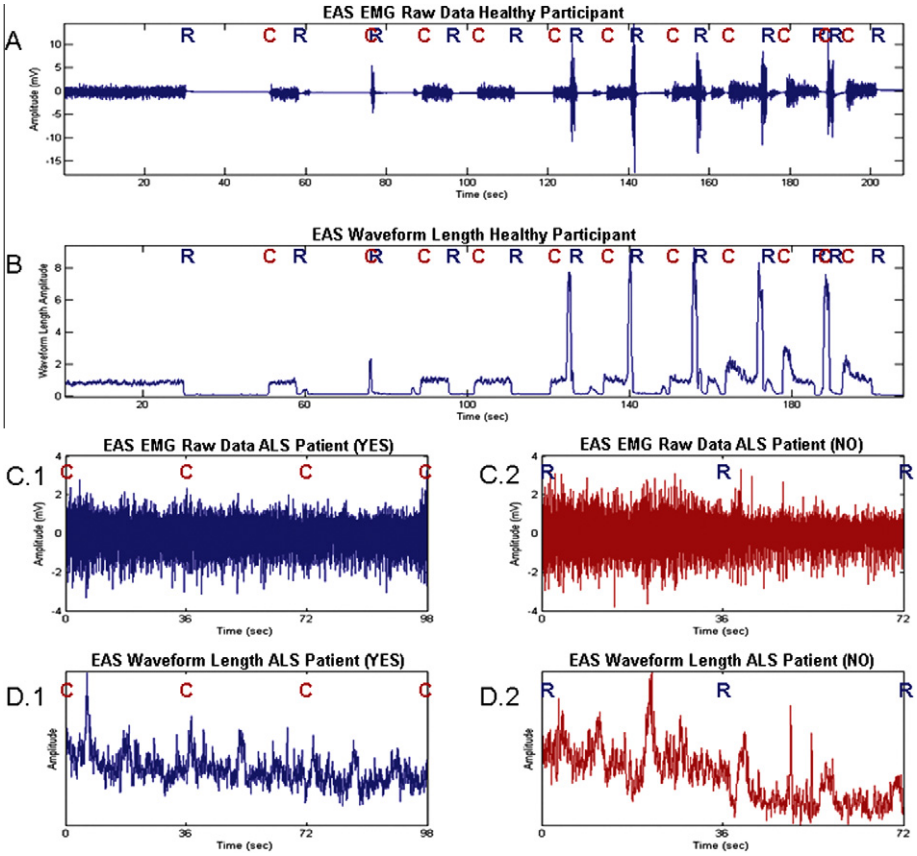

Fig. 4. *External anal sphincter:* (A) Raw EAS EMG activity of a healthy person following random auditory triggers. Red "C" indicates (contraction) and blues "R" (relaxation). The high peaks of EMG activity correspond to high intensity contractions. (B) WL extracted feature of activity in A. (C) Raw EAS EMG signal of the ALS patient and (D) WL extracted feature of activity in C. (C.1) and (D.1) "YES" answer trials (contraction) concatenated and (C.2) and (D.2) "NO" answer trials (relaxation) concatenated. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

versus foot, hand versus rest and foot versus rest from the ECoG data within the frequency domain focusing on motor imagery, passive movements and both simultaneously. We found statistically significant values ($p < 0.05$) while comparing hand versus foot passive movements and hand passive movements versus rest in specific frequency bins during LIS and in CLIS but no significant results were found for the foot versus rest conditions. Comparing foot versus hand passive movement cortical activity, significant results at 30 and 40 Hz frequency range were found in all the sessions and at 20 Hz frequency range for the simultaneous passive and imagery condition, while for the comparison hand versus rest, the significant activity was at 20 and 40 Hz frequency range for passive movements and at 40 Hz frequency range for passive movement and simultaneous motor imagery. No significant values where found for the comparisons during motor imagery only. However, when all the trials with passive movement were analyzed independent of being accompanied by imagery or not, AUC statistical analysis showed the significant frequencies during both LIS and CLIS to be at 20 Hz frequency range for Foot versus Hand and at 40 and 60 Hz frequency range for Hand versus rest. (See Table 1).

The electrodes showing high correlation with neural activity were located on the hand somatosensory area (see Fig. 5).

During the auditory oddball paradigm, two days after entering the CLIS state, clear averaged brain activity was detected. Shown in Fig. 6 are the averaged ERPs for 45 standard (blue) and deviant (magenta) tones from one representative run. Each line represents one of the 48 non-artefacted recording channels of the ECoG grid, re-referenced to another common grid channel (see Fig. 1A, G-105). The figure shows the N1/P2 component for the deviants, as well as the delayed P300 response in the range of 400–700 ms. These results demonstrate intact afferent auditory pathways and automatic attention, even though the N1/P2 complex is not clearly visible for the standard tones.

Table 1
Statistically significant frequency bins obtained from the area under the curve (AUC) analysis performed in the LIS (sessions 1 and 2) and CLIS (session 3) comparing different conditions. Passive stands for passive movement alone of the patients hand or foot, Pass&Imag stands for passive movement while the patient was asked to imagine the same movement and Pass&Imag + Pass stands for all the trials in which the patient's limb was passively moved independently of simultaneous imagery or not. Please note the stability of the statistically significant frequency ranges in the different conditions during locked-in state and completely locked-in state.

Conditions	Session 1 and 2 freq. bins (Hz) LIS	Session 3 freq. bins (Hz) CLIS
Foot versus hand	16–20 Hz	20–24 Hz
Pass&Imag		36–38 Hz
Foot versus hand	16–22 Hz and 30–32 Hz	20–42 Hz
Pass&Imag + Pass		60–62 Hz and 70–72 Hz
Foot versus hand	32–36 Hz and 40–42 Hz	29–36 and 40–42 Hz
Passive		56–58 Hz
Hand versus rest	6–8 Hz	22–26 Hz
Pass&Imag	40–42 Hz and 60–62 Hz	38–40 Hz
Hand versus rest	6–8 Hz	20–26 Hz and 30–34 Hz
Pass&Imag + Pass	40–46 Hz	40–44 Hz
	60–86 Hz and 92–94 Hz	60–66 Hz
Hand versus rest	24–26 Hz	22–28 Hz
Passive	40–42 Hz	40–44 Hz
	74–76 and 84–86 Hz	58–60 Hz

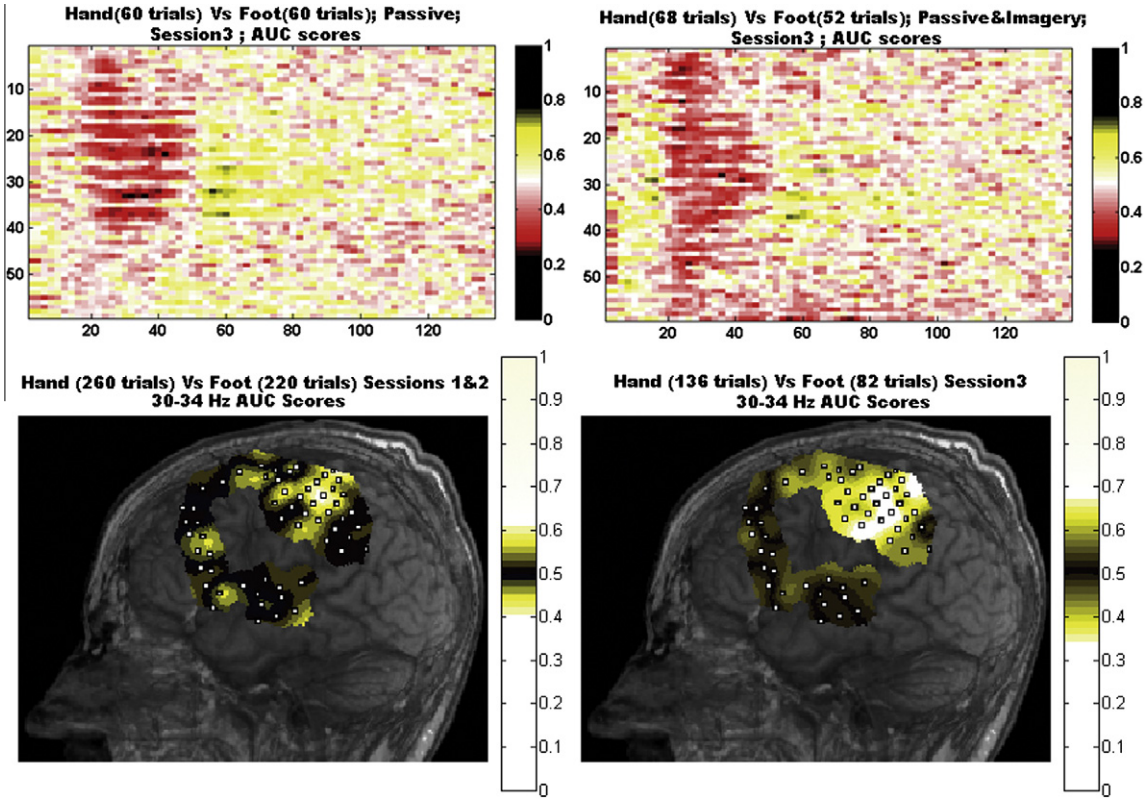

Fig. 5. *ECoG activity* during passive movement: on the top area under the curve (AUC) scores comparing foot versus hand movements of session 3 (CLIS); on the left side passive movements alone and on the right passive movements and motor imagery simultaneously AUC scores. In black the values that are statistically significant comparing hand versus foot movements. At the bottom the AUC scores in the frequency bin from 30 to 34 Hz plotted on the ALS patient X-rays (left, sessions 1 and 2 LIS and right, session 3 CLIS) comparing hand versus foot passive movements. In white the statistically significant AUC values. The hole in the center of the electrode grid is due to high impedances or lost channels.

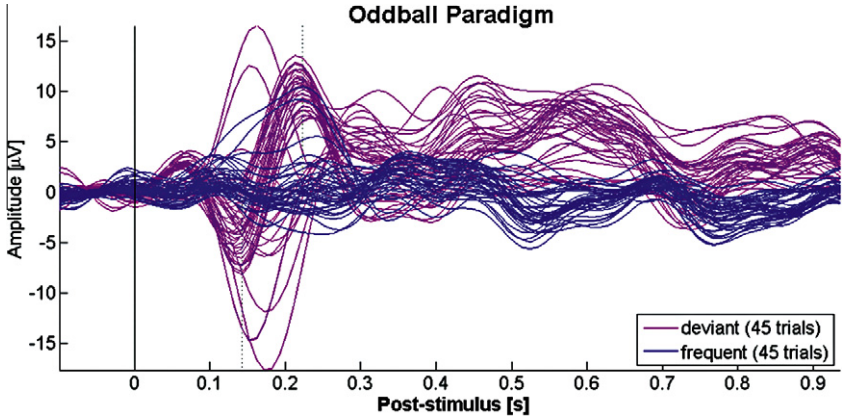

Fig. 6. *Averaged auditory evoked ERPs during CLIS* for 45 standard (blue) and deviant (magenta) tones. Each line represents one ECoG grid electrode recording. The vertical black line indicates beginning of the stimulus (time = 0 s). (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

4. Discussion and conclusion

LIS–CLIS transition was analyzed in one patient with ALS in several neurophysiological experiments that should provide information about the different stages of nervous system disorders. Between LIS and CLIS there are some clear physiological differences. Although the data presented in this paper are from one single patient only, who underwent the LIS–CLIS transition, this data set is unique and could be used as a first step towards a physiological data based instrument to define end stages of ALS and probably some other neurological diseases.

In the present patient the last remaining controllable muscles were the eye-muscles. This contradicts the hypothesis that the external anal sphincter is the last remaining controllable muscle in ALS. Jokelainen and Palo found no reports of rectal or bladder dysfunction in their review of 300 ALS patients (Jokelainen and Palo, 1976). But it was also reported that eye and sphincter muscles are affected by the disease (Carvalho et al., 1995; Pullen and Martin, 1995; Xu et al., 2007; Palmowski et al., 1995; Balaratnam et al., 2010; Okamoto et al., 1993) but we have not found any publication reporting the complete extinction of eye and sphincter movement control as observed in the present patient.

The fact that there is no cortical activation during vibrotactile stimulation may indicate that skin mechanoreceptors information is not reaching the cortical areas or that the skin mechanoreceptors do not function properly, at least in response to moderate to medium stimulus intensities. In contrast, proprioceptive information is processed in the brain as shown in the passive movement experiments. We hypothesize that joint receptors and muscle mechanoreceptors are less affected by the consequences of long-term immobilization than skin mechanoreceptors. Skin mechanoreceptors are located more at the body surface and therefore being more susceptible to damage from atrophy and skin deformation. These data suggest that muscle mechanoreceptors or joint receptors degenerate later than the skin mechanoreceptors and that at least some of the group Ia, II and Ib afferent fibers, muscle spindles and Golgi tendon organs are preserved in the CLIS. It has been proposed that joint receptor afferent input to the brain might be only significant when muscle spindle afferents do not contribute to proprioception (Burke et al., 1988). Taken together, the data indicate that if the above mentioned mechanoreceptors are not preserved then some of the joint receptors such as Ruffini endings, Pacinian endings and Golgi tendons and their respective fibers (slow and fast adapting fibers type II) should be preserved. On the other hand, the slowly adapting fibers type I and II (SAI and SAII), fast adapting type I and II (FAI and FAII) or their corresponding receptor types (Merkel cell, Ruffini ending, Meissners corpuscle and Pacinian ending, respectively) may be affected in CLIS.

Auditory information processing is preserved. Motor imagery did not elicit statistically significant brain activity when compared to rest. However, when motor imagery was performed accompanied by passive movements, the statistically significant frequency range obtained was different from the one obtained when passive movements without imagery were compared to rest. The classified activity decreased and shifted towards lower frequencies suggesting that some motor imagery might happen in CLIS.

We tried visual BCIs as described in Nijboer et al. (2008) without success.

Due to eye infection and to the ECoG location visual experiments were not performed after implantation. Due to the paralysis of eye-muscles in end-stage ALS, vision is severely compromised due to dryness of the cornea. The cornea of our patient was already seriously damaged months before the implantation, thus, significant results from visual experiments were not expected.

We believe that the reduced sensory information flow could play an important role in the extinction of motor imagery ability necessary for some BCI-based communication in CLIS as proposed by Kübler and Birbaumer (2008). Furthermore, the modality of elected BCI communication should consider the few remaining afferent flow information to avoid lack of feedback and thereby the above mentioned "extinction of thought" problem.

Kotchoubey et al. reported that all CLIS patients of the sample reported in (Kübler and Birbaumer, 2008) had normal ERP-responses to one or more of the complex cognitive tasks, indicating at least partially intact cognitive processing in LIS (Hinterberger et al., 2005b) and CLIS (Kotchoubey, 2005). We may conclude that somatosensory and visual processing is not intact in CLIS and the complete lack of motor control and lack of all kind of contingent external feedback for behavioral responses might be responsible for the cessation of voluntary cognitive activity and intention, goal directed thinking and imagery.

Since communication through a BCI seems to be the only way to avoid the extinction of thought it is necessary to know which afferent pathways would be more appropriate for feedback and reward. From these data reported here we conclude that passive movement or auditory based BCI are the only remaining possibilities. Other ways of afferent stimulation using nociceptors (pain receptors) and invasive stimulation is ethically problematic. Temperature stimulation or pH-communication (Wilhelm et al., 2006) could be another option and should be considered. Sniffing is not possible because it needs somato-muscular control. SCR regulation is not possible because of complete cessation of SCR in many if not in all ALS patients.

The last observed controllable muscle besides eye-muscles may vary between persons and usually involves facial muscles. The EAS is affected by the disease (Carvalho et al., 1995; Xu et al., 2007) and in spite of some fibrillation, no reliable control was possible. Once eye movements are lost the patient has reached the CLIS. In this state atrophy impairs mechanoreception of the skin while the auditory and muscle joint receptors pathways remain intact eliciting some cortical responses. BCI-based communication with auditory (Nijboer et al., 2008) and visual stimulation as described by our group (Nijboer et al., 2007) or imagery (Kübler and Birbaumer, 2008) did not result in reliable communication in CLIS. The fact that the auditory and proprioceptive systems still elicit brain responses suggests that BCI platforms for LIS and CLIS patients should avoid using feedback strategies that use visual or mechanoreceptive systems, and focus on disease-resistant systems such as auditory or proprioceptive systems that could be used in the LIS–CLIS transition. This might be the only way to prevent the "extinction of thought" in CLIS patients.

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