

EXPLORE THE IMPACT OF NOISE STRESS INDUCED BRAIN WAVE PATTERN AND BEHAVIOUR ALTERATIONS IN WISTAR ALBINO RATS

JEYAKUMARI P, RAVINDRAN RAJAN*

Department of Physiology, Dr. ALM Post Graduate Institute of Basic Medical Sciences, University of Madras, Chennai, Tamil Nadu, India.
Email: jeyakumari.p.s@gmail.com

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ABSTRACT

Objective: The aim of the study is to investigate the impact of noise-induced stress and electroencephalogram (EEG) with behavioral alteration in male Wistar albino Rats.

Methods: Adult albino rats were randomly divided into three groups. Each group contains six animals. Rats exposed to acute and sub-acute noise, stress (100 dB/4 h) were compared with control animals and assessed for learning and memory using an Eight-arm radial maze, Y-maze, T-maze and also monitoring of brain electrical activity showed by the electroencephalography.

Results: The reference memory and working memory error increases, in acute and sub-acute noise stress. The amplitude and frequency also increase in frontal and occipital lobar when compared to control animals.

Conclusion: Animals were exposed to noise stress showed learning and memory impairment and also changes in EEG wave pattern.

Keywords: Behavior, Electroencephalogram, Noise stress.

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INTRODUCTION

Sound may be a sort of automatic wave and is taken into account a useful tool for communication, whereas undesirable and hurtful levels of sound are referred to as noise. Acute stress affects several areas of the Pallium, Mild acute stress can cause a discount within the functioning of the prefrontal cortex, which causes large effects on our behavior [1]. The remembering ability is additionally decreased within the dorsolateral prefrontal cortex. Increased levels of cortisol when a private is stressed [2]. Decreased theta band synchronization when participants were subject to acute stressors [3]. Stress-response pathways affect the hippocampus and amygdala [4]. Therefore, if we are to look at brain dynamics in real-world conditions, to live the consequences of stress, we must even have reliable methods, especially those within the prefrontal cortex. Stress is one among the foremost important factors that intimidation homeostasis, among the strain, noise, stress that might not be avoided. Electroencephalogram (EEG) is employed for the activity of brain areas will be recorded [5]. The action of stress hormones on the receptors present within the brain region like the hippocampus [6].

Aim

The results reported that acute and sub-acute noise stress affects learning and memory, and also changes within the electrical activity of the brain.

METHODS

Experimental animals were all healthy and weighed about 180–200 g. The animals were kept within the Animal House of the Institute, University of Madras, Taramani, Chennai, India, and every one the animals were kept under standard laboratory conditions, housed three per cage (29 cm × 22 cm × 14 cm) and constant ambient temperature with 12-h light and 12-h dark cycle. The rats were allowed free access to food and water. Appropriate ethical clearance was obtained for this work from the Institutional Animal Ethical Committee (IAEC. No:01/27/2014) preceding the experiments.

Experimental design

The animals were divided into three groups; each group consists of six animals:

- Group I - Control animals
- Group II-Acute Stress, (4 h 1 day)
- Group III-sub acute stress (4 h/day for 15 days).

Noise stress procedure

Broadband (white) noise was elicited by a dissonance generator (0–26 KHZ) and amplified by an amplifier (40 W) connected to two loudspeakers (15 W) located at 30 cm above the animal cage. The intensity of the sound was measured by a sound level meter (Quest electronics cygnet systems D2023, serial No F02199, India) and maintained at 100 dB intensity. Animals were exposed to noise for 4 h/day.

EEG

German psychiatrist Hans Berger introduced the term EEG, which indicates a record of the variation in brain potential [7]. The electrical currents within the brain were discovered in 1875 by an English physician Richard Caton observed the EEG from the exposed brains of rabbits and monkeys. EEG complexity analyses, the complexity variations of 4 rhythms of EEG (delta, theta, and beta) are analyzed.

Physiological basis of the EEG signal

The EEG measures the electrical potentials of cortical neuronal dendrites near the brain's surface. Current derived from synapses move through the dendrites and cell body to a trigger zone within the axon base and labor under the membrane to the extracellular space along the way. Summation of potentials produced from the mixture of extracellular currents generated by populations of neurons is recorded as EEG [3]. The appearance of EEG rhythmic activity in scalp recordings is feasible as results of the synchronized activation of neurons and when their summated synaptic events become sufficiently large [8]. The rhythmic activity is also produced by both pacemaker neurons having inner capability of rhythmic oscillations and neurons that may synchronize

their activity through excitatory and inhibitory connections. The oscillations have their own discharge frequency, and counting on their internal connectivity. The neuronal oscillators start to act in synchrony after application of external sensory stimulation [9]. In 1924, Hans Berger, a German neurologist, used his ordinary radio instrument to boost the brain's electrical activity on the human scalp [10]. Later in 1934, Adrian and Matthews verifying concept of "human brain waves" and determined regular oscillations around 10–12 Hz which they called "alpha rhythm" The EEG measured directly from the cortical surface is termed electrocardiogram while when using depth probes it's called electrogram. The EEG denotes the electrical activity recorded from the scalp surface after being picked up by metal electrodes and conductive media [11]. Amplitude and frequency patterns in forebrain EEG activity constitute main defining characteristics of behavioral states [12].

When brain cells are activated, local currents are produced. EEG measures mostly the currents that flow during synaptic excitations of the many pyramidal neurons within the cortex. Differences of electrical potentials are caused by summed postsynaptic graded potentials from pyramidal cells that make electrical dipoles between soma and apical dendrites. Brain electrical current consists mostly of Na⁺, K⁺, Ca⁺⁺, and Cl⁻ ions that are pumped through channels in neuronal membranes within the direction governed by membrane potential. Three forms of EEG patterns, usually unite during one behavioral state, were detected.

1. Rhythmical slow activity or theta, it gives a behaviorally correlated fundamental peak frequency of 6–9 c/s, small bandwidth
2. Irregular slow activity it's a broad-band (0.5–25 c/s) spectrum almost like an occasional pass filter, low dorsoventral coherence
3. Fast waves of 20–60 c/s, which are larger in walking or rapid eye movement than awake. A model of the hippocampus circuitry indicates that three forms of EEG may require different afferent inputs and different bias on the hippocampal circuit [13] Experimental findings are mentioned which support the hypothesis that short-term (episodic) memory demands result in a synchronization (increase in band power) within the theta band, whereas long-term (semantic) memory demands cause a task-specific desynchronization (decrease or suppression of power) within the upper alpha band [14].

Stereotaxic implantation of electrodes

Male Wistar rats (180–200 g) are used for electrode implantation.

- All surgical procedures are performed stereo toxically under aseptic conditions
- Rats are anesthetized (ketamine and xylazine, intraperitoneally) and fixed in a stereotaxic apparatus so that the skull is positioned horizontally to keep the bregma and lambda in one plane
- The skull surface is cleaned so that the external landmarks of the surface become visible for the implantation of electrodes using bregma as the reference point
- Appropriate coordinates are selected based on the region of interest for electrode implantation.

To record electrical activity, that is, EEG, electrocorticogram electromyogram, and bipolar hippocampal EEG, electrodes (Nichrome, 28G; electrodes insulated with Epoxy resin) are implanted bilaterally.

Behavioral assessment

Preparation of animals for the radial eight-arm maze

Spatial learning and memory determined by employing a radial eight-arm maze apparatus [15]. Fig. 1 the weight of the rats was maintained at about 75–85% of their free-feeding level during behavioral training. Before starting experiment, a bunch of animals was trained, so they become habituated to the apparatus and a chunk of cereal for 3 days before each test starts. During this 3-day period, a 10-min period of habituation was repeated 3 times daily, at intervals of quite 1 h. Each rat was individually kept during a small cage after adaptation the variation and maze tests were performed between 10:00 and 12:00 h.

Rats were trained daily to search out a bit of cereal in four of the eight arms and were trained to locate four food rewards that were always



Fig. 1: Radial Eight-Arm Maze

placed within the same set of 4 arms. Rewarded arms got to every individual rat. The space contained several visual reference cues. Initially, animals were allowed to freely move the maze for 2 consecutive days with all arms baited with cereal. Only four arms (fixed for that animal) were always baited and food rewards placed at the top of the arms for training on the spatial task. Each trial starts with the position of the animal on the central platform facing arm much loved and ended when the rat had visited the four baited arms or after a period of 10 min.

According to Olton's definition (Table 1):

1. Number of reference memory errors, that is, each entry into a non-baited arm
2. Number of working memory errors, that is, re-entries into already visited baited arms
3. To go to all the baited arms at given time. Animal needs 25–28 training sessions to achieve the criterion of 0–2 errors. The animals were subjected to noise stress after the criterion 0–2 errors was reached.

Advantage

1. Different memory types can be tested: working memory, long-term (reference) memory, motor (egocentric) memory
2. Cheap, simple set-up, which might be dismantled and simply stored after use.

Control rats were kept within the above described cage during the corresponding period of your time, without noise stimulation. It is important to start allowing the animal to freely explore the maze (habituation) within the first 1–2 days, with no food available, for 5–10 min each trial. They will be introduced to the reward (baits), in an exceedingly limited number (e.g., no over 8–10 small pellets) in their home cage. On the subsequent day food trials were starts, and therefore the simplest training procedure to check spatial memory consists of baiting just one arm and training the animal to seek out it, for several trials. It is often called spatial delayed matching. When noise exposure of any kind exceeds 100 dB, noise becomes a stress. Above 30 cm of the cage, the noise was produced by two loudspeakers (15 W), throughout the cage uniformly expose noise stress was set as 100 dB and monitored by a sound level meter (Quest Electronics 215). This background level was selected supported the intensity. The animals were exposed for 4 h/day for 15 days. To avoid the influence of handling-stress on the evaluation of effects because of noise exposure, control rats were kept within the above-described cage during the corresponding period of your time, without noise stimulation.

Disadvantage

1. Animals must be well-handled and motivated to perform the tasks
2. Appetite task: the animals must be mildly food-deprived to be motivated to appear for baited arms; maybe an issue in some cases (e.g., Need full food supplies required the aged rat to remain healthy).

T maze apparatus

A T maze consists of a stem (50 cm × 50 cm) a choice aisle (10 cm × 10 cm) and two arms (50 cm × 10 cm) at the top of which was the goal area containing food. The side walls were 40 cm tall. Cloth curtains separated the goal area from the arm, so the animals cannot see the food from the selection area. Illumination is provided. The food reward should be palatable for the rats Fig. 2.

Procedure

Overnight fasted animals are only used, that is, Experiment was done under appetite motivation. The animal is left within the stem of the maze at the beginning point and allowed to explore the apparatus for 4–5 min. Then it's again put at the beginning point with reward being placed behind the curtain on the proper arm of the T maze.

Each animal is given ten trials and also the number of correct (i.e., moving toward the proper arm and eat the food) and therefore the number of wrong trials (i.e., to the left arm/return to the beginning point/return from light arm without taking the reward) is additionally noted. The result's expressed as percentage of correct trials. To attain reproducible results, the bias was eliminated by the experiment being done by another individual. Throughout the experiment, the experimenter stayed behind the beginning, hence handling gave no guidance to the animal. All sensory cues aside from the position of reward with relevancy the animal were rendered ineffective.

Y maze apparatus

In Fig. 3, Y-Maze with three identical arms of plexiglass (40 cm × 4.5 cm × 12 cm) 120° apart are going to be placed within the center of a space. The walls of every arm had distinct design that provided visual cues.

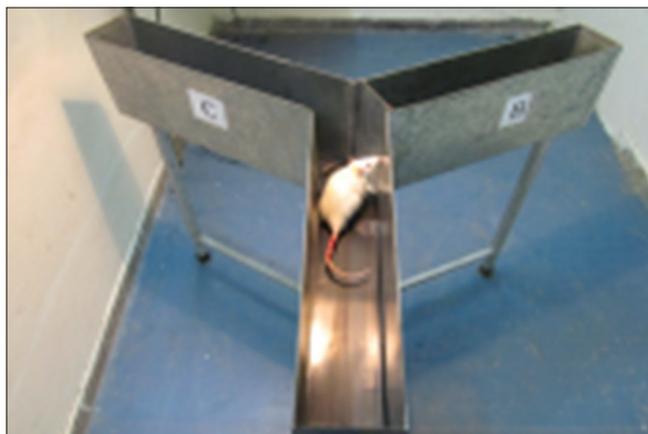


Fig. 2: Y- Maze



Fig. 3: T- Maze

Each mouse will place at the tip of 1 arm facing the middle and allowed to explore the maze for a period of 8 min. Sessions are video recorded and scored for entries into arms. Alternation behavior is going to be defined as consecutive entries into each of the arms without repetition. Percent of spontaneous alternation are going to be calculated as number of actual alternations divided by possible alternations (total arm entries-2) 100%.

RESULTS

EEG

The variance of frontal lobe amplitude (EEG) has been summarized in Fig. 4a and b.

The frontal lobe amplitude significant decreased in sub-acute noise stress when compared with control. Results shown are Mean±S.D when compared to control; the symbol represents statistical significance *p<0.05. The frontal lobe frequency significantly increased in sub-acute noise stress when compared with control. Results shown are Mean±S.D when compared to control; the symbol represents statistical significance *p<0.05.

The occipital lobe frequency decreased sub-acute noise stress when compared with control. Results shown are Mean±S.D when compared to control; the symbol represents statistical significance *p<0.05. The occipital lobe frequency increased sub-acute noise stress when compared with control. Results shown are Mean±S.D when compared to control; the symbol represents statistical significance *p<0.05 Fig. 5a and b.

Behavior

Figs. 6 and 7 in T maze, the time taken is increased sub-acute noise stress when compared with control. Results shown are Mean±S.D when compared to control; the symbol represents statistical significance *p<0.05. In y maze, noise stress the latency period of acute and sub-acute noise stress increased when compared with control. Results shown are Mean±S.D when compared to control; the symbol represents statistical significance *p<0.05.

Number of reference memory errors (each entry into a non-baited arm)

Number of reference memory errors in eight arm radial maze has been summarized: The reference memory error increased, acute and sub-acute noise stress when compared with control. Control is significant to acute and sub-acute noise stress. Results shown are Mean±S.D when compared to control; the symbol represents statistical significance *p<0.05 Fig. 8a.

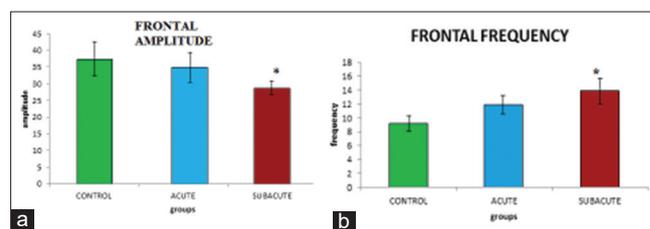


Fig. 4: (a) Frontal Amplitude, (b) Frontal Frequency (EEG)

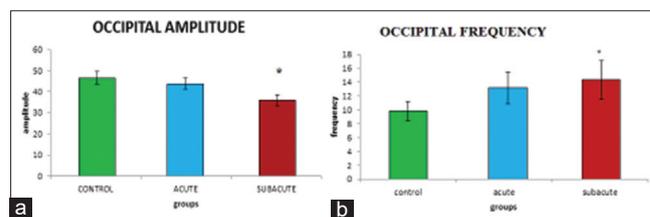


Fig. 5: (a) Occipital Amplitude, (b) Occipital Frequency (EEG)

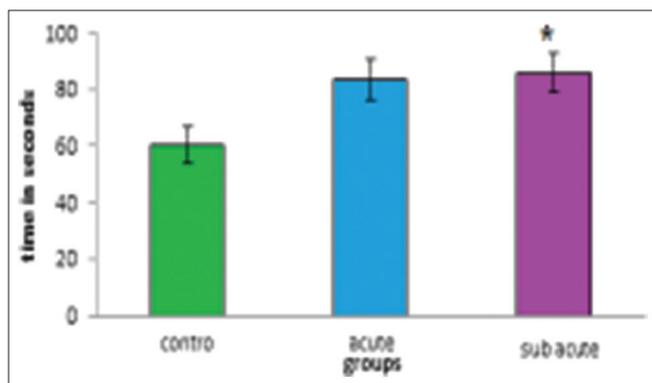


Fig. 6: T-Maze

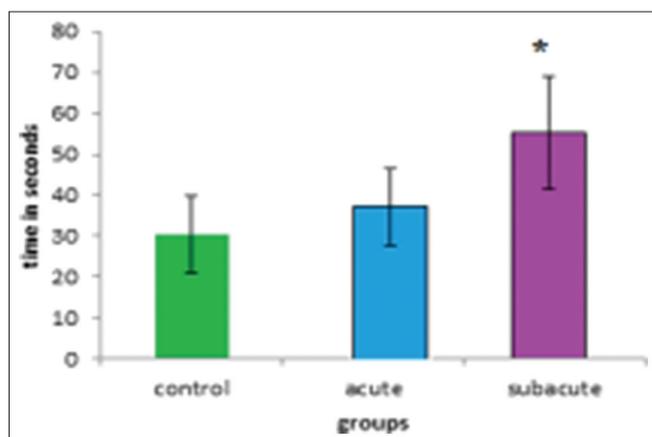


Fig. 7: Y-Maze

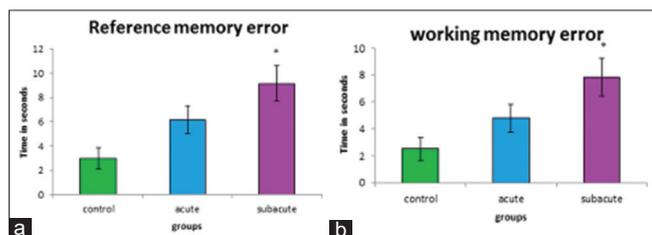


Fig. 8: Radial Eight-Arm Maze

Number of working memory errors (re-entries into already visited baited arms)

Fig. 8b number of working memory errors in eight arm radial maze has been summarized: Working memory error increases acute and sub-acute noise stress when compared with control. Control is significant to acute and sub-acute noise stress. Results shown are Mean±S.D when compared to control; the symbol represents statistical significance *p<0.05.

DISCUSSION

Stress has an opposing effect on memory consolidation and memory retrieval. There are many studies associated with consistent stress in animals leading to atrophy of the hippocampus, which plays a very important role in learning and memory. Learning new spatial associations could be a process highly protect to the effect of stress impairment within the hippocampus is related to defective spatial mentality [16]. This study shows that there is an impaired spatial mental capacity within the acute and sub-acute noise, stress, group in comparison to the control group which can be a result of noise-induced atrophic changes within the hippocampus in memory, experimental

Table 1: The dependent variables recorded are listed below

Variables	Definition
Working memory errors	Number of re-entries to an arm previously visited during the same trial
Reference memory errors	Number of entries into unbaited arms
Total memory errors	Number of working+reference memory errors
Path length	Distance travelled in centimeters
Velocity	Distance (cm) divided by total time (s)
Test session time	Total time taken to complete test (s)
Percent of time spent immobile	Time spent immobile (s) divided by test time (s)

performance is gathered within the type of information [9]. Thereby animals can perform well within the post-period. Whereas just in case noise stressed group there is impairment within the hippocampus thereby it alters the memory performance in these groups. Memory is also involved within the acquisition and recall of personal memory [17].

This study also shows that there is an increased remembering error in noise stressed group in comparison to regulate group animals because of impairment in prefrontal cortex function [10]. Duncko *et al.*, 2007 detected the possible involvement of memory in improved learning performance after exposure to cold pressor assay [4]. Exposure to cold pressor stress is related to signs of both enhanced and impaired remembering performance [18]. A stressful condition produces to the excessive production of free radicals, which results in oxidative stress [19]. In animal's hippocampus atrophy and associated memory deficits in stress and aging have implications for stress and aging in humans [10].

The accuracy of EEG mapping by examining the association between electrical activity and therefore the perfusion under each electrode as another measure of local cerebral function [6] the increased neither central nor adrenergic activity is related to impairment of prefrontal cortex functions involved in remembering performance [20]. Acute stress in humans disrupted functional brain activity and connectivity in fronto-parietal networks [21] chronic stress-induced depression, suggesting that microglia stimulators could function fast-acting anti-depressants in some sorts of depressive and stress-related conditions [22]. After noise-stress exposure, working and reference memory error increased [14].

Similar findings are noticed during this study even in 15 days of noise stress exposure. Sandi and Pinelo-Nava, 2007 explained that Glucocorticoids were found as standard mediating mechanisms for both the facilitating and impairing actions of stress in several memory processes and phases [23]. Shukitt-Hale *et al.*, 2004 rats made more errors when solving the eight arms radial water maze and specifically more reference memory errors and dealing memory errors [5]. Lupien *et al.*, 1994 explained that increasing cortisol excretion was related to declines in memory performance among the ladies [24]. In 2005 Elzinga and Roelofs was reported by a number of the pharmacological studies where high dose of cortisol was related to impaired spatial performance [16]. Theta frequency is smaller during slow wave sleep and awakening than during other waking behavioral states. The dorso ventral theta coherence increases within the order of slow wave sleep, awake-immobility,awakening, grooming [4]. In 2009 de Dovic *et al.*, has found that acute stress in humans disrupted functional brain activity and connectivity in fronto parietal networks [25].

Lupien *et al.* 1994 explained that increasing cortisol excretion was related to declines in memory performance among women [7]. In 2005 Elzinga and Roelofs was reported by some pharmacological studies where high doses of cortisol were related to impaired performance [26]. In the studies show that increasing the frequency amplitude in acute and sub-acute noise stress. EEG activity Spectral

power was less for the exercise compared to the control group within the delta band, but greater altogether other bands. The utmost power of brainwave appeared within the occipital region. Up to the 50th min after noise exposure, over two average power of EEG maximums of cortical potential appeared, and therefore the time points of maximum occurrence shifted forwards slowly following the rise of exposed noise frequency. The interval between 2 time points of maximum occurrence was reduced with the rise of the exposed noise frequency.

Bhoria 2012 explained that music of 100 dB sound level was applied alpha band power decreased, and beta band power increased [27]. Hence, the results of study showed that because the loudness of music increases to 100 db absolutely the power of beta band increases and absolute power of alpha band decreases. The brain recognizes the sound levels and discriminates the strain level. The brain is that the key organ involved in interpreting and responding to potential stressors [28].

Summation of potentials derived from the mixture of extracellular currents generated by populations of neurons is recorded as EEG [23]. Chronic stress is also reliably assessed by relative high beta EEG power at anterior temporal sites [29]. These results are in accordance noticed the possible involvement of remembering in improved learning performance after exposure to cold pressor check [30]. On the contrary, other studies testing the effect of acute stress exposure on remembering used the Trier Social assay or intense naturalistic stressors known to be related to hypothalamic-pituitary-adrenal axis activation at substantially higher level than the cold pressor assay [31].

CONCLUSION

Stress is ubiquitous and universally pervasive; however, its objective quantification has not been easy. In modern life, statistics show powerful effects of stress early in life, concurrent chronic stress, and socioeconomic status on both the morbidity and mortality of chronic disease. Present study is help us to bring out the varied changes within the EEG wave pattern particularly changes within the amplitude and frequency and behavioral performance of the noise stressed group thereby it reveals the noise induced effect in these groups. Further studies are recommended to prove the precise mechanism behind these changes within the EEG wave patterns. It is our duty to require necessary steps to forestall this noise stress which could help us to forestall of these pathological state changes.

AUTHOR'S CONTRIBUTIONS

The corresponding author has designed the work and critical revision of the manuscript. The first author carried out the behavioral assessment and paper writing.

CONFLICTS OF INTEREST

The authors declare that they no conflicts of interest concerning this research article.

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