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Ischemic Stroke and its Rehabilitation by low dose Direct Current Electromagneto Therapy

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Abstract*— Ischemic stroke is a neurological disease which ranks third as leading cause of death after heart disease and cancer. The present work reveals that electromagnetic stimulation may bring improvement after ischemic insult. We investigated the effects of low direct current electromagnetic field (DCEMF) i.e. below 600 Gauss, exposure on ischemic injury in a rat model of focal ischemia (15 minutes occlusion followed by reperfusion). An Electromagneto therapy unit was designed for the purpose. Ischemia induced rats were exposed to electromagnetic field (300-400 Gauss) for 1hour daily for 10 consecutive days. Exposure was initiated 20 minutes after the onset of ischemia and continued throughout reperfusion. Behavioral tests and Electroencephalogram (EEG) Signal analysis were used to measure the degree of ischemic injury. Exposure to the field demonstrated a clear rise in the rhythms of the EEG waves of the fronto-parietal region which had been attenuated due to onset of ischemia (P < 0.05). On histological examination, low DCEMF exposure reduced ischemic neuronal damage. Thus the preliminary data suggests that exposure to low DCEMF may have implications for the rehabilitation of ischemic neuronal insult in animal model.

Index terms - Electromagnetism, EEG, Stroke

I. INTRODUCTION

At its most basic level, the human body consists of cells, which are polarized. All cells contain various concentrations of positively and negatively charged ions. Furthermore, all molecules are made up of atoms consisting of electrons and protons. This basic understanding of the cellular and atomic structure of the human body suggests that the human body is an electrical machine. When considering the structure of the human body and the laws of physics, one can also assume that electricity and magnetism can alter the function of the human body. This simple relationship between the basic structure of the human body and the power of electricity and magnetism has provided a great deal of excitement in the fields of bio-magnetic and bio-electromagnetic research [1]. Bio-electromagnetic Therapy is the application of electromagnetic fields to treat and prevent disease and to promote health and longevity. Electromagnetism is a powerful clinical tool, simple to apply yet complex in its biological effects. Whenever current (I) passes through a wire, it induces a magnetic field (B). Although the electricity itself remains confined within the wire, the magnetic field induced moves outside. When a varying electromagnetic field is placed close to a conductive medium such as the human body, it will induce electrical currents [2]. A thorough review of the literature reveals that electro magneto therapy tends to have four major effects on the human body i.e. vasodilation, sensory reception, and change in hormone secretion and increase in cellular energy.

II. MATERIALS AND METHODS

A. Study design and the experimental set up

The Electro magneto Therapy Unit

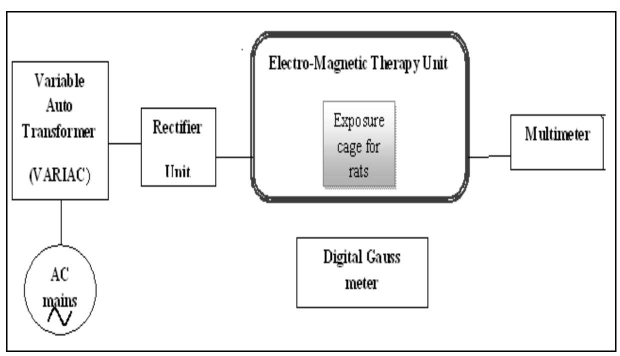

Figure 1. Schematic block diagram of the experimental set up

- The Magnetic Bed

The apparatus for exposing the rats in magnetic field is assembled in our laboratory. The setup compromises of two coils wound on bobbin of transformer [3]. Dimension of bobbin is 17.5 x 15.5 x 12 cm3, with a core gap of 7.7 x 7.7 cm2. Entire assemblies are mounted on a wooden base of dimension 63.5 x 27.5 cm (Fig 1). Height of wooden base is kept 5 cm from ground to avoid water/ chemical exposure. Wooden base facilitates rigid support to the structure and eliminate risk of shock hazard for user. The bobbin used here is nylon glass field 30% material. It is a thermo set polymer with temperature withstanding capability > 700 0F.

The bobbin was filled with ferrite core, which is used for both low power and high power transformer. 21.5 Kg of core was used to fill each core (total of 43 Kg for both cores). The core used was an ‘E’ shaped core with central leg of 7.7 cm and side leg of 3.8 cm. The two bobbin are filled with ‘E’ shaped core were placed facing each other on the wooden base. Two wooden rails of dimension 38.7 X 4.9 X 3.8 cm3 were nailed at distance of 15.5 cm on the wooden base to provide a firm support to side legs of both opposite facing core. It is also used as heat insulator. The side legs of two opposite facing cores were joined by ‘I’ shaped ferrite material of 15 cm in length. The ‘I’ shaped Ferrite is used to complete the magnetic circuit and to reduce the reluctance for flux path. The ‘E’ and ‘I’ shaped ferrite core are available in market for making transformer (Figure 2).

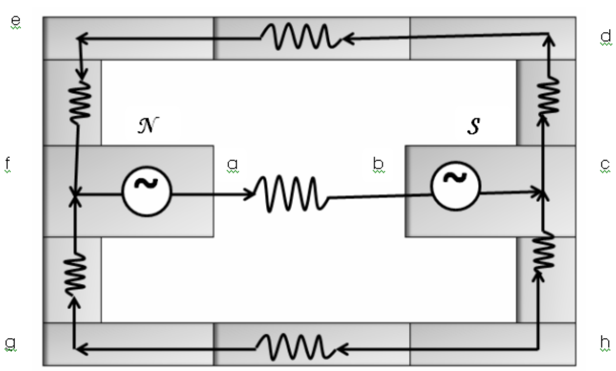

Figure 2. Magnetic circuit of magnetic bed

The cooling mechanism provided for electromagnet cooling is air cooling. The wire used for winding was SWG 16 gauge wire with diameter 0.064 inch (1.626 mm). (The maximum safe current to be flown in SWG 16 gauge wire is ~13Amp (by rule of 1 Amp/ 300 circular coils, for long wire)). Autotransformer provides a variable voltage over specified range. The overall dc resistance of both coil connected in series was 9.85 Ω.

- Exposure cage

The exposure cage used to expose rat was made in the laboratory from Perspex sheet with the dimension of 16.5 X 10.7 X 4.3 cm. The box was so close so as to provide isolation to animal from stray currents generated within the unit and restrain it within the magnetic premises. The plastic box was provided with many holes of 0.6 cm in diameter to facilitate air flow during the stipulated exposure time. The following tests were performed prior to experiment.

1. Continuity testing: Continuity of the coil was tested with the Simpson multi- meter in order to ensure constant flow of current.
2. Resistance testing: Static resistance of the coil was tested in order to calculate power requirement.
3. Insulation testing: Insulation testing was done in order to ensure safety for user and animal All the connection and joints were shielded with high insulator PVC tape. Testing was done at 10 Amp and results were satisfactory.

- Flux Vs Current

Flux versus current was calculated at center position of exposure cage by flux meter.

- Flux Vs Time

Flux versus time graph was measured to record variation of flux with time. Time was measured through a stop watch and flux is measured at 2 minutes interval over 22 minutes.

- Temperature Vs Time

Rise of temperature beyond certain limit is dangerous for the animal. Temperature of the setup rises due to flow of current in the coil. This is in turn due to Impedance of coil. Rise of temperature of coil increases the resistance of copper and thus further increases temperature. Temperature over 160 minutes was recorded at different flux densities at center of exposure cage.

B. The animal (rat) model of focal ischemia

Focal cerebral Ischemia was induced by occlusion of the middle cerebral artery (MCA) using a modification of the intraluminal technique [4]. Rats were anesthetized with ketamine (75mg/kg i.p) with xylazine (10 mg/kg i.p.) and maintained a constant body temperature of 37+- 0.5ºC. The left common carotid artery (CCA) was exposed through a midline incision in the neck region. The neck muscles were separated further to expose external carotid artery (ECA) and internal carotid artery (ICA). A 4.0 cm length 3-0 monofilament nylon suture (Ethicon) was introduced into the ECA lumen through a small nick and gently advanced from to the ICA lumen to block the origin of MCA. The approximate length of filament inserted near the bifurcation point to the MCA blockade site was about 18-22mm.The ECA stump was tightened by tread around the intraluminal nylon suture to prevent bleeding. Reperfusion was allowed by gently removing the monofilament after 15 minutes of ischemia. In the control animals, all procedures except for the insertion of the nylon filament were carried out. Animals were allowed to recover from anesthesia and on regaining the righting reflex, were transferred to cages in the animal room, with temperature maintained at 26 +- 2.5ºC. Animals were provided food and water ad-libitum.

C. Different behavioral tests and analysis

- Rota rod test

To assess effects on motor coordination, rats were trained to remain on a rotarod. All rats underwent a 3-day training program on the Rota rod apparatus. During the training period, each rat was placed on the rotarod at a constant speed (rotations per minute; rpm) for a maximum of 180 sec, and the latency to fall off the rotarod within this time period was recorded [5]. Rats received four trials per day for three consecutive days, by which time a steady baseline level of performance was attained. The test consisted of two trials at 5 increasing speed levels, ranging from 10 rpm to 25 rpm. The mean latency to fall of the rotarod at each speed level was recorded. The same training procedure was used for the animals that endured stroke. All animals were trained for 3 days, and were then tested at increasing speeds the day after surgery [6].

- Narrow Beam Walking Test

Beam walking was used as a measure of hind limb coordination by means of distance travelled across an elevated 100 cm beam (2.3 cm in diameter, 48 cm off the floor). Rats were systematically trained to walk along the elevated beam from start to finish with the aim of completing the task in 3 minutes. A safe location, i.e., a flat box, was placed at the end of the beam so that the rat was motivated to complete the task. Each rat was trained twice daily for a maximum of 3 minutes per trial for 3 consecutive days [7].

- Hang test

Neuromuscular strength was determined on the seventh day in the grid hang test. Rats were lifted by their tail and slowly placed on a horizontal grid (grid 12 cm2 opening 0.5 cm2) and supported until they grabbed the grid with both their fore and hind paws. The grid was then inverted so that the mice were allowed to hang upside down. The grid was mounted 20 cm above a hard surface, to discourage falling or injury in case of falling. The apparatus was equipped with a 3-inch wall to prevent animals from transverse to the upper side of the grid. Animals were allowed to stay on the grid on seventh day for 30 s and 10 chances were given with 1min interval and the best fall values were recorded [8].

- Histology

After neurological test, rat brain was perfused with 4 % PFA by transcardiac perfusion and isolated in chilled conditions. The cerebellum was removed and rest of the brain was immediately transferred at -20ºC. Frozen brain was cryo sectioned. The brain sections obtained with infarct were quantified by image analysis software. Further, infarct volume was calculated by linear integration of the infarct area of each slice multiplied by average thickness of brain section and expressed in mm3 [9].

- EEG Signal Acquisition and Analysis

EEG activity was measured in each rat under anesthesia immediately before MCAO, immediately before reperfusion at 20 minutes after occlusion, and again after 10days before euthanasia. EEG data were recorded for up to at least 30 min each time. This enabled us to establish an experimental exclusion criterion and to determine the effect of Electromagneto therapy to improve EEG activity in injured rats. Changes in EEG amplitude were quantified with the use of spectral analysis [10].

- Statistical Analysis

Data analyses were performed using Origin 8 and the Statistical Product and Service Solution 16.0 software package for Windows (SPSS Inc., Chicago, Illinois, USA). Behavioral data, volume of ischemia and EEG parameters were considered as continuous variables, stroke risk factors and outcome measures were dichotomized for analysis. Significance level was set at $p < 0.05$ and where appropriate lowered to avoid type I error inflation in multiple parameter testing.

III. RESULTS

A. Flux Vs Voltage

The Graph of Flux Vs Time shows variation of Flux over time is negligible. Thus there is a linear relationship between linear increments of Flux with increase in Voltage (Figure 3).

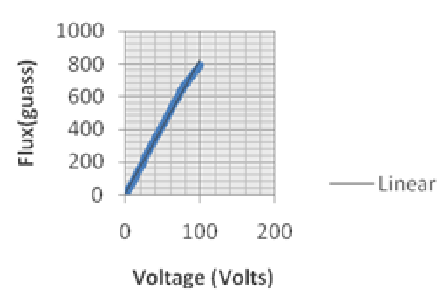

Figure 3: Flux Vs Voltage

B. Flux Vs Time

It is seen that the Flux lines obtained within the exposure cage is more or less static and so it can be termed as DCEMF and can be used as a site of therapy (Figure 4).

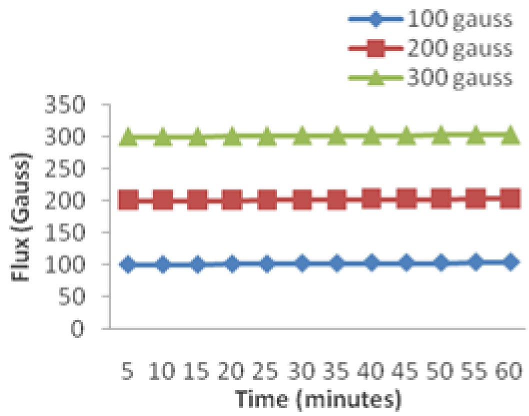

Figure 4: Flux Vs Time

C. Temperature vs. Time

The Graph revels that exposure below 400 Gauss can be given without causing any physiological damage to the animal as it are within the safe range (Figure 5).

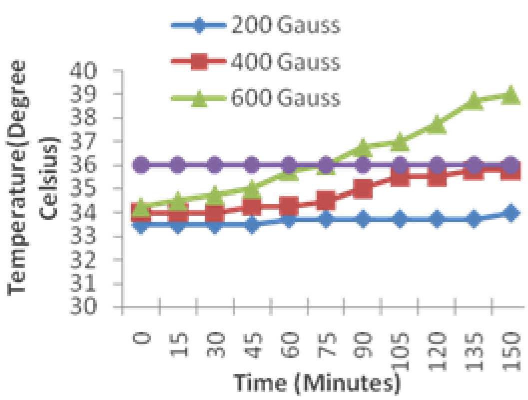

Figure 5: Temperature Vs Time

D. Rota rod Test

The time of retention on Rota rod at different RPM is greater in the focal ischemic rat model exposed to Electromagneto therapy. There is not much of a difference in control rats exposed to and not exposed to the therapy. At RPM=10, 15, 20 and 25 level recovery is found to be around 28%, 25%, 20% and 18% respectively. The significance was found to be $p<0.05$ (Figure 6).

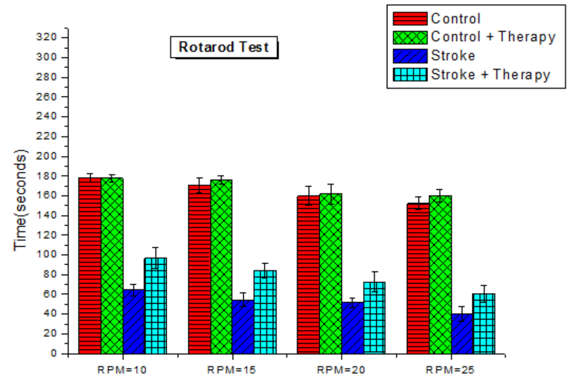

Figure 6: Graph of Time of Retention Vs different RPM levels expressed as mean ± SD

E. Narrow Beam walking Test

It was noticed that the rat that was exposed to the therapy took much lesser time in crossing the narrow beam as compared to them that were not exposed after focal ischemia.
The level of recovery was found to be 48% and $p>0.05$ significant (Figure 7).

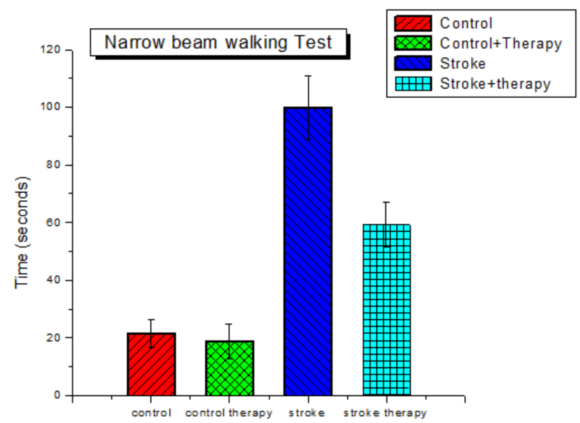

Figure 7: Graph of narrow beam walking test expressed as mean ± SD

G. Hang Test

The time of retention on the grid in upside down position for the stroke model of rat exposed to therapy was greater in comparison to the rat having focal ischemia without therapy. The hang time significantly decreased ($p<0.05$) in stroke induced group when compared with the control group. The treatment Group showed a marked improvement in hang time when compared with stroke group ($p<0.05$). The level of recovery was 43% (Figure 8).

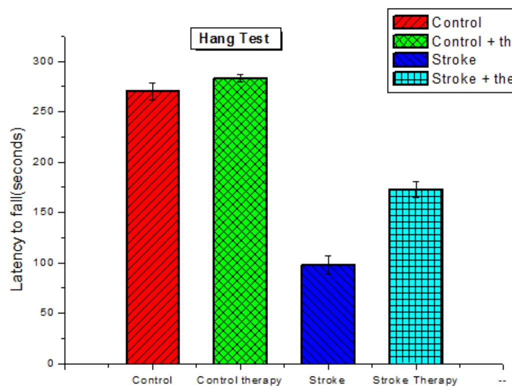

Figure 8: Graph of latency to fall from the grid in hang test expressed as mean ± SD

G. Histology

The coronal brain sections obtained and their further infarct analyses showed that there is no infarct in control rats and therapy exposed control rats, but the infarct size in the stroke model exposed to therapy has a reduction in the infarct volume as compared to the only stroke model (Figure 9). The result showed a recovery of 18%

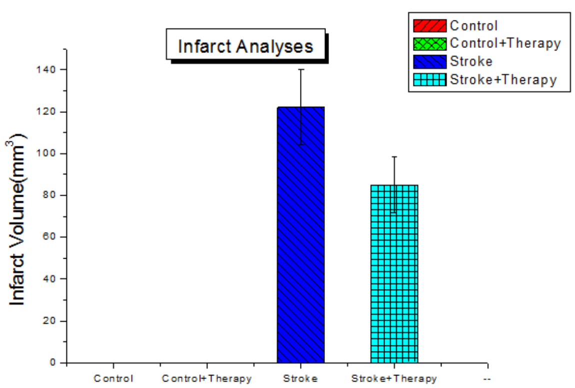

Figure 9: Graph of infarct volume found expressed as mean ± SD

H. EEG of Fronto-parietal region analyses

It was observed that Delta activity increased in rats those were kept at low static magnetic field relative to control rats and decreased remarkably in stroke induced rats relative to control. On treating depressed rats with low static magnetic field, Delta activity was increased relative to the rats which were not given therapy. A recovery of 32.5% was found at a significance of $p<0.05$. Theta activity was also decreased in stroke induced rats relative to control rats. On treating, Theta activity was seen to increase around 11.7% and significance of $p<0.05$. Alpha and beta rhythms were highly decreased in stroke induced rats and is hardly visible but on exposure to the electromagnetic radiation, their activity was noticeably increased.). This clearly states that recovery toward normal Alpha activity Therapy was very high relative to untreated rats. Recovery levels were found to be 42% and 26% respectively and were significant as well. Not much of a difference was noticed in the activity of different waves between control rats and the control rats that was exposed to Electromagneto therapy (Figure 10).

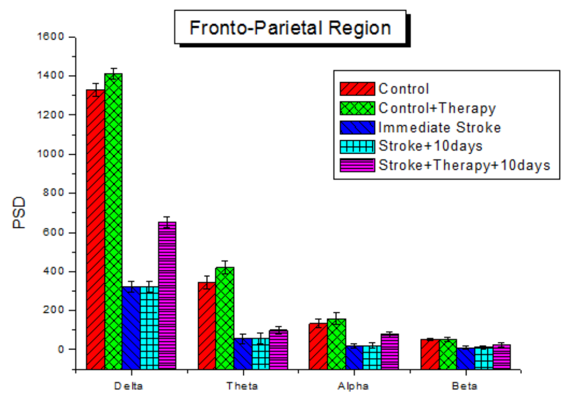

Figure 10: Graph of PSD values of Fronto-parietal region Vs different rhythms expressed as mean ± SD

I. EEG of Occipital region analyses

It was observed that Delta activity was highly increased in rats those in which stroke were induced relative to control. On treating depressed rats with low static magnetic field, Delta activity was decreased relative to the rats which were not given therapy. A recovery of 20% was found at a significance of $p<0.05$. Theta activity was also increased in stroke induced rats relative to control rats. On treating, Theta activity was seen to decrease around 39.5% and significance of $p<0.05$. Alpha and beta rhythms were decreased in stroke induced rats, on exposure to the electromagnetic radiation, their activity was increased but not much significant change is noticeable. Recovery levels were found to be 4.1% and 8.6% respectively and were thus not very significant. Not much of a difference was noticed in the activity of different waves between control rats and the control rats that was exposed to Electromagneto therapy (Figure 11).

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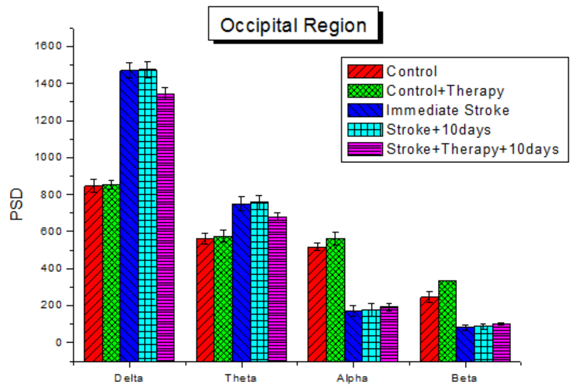

Figure 11: Graph of PSD values of Occipital region Vs different rhythms expressed as mean ± SD

J. EEG of Temporal region analyses

It was observed that Delta activity was highly increased in rats those in which stroke were induced relative to control. On treating depressed rats with low static magnetic field, Delta activity was decreased relative to the rats which were not given therapy. A recovery of 21% was found at a significance of $p<0.05$. Theta activity was also increased in stroke induced rats relative to control rats. On treating, Theta activity was seen to decrease around 18% and significance of $p<0.05$. Alpha and beta rhythms were decreased in stroke induced rats, on exposure to the electromagnetic radiation, their activity was increased but not much significant change is noticeable. Recovery levels were found to be 22.6% and 3.8% respectively. They were not that very significant. Not much of a difference was noticed in the activity of different waves between control rats and the control rats that was exposed to electromagneto therapy (Figure 12).

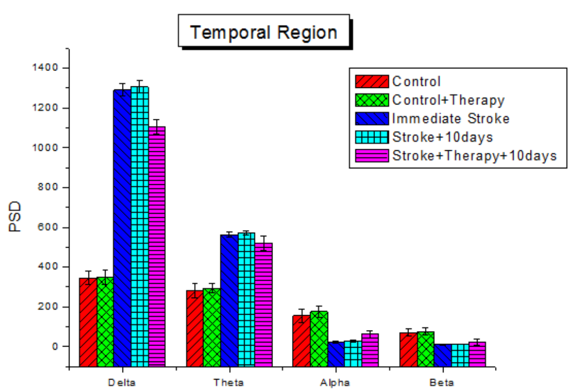

Figure 12: Graph of PSD values of temporal region Vs different rhythms expressed as mean ± SD

IV. DISCUSSION

The Graph of Flux Vs Time shows variation of Flux over time is negligible. Thus there is a linear relationship between linear increments of Flux with increase in Voltage. The Temperature Vs Time Graph revels the exposure below 400 Gauss can be given without causing any physiological damage to the animal as it is within the safe range. One can also expose the animal to 600 gauss for 60 minutes without harming it. It is seen that the Flux lines obtained within the exposure cage is more or less static and so it can be termed as DCEMF and can be used as a site of therapy.

Electromagnetic Therapy gives positive result in rehabilitation Focal Ischemia in behavioral testing compared to untreated stroked rats. There is an increase in the latency time of fall on the Rota rod performance test, decrease in the time to cross the required distance in narrow beam walking test and increase in the time of hanging from the grid in the hang test.

In the histological analyses it was found that there was a slight decrease in the infarct volume in therapy treated stroke rats.

There are specific changes in EEG. Stroked rats showed very high decrease in all the rhythmic activities of fronto parietal region and therapy treated rats showed very significant recovery toward achieving normal rhythms, by increasing the larger portion of decreased rhythms compare to untreated stroked rats. Stroked rats showed increase in the delta and theta rhythmic activities of occipital region, and there was noticeable difference in activities after therapy. But in case of beta and alpha rhythms not much noticeable conclusion can be drawn. Stroked rats showed increase in the delta and theta rhythmic, while there is a decrease in activities of alpha and beta was in the temporal region. There is an improvement seen in the same of therapy treated rats.

V. CONCLUSION

Electromagneto therapy is a non-invasive technique. It may be a very useful assistive modality to restore limbs movement along with conventional therapy after stroke. The results of this study suggests that the Electromagneto therapy is a promising therapeutic technique as is seen in stroke induced rats. The behavioral tests and histology showed significant positive changes after Electromagneto therapy intervention. The EEG analysis showed some improvement in the brain activity as well. Thus we can say that Electromagneto therapy seemed to be a good assistive treatment modality which can be given to get better results in stroke rehabilitation.

The study was conducted on rats, hence the effect on humans cannot be properly specified. Further detailed analysis should be done in finding out a safe limit of human exposure to electromagnetic field so that we can quantify the rehabilitative benefits of Electromagneto therapy in stroke patients and in their recovery process.

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