

ANTIEPILEPTIC DRUGS DELAY THE ONSET OF SEIZURES INDUCED BY AMINOPHYLLINE IN CONSCIOUS RATS

CHANDA KULKARNI*, THANGAM JOSEPH AND JOY DAVID

Department of Pharmacology,
St. John's Medical College,
Bangalore - 560 034

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Abstract : Aminophylline, 285.7±2.19 mg/kg infused intravenously in unanaesthetized rats produced onset of seizures within 3.2 ± 0.99 minutes. Seizures were repetitive and death occurred in 10.5 ± 1.75 minutes. Pretreatment of rats with carbamazepine, sodium valproate and diazepam at doses that prevented electroshock induced seizures were effective in significantly postponing seizures and death, but did not reduce mortality. Concomitant EEG studies in aminophylline infused rats showed that cortical excitability evidenced by initial cortical spiking occurred at 42 secs and polyspiking at 165 seconds. Following diazepam, the initial cortical spike was delayed 50 fold, appearing after 36 minutes. Antiepileptic drugs and EEG monitoring may prove useful in patients with status asthmaticus receiving intravenous aminophylline.

Key words : aminophylline seizures carbamazepine sodium valproate
diazepam pentobarbitone EEG rat

INTRODUCTION

A serious complication associated with intravenous aminophylline therapy for bronchial asthma is convulsions. Such seizures are difficult to control particularly in adults (1). In eight neurologically normal patients, intravenous aminophylline therapy (peak serum theophylline concentration of 53 ± 4.8 µg/mg) for status asthmaticus, induced focal or generalized seizures. These seizures occurred without any prior symptoms and were refractory to intravenous diazepam, diphenylhydantoin and phenobarbital (1). In subjects with brain impairment and coexistent bronchial asthma, seizures occurred at significantly lower serum theophylline levels (2). These repetitive, generalized seizures are commonly fatal and no specific, pharmacologically antagonistic drug is known (3). Barbiturate induced general anesthesia has also been used to control such intractable seizures (4). This experimental study was therefore undertaken to assess the hitherto undetermined effect of antiepileptic drugs, sodium valproate and carbamazepine in preventing aminophylline induced seizures in conscious rats. The effect of pentobarbitone as a general anesthetic was also

studied. Diazepam was the standard reference drug.

METHODS

Aminophylline (250 mg/10 ml, Harson Laboratory) was given intravenously to unanaesthetized Wistar rats, 200-230 g of either sex, using the method of tail vein infusion. A 24G needle from a scalp vein infusion set was inserted into the lateral tail vein. A stop watch was started to time the infusion at the instant the column of blood entered the needle and was stopped at the onset of tonic extension of the hind limbs. This method ensured that the time taken to seizure and the dose of aminophylline required to induce the seizure was obtained within relatively narrow limits. Since aminophylline induced seizures were invariably fatal, different groups of rats (n=10) were used for the control and those pretreated with drugs.

Aminophylline was administered intravenously over 2 to 3 min, uptill a dose level which would result in repeated seizures and ultimately, death. In drug treated groups, diazepam, 10 mg/kg, ip (reference drug), carbamazepine, 10 mg/kg, ip and sodium valproate, 300 mg/kg, ip were given 45 min before the

*Corresponding Author

challenge dose of aminophylline. In some rats, diazepam 2 mg/kg, iv was given 15 min before aminophylline. In previous experiments, these doses of afore mentioned antiepileptic drugs were shown to afford 100% protection to rats subjected to maximal electroshock, (150 mA for 0.2 sec) 45 min after their administration (5). The parameters determined in this study were: time taken to the: (a) onset of generalized convulsions and (b) duration of intermittent seizures till the occurrence of death, these could serve as visual indices of pharmacological antagonism of aminophylline induced seizures.

Effect of pentobarbitone on aminophylline induced seizures was studied separately on a group of four rats. Aminophylline challenge was immediately attempted when pentobarbitone 25 mg/kg, ip produced anaesthesia accompanied with loss of righting reflex. The resultant effects were observed.

For EEG studies in some rats, two pairs of stainless steel screw electrodes were placed overlying the dura in both centroparietal areas (see electrode montage diagram in Fig. 1) under pentobarbitone anaesthesia, (35 mg/kg, ip). The reference electrode was secured in the frontal sinus. Two weeks were allowed for recovery from surgery, prior to continuous EEG monitoring during drug antagonism studies. For bipolar recording of EEG activity with a Grass Model 7 Polygraph, rats were placed in suitably

shielded plastic holders. Baseline EEGs were recorded for 10 to 20 min and thereafter, following aminophylline infusion. The latency to: (A) the initial cortical spike and (B) repetitive polyspiking, heralding visible seizures were determined. These were considered EEG indices of aminophylline induced cortical excitability. Prolongation of spike latencies for (A) and (B) were assessed only in rats treated with diazepam, 10 mg/kg, ip. Statistical analysis was done using Student's 't' test.

RESULTS

Effect of iv aminophylline in rats : Aminophylline produced tonic extension of hind limbs caudalwards, associated with hyperacousticity and intermittent gasping. The onset of seizures following aminophylline occurred at a baseline value of 3.2 ± 0.99 min at a dose of 285.7 ± 2.19 mg/kg, iv. Bouts of generalized seizures followed and lasted till death due to persistent apnoea ensued at 10.5 ± 1.75 min. The reference agent diazepam, 10 mg/kg, ip, significantly prolonged the onset of aminophylline seizures 12 fold, from baseline values to 38.5 ± 9.07 min ($P < 0.01$), sodium valproate was equieffective with diazepam ($P < 0.001$), while carbamazepine was the most effective agent prolonging the onset of seizures 32 times, from baseline values to 104.1 ± 7.27 min ($P < 0.001$). Carbamazepine and sodium valproate delayed seizures without any behavioral change

TABLE I : Effect of antiepileptic drug pretreatment on aminophylline induced seizures in rats.

Drug and Route	n	Dose mg/kg	Time of Onset of seizures (min)	% Dead time of death*
Aminophylline, iv	10	285.7±2.19	3.2±0.99	100% in 10.5 ± 1.75 min
Aminophylline after pretreatment with:				
Diazepam, ip	10	10	38.5±9.07*	30% in 1 hr 70% in 24 hr
Diazepam, iv	4	2	58.66±45.36**	50% in 1 hr 50% in 24 hr
Sodium valproate, ip	10	300	42±9.17**	20% in 1 hr 60% in 4.5 hr 20% in 5 hr
Carbamazepine, ip	10	10	104.1±17.27**	20% in 1 hr 80% in 24 hr

*± SEM : * $P < 0.01$, ** $P < 0.001$

*: Duration of seizures preceding death.

whereas similar protective effects with diazepam occurred with drowsiness, hypotonia and ataxia. None of the antiepileptic drugs were able to prevent death and could only reduce the seizure intensity and prolong intervals. 20% 30% of animals pretreated with the different antiepileptic drugs died within one hr and the rest died in 24 hr, including 50% rats following intravenous diazepam (Table I).

The effect of pentobarbitone-induced general anesthesia on aminophylline-induced seizures : Pentobarbitone in a dose of 25 mg/kg, ip, produced loss of corneal reflex, pain sensation and abolition of righting reflex within 5-6 minutes. Following the administration of iv aminophylline, one rat showed instantaneous convulsions and died in 1 hr. The remaining three rats exhibited bouts of spontaneous intermittent convulsions as well as seizures precipitated by auditory and tactile stimulation. These rats died in 12-21 hours.

EEG studies in rats: Fig. 1 shows the EEG with baseline low voltage activity, 17 to 18 Hz, in the

conscious, minimally restrained rat. Immediately following aminophylline infusion (arrow in Fig. 1) fast activity, 22-24 Hz was seen and the initial cortical spike occurred at a latency of 42 sec. Thus the first spike in the EEG appears 2.5 min earlier than the visible onset of seizures (3.2 min). Thereafter, intermittent spiking, from all cortical leads interspersed with bursts of sharp waves, often accompanied by jerks of body and limbs were seen. The latency to polyspiking was 165 sec and was associated with repeated myoclonic jerking followed by repetitive seizures and high frequency and amplitude EEG epileptiform discharges. The EEG showed isoelectricity when apnoea preceded death. Following diazepam, 10 mg/kg, ip the initial spike appeared after 36 min, about 50 times prolonged from baseline of 42 seconds. Polyspikes were seen after 1 hr.

DISCUSSION

The present study shows that aminophylline given intravenously to conscious rats induces status epilepticus like condition, terminating fatally. Meth-

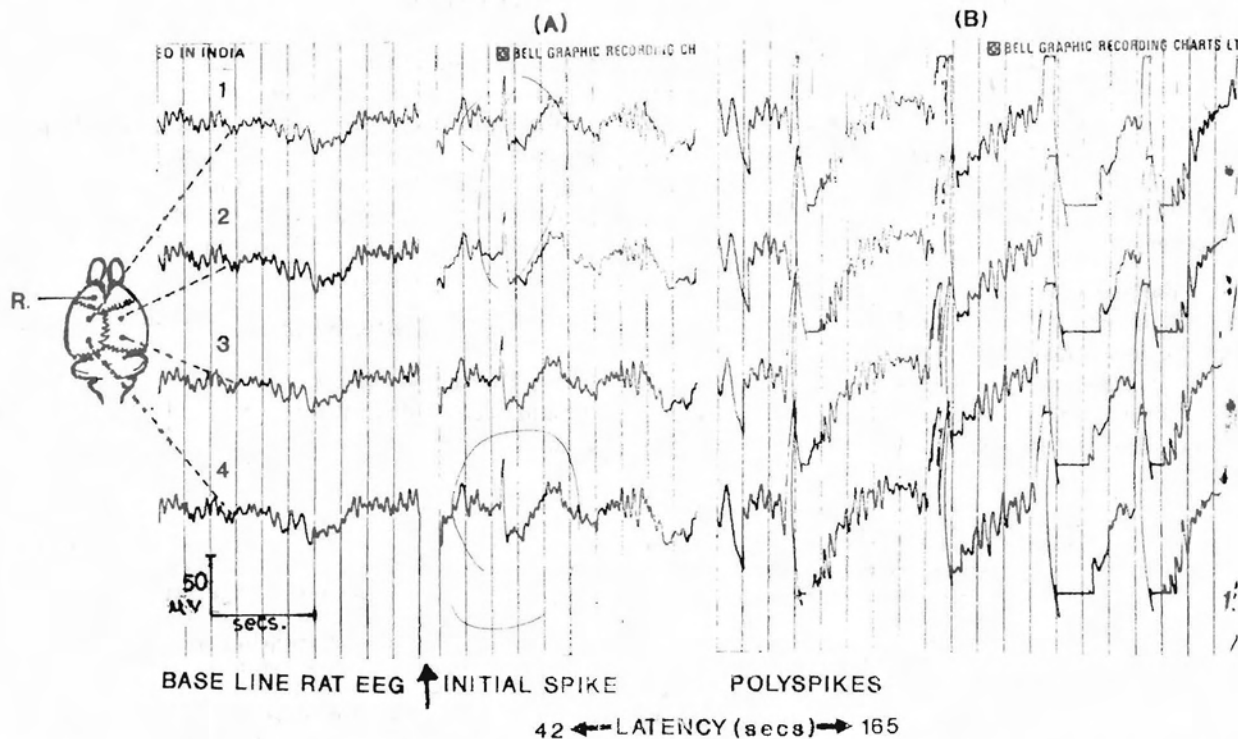


Fig. 1 : Electroencephalogram(EEG) of rat showing baseline EEG and latency (secs) to (A) initial spike and (B) polyspikes following intravenous aminophylline (↑). R : common reference electrodes; and 1-4 represent cortical areas in the electrode montage diagram.

ylxanthines such as aminophylline, is reported to stimulate all parts of the CNS, provided that high enough brain/CSF concentrations are attained (3). At low concentrations of infused aminophylline, there were no associated behavioural changes but cortical excitability was evidenced by fast activity in the rat EEG, 22-24 Hz, and intermittent spiking. At higher concentrations, there was stimulation of the medullary respiratory center, as seen by symptoms of gasping, anoxia and apnoea (3). Epileptiform polyspikes and high voltage, fast frequency discharges in the EEG coincided with behavioral convulsions.

Diazepam has remained the drug of first choice for status epilepticus (6) after it was first introduced for the treatment of this condition by Gastaut et al (7). The rapid equilibration of diazepam in the brain led to immediate control of EEG and motor symptoms of epileptic activity. The use of diazepam and other antiepileptic drugs for aminophylline induced seizures in experimental animals (8) as well as in patients with status asthmaticus who had developed seizures following intravenous aminophylline (2,9) has not always been successful. In rats Walker (8) produced repeated seizures with aminophylline, 351 ± 62 mg/mg, iv and could elevate seizure threshold about two fold with diazepam, 10 mg/kg, ip. However at this high dose, diazepam produced marked behavioral somnolence and ataxia. In our study, diazepam, 10 mg/kg, ip also produced drowsiness and ataxia and significantly delayed the onset of seizure twelvefold but did not reduce mortality.

Barbiturate induced general anesthesia has been reported to be one of the modes of successful treatment for intractable status epilepticus (4). However, in our study pentobarbitone anaesthesia failed to protect rats against aminophylline induced seizures. Though the mortality was prolonged by 12 to 24 hr, in 75%

of the animals the protection against seizure episodes provoked by auditory or tactile stimulation was absent. Further, these seizures following the drug interaction appeared to resemble those induced by strychnine.

Interestingly, EEG studies in diazepam pretreated rats showed that the initial cortical spike was delayed 50 times (42 sec to 36 min), compared to the aminophylline infused group. Therefore EEG monitoring may be a valuable predictor of seizure predisposition to aminophylline in animals and humans. There was however no differentiation for EEG polyspikes and visible seizures as both appeared almost simultaneously.

Carbamazepine was the most effective agent in antagonizing the aminophylline induced seizures in rats and delayed the onset of seizures thirtytwo fold, while sodium valproate was as effective as diazepam. Sodium valproate was the least effective in reducing mortality while carbamazepine was superior to diazepam. To the best of our knowledge, carbamazepine and sodium valproate have not been used clinically for aminophylline intoxication.

In conclusion, the results of this study have practical clinical implications and suggest that carbamazepine in particular or sodium valproate, combined with routine EEG monitoring, may be of value in the treatment of status asthmaticus patients given intravenous aminophylline. This precautionary monitoring is of significance, since there are no warning symptoms of impending seizures and no detectable signs of neurological impairment in patients with acute respiratory insufficiency.

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