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Comparison of Anticonvulsant Effect of Pentobarbital and Phenobarbital Against Seizures Induced by Maximal Electroshock and Picrotoxin in Rats

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MEHTA, A. K. AND M. K. TICKU. *Comparison of anticonvulsant effect of pentobarbital and phenobarbital against seizures induced by maximal electroshock and picrotoxin in rats.* PHARMACOL BIOCHEM BEHAV **25**(5) 1059–1065, 1986.—Pentobarbital and phenobarbital exhibited anticonvulsant effect against maximal electroshock (MES) and picrotoxin-induced seizures in rats. Bicuculline, a $GABA_A$ receptor antagonist, reversed the anticonvulsant effect of pentobarbital, but not of phenobarbital, at a dose having no effect *per se*. Although picrotoxin (2 mg/kg, IP) potentiated MES seizures, it did not reverse the anticonvulsant effect due to either pentobarbital or phenobarbital. $GABA_B$ receptor antagonists such as δ-amino-n-valeric acid and homotaurine failed to modify the anticonvulsant effect due to pentobarbital or phenobarbital. Furthermore, $GABA_A$ agonist muscimol but not baclofen, a $GABA_B$ receptor agonist, exhibited the anticonvulsant effect against MES-induced seizures. However, baclofen when combined with sub-effective dose of pentobarbital or phenobarbital offered protection against MES seizures. Pentobarbital and phenobarbital were effective in almost equivalent doses against MES, as well as against picrotoxin-induced seizures. These observations indicated that pentobarbital exhibits anticonvulsant effect against MES seizures through the involvement of $GABA_A$ receptors, and activation of $GABA_B$ receptors alone does not seem to play any significant role in MES seizures and in the anticonvulsant effect of pentobarbital. However, activation of $GABA_B$ receptor does potentiate the facilitatory effect of barbiturates on $GABA_A$ergic transmission and in their anti-MES effect. Moreover, these results also suggest that the anticonvulsant effect of barbiturates against MES-seizures may involve other mechanisms in addition to $GABA_A$ergic transmission.

Pentobarbital Phenobarbital Convulsions Bicuculline Muscimol $GABA_A$ receptors Baclofen Picrotoxin

GABA-MEDIATED inhibitory transmission has been implicated in the action of a variety of centrally acting convulsant, anticonvulsant and anxiolytic drugs. GABA-receptors have been classified into subtypes, namely $GABA_A$ and $GABA_B$ receptors [9]. $GABA_A$ receptors, unlike $GABA_B$ receptors, are susceptible to blockade by bicuculline. On the other hand, $GABA_B$ receptors are antagonized by δ-aminovaleric acid [11,18], and activated by baclofen [6,9]. Furthermore, homotaurine (3-aminoproprane sulphonic acid), a potent and specific agonist of $GABA_A$ receptors [3, 4, 6], has also been demonstrated to be a $GABA_B$ receptor antagonist [7]. GABAergic system has been implemented in various neurological disorders [5, 8, 10, 14, 26, 29] including epilepsy [19]. There is also evidence that the anticonvulsant activity of pentobarbital against maximal electroshock (MES)-induced seizures in rats may be mediated through GABAergic mechanism [26]. However, the role of GABA-receptor subtypes in maximal electroshock-induced seizures (MES), and the potential mechanisms involved in the anticonvulsant effect of phenobarbital are unknown. The present study was undertaken to (i) define the involvement of GABA receptor subtypes in MES, and (ii) compare the anticonvulsant profile of pentobarbital against MES-induced seizures, as well as against a chemoconvulsant, which inhibits $GABA_A$ergic transmission.

METHOD

Male Sprague-Dawley rats, weighing 180–200 g, were used. The animals were kept at a constant room temperature (25°C), and had free access to water and food.

For intracerebroventricular (ICV) drug administration, a polyethylene cannula was chronically implanted in the left lateral ventricle [23]. The drugs were slowly injected into the ventricle in a volume of 10 μl, with the help of a Hamilton microsyringe.

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TABLE 1

EFFECT OF VARIOUS DRUGS ON THE ANTICONVULSANT EFFECT OF PENTOBARBITAL AGAINST MES-INDUCED SEIZURES

Drug	Dose (mg/kg, IP)	n	Mean duration (sec ± S.D.) Tonic limb flexion	Tonic extensor	Clonus
Control	—	12	5.3 ± 1.22	13.8 ± 1.91	14.8 ± 2.55
Pentobarbital	10	5	5.8 ± 0.84	12.0 ± 1.22	14.0 ± 2.00
	20	6	2.2 ± 0.41*	4.0 ± 1.26*	7.2 ± 1.83*
	40	8	0.0 ± 0.00*	0.0 ± 0.00*	4.5 ± 1.93*
Bicuculline	2				
+pentobarbital	20	6	3.5 ± 0.55†	8.0 ± 1.09†	12.3 ± 1.37†
Picrotoxin	2				
+pentobarbital	20	7	2.3 ± 0.49NS	4.1 ± 1.46NS	9.4 ± 1.81†
δ-Amino-n-valeric acid	10				
+pentobarbital	20	5	2.2 ± 0.45NS	4.2 ± 0.84NS	8.0 ± 1.58NS
δ-Amino-n-valeric acid	10 μg, ICV				
+pentobarbital	20	5	2.0 ± 1.22NS	3.4 ± 1.67NS	6.4 ± 1.14NS
Homotaurine	10				
+pentobarbital	20	6	2.0 ± 1.10NS	3.3 ± 1.21NS	8.2 ± 1.33NS
Homotaurine	1 μg, ICV				
+pentobarbital	20	5	1.8 ± 0.84NS	3.2 ± 1.30NS	6.6 ± 0.89NS

Drugs were administered 30 min prior to MES-induced seizures. The results are mean ± SD of numbers of animals, as indicated (n).

*†$p<0.05$ as compared to control and pentobarbital (20 mg/kg, IP) groups, respectively, by one way analysis of variance, using Student-Newman-Keuls procedure.

NS: Not significant as compared to pentobarbital (20 mg/kg, IP) group.

TABLE 2

PER SE EFFECT OF VARIOUS ANTAGONISTS EMPLOYED AGAINST MES-INDUCED SEIZURES

Drug	Dose (mg/kg, IP)	n	Mean duration (sec ± S.D.) Tonic limb flexion	Tonic extensor	Clonus
Control	—	12	5.3 ± 1.22	13.8 ± 1.91	14.8 ± 2.55
Bicuculline	2	6	5.0 ± 0.89	11.5 ± 1.38	14.8 ± 1.72
Picrotoxin	2	6	7.3 ± 1.37*	21.2 ± 1.33*	24.2 ± 2.48*
δ-Amino-n-valeric acid	10	5	5.6 ± 1.52	12.0 ± 1.58	14.0 ± 1.58
	10 μg, ICV	5	4.2 ± 0.45	12.2 ± 1.30	14.6 ± 2.41
Homotaurine	10	6	4.2 ± 1.33	10.7 ± 3.33*	12.3 ± 1.97
	1 μg, ICV	5	4.0 ± 1.00	12.0 ± 2.00	14.6 ± 2.61

Each value represents the mean ± SD of number of animals, as indicated (n).

*$p<0.05$ as compared to control group by one way analysis of variance, using Student-Newman-Keuls procedure.

Maximal Electroshock (MES) Seizures

Maximal electroshock seizures were produced in rats by passing an alternating electrical current (150 mV, 50 pulses/sec, 0.2 sec) via corneal electrodes. Various phases of convulsions, i.e., tonic flexion, extensor and clonus, were timed, and the protection against tonic phase was considered as anticonvulsant activity [12].

Picrotoxin-Induced Seizures

A preliminary study demonstrated that a 10 mg/kg, IP

TABLE 3

EFFECT OF GABAERGIC DRUGS AND THEIR INTERACTION WITH PENTOBARBITAL AGAINST MES-INDUCED SEIZURES

Sr. No.	Drug	Dose	n	Mean duration (sec ± S.D.) Tonic limb flexion	Tonic extensor	Clonus
1	Control	—	7	4.4 ± 0.53	12.0 ± 1.15	14.6 ± 3.10
2	Muscimol					
		(a) 10 ng, ICV	5	4.4 ± 0.55	14.2 ± 2.28	14.8 ± 3.56
		(b) 100 ng, ICV	5	1.4 ± 0.55†	1.8 ± 0.45†	9.8 ± 1.48†
3	Bicuculline	2 mg/kg, IP				
	+muscimol	100 ng, ICV	6	4.0 ± 0.89§	12.3 ± 2.58§	13.5 ± 2.17‡
4	Baclofen	10 mg/kg, IP	9	5.4 ± 1.13	9.9 ± 2.71	13.9 ± 1.27
		20 mg/kg, IP	5	5.2 ± 0.84	12.2 ± 1.48	14.6 ± 1.14
		100 ng, ICV	5	4.4 ± 0.55	12.2 ± 1.92	14.0 ± 2.00
		1 μg, ICV	5	4.6 ± 0.55	13.4 ± 2.51	12.8 ± 1.92
5	Pentobarbital					
	+muscimol	10 mg/kg, IP				
		10 ng, ICV	5	0.8 ± 0.45†	1.0 ± 0.71†	7.8 ± 1.92†
6	Bicuculline	2 mg/kg, IP				
	+pentobarbital	10 mg/kg, IP	5	3.5 ± 0.78§	7.0 ± 0.55§	12.2 ± 1.78*
	+muscimol	10 ng, ICV				
7	Pentobarbital	10 mg/kg, IP				
	+baclofen	10 mg/kg, IP	6	3.0 ± 0.63†	7.5 ± 1.05†	9.3 ± 0.82†
8	Pentobarbital	10 mg/kg, IP				
	+baclofen	100 ng, ICV	8	1.0 ± 0.00†	1.4 ± 0.52†	11.6 ± 4.00
9	δ-Amino-n-valeric					
	acid	10 mg/kg, IP				
	+pentobarbital	10 mg/kg, IP	5	5.8 ± 1.30‡	12.4 ± 1.52§	14.6 ± 2.07‡
	+baclofen	10 mg/kg, IP				
10	δ-Amino-n-valeric					
	acid	10 mg/kg, IP				
	+pentobarbital	10 mg/kg, IP	5	4.2 ± 0.84*	9.8 ± 1.10*	13.4 ± 1.67NS
	+baclofen	100 ng, ICV				

†$p<0.05$ as compared to control group by one way analysis of variance, using Student-Newman-Keuls procedure.
*$p<0.05$ when comparison was made between groups 6 and 5; 10 and 8 by Student t-test.
‡$p<0.01$ when comparison was made between groups 3 and 2b; 9 and 7 by Student t-test.
§$p<0.001$ when comparison was made between groups 3 and 2b; 6 and 5; 9 and 7 by Student t test.
NS: not significant when comparison was made between groups 10 and 8 by Student t-test.

dose of picrotoxin induced severe tonic seizures followed by mortality in 100% animals. This dose was employed to compare the efficacy of pentobarbital and phenobarbital against picrotoxin-induced seizures. Onset of seizures, severity and mortality rate were recorded after administration of picrotoxin within a 90 min test session.

Statistics

The data is expressed as mean±S.D. The results were analyzed by one-way analysis of variance, and the level of significance was determined by Student-Newman-Keuls procedure or as mentioned. A value of $p<0.05$ was considered statistically significant.

Drugs

Pentobarbital sodium, phenobarbital, picrotoxin, (+) bicuculline, δ-amino-n-valeric acid HCl, and muscimol were purchased from Sigma Chemicals (St. Louis, MO), and homotaurine sodium from Aldrich Chem. Co. (±)Baclofen was a gift from Ciba-Geigy (Basel, Switzerland) and diazepam from Hoffman-La Roche, Inc. (Nutley, NJ).

Picrotoxin was dissolved in dimethylsulphoxide (DMSO) and bicuculline was dissolved in a few drops of 0.1 N HCl, and adjusted to pH 5 with 0.1 N NaOH. Diazepam was dissolved in propylene glycol, and phenobarbital was dissolved in a few drops of 0.1 N NaOH, after which the final volume was made up with 0.9% w/v NaCl. All other drugs were dissolved in 0.9% w/v NaCl, and drugs were injected intraperitoneally (IP) in a volume of 1 ml/kg body weight or intracerebroventricularly (ICV) in a volume of 10 μl 30 min prior to electroshock or chemoconvulsant exposure. The control animals received the equivalent volume of vehicle. Doses refer to the salt or base as specified above.

RESULTS

Maximal Electroshock-Induced Seizures

Pentobarbital (20 and 40 mg/kg, IP) offered a protection against the extensor phase of the electroshock-induced convulsions (Table 1). Bicuculline (2 mg/kg, IP), when adminis-

TABLE 4

EFFECT OF VARIOUS ANTAGONISTS ON THE ANTICONVULSANT EFFECT OF PHENOBARBITAL AGAINST MES-INDUCED SEIZURES

Drug	Dose (mg/kg, IP)	n	Mean duration (sec ± S.D.) Tonic limb flexion	Tonic extensor	Clonus
Control	—	12	5.3 ± 1.22	13.8 ± 1.91	14.8 ± 2.55
Phenobarbital	10	5	4.4 ± 0.55	12.2 ± 3.27	13.2 ± 2.68
	15	5	2.6 ± 0.55*	8.0 ± 1.58*	13.0 ± 2.24
	30	5	1.6 ± 0.55*	2.2 ± 0.45*	13.6 ± 3.85
	45	6	0.3 ± 0.52*	0.3 ± 0.52*	11.3 ± 2.73
	60	5	0.2 ± 0.45*	0.2 ± 0.45*	9.2 ± 6.38*
Bicuculline	2				
+phenobarbital	30	7	1.1 ± 0.69NS	1.3 ± 0.76NS	12.9 ± 2.73NS
Picrotoxin	2				
+phenobarbital	30	6	1.5 ± 0.84NS	1.8 ± 1.17NS	14.2 ± 1.60NS
δ-Amino-n-valeric acid	10				
+phenobarbital	30	5	1.8 ± 0.45NS	2.4 ± 0.55NS	13.0 ± 3.08NS
Homotaurine	10				
+phenobarbital	30	5	1.6 ± 0.55NS	2.0 ± 0.71NS	12.6 ± 3.21NS

Drugs were administered 30 min prior to MES-induced seizures. The results are mean ± S.D. of number of animals, as indicated (n).

*$p<0.05$ as compared to control group by one way analysis of variance, using Student-Newman-Keuls procedure.

NS: Not significant as compared to phenobarbital (30 mg/kg, IP) group.

TABLE 5

INTERACTION OF GABAERGIC DRUGS WITH PHENOBARBITAL AGAINST MES-INDUCED SEIZURES

Sr. No.	Drug	Dose	n	Mean duration (sec ± S.D.) Tonic limb flexion	Tonic extensor	Clonus
1	Control	—	7	4.4 ± 0.53	12.0 ± 1.15	14.6 ± 3.10
2	Phenobarbital	10 mg/kg, IP				
	+muscimol	10 ng, ICV	5	1.6 ± 0.55†	2.8 ± 1.92†	8.2 ± 1.92†
3	Bicuculline	2 mg/kg, IP				
	+phenobarbital	10 mg/kg, IP	5	3.2 ± 0.84‡	5.4 ± 1.14*	14.0 ± 3.16*
	+muscimol	10 ng, ICV				
4	Phenobarbital	10 mg/kg, IP	5	1.8 ± 0.45†	3.0 ± 2.00†	8.0 ± 1.58†
	+baclofen	100 ng, ICV				
5	δ-Amino-n-valeric acid	10 mg/kg, IP				
	+phenobarbital	10 mg/kg, IP	5	3.8 ± 0.84‡	5.8 ± 1.48*	13.2 ± 2.28*
	+baclofen	100 ng, ICV				

†$p<0.05$ as compared to control group by one way analysis, using Student-Newman-Keuls procedure. The results are mean ± SD of number of animals, as indicated (n). *$p<0.05$ when comparison was made between groups 3 and 2; 5 and 4 by Student t-test. ‡$p<0.01$ when comparison was made between groups 3 and 2; 5 and 4 by Student t-test.

tered 30 min prior to pentobarbital, reversed the anticonvulsant effect without exhibiting any effect *per se* (Tables 1 and 2). The effect of various antagonists employed in this study on the MES-seizures is shown in Table 2. Picrotoxin (2 mg/kg, IP), although potentiated MES-induced seizures (Table 2), did not reverse the anticonvulsant effect due to pentobarbital (Table 1). δ-Amino-n-valeric acid (10 mg/kg, IP or 10 μg, ICV) as well as homotaurine (10 mg/kg, IP or 1 μg, ICV) also failed to modify the anticonvulsant effect of pentobarbital (Table 1). (±)Baclofen alone did not modify the MES-seizure activity (Table 3). However, (±)baclofen (10 mg/kg, IP or 100 ng, ICV) when combined with sub-effective

TABLE 6

EFFECT OF PENTOBARBITAL AND PHENOBARBITAL AGAINST CONVULSION INDUCED BY PICROTOXIN

Drug	Dose (mg/kg, IP)	n	Mean duration (min ± S.D.) Clonic	Tonic	Mortality within 90 min
Picrotoxin	10	8	8.1 ± 0.51	16.7 ± 2.27	8/8†
Pentobarbital	5	5	8.3 ± 0.50	17.2 ± 0.98	4/5
	7.5	6	9.6 ± 0.95	22.2 ± 2.73*	2/6‡
	10	6	12.3 ± 0.81*	28.7 ± 1.70*	0/6‡
	20	6	16.2 ± 2.83*	—‡	0/6‡
Phenobarbital	5	5	7.9 ± 0.64	15.7 ± 1.25	5/5
	10	5	10.1 ± 0.24*	17.8 ± 0.92	2/5‡
	15	5	12.1 ± 0.43*	31.0 ± 1.07*	0/5‡
	25	5	14.0 ± 0.79*	—‡	0/5‡

Barbiturates were administered 30 min prior to picrotoxin (10 mg/kg, IP). The results are the mean ± SD of number of animals indicated (n). †Mortality rate was monitored for 90 min following picrotoxin administration. In the case of picrotoxin group all the animals died within 30 min, following its administration. *$p<0.05$ as compared to picrotoxin group, by one way analysis of variance using Student-Newman-Keuls procedure. ‡$p<0.01$ as compared to picrotoxin group using Fisher's exact test. —: implies no convulsion.

dose of pentobarbital (10 mg/kg, IP) offered protection (Table 3). This effect could be blocked by prior treatment of animals with δ-amino-n-valeric acid (Table 3). Similarly, a combination of sub-effective doses of muscimol (10 ng, ICV) and pentobarbital (10 mg/kg, IP) exhibited anticonvulsant activity, which was susceptible to reversal by prior treatment of the animals with bicuculline (Table 3).

Like pentobarbital, phenobarbital (15–60 mg/kg, IP) exhibited protection against the electroshock-induced seizures (Table 4). This effect was resistant to blockade by bicuculline, picrotoxin, δ-amino-n-valeric acid and homotaurine (Table 4). A sub-effective dose of phenobarbital (10 mg/kg, IP) when combined with baclofen (100 ng, ICV) also elicited anticonvulsant effect. This effect could be blocked by prior treatment of animals with δ-amino-n-valeric acid (Table 5). Similarly, a combination of sub-effective doses of phenobarbital (10 mg/kg, IP) and muscimol (10 ng, ICV) also offered protection, susceptible to blockade by bicuculline, against MES-induced seizures (Table 5).

Picrotoxin-Induced Seizures

Picrotoxin (10 mg/kg, IP) induced severe tonic intermittent convulsions followed by death in 100% animals (Table 6). Pentobarbital (7.5–20 mg/kg, IP) as well as phenobarbital (10–25 mg/, IP) exhibited anticonvulsant activity in a dose-dependent manner against picrotoxin-induced convulsions (Table 6).

DISCUSSION

The GABA receptor complex, with which various drugs interact to either facilitate or inhibit GABAergic transmission, is an oligomeric complex with multiple sites. Barbiturates have been shown to bind to a distinct allosteric site coupled to GABA, benzodiazepine and picrotoxin sites of the benzodiazepine GABA receptor-ionophore complex [17,34]. Pentobarbital has been reported to enhance the binding of [^{3}H]GABA and [^{3}H]-benzodiazepine agonists and inhibit the binding of picrotoxin like convulsants to brain membranes [1, 13, 17, 24, 31–34, 36]. Neurophysiological studies also lend support to the concept that barbiturates mediate their effects via GABAergic mechanisms [16, 21, 22, 28, 30]. Thus, while both pentobarbital and phenobarbital facilitate GABAergic transmission, only pentobarbital activates $GABA_A$ receptor coupled chloride channels directly [16, 21, 22]. Further, both these barbiturates decrease glutamate-mediated excitatory response [16, 21, 22, 28]. Several studies have reported that pentobarbital is much more potent than phenobarbital in enhancing GABA responses [16, 21, 22], muscimol responses [30], and in reducing the potency of picrotoxin as a GABA antagonist [22].

There also exist differences in the ability of pentobarbital and phenobarbital to interact with various binding sites on the oligomeric GABA receptor complex. Thus, while both barbiturates inhibit [^{3}H]α-dihydropicrotoxinin (DHP) and [^{35}S]t-butylbicyclophosphorothionates (TBPS) binding (e.g., [34]), accelerate the dissociation of [^{35}S]TBPS binding as compared to the convulsant dissociation pattern [17], and only pentobarbital enhances both [^{3}H]GABA and [^{3}H]benzodiazepine agonist binding [1, 13, 32, 33, 36]. Interestingly, phenobarbital inhibits this enhancing effect of pentobarbital [13], suggesting that the two barbiturates must bind to the same sites.

The present study demonstrated involvement of $GABA_A$ receptor system in the MES-induced seizures, and in the anticonvulsant effect of barbiturates against MES-induced seizures. This is supported by the following findings: (i) $GABA_A$ agonist muscimol protects against MES-induced seizures; (ii) pentobarbital and phenobarbital produce a dose-dependent anticonvulsant effect against MES-induced seizures; (iii) pentobarbital's anticonvulsant effect was blocked by bicuculline; and (iv) sub-effective doses of muscimol and pentobarbital or phenobarbital produced an anticonvulsant effect, which was blocked by bicuculline. Our studies are in disagreement with a recent report which indicated that $GABA_A$ergic transmission does not play a role in the anti-MES activity of barbiturates [35]. Their conclusions are based on the observation that synergistic effect could not

be observed between barbiturates and $GABA_A$ agonists such as progabide and 4,5,6,7-tetrahydroisoxazolo [5,4-C]pyridine-3-01 (THIP; [35]). This difference may be due to different agonists used in the two studies (e.g., muscimol vs. THIP) or the species difference (rats vs. mice). However, the above lines of evidence indicate clearly a role for $GABA_A$ergic transmission in the anti-MES activity of barbiturates. Anticonvulsant effect of $GABA_A$ agonists like muscimol [15,18] and pentobarbital [27] against seizures induced by impairment of GABA-mediated transmission are well documented. A previous report from our laboratory has also shown that pentobarbital at sub-effective doses when combined with sub-effective doses of other facilitators of GABAergic transmission, potentiated the anticonvulsant effect against MES-induced seizures [26] or against $GABA_A$ antagonists [27].

$GABA_B$ receptors do not seem to be involved directly in the anticonvulsant effect of pentobarbital, since $GABA_B$ receptor antagonist such as δ-amino-n-valeric acid [11,20] and homotaurine [7] failed to modify the effect of barbiturates. Furthermore, baclofen, $GABA_B$ receptor agonist [6,9], did not exhibit anticonvulsant effect by itself against MES-induces seizures, whereas $GABA_A$ agonist muscimol does provide protection. The inability of $GABA_B$ antagonists to modify the anticonvulsant effect cannot be due to their inability to pass the blood brain barrier, since similar results were obtained when these drugs were administered by ICV route. However, a sub-effective dose of pentobarbital, or phenobarbital, when combined with baclofen offered protection against MES-induced seizures. This effect could be blocked by $GABA_B$ antagonists. This is an interesting observation and suggests that activation of $GABA_B$ receptors potentiates the anticonvulsant effect of barbiturates. This potentiating effect could be due to the ability of baclofen to inhibit the release of excitatory transmitters such as glutamate and aspartate [25]. These results also indicate that activation of $GABA_B$ receptors alone does not elicit anticonvulsant effect, since baclofen by itself was ineffective as an anticonvulsant. However, when combined with drugs like pentobarbital and phenobarbital, which facilitates $GABA_A$ergic transmission and inhibit glutamate-mediated excitatory responses, the overall CNS activity is inhibited further, probably due to decreased release of excitatory transmitters by baclofen. These observations would tend to support the notion that barbiturates produce their anticonvulsant effect both by facilitation of $GABA_A$ergic transmission and inhibition of excitatory transmission. Recently, baclofen has also been reported to potentiate the anti-MES activity of phenytoin [2].

The anticonvulsant effect of phenobarbital against MES-seizures could not be blocked either by $GABA_A$ or $GABA_B$ receptor antagonists. It is, however, difficult to conclude that the anticonvulsant activity of phenobarbital is not mediated through $GABA_A$ receptors, based on this observation alone, since the effect of higher doses of bicuculline (and picrotoxin) on the phenobarbital-anticonvulsant effect could not be studied as it exhibits convulsions at higher doses. Furthermore, baclofen, like muscimol, when combined with sub-effective dose of phenobarbital exhibited anticonvulsant effect, results similar to that obtained with pentobarbital. It may be pointed out that both pentobarbital and phenobarbital antagonize bicuculline-induced paroxymal depolarizing events [28], reverse the picrotoxin antagonism of GABA responses [22], and prevent picrotoxin-induced convulsions (Table 6). Phenobarbital and pentobarbital were almost equi-effective anticonvulsants against MES, as well as picrotoxin-induced seizures. These agents, however, offered protection at lower doses against picrotoxin-induced seizures as compared to MES seizures.

In conclusion, the anticonvulsant effect of barbiturates appears to be mediated through $GABA_A$ receptors, and $GABA_B$ receptors do not directly play a significant role in MES-induced seizures and in the anticonvulsant effect of barbiturates as well. However, activation of $GABA_B$ receptors, does potentiate the anti-MES activity of drugs like barbiturates which facilitate GABAergic transmission and inhibit glutamate-mediated excitation.

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