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Anticonvulsant Activity of 5,7DCKA, NBQX, and Felbamate Against Some Chemoconvulsants in DBA/2 Mice

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DE SARRO, G. B., E. ONGINI, R. BERTORELLI, U. AGUGLIA AND A. DE SARRO. *Anticonvulsant activity of 5,7DCKA, NBQX, and felbamate against some chemoconvulsants in DBA/2 mice.* PHARMACOL BIOCHEM BEHAV **55**(2) 281–287, 1996.—The anticonvulsant effects of felbamate (10–300 mg/kg, intraperitoneally, IP), and those of two representative antagonists of the excitatory amino acid receptors, 5-7dichlorokynurenic acid (5-7DCKA; 0.6–30 nmol/mouse, intracerebroventricularly, ICV), and 2,3-dihydroxy-6 nitro-7-sulfamoylbenzo(F)quinoxoline (NBQX; 1.1–33.6 mg/kg, IP) were studied in the DBA/2 mice. All drugs protected the animals from sound-induced seizures. The drugs were also effective against seizures induced by stimulation of the excitatory amino acid receptor complex using the agonists *N*-methyl-D-aspartate (NMDA) or α-amino-3-hydroxy-5 methyl-4-isoxazolepropionic acid (AMPA). In separate studies, felbamate protected mice from seizures induced by ICV administration of the activator of dihydropyridine-sensitive calcium channels, methyl-1,4-dihydro-2,6-dimethyl-3-nitro-4-(2-trifluoromethylphenyl) pyridine-5-carboxylate (Bay k 8644), with ED_{50} values of 26 and 46.9 mg/kg for tonus and clonus, respectively. Using Bay k 8644, NBQX (1–40 mg/kg IP) was uneffective, while 5,7DCKA (5–90 nmol/mouse, ICV) protected mice against tonus. Moreover, felbamate prevented seizures induced by blocking voltage-dependent K^+ channels using α-dendrotoxin, with ED_{50} values of 22.6 mg/kg for tonus and of 34.8 mg/kg for clonus. Conversely, 5,7DCKA or NBQX did not significantly antagonize seizures induced by α-dendrotoxin. The present data indicate that felbamate is an effective anticonvulsant drug in DBA/2 mice with a broader anticonvulsant spectrum than 5,7DCKA and NBQX. **Copyright © 1996 Elsevier Science Inc.**

Felbamate 5,7DCKA (5,7dichlorokynurenic acid) NBQX (2,3-dihydroxy-6-nitro-7-sulphamoylbenzo(F)quinoxoline)
Audiogenic seizures Voltage-dependent K^+ channels Dihydropyridine-sensitive Ca^{2+} channels

FELBAMATE was found to exhibit a wide spectrum of anticonvulsant activity in animal seizure models (5,16,36), but little was initially known regarding the biological mechanism underlying these effects (21). Recently, some studies have shown that felbamate has a novel mechanism of action compared with established antiepileptic drugs. Felbamate appears to interact with the $GABA_A$ receptor complex (22) but its activity may principally depend on interaction with excitatory amino acids, especially the glycine binding site of the NMDA receptor (19,33). Thus, felbamate has been reported to displace [^{3}H]5,7-dichlorokynurenic acid (5,7DCKA), a competitive antagonist at the strychnine-insensitive glycine site of the NMDA receptor (20). Felbamate has also been demonstrated to inhibit NMDA receptor-mediated cationic currents in cultured rat hippocampal neurons (22). Further support comes from studies in rodent seizure models showing that glycine can selectively reverse the anticonvulsant effects of felbamate. In fact, glycine itself or D-serine, a glycine agonist, both produce a parallel rightward shift of the dose–response curve of felbamate against seizures induced by either maximal electro-

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shock or NMDA in mice (6) and in Fring's audiogenic seizure-susceptible mice (35), respectively. In keeping with these data, we have demonstrated that glycine reduces the protective effects of felbamate against seizures induced by NMDA and that D-serine markedly attenuates the anticonvulsant properties of felbamate also in the genetic model of the audiogenic seizure-prone DBA/2 mouse (10). More recently, Wamsley and co-workers (34) have shown that strychnine-insensitive glycine receptors represent a site of action of felbamate in the human brain.

In the attempts to better understand the mode of action of felbamate, we have studied its activity in the DBA/2 mice model, in comparison with two different antagonists at the excitatory amino acid receptor complex, namely 5,7DCKA, and 2,3-dihydroxy-6-nitro-7-sulfamoylbenzo(F)quinoxoline (NBQX). 5,7DCKA anticonvulsant effects are principally mediated at the strychnine-insensitive glycine sites of the NMDA receptors (15). NBQX, a non-NMDA receptor antagonist, has anticonvulsant activity in genetic seizure models (4,27) as well as neuroprotective effects in animal models of cerebral stroke (26). In previous studies, we have investigated the role of the excitatory amino acids in the anticonvulsant activity of felbamate by evaluating the protective effects of the drug against seizures induced by either audiogenic stimulation or treatment with NMDA or α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA), the latter being an agonist at the non-NMDA receptor complex (10). The aim of the present study was to complete the approach used previously comparing the anticonvulsant effects of felbamate with those of 5,7DCKA and NBQX, under the same experimental conditions. Because antiepileptic drugs may also act on ion channels, we studied whether the seizures induced by compounds acting on voltage-dependent K^+ channels as well as the dihydropyridine Ca^{2+} channels are antagonized by felbamate. For this purpose, felbamate, 5,7DCKA and NBQX were examined for their effects against seizures induced by methyl-1,4-dihydro-2,6-dimethyl-3-nitro-4-(2-trifluoromethylphenyl)-pyridine-5-carboxylate (Bay k 8644) an activator of the dihydropyridine calcium channels (24), and also against the seizures induced by α-dendrotoxin, a snake venom peptide that acts as a blocker of the voltage-dependent K^+ channels.

METHOD

Animals

Male and female DBA/2 mice weighing 8–12 g (22–26 days old) were used (Charles River, Calco, Como, Italy). Animals were housed in groups of 8–10 under a 12 L:12 D cycle (lights on at 0700 h) with food and water available ad lib.

Procedures involving animals and their care were conducted in conformity with the istitutional guidelines, in compliance with national and international laws and policies.

Experimental Design

Seizures were induced by auditory stimulation or intracerebroventricular (ICV) injections of NMDA, AMPA, α-dendrotoxin, or Bay k 8644.

DBA/2 mice were exposed to auditory stimulation 45 min following intraperitoneal (IP) administration of vehicle or felbamate, 30 min following ICV injection of vehicle or 5,7DCKA, 20 min following IP injection of vehicle or NBQX. Each mouse was placed under a hemispheric perspex dome (diameter 58 cm) and 1 min allowed for habituation and assessment of locomotor activity. Auditory stimulation (12–16 kHz, 109 dB) was applied for 1 min or until tonic extension occurred. Seizure response as previously reported (7) was assessed using the following scale: 0 = no response, 1 = wild running, 2 = clonus, 3 = tonus, 4 = respiratory arrest. The maximum response was recorded for each animal. Rectal temperature was recorded immediately prior to auditory testing using an Elektrolaboratoriet thermometer type T.E.3. Behavioral changes were observed during the period between drug administration and auditory testing.

For ICV injections, mice were anesthetized with ether and injections were made in the left or right lateral ventricle (coordinates 1 mm posterior and 1 mm lateral to the bregma; depth 2.4 mm) using a 10 μl Hamilton microsyringe (type 701N) fitted with a nylon cuff on the needle as described elsewhere (8). Injections of vehicle (phosphate buffer) or drugs by this procedure led to an uniform distribution throughout the ventricular system within 10 min (8). The second ICV injection in both cases was performed into the same site of the first injection. In particular, DBA/2 mice were placed individually for 30–40 s on a cotton surface into a glass cilinder dome (diameter 15 cm) where ether was present on the floor. The mice usually recovered from anesthesia within 90–150 s and at this time they were able to walk into the cages. Five minutes after ether anesthesia DBA/2 mice showed audiogenic seizures following auditory stimulation and were normally responsive to chemoconvulsants as previously documented (8–12). The seizure score recorded after ICV injection of Bay k 8644, previously described (9), was the following: 0 = no response; 1 = scratching and twisting of the fore limbs; 2 = rearing and walking; 3 = intermittent clonic jerks of limbs with tonic flexion of fore limbs and tail flexion; 4 = head bobbing with complex grooming actions (licking of fur and scratching); 5 = jumping, squeaking, and tonic extension of hind limbs; 6 = barrel rolling. The seizure score observed following ICV injection of α-dendrotoxin was the following: 0 = no response; 1 = compulsive grooming, forelimb stereotypies, and/or jaw opening; 2 = trembling; 3 = opistotonus, straub tail; 4 = hind limb myoclonus and falling down; 5 = wild running followed by clonic seizures; 6 = hind limb tonic extension; 7 = death. The animals were placed singly in a 30 × 30 × 30 cm box and the observation time was 20 min after NBQX injection, 30 min after the administration of 5,7DCKA, 60 min after the injection of convulsants: NMDA, α-dendrotoxin, AMPA, or Bay k 8644 and 60 min after felbamate. The occurrence of clonic and tonic seizure signs and their latency were recorded.

Effects on Behavior and Motor Movements

Behavioral changes and their onset and duration were recorded after drug injection until the time of convulsant test. In particular, two independent observers followed gross behavioural changes consisting of locomotor activity, ataxia, squatting posture, and possible piloerection. These behavioral changes were noted but not statistically analyzed.

Statistical Analysis

Statistical comparison between groups of control and drug-treated DBA/2 mice was made using Fisher's exact probability test (incidence of the seizure phases) or ANOVA with Dunnett's *t*-test (rectal temperatures). The percentage incidence of each phase of the audiogenic seizure was determined for each drug. These values were plotted against the corre-

TABLE 1

EFFECTS OF FELBAMATE, 5,7 DCKA, AND NBQX AGAINST BAY K 8644- OR α-DENDROTOXIN-INDUCED SEIZURES IN DBA/2 MICE

Treatment	Dose Range	ED_{50} (95% Confidence Limits)	
		Tonic Extension	Clonic Seizures
Felbamate + Bay k 8644	3–100 mg/kg	26.7 (13.1–51.4)	46.9 (19.5–113.3)
5,7 DCKA + Bay k 8644	5–90 nmol/mouse	77.9 (51.3–118.1)	NA
NBQX + Bay 8644	1–40 mg/kg	NA	NA
Felbamate + α-dendrotoxin	3–100 mg/kg	22.6 (11.2–45.4)	34.4 (16.3–72.3)
5,7 DCKA + α-dendrotoxin	5–90 nmol/mouse	NA	NA
NBQX + α-dendrotoxin	1–40 mg/kg	NA	NA

Bay k 8644 or α-dendrotoxin were administered intracerebroventricularly at the CD_{97} for either clonus or forelimb tonic extension, 1 h, 30, and 20 min after felbamate, 5,7 DCKA, or NBQX injection, respectively. All data were calculated according to the method of Litchfield and Wilcoxon (18).

NA = not active or weakly active up to 90 nmol/mouse for 5,7, DCKA and 40 mg/kg for NBQX.

sponding doses by a computer construction of the dose–effect curves for calculation of ED_{50} (with 95% confidence limits). The ED_{50} values for each compound were calculated using a computer program of the method of Litchfield and Wilcoxon (18). At least 32 animals were used to calculate each ED_{50} value.

Drugs

Felbamate was supplied by Schering-Plough (Kenilworth, NJ), NBQX by Novo Nordisk (Malov, Denmark), and Bay k 8644 by Bayer (Milan, Italy), NMDA was purchased from Sigma (St. Louis, MO), AMPA and 5,7DCKA were purchased from Tocris (Buckhurst Hill, UK), and α-dendrotoxin from Calbiochem (San Diego, CA).

For systemic injections, all compounds were given intraperitoneally (0.1 ml/10 g of body weight of the mouse). Felbamate was administered as a freshly prepared solution in 50% dimethylsulphoxide and 50% sterile saline (0.9% NaCl). 5,7DCKA, AMPA, NMDA, and NBQX were dissolved in a minimum quantity of NaOH 1 N. The final volume was made up with sodium phosphate buffer (67 mM). When necessary, the pH was adjusted to 7.3–7.4 by adding HCl 0.2 N. Doses and time of administration are reported in the Table 1 or in the figures. Previous experiments have shown that these vehicles, when administered either IP or ICV, do not affect both behavior and responses to auditory stimulation of DBA/2 mice and to chemoconvulsants.

All drugs administered ICV were dissolved in sodium phosphate buffer 67 mM, microinjected in a volume of 5 or 10 μl per mouse. Doses and time of administration are reported in Table 1 or in the figures. Because of the light sensitivity of some compounds, weighing and handling were carried out under sodium vapor lamps and the substances were protected from light during the experiments.

RESULTS

Effects of Felbamate, 5,7DCKA, and NBQX on Audiogenic Seizures

As previously reported, felbamate (10–300 mg/kg IP, n = 80) and 5,7DCKA (0.6–30 nmol/mouse ICV, n = 50), administered 45 and 30 min before auditory stimulation, respectively, dose dependently reduced the severity of the audiogenic seizures in DBA/2 mice (10). Felbamate antagonizes audiogenic seizures with an ED_{50} value of 23.1 (12.1–44.0) against tonus and 48.8 (35.4–67.2) against clonus, respectively. The ED_{50} values of 5,7DCKA against tonus were 2.2 (1.2–3.9) nmol/mouse and against clonus 2.4 (1.2–5.0) nmol/mouse, respectively.

Like felbamate and 5,7DCKA, pretreatment (20 min) with NBQX (1.1–33.6 mg/kg IP, n = 60) was able to suppress, in a dose-dependent manner, the severity of the audiogenic seizures (11). The ED_{50} values of NBQX against tonic and clonic seizures were 4 (2.1–7.6) and 6.1 (3.2–11.9), respectively.

Stimulation of Excitatory Amino Acid Receptors

NMDA by itself (0.2–10 nmol/mouse, ICV, n = 50) produced generalized seizures. In particular, tremor and headbobbing, hypermotility, jumping, and circling preceded the first clonic episode, which consisted of wild running, jumping, and loss of righting.

As previously reported, felbamate (1–100 mg/kg, IP, n = 50) and 5,7DCKA (5–60 nmol/mouse ICV, 30 min before, n = 50), dose dependently reduced the incidence of clonus and tonus produced by NMDA (10). NBQX pretreatment (1–40 mg/Kg IP, n = 60), 20 min before, prevented the incidence of clonus and tonus induced by the calculated NMDA CD97 for clonic seizures. The ED_{50} values of NBQX against tonus were 18.9 (10.7–33.2) mg/kg and against clonus 22.1 (12.6–38.8) mg/kg, respectively. In particular, significant protection ($p < 0.01$) against tonus was evident following NBQX 20 and 40 mg/kg IP, while significant antagonism of clonus was obtained after NBQX 20 ($p < 0.05$) and 40 ($p < 0.01$) mg/kg IP.

Injections of AMPA (1–10 nmol/mouse, ICV), an agonist at non-NMDA receptors, induced generalized seizures, similar to those caused by NMDA administration (10). As previously reported, felbamate (3–100 mg/kg, IP, n = 50) or NBQX (1–40 mg/kg IP, n = 60) pretreatment (1 h or 20 min, respectively) reduced the incidence of all limb clonus and forelimb tonic extension seizures induced by the ICV administration of CD97 of AMPA for either clonus or forelimb tonic extension (10). Similarly to felbamate and NBQX, ICV pretreatment with 5,7DCKA (5–60 nmol/mouse, 30 min before, n = 60) was able to suppress, in a dose-dependent manner, the tonic and clonic component of seizures induced by AMPA. The ED_{50} values of 5,7DCKA against tonus were 44.0 (33.7–57.6) mg/kg and

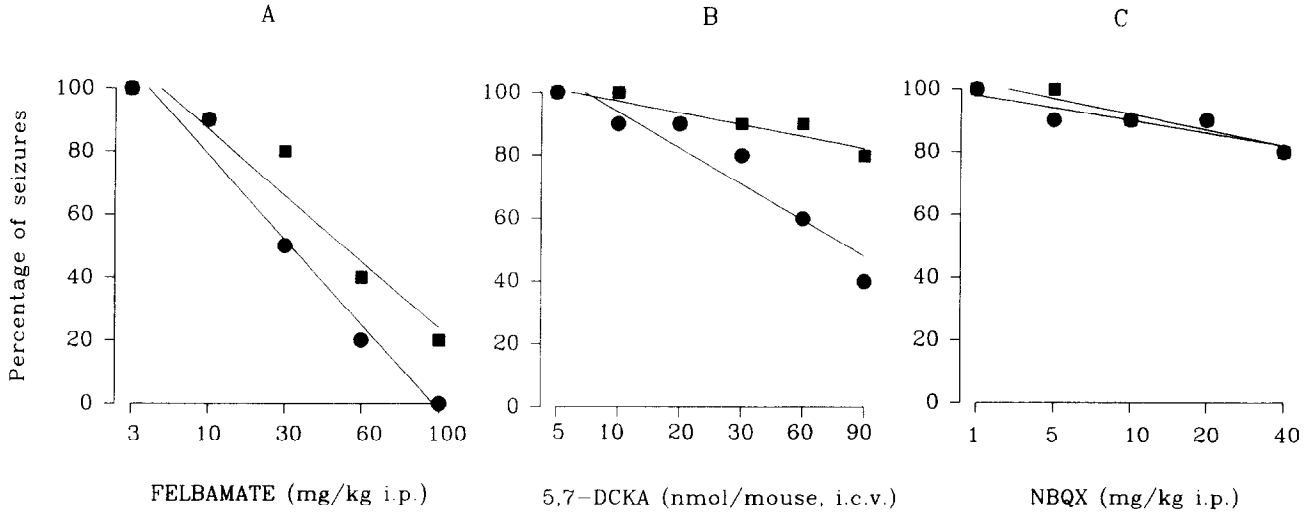

FIG. 1. Effects of felbamate (A), 5,7DCKA (B), and NBQX (C) on clonic and tonic phases of the seizures induced by ICV injection of Bay k 8644 in DBA /2 mice. Groups of 10 mice were pretreated with increasing felbamate (3–100 mg/kg, IP; 60 min before) 5,7DCKA (5–90 nmol/mouse ICV, 30 min before) or NBQX (1–40 mg/kg, IP; 20 min before) and then individual mice were challenged with either the CD97 of Bay k 8644 for clonus (48.9 nmol/mouse, ICV) or tonus (53.9 nmol/mouse, ICV). Mice were observed for the presence or absence of seizure activity for 30 min. ■ clonus; ● tonus.

against clonus 51.9 (40.5–66.6) mg/kg, respectively. In particular, significant antagonism of tonus was observed following 5,7DCKA 30 ($p < 0.05$) and 60 ($p < 0.01$) nmol/mouse ICV, while a significant protection ($p < 0.01$) against clonus was seen following 5,7DCKA 60 nmol/mouse.

Dihydropyridine Ca^{2+} Channels

Injection of Bay k 8644 (28–112 nmol/ mouse, ICV, n = 60), an activator of dihydropyridine calcium channel, induced clonic–tonic seizures. The behavioral pattern of Bay k 8644 and the incidence of seizures was previously described to be dose dependent within the dose range studied (9). The CD97 for clonus was 48.9 nmol/mouse, while that for forelimb tonic extension was 53.9 nmol/mouse. All mice treated with the latter dose showed generalized seizures and 9 out of 10 mice died following tonic extension.

As shown in Fig. 1A, felbamate pretreatment (3–100 mg/kg IP, 1 h, n = 50) was able to reduce the incidence of both clonus and forelimb tonic extension seizures induced by the ICV injection of the CD97 of Bay k 8644 for either clonus or forelimb tonic extension. The ED_{50} values of felbamate against tonic and clonic seizures are reported in Table 1. Pretreatment (30 min) with 5,7DCKA (5–90 nmol/mouse ICV, n = 60) was able to significantly decrease the incidence of tonus induced by the ICV injection of Bay k 8644 (Fig. 1B). Observation of the clonic component of seizures induced by Bay k 8644 was not feasible for the presence of side effects (ataxia), which appeared after the administration of the highest doses (5,7DCKA 60 and 90 nmol/mouse ICV). The ED_{50} values of 5,7DCKA against tonic seizures are reported in Table 1. As shown in Fig. 1C, pretreatment (20 min) with NBQX (1–40 mg/kg IP, n = 60) did not significantly antagonize the incidence of forelimb clonus and tonus induced by ICV injection of the CD97 of Bay k 8644 for either tonus (53.9 nmol/mouse) or clonus (48.9 nmol/mouse).

Voltage-Dependent K^+ Channels

The ICV administration of α-dendrotoxin (0.08–0.24 nmol/mouse, n = 60) produced a typical sequence of epileptic signs. In particular, within 5 min following the injection of α-dendrotoxin, compulsive grooming, jaw opening and forelimb stereotypies were observed. These signs were followed by trembling, hindlimb myoclonus, straub tail, and opistotonus. Following the highest doses of α-dendrotoxin (0.2 and 0.24 nmol/mouse, ICV) most of the mice showed hindlimb myoclonus, falling down, wild running, and hindlimb tonic extension, sometimes followed by death. The calculated CD97 of α-dendrotoxin for tonic extension was 0.26 nmol/mouse, while that for clonus was 0.21 nmol/mouse. All mice treated with the former dose showed tonic–clonic seizures, and 9 out of 10 animals died following tonic extension.

As shown in Fig. 2A, felbamate pretreatment (3–100 mg/kg, IP, 1 h, n = 50) reduced in a dose-dependent manner the incidence of both clonus and forelimb tonic extension induced by the ICV injection of the CD97 of α-dendrotoxin for either clonus or tonic extension. On the contrary, pretreatment with 5,7DCKA (5–90 nmol/mouse, ICV, n = 60) or with NBQX (1–40 mg/kg IP, n = 50) did not significantly antagonize the incidence of forelimb clonus and tonus induced by ICV injection of the CD97 of α-dendrotoxin (Fig. 2B and C). The ED_{50} values of felbamate, 5,7DCKA and NBQX against tonic extension and clonus are reported in Table 1.

Effects on Behavior and Motor Movements

No impairment of coordinated motor movements was present in DBA/2 mice receiving NBQX or felbamate, while mild piloerection was observed following IP injection of the highest dose of NBQX (40 mg/kg). Neurological side effects such as mild ataxia, squatting posture, and piloerection appeared after the ICV administration of the highest doses of 5,7DCKA (60 and 90 nmol/mouse).

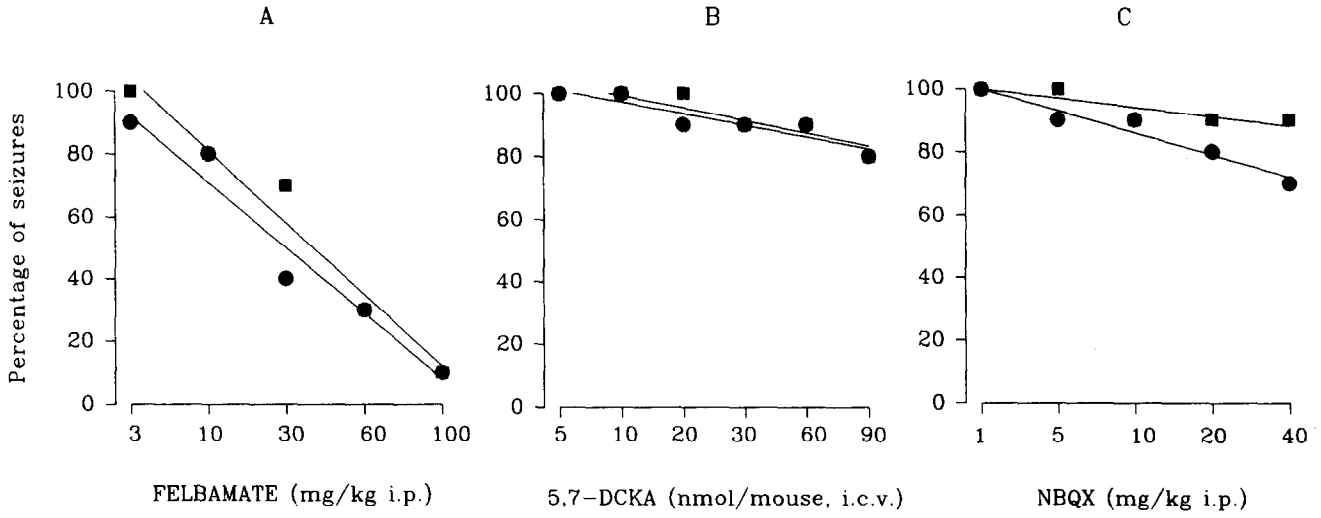

FIG. 2. Effects of felbamate (A), 5,7DCKA (B), and NBQX (C) on clonic and tonic phases of the seizures induced by ICV injection of α-dendrotoxin in DBA/2 mice. Groups of 10 mice were pretreated with increasing doses of felbamate (3–100 mg/kg, IP; 60 min before) 5,7DCKA (5–90 nmol/mouse ICV, 30 min before), or NBQX (1–40 mg/kg, IP; 20 min before) and then individual mice were challenged with either the CD97 of α-dendrotoxin for clonus (0.21 nmol/mouse, ICV) or tonus (0.26 nmol/mouse, ICV). Mice were observed for the presence or the absence of seizure activity for 30 min. ■ clonus; ● tonus.

DISCUSSION

The present results show that felbamate, 5,7DCKA, and NBQX possess anticonvulsant properties in DBA/2 mice, a genetic model used for its susceptibility to seizures, specifically, reflex epilepsy, which may counterpart in human to brainstem or centrencephalic epilepsy (2,3,12,14,25). We have previously demonstrated that these drugs fully protected DBA/2 mice from sound-induced tonic extension (10,11) with ED_{50} values lower than those observed in the maximal elettroshock seizure model. For example, felbamate had an ED_{50} of 23 mg/kg IP against tonus while in the electroshock model it has anticonvulsant activity with an ED_{50} value of 50.1 mg/kg, IP (29) or 59 mg/kg, SC (6). 5,7DCKA possesses an ED_{50} value of 2.2 nmol/mouse ICV against tonus (10). NBQX showed an ED_{50} of 4 mg/kg IP against tonus (11), which is markely lower than that found in the maximal electroshock seizure model (0.51 nmol/kg, equivalent to 174.4 mg/kg) by Turski and co-workers (31). Over a low dose range, felbamate was equipotent against tonic seizures induced by the two different agonists for the excitatory amino acids receptor complex, NMDA and AMPA. As expected, 5,7DCKA was found to be twice as potent against seizures induced by NMDA as against those induced by AMPA. Conversely, NBQX had the most pronounced anticonvulsant activity against seizures induced by ICV injection of AMPA. The drug, which has higher affinity for AMPA receptors (26), had a protective action against NMDA-induced seizures by in DBA/2 mice, further confirm the mutual interaction that occurs between NMDA and nonNMDA receptors (17). However, some authors have suggested that clear evidence for receptor selectivity of the specific antagonists cannot be easily obtained by simple comparison of their potency in antagonizing seizures induced by AMPA or NMDA (1). In this regard, the nonselective anticonvulsant effects of 5,7DCKA observed in the present study further support such conclusions.

The comparison of effective doses of NBQX in various models of experimental epilepsy indicates that this compound is almost equipotent against seizures induced by 3-mercaptopropionic acid, AMPA, or audiogenic stimulation. The anticonvulsant effects were observed in absence of clear-cut muscle relaxation, while effects on muscle relaxation were evident when NBQX prevented seizures induced by compounds interacting with GABA or in maximal electroshock (31). In previous studies, it has been demonstrated that the mechanisms involved in the reflex activity of the spinal cord are either monosynaptic (AMPA-mediated) and polysynaptic (NMDA-mediated), and malfunction of different glutamate receptors subtypes may result in diverse neurological disorders of the spinal cord (31,32). The antispastic action of NBQX was observed in the absence of sedation.

Pharmacological Profile of Felbamate

The present data clearly show that felbamate has different pharmacological profile than that of 5,7DCKA, CPPene, and NBQX. In particular, felbamate has a broader anticonvulsant spectrum than 5,7DCKA, CPPene, and NBQX. Comparing effective doses of felbamate in various convulsion models, it emerges that the drug is equipotent against seizures induced by ICV injection of either NMDA or non-NMDA receptors. Although it is difficult to infer the mechanism of action from in vivo models, it appears that felbamate interacts with more mechanisms activated by the excitatory amino acids, a finding in agreement with other electrophysiological findings (13). In addition, felbamate was much more potent against seizures induced by NMDA or AMPA than against Bay k 8644 or α-dendrotoxin–induced seizures. Conversely, the drug is much less effective against a variety of other chemical agents that induce seizures through the blockade of different mechanisms, for instance, blockade of the GABAergic transmission (21). A quantitative comparison of the anticonvulsant efficacy of felbamate in DBA/2 mice against audiogenic seizures and those induced by chemoconvulsants acting on excitatory amino acid neurotransmission or ionic channels shows that this compound possess a broader spectrum of anticonvulsant activity than antagonists acting on excitatory amino acid receptors or classical antiepileptics (4,9,11,13). The differences ob-

served between felbamate and other compounds examined in the present study suggest that felbamate could act by a channel blocking mechanism as well as an antagonist of excitation induced by excitatory amino acids. Very recently, it has been suggested that felbamate may principally act by a channel-blocking action and also possibly by distinct effects on channel gating linked to NMDA receptors (28,30).

The low toxicity of felbamate in comparison to compounds acting on specific receptors furtherly suggest that it may exert its therapeutic effects via low-affinity interaction with more than one receptor system. This contradicts the conventional view in drug development that side effect reduction is best achived by high selectivity and potency. However, in the case of anticonvulsant compounds, which modulate the activity of excitability mechanisms critical to normal brain functions, drugs with high potency and efficacy may have usually unaceptable toxicity. In fact, some of the most favorable antiepileptic drugs appear to be of modest potency or exert anticonvulsant effects at low levels of receptor occupancy (28). Moreover, as in the case of felbamate, it may be possible to achieve enhanced anticonvulsant activity by a combination of actions that, by themselves, are below the threshold for side effects.

Felbamate provides an interesting example of research efforts to discover new drugs that interact with excitatory amino acid effects as well as with ionic channels (23). However, the mechanism underlying felbamate's action requires further investigation. Molecular biology techniques, which permit the cloning and expression of genes encoding ion channels, could be useful to study more deeply the interactions between felbamate, receptors, and ion channels.

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