Anesthesiology
V 43, No 4, Oct 1975

Barbiturates and Aortic and Venous Smoooth-muscle Function

Bella T. Altura, Ph.D. and Burton M. Altura, Ph.D.†*

Using isolated rat aortic strips (AS) and portal veins (PV), it was found that all of the barbiturates studied (thiopental, secobarbital, pentobarbital, amobarbital, phenobarbital, and barbital): a) inhibit development of spontaneous mechanical activity (vasomotion) in AS and PV in concentrations used to induce surgical anesthesia or concentrations used for anticonvulsive therapy; b) dose-dependently attenuate contractions induced by epinephrine and potassium (K^+); c) cause non-competitive displacement of the dose–response curves of these vasoactive compounds; d) attenuate calcium (Ca^{++})-induced contractions of K^+-depolarized AS and PV; e) rapidly relax drug-induced, as well as Ca^{++}-induced, contractions of AS and PV. In addition, the data indicate that: a) rat portal venous smooth muscle is more sensitive to the inhibitory actions of barbiturates than is rat aortic smooth muscle, and b) thiopental, but none of the other barbiturates, can elicit dose-dependent contractions of AS. Concentrations of barbiturates known to be present during induction of surgical anesthesia can exert depressant effects on at least two types of vascular smooth muscle that may be related to actions on movement and/or translocation of Ca^{++}. (Key words: Hypnotics, barbiturates; Arteries, barbiturates; Veins, barbiturates.)

CONSIDERABLE CONTROVERSY currently exists as to the effects of barbiturates on the peripheral vasculature.[1–3] We demonstrated in a preliminary study[4] that anesthetic and anticonvulsive concentrations of pentobarbital produce nonspecific inhibition of contraction of isolated vascular smooth muscle. Price and Price,[5] as well as Burn and Hobbs,[6] reported that thiopental induces contractions of rabbit aortic strips. It was therefore of interest to expand our preliminary studies with pentobarbital to additional types of barbiturates (short-acting, intermediate, and long-acting). Such direct *in-vitro* studies of arterial and venous smooth-muscle segments eliminate the possible contributions that *in-vivo* reflex, metabolic and cardiac components may exert on intact blood vessels. The results indicate that concentrations of barbiturates used to induce surgical anesthesia can exert profound depressant effects on vascular smooth muscle.

Methods

Thoracic aortas and portal veins were obtained from male rats (Wistar strain, 275–425 g) after sacrifice by guillotine. The aortas were cut helically into vascular strips (1.5–2.0 mm wide by 25 mm long) and set up isometrically, *in vitro*, under a resting tension of 1.5 g, essentially as described previously.[7] Ten-millimeter segments of portal veins were tied at both ends with sutures and arranged isometrically, *in vitro*, under a resting tension of 500 mg. Both aortic strips and portal vein segments were equilibrated for 2 hours in muscle chambers containing Krebs-Ringer bicarbonate (KRB) solution.[7] The KRB solution was oxygenated continuously with 95 per cent O_2–5 per cent CO_2 and kept at 37 C (*p*H 7.4–7.5). The loading tensions were maintained and periodically adjusted throughout the experiments. The incubation media were routinely changed every 10–15 min as a precaution against interfering metabolites.[8] The recording equipment was identical to that described previously.[8]

* Assistant Professor of Physiology, Downstate Medical Center.

† Professor of Physiology, Downstate Medical Center, and Visiting Professor of Anesthesiology and Physiology, Albert Einstein College of Medicine.

Received from the Department of Physiology, State University of New York, Downstate Medical Center, Brooklyn, New York 11203, and the Departments of Anesthesiology and Physiology, Albert Einstein College of Medicine of Yeshiva University, Bronx, New York 10461. Accepted for publication May 27, 1975. Supported in part by grant HL-12462 of the National Heart and Lung Institute and by grant MH-26236 from the National Institute of Mental Health. A preliminary report of a portion of this work was presented before the Fall Meeting of the American Society for Pharmacology and Experimental Therapeutics, Montreal, 1974.

Address reprint requests to Burton M. Altura, Ph.D., Department of Physiology, State University of New York, Downstate Medical Center, 450 Clarkson Avenue, Brooklyn, New York 11203.

After the 2-hour incubation period, the following types of experiments were run on the aortic strips and portal veins: 1) Aortic strips and portal veins were exposed to KRB solution containing various concentrations of the barbiturates (*i.e.*, 2.5×10^{-5} to 2×10^{-3} M) for 5- to 15-min periods to determine whether barbiturates affected baseline tension and/or development of spontaneous mechanical activity. The time period of 5 to 15 min was chosen since systemic administration of barbiturates to intact rats induces general anesthesia within 15 min. In fact, longer equilibration times (*i.e.*, 45–60 min) did not alter the results. 2) In other experiments different aortas and portal veins were exposed to epinephrine (Adrenalin chloride, Parke-Davis and Co.) and potassium (potassium chloride, A.C.S. certified) before and after exposure to different concentrations of barbiturates (*i.e.*, 5×10^{-5} to 2×10^{-3} M) (10 min preincubation period); these agonists were chosen since they are thought to produce contraction of vascular smooth muscle by different mechanisms.[8] The latter experiments employed single agonist doses (*i.e.*, submaximal and maximal), as well as complete cumulative dose–response curves. The results of these experiments are expressed as developed isometric tension (mg) and as percentages of maximal agonist-induced contractile responses. Barbiturate-induced changes in baseline tension of aortic strips were adjusted to control level before addition of the agonists. 3) In addition, various doses of barbiturates were added after the establishment of single-dose agonist-induced contractile responses. 4) Paired aortic strips from the same animal and portal vein segments equilibrated in normal KRB solution were exposed to two successive supramaximal doses of epinephrine, 10 μg/ml. Upon relaxation of the last contraction, in normal KRB solution, these paired tissues were exposed for 30 min to Ca^{++}-free KRB solution, followed by exposure to a Ca^{++}-free potassium chloride depolarizing solution for 45 min. The latter solution had 118 mM NaCl iso-osmotically replaced with KCl (total $K^+ = 123.9$ mM). Such tissues contract in response to added Ca^{++}.[7] Cumulative $CaCl_2$ dose–response curves were therefore obtained for paired, depolarized Ca^{++}-depleted aortic strips in the absence or presence of the barbiturates (10 min preincubation). These results are expressed as percentages of maximal epinephrine-induced contractile responses, since it has been demonstrated that catecholamines produce the greatest maximal response in rat aorta[7] and portal veins (unpublished experiments). Where appropriate, the means (±SE) of the responses in control and experimental (with barbiturate) vascular tissues were compared for statistical significance by means of Student's t test, multiple-range t test, t test for paired data, or analysis of variance. In all cases, a minimum of six different preparations was utilized for each experiment. The barbiturates employed in these studies were: amobarbital sodium (Amytal sodium [powder], E. Lilly and Co.), thiopental sodium (Pentothal sodium [powder], Abbott Laboratories), pentobarbital sodium (Nembutal sodium, Abbott Laboratories), secobarbital sodium (sodium secobarbital injection, Interstate Drug Exchange), phenobarbital sodium (sodium phenobarbital injection, U.S.P., Parke-Davis), and sodium barbital (sodium diethylbarbiturate, purified powder, Fisher Scientific Co.). In those instances where the barbiturates contained preservatives, *e.g.*, 40 per cent propylene glycol and 10 per cent alcohol (pentobarbital sodium) and 60 per cent propylene glycol and 2 per cent benzyl alcohol (secobarbital sodium), appropriate control studies with preservatives alone were utilized. None of these vehicles, in the concentrations employed, exerted any of the vascular effects produced by the barbiturates.

Results

Influence of Barbiturates on Baseline Tension and Spontaneous Mechanical Activity of Rat Aortic Strips and Portal Veins

Figures 1 and 2 show recordings of typical changes in resting tension and spontaneous mechanical activity in isolated aortas and portal veins, after the addition of different concentrations of barbiturates. Concentrations of as little as 1×10^{-4} M of secobarbital, amobarbital, or phenobarbital partially inhibited spontaneous mechanical activity as

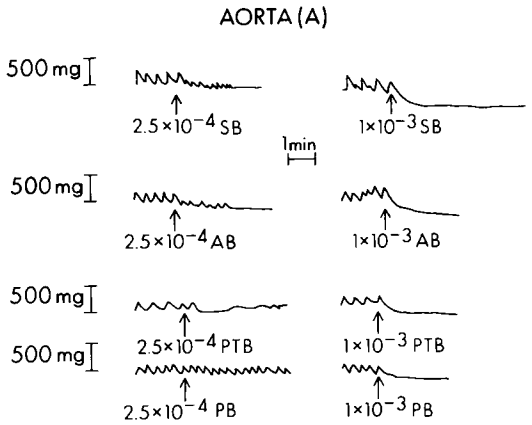

FIG. 1. Effects of various concentrations of secobarbital (SB), amobarbital (AB), phenobarbital (PB) and pentobarbital (PTB) on spontaneous mechanical activity and resting tension in four isolated rat aortic strips (A). The arrows indicate the point at which the tissues were exposed to the barbiturates (molar concentration). The values on the left of this figure and the others represent tension.

well as lowered resting tension in rat aortic strips; less than 1×10^{-4} M did not influence spontaneous activity or resting tension. Slightly higher concentrations (*e.g.*, $2\text{–}5 \times 10^{-4}$ M) of pentobarbital and barbital were required to exert similar actions. In all cases, the greater the concentration of barbiturate, the more rapidly the inhibition became manifest and the more the resting tension was lowered (fig. 1). Figure 2 demonstrates that barbiturates also inhibited spontaneous mechanical activity in rat portal venous smooth muscle, but that the threshold inhibitory concentrations for the barbiturates, in all cases, were less than in the aorta, as in figure 1 (*e.g.*, $0.25\text{–}5 \times 10^{-5}$ M *vs* $1\text{–}2.5 \times 10^{-4}$ M). In addition, although barbiturates lowered resting tension in rat aortic strips (fig. 1), they had no such effect in rat portal veins (fig. 2). Exposure of the aorta to increasing concentrations of thiopental resulted in dose-dependent contractions (fig. 3). These contractions could be elicited repetitively in the same aortic strips and did not manifest tachyphylaxis. Thiopental did not induce contraction in veins but did inhibit spontaneous activity (fig. 3).

Influence of Barbiturates on Contractions of Rat Aortic Strips and Portal Veins Induced by Single Doses of Vasoactive Agents

Figure 4 demonstrates that exposure of rat aorta and portal vein to different concentrations of the barbiturates, 10 min prior to the addition of epinephrine or potassium chloride, inhibited in a dose-related manner the contractile responses of both vasoactive agents. A plot of the log of barbiturate concentration versus percentage inhibition of the ED_{50} isometric contractile responses of potassium chloride and epinephrine (fig. 5) reveals that: 1) the regression lines for all of the barbiturate-induced inhibitory effects, with the exception of barbital, are parallel, thereby suggesting that all of the barbiturates (except barbital) may be acting on vascular smooth muscle by the same mechanism; 2) barbiturates have a distinct relative order of inhibitory potency: thiopental > secobarbital ≅ pentobarbital > amobarbital > phenobarbital >> barbital.

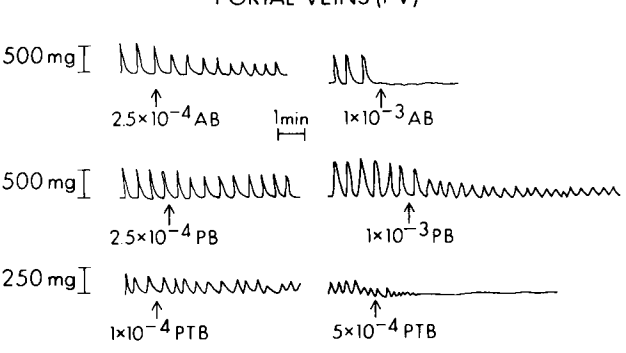

FIG. 2. Effects of various concentrations of barbiturates on development of spontaneous mechanical activity in three isolated rat portal veins (PV). Symbols and conventions are similar to those in figure 1.

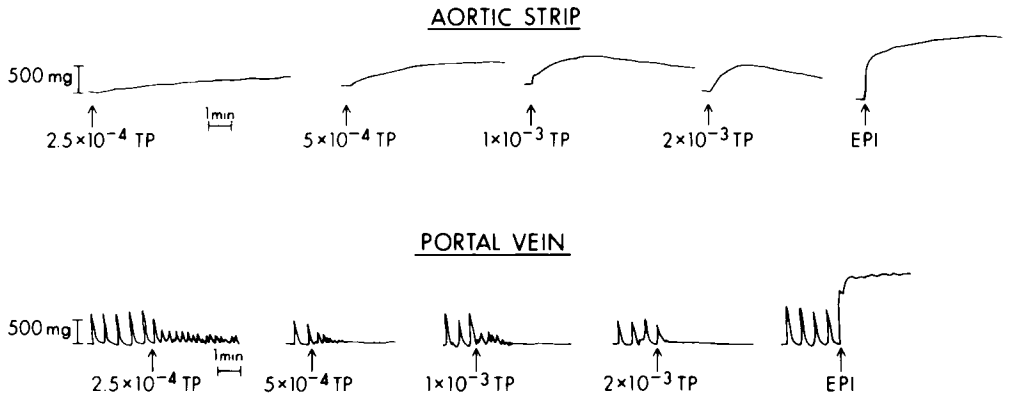

FIG. 3. Effects of various concentrations of thiopental (TP) on a single aortic strip and a single portal vein. For comparative purposes, a maximum epinephrine (EP)-induced contraction (5 μg/ml) in each blood vessel is also shown.

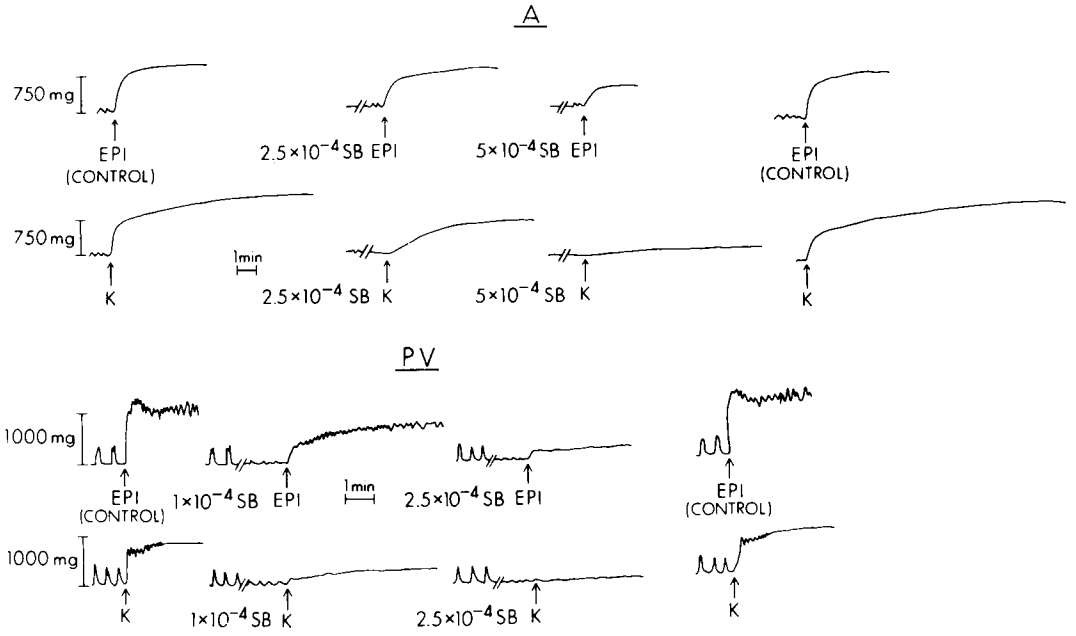

FIG. 4. Effects of barbiturates on epinephrine (EPI)-and potassium chloride (K)-induced contractions in rat aortic strips (A) and portal veins (PV). The barbiturate, SB, remained in contact with the vascular strips during addition of EPI (2.5 μg/ml—A; 5 μg/ml—PV) and K (20 mM—A; 80 mM—PV). A different aorta and a different portal vein were used for each agonist. Note that the K-induced responses appear to be more sensitive to the barbiturates than are the EPI-induced responses. In addition, this figure demonstrates that barbiturate inhibition is completely reversible upon washing.

EFFECTS OF BARBITURATES ON EPINEPHRINE AND POTASSIUM DOSE–RESPONSE CURVES OF RAT AORTIC STRIPS AND PORTAL VEINS

The above-described findings with single doses of agonists suggest that barbiturates might act as depressants of vascular muscle by nonspecifically inhibiting the action of vasoactive stimulants. We turned our attention to studying the effects of barbiturates on the complete dose–response curves of the two agonists.

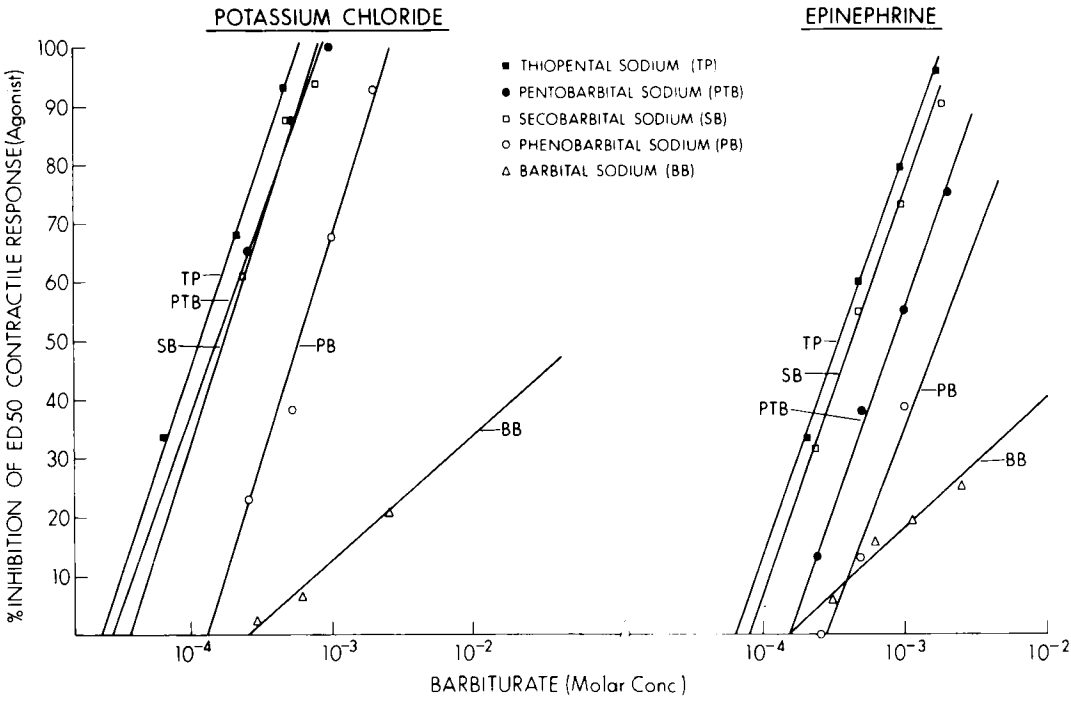

FIG. 5. Log concentration–effect curves of the percentage inhibition to ED_{50} isometric contractile responses of potassium chloride and epinephrine in aortic strips produced by different barbiturates.

Figure 6 compares catecholamine concentration–effect curves for rat aortas in the presence of a very potent barbiturate (*i.e.*, thiopental) and a weakly potent barbiturate (*i.e.*, phenobarbital). This figure demonstrates that increasing concentrations of barbiturates shifted the epinephrine dose–response curves to the right concomitant with reduction in maximum response and that thiopental was more potent as an inhibitor of epinephrine-induced contractions than was phenobarbital. Figure 6 also indicates that the difference between the potencies of these two barbiturates was more pronounced in venous smooth muscle. In addition, it is obvious from figure 6 that rat portal venous muscle was more sensitive to the inhibitory actions of the barbiturates than aortic (arterial) smooth muscle. For example, 1) 1×10^{-4} M thiopental failed to inhibit epinephrine-induced responses in aortic smooth muscle, but depressed the maximum portal venous smooth muscle responses by almost 55 per cent, and 2) although $1\text{–}5 \times 10^{-4}$ M phenobarbital also failed to inhibit catecholamine-induced responses in the arterial muscle, these concentrations significantly shifted the catecholamine dose–response curves for the portal veins to the right and reduced the maximum contractile tension by as much as 30 per cent. Tables 1 and 2 compare the relative potencies of several other intermediately acting barbiturates with those of thiopental and phenobarbital in the two types of vascular muscle. These comparative data, especially those in the last columns in tables 1 and 2, indicate a relative rank order of inhibitory potency: thiopental > secobarbital ≅ pentobarbital > amobarbital >> phenobarbital.

Potassium-induced contractions of both aortic strips and portal veins were also markedly inhibited by barbiturates (fig. 7). Here, too, the potassium-induced responses in venous muscle were more sensitive than aortic strips to inhibitory actions of the barbiturates (fig. 7, tables 3 and 4); this is evident in the fact that the ED_{50} concentrations and maximum tensions were relatively more depressed in the veins. In addition, tables 3 and 4 indicate for the

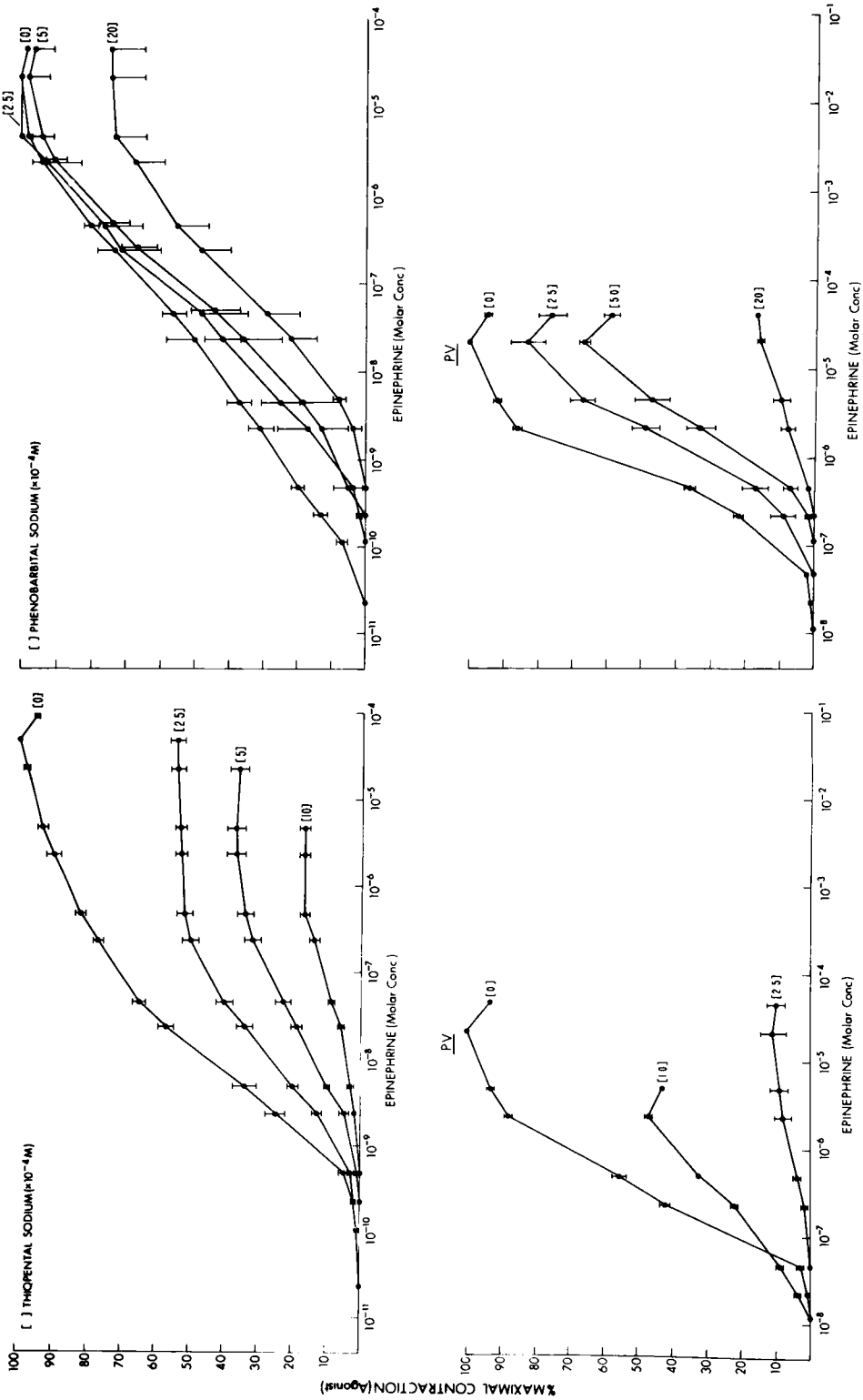

FIG. 6. Effects of thiopental and phenobarbital ($\times 10^{-4}$ M) on epinephrine dose–response curves in rat aortic strips (A) and portal veins (PV). Values are means ± 1 SEM. At least six different strips were utilized for each concentration of barbiturate.

TABLE 1. Influence of Barbiturates on Epinephrine-induced Contractile Responses of Isolated Rat Aortic Strips

Barbiturate (M)	Epinephrine Responses			
	Threshold Concentration (M)*	ED_{50}(M)	Maximum Tension (mg)	Inhibition of Control Tension (Per Cent)
Thiopental				
Controls	$1.2 \times 10^{-10} \pm 0.1$	$1.5 \times 10^{-8} \pm 0.4$	990 ± 45	
2.5×10^{-4}	$1.2 \times 10^{-10} \pm 0.1$	$1.1 \times 10^{-8} \pm 0.4$	534 ± 68†	36.1
2×10^{-3}	$2.4 \times 10^{-9} \pm 0.1$†	$4.0 \times 10^{-8} \pm 0.2$†	168 ± 32†	83.0
Secobarbital				
Controls	$1.2 \times 10^{-10} \pm 0.2$	$2.0 \times 10^{-9} \pm 0.5$	$1,150 \pm 28$	
2.5×10^{-4}	$4.8 \times 10^{-10} \pm 0.1$†	$1.5 \times 10^{-8} \pm 0.4$†	943 ± 60†	18.0
2×10^{-3}	$2.3 \times 10^{-9} \pm 0.2$†	$7.5 \times 10^{-9} \pm 0.2$†	172 ± 15†	85.0
Pentobarbital				
Controls	$1.2 \times 10^{-10} \pm 0.2$	$9.0 \times 10^{-9} \pm 0.7$	$1,370 \pm 42$	
2.5×10^{-4}	$4.7 \times 10^{-10} \pm 0.4$†	$2.4 \times 10^{-8} \pm 0.3$†	$1,227 \pm 36$	10.0
2×10^{-3}	$2.4 \times 10^{-9} \pm 0.3$†	$4.7 \times 10^{-8} \pm 0.2$†	382 ± 20†	72.1
Amobarbital				
Controls	$1.2 \times 10^{-10} \pm 0.1$	$1.6 \times 10^{-8} \pm 0.5$	$1,177 \pm 32$	
2.5×10^{-4}	$2.4 \times 10^{-9} \pm 0.2$†	$7.0 \times 10^{-8} \pm 0.5$†	$1,153 \pm 35$	2.0
2×10^{-3}	$2.4 \times 10^{-9} \pm 0.1$†	$4.0 \times 10^{-8} \pm 0.5$†	529 ± 21†	55.1
Phenobarbital				
Controls	$1.2 \times 10^{-10} \pm 0.2$	$5.0 \times 10^{-8} \pm 0.7$	931 ± 78	
2.5×10^{-4}	$1.2 \times 10^{-10} \pm 0.4$	$4.6 \times 10^{-8} \pm 0.9$	930 ± 88	0
2×10^{-3}	$2.2 \times 10^{-9} \pm 0.3$†	$8.8 \times 10^{-8} \pm 0.7$†	688 ± 61†	26.1

* Values in the table represent means (± SEM) for at least six different aortic strips.
† Significantly different from control values, $P < 0.01$.

potassium-induced responses a relative rank order of inhibitory potencies similar to that observed for the epinephrine-induced contractions.

COMPARATIVE EFFECTS OF BARBITURATES ON CALCIUM CHLORIDE-INDUCED CONTRACTIONS OF AORTIC STRIPS AND PORTAL VEINS

Not only did barbiturates reduce the magnitude of the calcium-induced contractions of potassium-depolarized aortic strips and portal veins, but increasing barbiturate concentrations reduced the magnitude of the maximum tension as well (fig. 8). In addition, the barbiturates caused a dose-dependent displacement to the right and shallowing of the sigmoid log concentration–effect curves. It is evident from these dose–response curves that not only were the threshold concentrations of Ca^{++} increased by the barbiturates, but the ED_{50} Ca^{++} concentrations were also increased in the presence of barbiturates. It should also be noted that: 1) of all the barbiturates tested, thiopental was the most potent in inhibiting Ca^{++}-induced contractions in both types of vascular muscle, whereas phenobarbital was the least potent (the other barbiturates were intermediate in potency: secobarbital > pentobarbital > amobarbital, table 5); 2) the calcium-evoked responses in portal venous smooth muscle were more sensitive to the inhibitory actions of the barbiturates than were the corresponding responses in aortic smooth muscle (*e.g.*, compare aorta *vs.* portal vein in fig. 8); 3) Ca^{++}-evoked responses in the two types of vascular tissue were more sensitive to the inhibitory actions of the barbiturates than were the responses to the other vasoactive agents used in this study (table 5).

Relaxation of Established Epinephrine-, Potassium-, and Calcium-induced Contractions of Aortic Strips and Portal Veins

Only single-dose experiments were performed. Submaximal and maximal concentrations of epinephrine and KCl were utilized in normal Krebs-Ringer bicarbonate solution. $CaCl_2$ in a Ca^{++}-free potassium-depolarizing solution (see Methods) was also used as an agonist in these experiments. All aortic strips and portal veins contracted by treatment with the three agonists were dose-dependently relaxed upon addition of barbiturates (fig. 9). This relaxant action became more pronounced as the concentration of barbiturate, in the isolated organ bath, was increased. These relaxant actions, as well as the inhibitory actions of the barbiturates noted above (*i.e.*, from 2.5×10^{-5} to 2×10^{-3}M), were completely and rapidly reversible after washing in normal KRB solution.

Discussion

The present findings demonstrate that barbiturates can potently depress contractility of at least two types of mammalian vascular smooth muscles. Furthermore, these inhibitory actions: 1) take place with concentrations of barbiturates known to be present during surgical anesthesia or anticonvulsive therapy (*i.e.*, $0.25–2.5 \times 10^{-4}$ M),[9–12] and 2) show a relative order of ptoency similar to the anesthetic–hypnotic potencies of these barbiturates, *i.e.*, thiopental > secobarbital ≅ pentobarbital > amobarbital > phenobarbital > barbital. It is now thought that most of the barbiturates used in our study can produce peripheral vasodilatation.[1–3] Although administration of anesthetic doses of some of these barbiturates

TABLE 2. Influence of Barbiturates on Epinephrine-induced Contractile Responses of Isolated Rat Portal Veins

Barbiturate (M)	Epinephrine Responses			
	Threshold Concentration (M)*	ED_{50}(M)	Maximum Tension (mg)	Inhibition of Control Tension (Per Cent)
Thiopental				
Controls	$2.4 \times 10^{-8} \pm 0.1$	$3.5 \times 10^{-7} \pm 0.4$	800 ± 84	
1×10^{-4}	$2.4 \times 10^{-8} \pm 0.2$	$2.7 \times 10^{-7} \pm 0.3$	376 ± 26†	53.0
2.5×10^{-4}	$2.4 \times 10^{-7} \pm 0.1$†	$8.3 \times 10^{-7} \pm 0.4$†	88 ± 47†	89.0
Secobarbital				
Controls	$4.6 \times 10^{-8} \pm 0.2$	$2.0 \times 10^{-7} \pm 0.2$	$1,050 \pm 67$	
1×10^{-4}	$4.6 \times 10^{-8} \pm 0.4$	$3.7 \times 10^{-7} \pm 0.4$†	832 ± 29†	20.8
2.5×10^{-4}	$2.4 \times 10^{-7} \pm 0.2$†	$4.8 \times 10^{-7} \pm 0.4$†	250 ± 36†	76.2
Pentobarbital				
Controls	$4.6 \times 10^{-8} \pm 0.3$	$2.7 \times 10^{-7} \pm 0.2$	$1,122 \pm 52$	
1×10^{-4}	$4.6 \times 10^{-8} \pm 0.2$	$3.2 \times 10^{-7} \pm 0.1$†	841 ± 36†	25.0
2.5×10^{-4}	$4.6 \times 10^{-8} \pm 0.2$	$4.5 \times 10^{-7} \pm 0.3$†	237 ± 10†	78.9
Amobarbital				
Controls	$4.6 \times 10^{-8} \pm 0.1$	$2.0 \times 10^{-7} \pm 0.3$	860 ± 87	
1×10^{-4}	$4.6 \times 10^{-8} \pm 0.1$	$5.0 \times 10^{-7} \pm 0.3$†	670 ± 24†	22.1
2.5×10^{-4}	$2.4 \times 10^{-7} \pm 0.4$†	$3.0 \times 10^{-7} \pm 0.2$†	438 ± 37†	49.1
Phenobarbital				
Controls	$2.4 \times 10^{-8} \pm 0.1$	$7.2 \times 10^{-7} \pm 0.4$	781 ± 98	
1×10^{-4}	$2.4 \times 10^{-8} \pm 0.2$	$7.5 \times 10^{-7} \pm 0.6$	775 ± 107	0.8
2.5×10^{-4}	$2.4 \times 10^{-7} \pm 0.6$	$1.5 \times 10^{-6} \pm 0.6$†	648 ± 81	17.0

* Values in the table represent means (± SEM) for at least six different portal veins.
† Significantly different from control values, $P < 0.01$.

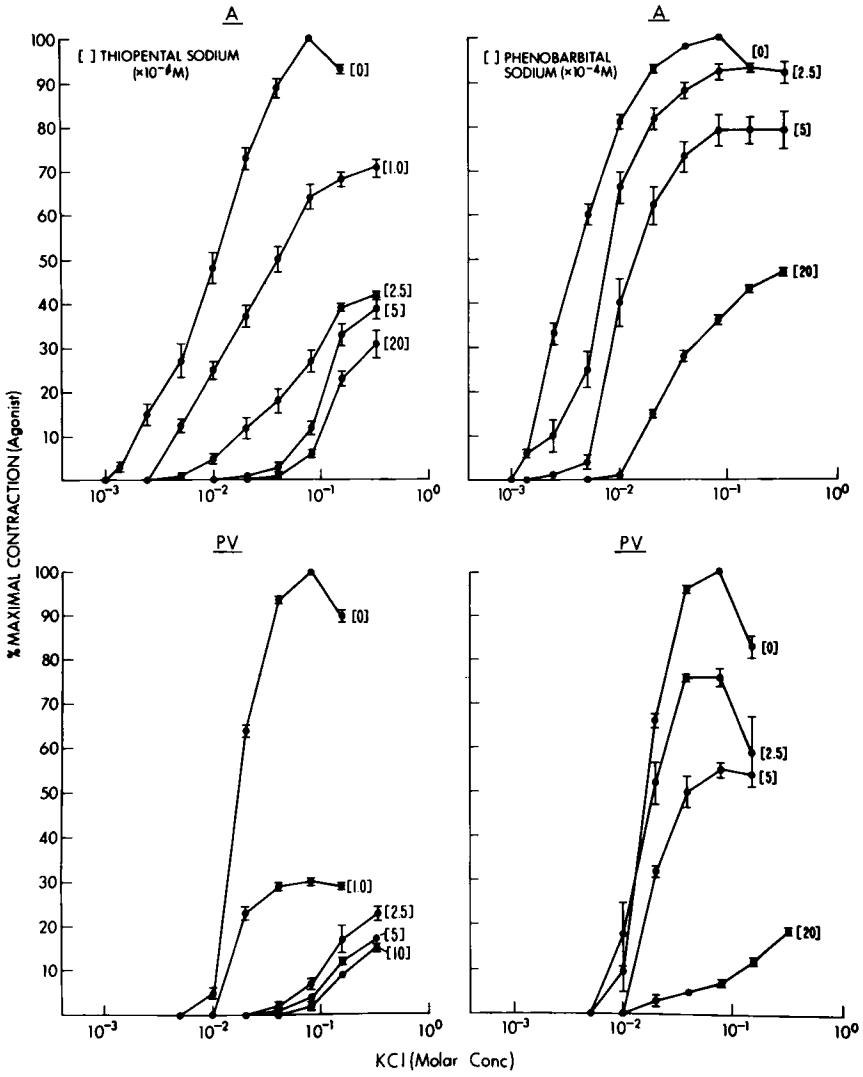

FIG. 7. Effects of thiopental and phenobarbital (× 10^{-4} M) on potassium chloride dose–response curves in rat aortic strips (*upper panels*) and rat portal veins (*lower panels*). Symbols and conventions similar to those in figure 6.

may not always result in peripheral vasodilatation,[1–3,12,13] it is generally thought that deep anesthesia, when induced by these hypnotic agents, will result in such dilatation.[1–3,12,14–17] In this context, one must consider the possiblity that the direct vasodepressant actions of the barbiturates observed here in isolated vascular smooth muscles may: 1) play a significant role in barbiturate-induced peripheral vasodilatation; and 2) be similar to the phenomenon observed in the arterioles and venules in the microcirculation.[14–16]

Our data could be used to suggest that the direct vascular actions of barbiturates may contribute to the decrease of vasoconstrictor tone associated with induction of anesthesia by barbiturates. The present findings suggest that at least two vascular mechanisms could contribute to the latter: 1) depression of vasomotor tone via an inhibition of the normal rhythm or vasomotion (*i.e.*, spontaneous mechanical activity) of vascular smooth muscle; 2) depression of vasomotor tone via an inhibition of the contractile (constrictor) action of endogenous neurohumoral substances that are believed to play roles in maintaining vascular tone.[18]

Any explanation proposed to account for the

TABLE 3. Influence of Barbiturates on Potassium-induced Contractile Responses of Isolated Rat Aortic Strips

Barbiturate (M)	Potassium Responses			
	Threshold Concentration (M)*	ED_{50} (M)	Maximum Tension (mg)	Inhibition of Control Tension (Per Cent)
Thiopental				
Controls	$1.4 \times 10^{-3} \pm 0.2$	$1.1 \times 10^{-2} \pm 0.5$	816 ± 77	
1×10^{-4}	$5.0 \times 10^{-3} \pm 0.3$†	$2.0 \times 10^{-2} \pm 0.4$	579 ± 38†	29.0
5×10^{-4}	$2.0 \times 10^{-2} \pm 0.3$†	$5.4 \times 10^{-2} \pm 0.2$†	326 ± 19†	60.0
Secobarbital				
Controls	$1.4 \times 10^{-3} \pm 0.2$	$6.9 \times 10^{-3} \pm 0.3$	693 ± 83	
1×10^{-4}	$5.0 \times 10^{-3} \pm 0.5$†	$1.2 \times 10^{-2} \pm 0.5$†	637 ± 47	8.1
5×10^{-4}	$1.0 \times 10^{-2} \pm 0.1$†	$8.5 \times 10^{-2} \pm 0.3$†	277 ± 35†	60.0
Pentobarbital				
Controls	$5.5 \times 10^{-4} \pm 0.4$	$1.3 \times 10^{-2} \pm 0.1$	715 ± 37	
1×10^{-4}	$1.2 \times 10^{-3} \pm 0.3$†	$2.1 \times 10^{-2} \pm 0.2$†	608 ± 25†	15.0
5×10^{-4}	$5.0 \times 10^{-3} \pm 0.3$†	$3.8 \times 10^{-2} \pm 0.1$†	436 ± 22†	39.0
Amobarbital				
Controls	$1.4 \times 10^{-3} \pm 0.1$	$5.5 \times 10^{-3} \pm 0.3$	860 ± 92	—
1×10^{-4}	$2.5 \times 10^{-3} \pm 0.4$†	$8.0 \times 10^{-3} \pm 0.4$†	765 ± 47	11.0
5×10^{-4}	$1.0 \times 10^{-2} \pm 0.1$†	$4.0 \times 10^{-2} \pm 0.3$†	387 ± 27†	55.0
Phenobarbital				
Controls	$1.4 \times 10^{-3} \pm 0.2$	$3.9 \times 10^{-3} \pm 0.3$	792 ± 65	
1×10^{-4}	$1.4 \times 10^{-2} \pm 0.3$	$4.1 \times 10^{-3} \pm 0.4$	783 ± 69	1.1
5×10^{-4}	$2.3 \times 10^{-2} \pm 0.1$†	$1.2 \times 10^{-2} \pm 0.6$†	625 ± 57†	21.1

* Values in the table represent means (± SEM) for at least six different aortic strips.
† Significantly different from control values, $P < 0.01$.

TABLE 4. Influence of Barbiturates on Potassium-induced Contractile Responses of Isolated Rat Portal Veins

Barbiturate (M)	Potassium Responses			
	Threshold Concentration (M)*	ED_{50} (M)	Maximum Tension (mg)	Inhibition of Control Tension (Per Cent)
Thiopental				
Controls	$1 \times 10^{-2} \pm 0.2$	$1.7 \times 10^{-2} \pm 0.2$	720 ± 86	
1×10^{-4}	$2 \times 10^{-2} \pm 0.2$†	$1.6 \times 10^{-2} \pm 0.3$	216 ± 9†	70.0
5×10^{-4}	$4 \times 10^{-2} \pm 0.2$†	$1.2 \times 10^{-1} \pm 0.3$†	129 ± 8†	82.1
Pentobarbital				
Controls	$5 \times 10^{-3} \pm 0.1$	$2 \times 10^{-2} \pm 0.2$	$1,270 \pm 49$	
1×10^{-4}	$2 \times 10^{-2} \pm 0.1$†	$2 \times 10^{-2} \pm 0.2$	817 ± 19†	35.7
5×10^{-4}	$8 \times 10^{-2} \pm 0.3$†	$1.5 \times 10^{-2} \pm 0.2$†	266 ± 8†	79.1
Amobarbital				
Controls	$1 \times 10^{-2} \pm 0.4$	$1.7 \times 10^{-2} \pm 0.3$	860 ± 92	
1×10^{-4}	$1 \times 10^{-2} \pm 0.3$	$1.3 \times 10^{-2} \pm 0.4$	602 ± 27†	30.0
5×10^{-4}	$8 \times 10^{-2} \pm 0.1$†	$1.6 \times 10^{-1} \pm 0.2$†	154 ± 11†	82.1
Phenobarbital				
Controls	$1 \times 10^{-2} \pm 0.3$	$1.7 \times 10^{-2} \pm 0.3$	740 ± 67	
1×10^{-4}	$1 \times 10^{-2} \pm 0.5$	$1.6 \times 10^{-2} \pm 0.4$	532 ± 12†	28.1
5×10^{-4}	$2 \times 10^{-2} \pm 0.2$	$1.8 \times 10^{-2} \pm 0.4$	407 ± 32†	45.0

* Values in the table represent means (± SEM) for at least six different veins.
† Significantly different from control values, $P < 0.01$.

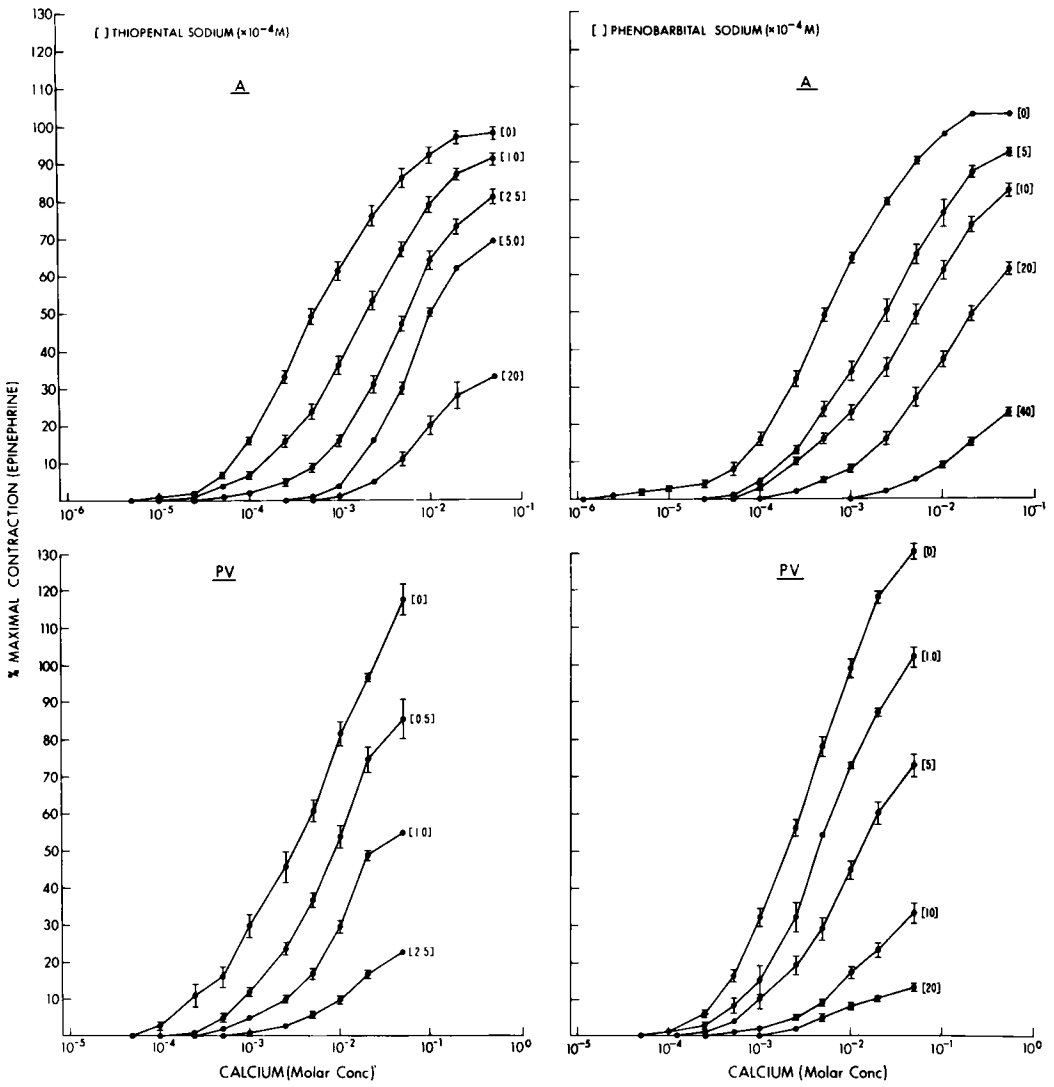

FIG. 8. Effects of thiopental and phenobarbital on calcium chloride-induced contractions of potassium-depolarized rat aortas (*upper panels*) and portal veins (*lower panels*) (see Methods). n = 6–10 each. Values are means ± SEM.

inhibitory effects of barbiturates on rat aortic and portal venous muscle must take into account the following facts and observations. The inhibition of smooth muscle: 1) is directed against development of spontaneous mechanical activity (vasomotion) and baseline tension (tone), as well as against development of drug-induced contractile tension; 2) develops rapidly (*i.e.*, within seconds); 3) is nonspecific (*i.e.*, epinephrine-, potassium-, and calcium chloride-induced contractions are attenuated); 4) is seen in at least two types of vascular smooth muscle; 5) is reversed rapidly with washing (*i.e.*, within a few minutes); 6) All of the barbiturates, with the possible exception of barbital, appear to act via the same mechanism (*e.g.*, note parallel regression lines in figure 5).

That all of the barbiturates dose-dependently inhibit potassium and calcium chloride-induced contractions of isolated aortic and portal venous smooth muscle suggests that barbiturates may act directly to reduce the availability of Ca^{++} to the contractile machinery either at and/or beyond the cell membrane. If so, one would anticipate that

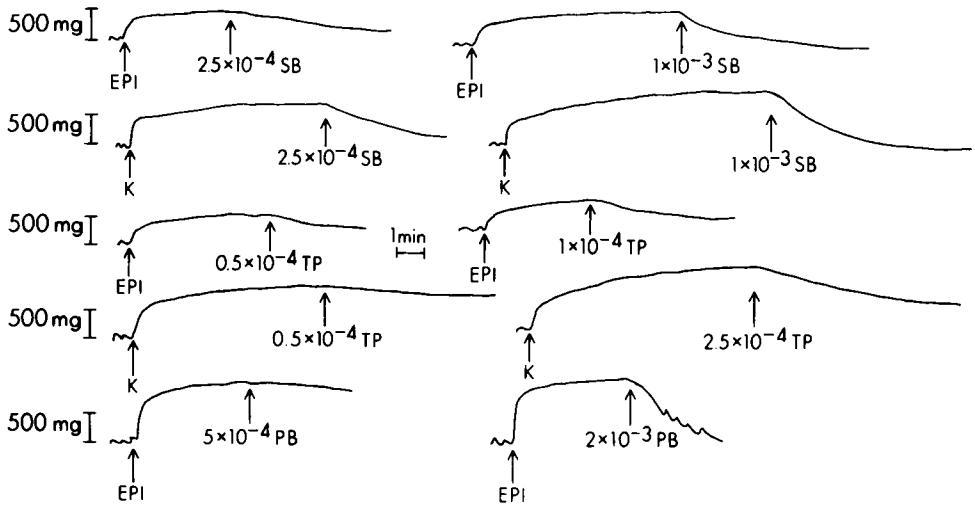

FIG. 9. Relaxation of epinephrine (EPI) (5 ng/ml)-induced and KCL (K) (10 mM)-induced contractions of three isolated rat aortas by secobarbital (SB), thiopental (TP), and phenobarbital (PB).

calcium-induced contractions would be the most sensitive of all to barbiturate inhibition, exactly as seen in the present study (table 5). It is known that calcium chloride- and potassium-induced contractions of vascular smooth muscle are dependent on extracellular calcium ions, which presumably pass through the membrane, which has become highly permeable in the presence of a high potassium ion concentration.[19,20] Barbiturates may block the passage of calcium ions across the vascular membrane as do agents such as cinnarizine, SKF 525A, Verapamil, and Lanthanum.[19,20] Nayler and Szeto have, in fact, recently demonstrated that pentobarbital can affect calcium uptake and its distribution in mammalian heart muscle.[21] This could explain why barbiturates inhibit development of spontaneous mechanical activity in the rat aorta and portal vein (figs. 1 and 2). It is thought that such contractile spike activity is dependent on influx of extracellular calcium ions.[22,23] Although other vasoactive agents such as catecholamines may utilize an intracellular and/or tightly bound membrane pool in contraction of vascular muscle,[19,20] barbiturates could interfere with mobility of these intracellular calcium ions, since barbiturates penetrate both membrane and cells rapidly.[24] This would suggest that barbiturates may act at the vascular muscle membrane level as well as intracellularly. The observed shifts of the log dose–response curves for the agonist, as well as the Ca^{++}-induced contractions, in the presence of barbiturates, concomitant with the reduction in maximum developed tensions, would be consistent with this hypothesis. Furthermore, the available data using barbiturates in other cell systems are in agreement with the hypothesis of two cellular sites of action.[21,24,25,26,27] In this context, it is of interest that Price[27] has recently suggested that another

TABLE 5. Relative Sensitivities of $CaCl_2$-, Epinephrine-, and Potassium-induced Contractions of Rat Aortic Strips to the Inhibitory Action of Barbiturates

Vasoactive Stimulus	Dose of Barbiturate Required to Exert Threshold Inhibitory Action (× 10^{-5}M)*				
	Thiopental	Secobarbital	Pentobarbital	Amobarbital	Phenobarbital
$CaCl_2$	5	5	7.5	10	25
Epinephrine	10	10	25	25	100
Potassium	5	10	10	10	50

* The lowest concentration of barbiturate either needed to increase significantly the threshold concentration of vasoactive stimulant for contraction, ED_{50}, and/or needed to reduce significantly the maximum tension of the agonist.

general anesthetic, halothane, may produce depression of cardiac muscle also by interfering with Ca^{++} at two cellular sites. Similarly, Duggan[28] has recently demonstrated that the calcium pumps of both rabbit and frog skeletal muscle sarcoplasmic reticulum are stimulated by barbiturates in low concentrations. The latter mechanism, if present in rat aortic and portal venous smooth muscle, could also aid in explaining why barbiturates rapidly relax established drug-induced contractions in these vascular smooth muscles (fig. 9).

The finding that thiopental, but not other barbiturates, can dose-dependently contract arterial smooth muscle is consistent with results of other investigators,[5,6] and may, as suggested by others, account for the arterial spasm seen when thiopental sodium is administered intra-arterially.[5,6] In addition, since portal venous smooth muscle cannot undergo contraction in response to thiopental, this may explain why venous spasm is not seen clinically after intravenous administration of thiopental, and serves to demonstrate another example of the well-known heterogeneity of vascular muscles.[20]

References

1. Price HL: General anesthesia and circulatory homeostasis. Physiol Rev 40:187–218, 1960
2. Greisheimer EM: The circulatory effects of anesthetics, Handbook of Physiology, Section 2, Circulation, Volume III. Edited by WF Hamilton, P Dow. Washington, D.C., Am Physiol Soc, 1965, pp 2485–2487
3. Sharpless SK: Hypnotics and sedatives. I. The barbiturates, The Pharmacological Basis of Therapeutics. Fourth edition. Edited by LS Goodman, A Gilman. New York, Macmillan, 1970, pp. 98–120
4. Altura BT, Altura BM: Effects of pentobarbital sodium on isolated rat arterial and venous smooth muscle. Pharmacologist 16:303, 1974
5. Price ML, Price HL: Effects of general anesthetics on contractile responses of rabbit aortic strips. ANESTHESIOLOGY 23:16–20, 1962
6. Burn JH, Hobbs R: Mechanism of arterial spasm following intra-arterial injection of thiopentone. Lancet 1:1112–1115, 1959
7. Altura BM, Altura BT: Peripheral vascular actions of glucocorticoids and their relationship to protection in circulatory shock. J Pharmacol Exp Ther 190:300–315, 1974
8. Altura BM, Altura BT: Differential effects of substrate depletion on drug-induced contractions of rabbit aorta. Am J Physiol 219: 1698–1705, 1970
9. Larrabee MG, Posternak JM: Selective action of anesthetics on synapses and axons in mammalian sympathetic ganglia. J Neurophysiol 15:91–114, 1952
10. Brodie BB, Burns JJ, Mark LC, et al: The fate of pentobarbital in man and dog and a method for its estimation in biological material. J Pharmacol Exp Ther 109:26–34, 1953
11. Vesell ES, Passananti GT: Utility of clinical chemical determinations of drug concentrations in biological fluids. Clin Chem 17: 851–866, 1971
12. Wylie WD, Churchill-Davidson HC: A Practice of Anesthesia. Third edition. London, Lloyd-Luke Medical Books Ltd., 1972
13. Olmstead F, Page IH: Hemodynamic changes in dogs caused by sodium pentobarbital anesthesia. Am J Physiol 210:817–820, 1966
14. Hershey SG, Zweifach BW, Rovenstein EA: Effects of depths of anesthesia on behavior of peripheral vascular bed. ANESTHESIOLOGY 14:245–254, 1953
15. Hershey SG, Zweifach BW: Peripheral vascular homeostasis in relation to anesthetic agents. ANESTHESIOLOGY 11:145–154, 1950
16. Baez S., Orkin LR: Microcirculatory effects of anesthesia in shock, Shock. Edited by SG Hershey. Boston, Little, Brown and Co., 1964, pp 207–225.
17. MacCannell KL: The effects of barbiturates on regional blood flows. Can Anaesth Soc J 16: 1–6, 1969
18. Mellander S, Johansson B: Control of resistance, exchange and capacitance functions in the peripheral circulation. Pharmacol Rev 20: 117–196, 1968
19. Godfraind T, Kaba A: Blockade or reversal of the contraction induced by calcium and adrenaline in depolarized arterial smooth muscle. Br J Pharmacol 36:549–560, 1969
20. Bohr DF: Vascular smooth muscle updated. Circ Res 32:665–672, 1973
21. Nayler WG, Szeto J: Effect of sodium pentobarbital on calcium in mammalian heart muscle. Am J Physiol 222:339–344, 1972
22. Biamino G, Kruckenberg P: Synchronization and conduction of excitation in the rat aorta. Am J Physiol 217:376–382, 1969
23. Biamino G, Johansson B: Effects of calcium and sodium on contracture tension in the smooth muscle of the rat portal vein. Pfluegers Arch 321:143–158, 1970
24. Seeman P: The membrane actions of anesthetics and tranquilizers. Pharmacol Rev 24:583–655, 1972
25. Alper MH, Flacke W: The peripheral effects of anesthetics. Annu Rev Pharmacol 9:273–296, 1969
26. Blaustein MP: Barbiturates block sodium and potassium conductance increases in voltage-clamped lobster axons. J Gen Physiol 51: 293–307, 1968
27. Price HL: Calcium reverses myocardial depression caused by halothane. ANESTHESIOLOGY 41:576–579, 1974
28. Duggan PF: Stimulation of calcium uptake of muscle microsomes by phenothiazines and barbiturates. Eur J Pharmacol 13:381–386, 1971