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Cardiovascular Responses to Three Prostaglandin Endoperoxide Analogs in the Dog (39512)[1]

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In the dog, the bisenoic prostaglandin (PG) precursor, arachidonic acid (AA), in an acute intravenous dose of 300 μg/kg, has a profound depressor effect of short duration which is blocked by PG synthetase inhibitors (1). Tachyphylaxis does not develop. AA is directly vasoconstrictor, however, in the canine pulmonary circulation (2) and in the isolated canine hindlimb (3). Myocardial contractile force in the dog is not directly affected by AA, although a reflex positive inotropic effect may be observed (4).

A measurable delay in the time of onset of AA-induced hypotension and the inhibition of this response by aspirin suggested that these cardiovascular effects were due to products of AA metabolism. However, these effects are not those observed with PGE_2, which has a direct positive inotropic effect (4), or $PGF_{2\alpha}$, which is predominantly pressor in its blood pressure effect. We proposed, therefore, that the cardiovascular actions of AA were due to endoperoxide intermediates formed transiently in the biosynthesis of PGE_2 and $PGF_{2\alpha}$.

Stable analogs of PG endoperoxides have now become available in limited quantities for biologic testing (5, 6). We have examined the effects of these newly synthesized analogs in the dog circulation, in a manner similar to that in which the properties of AA, PGE_2, and $PGF_{2\alpha}$ have been characterized, in order to gain insight into the pharmacologic properties of the intermediate compounds derived from AA.

Materials and methods. Mongrel dogs of either sex were anesthetized with sodium pentobarbital (30 mg/kg) and maintained on intermittent positive pressure respiration with a Harvard respirator. A left thoracotomy was performed. In three dogs, a Walton-Brodie strain gauge arch was sutured to the right ventricular wall for measurement of myocardial contractile force. In three additional dogs, a small catheter was inserted into a branch of the left pulmonary artery for direct recording of pulmonary arterial pressure. In each dog, a femoral artery and vein were catheterized for direct measurement of systemic arterial pressure and for administration of test substances directly into the inferior vena cava.

Two cyclic ether endoperoxide analogues were generously provided by Dr. G. L. Bundy of the Upjohn Company. These are: (15*S*)-hydroxy-9α,11α-(epoxymethano)-prosta-5*Z*,13*E*-dienoic acid, referred to as U44069, and (15*S*)-hydroxy-11α,9α-(epoxymethano)-prosta-5*Z*,13*E*-dienoic acid, referred to as U46619. Dr. E. J. Corey of Harvard University kindly provided 0.3 mg of an *azo*-endoperoxide analog: (15*S*)-hydroxy-9α,11α-(azo)-prosta-5*Z*,13*E*-dienoic acid. (We have referred to this compound in this paper as *azo*.)

The three endoperoxide analogs were dissolved in ethanol to provide 1 mg/ml solutions. These were diluted in saline to 20 μg/ml. Hexamethonium chloride (Schwarz/Mann) was prepared in aqueous solution in a concentration of 50 mg/ml. Indomethacin (Merck) was prepared in dilute sodium carbonate as a 2 mg/ml solution for use intravenously.

Following observation of the initial cardiovascular response to each of the endoperoxides, ganglionic blockade was induced with hexamethonium (2 mg/kg) in three dogs and the test doses were repeated. Similarly, after initial observations, two animals received indomethacin to block PG synthetase (2 mg/kg), and the endoperoxides in the test doses were repeated. Fewer test doses of the *azo* compound were given owing to the limited amount available.

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Results. Systemic arterial pressure. Each of the three endoperoxide analogs is predominantly pressor (Table I). One injection of the *azo* compound at a dose of 0.5 μg/kg increased systolic and diastolic arterial pressure by 16 and 20%, respectively. Administration of 2.5 μg/kg increased the systolic pressure in four dogs by 25% (SE ±3.5) and the diastolic pressure by 23% (SE ±1.0).

In six dogs, 1.25 μg/kg of U46619 caused a mean systolic rise of 13.7% (SE ±5.5) and a mean diastolic rise of 17.5% (SE ±4.7). A larger dose of 2.5 μg/kg caused 40 and 38% increases in systolic and diastolic pressures, respectively, in one dog. In seven dogs, 1.25 μg/kg of U44069 caused a mean systolic rise of 17.0% (SE ±4.3) and a mean diastolic rise of 17.3% (SE ±2.8). A single dose of 2.5 μg/kg caused 47 and 49% increases in systolic and diastolic pressures, respectively. The duration of these pressor responses ranged from 5 to 10 min. The typical manner of blood pressure rise, following a transient fall of several seconds duration, for each of these three endoperoxide analogs is shown in Figs. 1 and 2.

Pulmonary arterial (PA) pressure. Each one of the endoperoxide analogs is markedly vasoconstrictor in the pulmonary circulation. Immediate elevations of PA pressure following intravenous administration were as follows: Injection of *azo* (2.5 μg/kg) caused a 50% rise in systolic and 140% rise in diastolic pressure in the PA. Three injections of U46619 (1.25 μg/kg intravenously) caused a mean PA systolic pressure rise of 55% (SE ±7.7) and a diastolic rise of 158% (SE ±78). Three injections of U44069 (1.25 μg/kg) caused a mean PA systolic pressure rise of 83% (SE ±36) and diastolic rise of 132% (SE ±47). The configuration of the PA pressure rise is shown in Fig. 1.

Myocardial contractile force (MC). Each

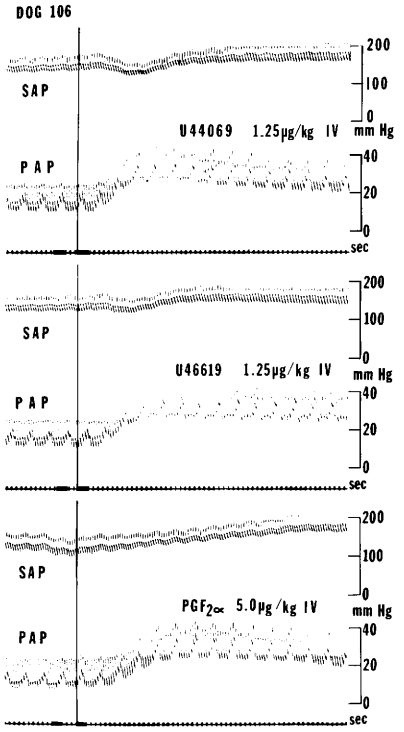

FIG. 1. Simultaneous recordings of systemic arterial pressure (SAP) and pulmonary arterial pressure (PAP) in a dog receiving the doses of U44069, U46619, and $PGF_{2\alpha}$ shown. In each case the vertical line represents the instant of introduction of the substance into the inferior vena cava. The transient drop in SAP in response to the endoperoxide analogs is interpreted as due to momentarily decreased return to the left heart because of rapid and intense pulmonary vasoconstriction. This is sometimes also observed with $PGF_{2\alpha}$.

TABLE 1. SYSTEMIC ARTERIAL PRESSURE (SAP) RESPONSES TO ENDOPEROXIDES AND $PGF_{2\alpha}$.

Compound (dose (*n*))	Initial blood pressure (systolic/diastolic, mm Hg)	Highest BP response (systolic/diastolic, mm Hg)	Percentage of change (systolic/diastolic, ± SE)
Azo (Corey) 2.5 μg/kg (4)	159 ± 6.6/122 ± 2.5	199 ± 10.7/150 ± 3.5	25.2 ± 3.5/22.8 ± 1.0
U46619 1.25 μg/kg (6)	168 ± 4.2/126 ± 4.0	191 ± 13.1/148 ± 10.4	13.7 ± 5.5/17.5 ± 4.7
U44069 1.25 μg/kg (7)	165 ± 8.4/127 ± 6.4	193 ± 10.0/149 ± 8.4	17.0 ± 4.3/17.3 ± 2.8
$PGF_{2\alpha}$ 5 μg/kg (7)	172 ± 6.1/131 ± 4.6	199 ± 10.5/154 ± 8.4	15.7 ± 4.7/17.6 ± 5.2

of the endoperoxide analogs is directly positively inotropic (Fig. 2). In three dogs, *azo* (2.5 μg/kg) caused a mean 13.5% increase in MC (SE ±2.9). Following ganglionic blockade with hexamethonium, in one dog MC increased 25%. In three dogs, U46619 (1.25 μg/kg) caused a mean MC increase of 15% (SE ±7.2). MC increased 16.7% in one dog given U46619 (1.25 μg/kg) after ganglion blockade. In three dogs, U44069 (1.25 μg/kg) caused a mean MC increase of 7.7% (SE ±4.2). Two of these dogs were given hexamethonium, following which U44069 in the same dose caused MC increases of 35 and 13%.

Heart rate. The three endoperoxide analogs caused moderate reduction in heart rate during the pressor responses. These changes were as follows: *azo* (2.5 μg/kg), 15% in four dogs (SE ±4.8); U46619 (1.25 μg/kg), 7.5% in six dogs (SE ±2.4); U44069 (1.25 μg/kg), 8.9% in seven dogs (SE ±2.3). Heart rate alterations were reduced or abolished following ganglionic blockade with hexamethonium to 4% for *azo* (one dog); 3% for U46619 (one dog); and no change in heart rate for U44069 (two dogs).

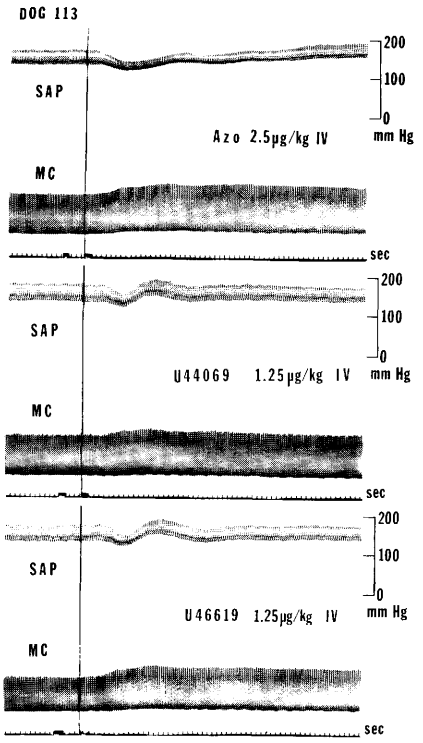

FIG. 2. Simultaneous recordings of SAP and myocardial contractile force (MC) in a dog receiving the doses of endoperoxide analogs shown. An increase in myocardial contractility occurs, which persists after ganglionic blockade produced by hexamethonium (see text).

Pretreatment with indomethacin. In two dogs, U46619 and U44069 were given before and after indomethacin (2 mg/kg) which was previously shown in this laboratory to block completely the vascular responses to arachidonic acid. The systemic arterial pressor responses to each of the cyclic ether analogues following indomethacin were as follows: U46619 at 2.5 μg/kg, 170/135 to 235/195 (38% systolic, 44% diastolic), at 1.25 μg/kg, 165/130 to 190/155 (15% systolic, 19% diastolic); U44069, 2.5 μg/kg, 165/135 to 220/185 (33% systolic, 37% diastolic), at 1.25 μg/kg, 170/135 to 210/170 (23.5% systolic, 26% diastolic).

Comparative studies with $PGF_{2\alpha}$. Since the endoperoxide analogs were predominantly pressor, $PGF_{2\alpha}$ (5 μg/kg) was given to seven dogs that had received endoperoxide analogs to compare effects (Table I). The mean systolic and diastolic systemic pressure elevations were 15.7 and 17.6%, respectively, (SE ±4.7 and ±5.2). Heart rate was reduced during the pressor response, mean 5.2% (SE ±2.2). PA pressures in three were raised 45, 26, and 62% systolic, and 26, 20, and 242% diastolic (Fig. 1).

Discussion. These first syntheses of stable analogs of PG endoperoxides by Corey *et al.* (5) and by Bundy (6) are of importance to investigators of the PG system, and particularly those who have studied the physiologic and pharmacologic effects of the PG and endoperoxide precursors, since the naturally occurring endoperoxides PGG_2 and PGH_2 are unstable and not generally available. The only information published on the blood pressure effects of PGG_2 and PGH_2 is that these endoperoxides produce a "triphasic response" in the anesthetized guinea pig (7).

The three analogs which were available in limited amounts for this study are extremely active biologically, in some ways contrary to expectations. For example, each is predomi-

nantly pressor, although the precursor of the bisenoic PGs, AA, is depressor. The slight and transient drop in systemic arterial pressure following intravenous injection of the three analogues, as shown in Figs. 1 and 2, we have interpreted as due to the marked pulmonary vasoconstriction which transiently reduces left ventricular output.

Unlike AA, but similar to the primary PGs, PGE_2 and $PGF_{2\alpha}$, the three endoperoxide analogs have directly positive inotropic effects on the dog heart (Fig. 2). This response is not blocked by interruption of the baroreceptor reflexes as is the variable myocardial contractile force response to AA.

The molecular manipulations required to make these compounds stable enough for use in studies such as this make it inappropriate to say that the observed responses represent those elicited by the naturally occurring endoperoxides that are formed in the biosynthetic pathway from AA to PGE_2 and $PGF_{2\alpha}$. In fact, Corey and his colleagues use the expression "highly active biochemical mimic of prostaglandin endoperoxides" in reference to the *azo* compound (5). However, these analogs are potent vasoactive agents and more so than $PGF_{2\alpha}$ which exerts comparable effects on the canine circulation only in larger doses (1, 4). Moreover, these preliminary investigations indicate that the cyclic ether endoperoxide analogs of Bundy are approximately twice as potent in their vasoconstrictor effects, both systemic and pulmonary, as the *azo* analog of Corey and his colleagues.

Summary. Three stable analogs of prostaglandin endoperoxides have been studied in limited quantities for their effects on the canine cardiovascular system. They are potent systemic pressor agents and powerful pulmonary vasoconstrictors. They directly increase myocardial contractile force. These responses are not altered by indomethacin. In their blood pressure and myocardial effects they are considerably more potent than arachidonic acid and may be estimated to be more potent than the primary bisenoic PGs, PGE_2 and $PGF_{2\alpha}$. The cyclic ether endoperoxide analogs of Bundy are approximately twice as potent in their effects on these parameters as the *azo* analog of Corey *et al.*

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