Two Genetically Selected Strains of Rats Exhibit Hypersensitivity or Resistance to Cocaine-Induced Fatal Arrhythmias

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ABSTRACT

We identified for the first time two genetically selected strains of rats that differ markedly in sensitivity to cocaine-induced life-threatening cardiac arrhythmias and arrest. The two strains of rats, designated as Fast and Slow, were bred for sensitivity (Fast) or resistance (Slow) to electrically kindled seizures. Studies were performed on halothane-anesthetized, mechanically ventilated rats. Animals were given cocaine (3 or 4 mg/kg/min i.v.) until they died. Arrhythmias (atrioventricular conduction block) developed at much lower cumulative cocaine doses in Slow-kindling rats than in Fast-kindling rats (15 ± 1 versus 42 ± 3 mg/kg, p < .01). The lethal cocaine dose (the dose that caused cardiac arrest) was also markedly lower in Slow than in Fast strains (32 ± 2 versus 62 ± 6 mg/kg, p < .01). These differences between the two strains were not significantly altered by pretreatment of animals with either ganglionic blockers, hexamethonium (20 mg/kg i.v.) or chlorisondamine (5 mg/kg i.v.), or a nonselective beta adrenergic receptor blocker, propranolol (1 mg/kg i.v.). A nonselective alpha adrenergic receptor blocker, phentolamine (10 mg/kg i.v.), however, abolished the differences between the Fast and Slow strains in the doses of cocaine required to produce atrioventricular conduction block and cardiac arrest. The results provide the first evidence of genetically determined susceptibility or resistance to cocaine-induced cardiotoxicity. There appears to be a genetically determined difference in the alpha adrenergic receptor system between the two strains that is responsible for the differential sensitivity to cocaine-induced arrhythmias and cardiac arrest.

The cardiotoxicity of cocaine represents a serious hazard that sometimes is difficult to prevent and manage in certain individuals who appear to be particularly sensitive to cocaine. These individuals experience fatal cardiotoxic effects of cocaine at much lower than predicted doses (Schachne et al., 1984; Isner et al., 1986; Lange and Willard, 1993). Compelling evidence indicates that deleterious cardiovascular events such as stroke, myocardial infarction, and lethal cardiac arrhythmias associated with cocaine use appear to be unrelated to the dose of cocaine and frequency of use in some individuals (Mittleman and Wetle, 1984; Minor et al., 1991). Acute myocardial infarction and sudden death have been associated with the initial use of small doses of cocaine (Wehbie et al., 1987). In several case reports, patients had been administered cocaine for nasal surgery. Two of the patients experienced conduction defects, and one patient developed ventricular arrhythmias; all resulted in sudden death (Young and Glauber, 1947; Benchimol et al., 1978; Nanji and Filipenko, 1984). The lethal dose has been reported to be 1.2 g, but severe toxicity has been reported with a dose as low as 20 mg (Estroff and Gold, 1986). This evidence strongly suggests that predisposition of certain individuals plays an important role in the fatal cardiac complications of cocaine. However, what is responsible for the predisposition has not been determined.

We report here that selective breeding can alter the sensitivity of animals to the arrhythmogenic and lethal effects of cocaine. Two strains of rats, genetically fast- and slow-amygda kindling (Fast and Slow, respectively) rats, were originally derived in the 1980s from an F1 cross between Wistar and Long-Evans rats by breeding for sensitivity or resistance

ABBREVIATIONS: Fast, genetically fast-amygda kindling rats; Slow, genetically slow-amygda kindling rats; ECG, electrocardiogram; EEG, electroencephalogram; dp/dt, first derivative of left ventricular pressure; LVEDP, left ventricular end-diastolic pressure; AV, atrioventricular; MABP, mean arterial blood pressure; LVSP, left ventricular systolic pressure; MAC, minimum alveolar concentration.
to kindled seizures induced by electrical stimulation of the amygdala (Steingart, 1983). The “fast-kindling” and “slow-kindling” strains exhibit a 4-fold difference in amygdala kindling rates (Dufresne et al., 1989; Elmer et al., 1997). This report follows the observation that 100% of Slow rats died at their cocaine seizure threshold in contrast to a less than 17% lethality rate in Fast rats (Reigel et al., 1997). We further found that when given an acute i.v. infusion of cocaine, rats from the Slow strain had life-threatening cardiac arrhythmias [mostly atrioventricular (AV) conduction block] and cardiac arrest much sooner than did rats from the Fast strain. The cocaine dose required to produce arrhythmias was approximately three times lower in the Slow than in the Fast strain. Because the cardiotoxic effects of cocaine are largely attributed to its stimulatory effects on the autonomic nervous system (Tella et al., 1992; de Jong, 1994; Chen et al., 1995), we tested the effects of ganglionic blockers and selective receptor antagonists to examine the contribution of the autonomic nervous system to the differential sensitivity to cocaine-induced arrhythmias and cardiac arrest between Fast and Slow rats. Our studies provide the first evidence of genetically determined differential sensitivity to cocaine-induced cardiotoxicity in animals, and the results suggest the involvement of the alpha adrenergic receptor system in the differential sensitivity.

**Experimental Procedures**

**Animals.** Fast-kindling and Slow-kindling rats were originally derived from an F1 cross between Wistar and Long-Evans rats by breeding for sensitivity or resistance to kindled seizures induced by electrical stimulation of the amygdala (Steingart, 1983; Dufresne et al., 1989). Since their initial isolation, Fast and Slow rats have undergone further differentiation by sister-brother breeding. Fast rats are now approximately four times more sensitive to kindled seizures than are Slow rats (Elmer et al., 1997). Except for the marked difference in kindling rates, both strains grow and reproduce normally (Elmer et al., 1997). Fast and Slow rats were reared at Carleton University (Ottawa, Canada). At 5 weeks of age, the animals were shipped to and housed in the Laboratory Animal Resource Center of Texas Tech University Health Sciences Center. Male animals (70 ± 5 days of age) were used. Wistar and Long-Evans rats, matched for sex, age, and weight, were purchased from SASCO Animal Laboratory (Houston, TX) and used as control strains. Animals were housed three per cage and fed ad libitum.

**Surgical Preparation.** Experiments were done in lightly anesthetized, mechanically ventilated rats. This was done to eliminate confounding systemic effects of hypoxia and hypercapnia due to respiratory failure induced by cocaine. Experiments were done in intact animals using both closed-chest and open-chest preparations.

All rats were anesthetized with 1.75% halothane in oxygen during surgical procedures. The trachea was cannulated, and mechanical ventilation was instituted using a rodent ventilator (Harvard Apparatus, South Natick, MA). A polyethylene catheter (PE 50) was placed through the left femoral vein into the vena cava for test drug infusion, and another cannula was placed into the right femoral vein for administration of neuromuscular blocking agent. The right and left femoral arteries were cannulated for arterial pressure measurements and for blood sampling, respectively.

Measurement of cardiac function was done in the open-chest preparation in which a midline thoracotomy was done to expose the heart and ascending aorta. An electromagnetic flowprobe (model FM 501D; Carolina Medical, King, NC) was positioned on the ascending aorta for cardiac output measurements. A fluid-filled catheter (PE 50) was placed via a stab wound in the apex of the heart into the left ventricle to measure left ventricular pressure and the first derivative of left ventricular pressure (dp/dt). Left ventricular end-diastolic pressure (LVEDP) was obtained from high amplification of left ventricular pressure.

After the surgical preparation, halothane concentration was decreased to 0.5% or 0.7% to provide postoperative analgesia and subdued consciousness; 0.5% halothane plus 70% N2/O30% O2 was used in the closed-chest experiments. This concentration of halothane/N2/O provided an approximately 1.0 minimum alveolar concentration (MAC) of anesthesia, adequate analgesia, and subdued consciousness in rats (Lawrence and Livingston, 1981; Heavner, 1986). For open-chest experiments, halothane, N2/O, and O2 concentrations were set at 0.7, 50, and 50%, respectively. This higher halothane concentration was used to offset reduced N2/O. Reduced functional residual capacity of the lungs associated with thoracotomy requires higher O2 concentrations to maintain PaO2 above 90 mm Hg. Doxazurin (0.2 mg/kg i.v.), a neuromuscular blocking agent, was administered as needed to prevent spontaneous breathing. Rectal temperature was maintained at 38 ± 0.2°C using a warming blanket and radiant heat.

**Physiological Measurements.** Electrocardiogram (ECG) leads I, II, and V1 and fronto-occipital electroencephalography (EEG) were recorded with subcutaneously placed needle electrodes. ECGLs, EEG, arterial blood pressure, and cardiac functional indices were recorded on a chart recorder (7758A recorder; Hewlett Packard, Waltham, MA) throughout the experiment.

Arterial blood was taken from all animals before cocaine administration (25 min after surgical preparation was completed) for baseline blood gas analysis (NOVA STAT Profile 5 Blood Gas Analyzer; NOVA Biomedical, Waltham, MA). The experiment did not proceed until blood gas values were within the normal physiological range (pH 7.35–7.45, pCO2 28–40 mm Hg, paO2 ≥ 90 mm Hg).

### TABLE 1

Baseline hemodynamic and ECG values of four strains of rats

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Wistar</th>
<th>Long-Evans</th>
<th>Fast</th>
<th>Slow</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>MABP (mm Hg)</td>
<td>HR (beats/min)</td>
<td>HR × MABP (mm Hg · beats/min -1 · 10−2)</td>
<td>LVSP (mm Hg)</td>
</tr>
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<td>---------------------</td>
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<td>------------</td>
<td>--------</td>
<td>--------</td>
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<td>92 ± 3</td>
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<td>88 ± 2</td>
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<td>113 ± 3***</td>
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</tr>
<tr>
<td></td>
<td>102 ± 4***</td>
<td>358 ± 7</td>
<td>369 ± 20</td>
<td>123 ± 4***</td>
</tr>
</tbody>
</table>

Fast and Slow refer to genetically fast- and slow-amygdala kindling rats, respectively. Wistar and Long-Evans rats were control strains.

HR, heart rate; the number of QRS complexes per minute appearing on the ECG.

Baseline values were obtained after 30-min stabilization and before the administration of cocaine hydrochloride.

Values are mean ± S.E.M.

*p < .05 versus Wistar, **p < .05 versus Long-Evans, ***p < .05 versus Fast.
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At 30 min after surgery, baseline hemodynamic and electrophysiological data were obtained. For the closed-chest experiments, animals were pretreated with autonomic blocking agents before cocaine infusion. One of the following four autonomic blocking agents were used: 1) a competitive ganglionic blocker, hexamethonium (20 mg/kg, n = 7 or 8 each strain) (Abdel-Rahman, 1989; Tella et al., 1992); 2) a noncompetitive ganglionic blocker, chlorisondamine (5 mg/kg, n = 7–9 each strain) (Abdel-Rahman, 1989; Tella et al., 1992); 3) a nonselective alpha adrenergic receptor blocker, phentolamine (10 mg/kg, n = 6 or 7 each strain) (Williams et al., 1978; Murphy et al., 1991); and 4) a nonselective beta adrenergic receptor blocker, propanolol (1 mg/kg, n = 6 or 7 each strain) (Murphy et al., 1991; Branch and Knuepfer, 1992). All of the autonomic blocking agents were given 5 or 10 min (chlorisondamine) before the initiation of infusion of cocaine as a bolus i.v. injection (30 sec, 1 ml/kg). Drug doses were selected based on literature reports (Williams et al., 1978; Murphy et al., 1991; Abdel-Rahman, 1989; Branch and Knuepfer, 1992; Tella et al., 1992) and our preliminary data. A similar dose of hexamethonium was demonstrated in a previous study to cause 36 to 42% reduction in arterial pressure and a significant attenuation of the pressor response of cocaine (Tella et al., 1992). Chlorisondamine was chosen because of its reported complete ganglionic blockade (Abdel-Rahman, 1989; Tella et al., 1992). The doses of phentolamine and propanolol were reported to produce complete alpha and beta adrenergic blockade, respectively (Williams et al., 1978; Branch and Knuepfer, 1992; Tella et al., 1992).

Sensitivities to seizures, arrhythmias, or cardiac arrest induced by cocaine were expressed in terms of the cumulative doses of cocaine administered when seizures, arrhythmias, or cardiac arrest occurred. Seizures were defined as the first appearance of bursts of multiple sharp spikes of greater than 100 μV on the EEG. Cardiac arrhythmias were defined as the first appearance of at least three consecutive abnormal beats/10 s accompanied by changes in arterial blood pressure, including second- and third-degree AV conduction block, sinus arrest (disappearance of the P wave), and ventricular arrhythmias (ventricular tachycardia or fibrillation). First-degree AV conduction block (prolongation of the P-R interval), and isolated premature contractions were not defined as arrhythmias. Cardiac arrest was defined as complete disappearance of the QRS complex.

Cocaine Concentration Measurements in Plasma and Myocardium. Arterial blood samples (~0.4 ml each sample) were obtained for analysis of plasma cocaine concentrations at the onset of arrhythmias and at various intervals during cocaine infusion. Blood was collected in a heparinized tube containing 1 drop of saturated sodium fluoride to inhibit the activity of cholinesterase that hydrolyzes cocaine. Plasma was obtained by immediate centrifugation at 4°C and was stored at −80°C until analysis. Plasma cocaine and its major metabolite benzoylecgonine levels were determined by high-performance liquid chromatography using UV detection as described previously (Heavner et al., 1995). Six to eight animals from each strain were sacrificed by exsanguination at the onset of AV conduction block, and the hearts were rapidly removed for the measurement of heart cocaine and benzoylecgonine concentrations. Ventricles were weighed and homogenized in 10× volume (g/ml) of 0.05 M KH₂PO₄, using a Brinkmann Polytron homogenizer. The homogenate was stored at −80°C until assay. The same procedure was used for tissue as for plasma cocaine concentration measurement.

Materials. Drugs used included cocaine, hexamethonium, chlorisondamine, phentolamine, and propranolol; all were hydrochloride salt and were dissolved in 0.9% sterile saline. All drugs, with the exception of chlorisondamine, were obtained from Sigma Chemical Co. (St. Louis, MO). Chlorisondamine was obtained from Novartis Pharmaceuticals Corporation (Summit, NJ).
Results

Baseline Values

Both Fast and Slow rats grew normally and showed no gross developmental abnormalities. Body weight was not significantly different among the four strains of rats (Wistar, 277 ± 30 g; Long-Evans, 271 ± 24; Fast, 277 ± 57; Slow, 312 ± 28). Baseline blood gases were within normal ranges in all animals. The MAC for halothane was similar between Fast and Slow rats (1.11 ± 0.09% versus 1.03 ± 0.04%; \( p < .05 \)).

As shown in Table 1, baseline mean arterial blood pressures (MABPs) of Slow and Fast rats were similar but were significantly higher than those of control rats. Baseline heart rate in Fast rats was significantly lower than that in Wistar rats. Baseline values for the rate-pressure product (heart rate \( \times \) MABP), however, were similar among all strains. Left ventricular systolic pressure (LVSP) was significantly higher in Fast than in all other strains. There were no significant differences in dp/dt\(_{\text{max}}\), LVEDP, and cardiac output among strains. Baseline P-R interval was slightly longer in Fast than in Long-Evans rats, which may reflect a slower heart rate in Fast rats. Baseline QRS complexes did not differ among strains. Fewer than 5% of the animals had premature ventricular contractions during the stabilization period. We did not proceed with the experiment in rats in which premature contractions were sustained throughout the stabilization period. No other arrhythmias were noted.

Under Cocaine Challenge

No significant differences were observed for any parameter measured between the two control strains (Wistar and Long-Evans) in response to cocaine infusion.

Hemodynamic Responses to Acute i.v. Cocaine Infusion. During cocaine infusion, arterial blood pressure, heart rate, LVSP, and LVEDP increased initially, peaked between 0.3 and 0.6 min, and then gradually declined in all animals (Fig. 1). The peak rate-pressure product (beats/min \( \cdot \) mm Hg\(^{-1} \cdot 100^{-1} \)) was significantly higher in Slow than in other strains (653 ± 26 versus 557 ± 16 in Fast, \( p < .01 \), versus 494 ± 31 in Wistar, \( p < .01 \), versus 448 ± 32 in Long-Evans, \( p < .01 \)). After the initial increase, hemodynamic indices declined as infusion continued. The rate of decline in MABP and heart rate was greater in Slow than in Fast rats. The
dramatic decline in heart rate after 4 min in Slow rats was associated with an earlier occurrence of AV conduction block in this strain (~4 min in Slow strain versus ~11 min in Fast strain). There were no significant differences in the changes of LVSP, dp/dt, LVEDP, and cardiac output during cocaine infusion between Slow and Fast strains. At the time of onset of arrhythmias, no significant differences in MABP and heart rate were observed among strains (MABP 55 ± 4 mm Hg in Wistar, 39 ± 4 in Long-Evans, 58 ± 4 in Fast, and 61 ± 7 in Slow; heart rate, 326 ± 11 beats/min in Wistar, 298 ± 6 in Long-Evans, 282 ± 4 in Fast, and 322 ± 15 in Slow).

**ECG Changes and Arrhythmia Patterns.** The most profound ECG changes during acute i.v. cocaine infusion in all animals were marked prolongation of the P-R interval and widening of the QRS complex. These changes were greater in Slow than in Fast rats (Fig. 1). Prolongation of the P-R interval gradually proceeded to second-degree AV conduction block (Fig. 2), the predominant arrhythmia induced by cocaine under our experimental conditions (halothane/N₂O anesthesia plus ventilation). Only 1 rat (of 25 Slow rats) had transient ventricular tachycardia. Eight rats (2 of 21 Wistar, 4 of 20 Long-Evans, and 2 of 20 Fast rats) had sinus arrest as their first arrhythmia. Arrhythmias occurred either before or at the same time as seizures in Slow and after seizures in all other strains.

**Cocaine Doses Required to Produce Arrhythmias and Cardiac Arrest.** Cocaine doses required to produce AV conduction block and cardiac arrest were markedly lower in Slow than in Fast rats (Fig. 3). Slow rats developed AV conduction block at about one third the dose of cocaine required for the same endpoint in the Fast strain. Similarly, cocaine doses for cardiac arrest were markedly lower in Slow than in Fast rats. Cocaine doses required to produce AV conduction block and cardiac arrest in control animals were between the doses for Slow and Fast rats. Seizure-inducing doses of cocaine were similar among strains (Fig. 3) despite marked differences in amygdala kindled seizure rates (Elmer et al., 1997).

**Plasma and Heart Cocaine Concentrations.** As shown in Fig. 4A, plasma cocaine concentrations increased in all animals as cocaine infusion continued. However, plasma cocaine concentrations were much higher per amount infused in the Slow strain than in all other strains. The slope for the increase in plasma levels per unit infused was three times greater for the Slow than for the Fast strains (1.86 ± 0.13 versus 0.55 ± 0.09, p < .01). The cocaine concentrations in plasma and myocardial tissues at arrhythmia onset were significantly lower in Slow than in Fast rats (Fig. 4B). The plasma levels of cocaine metabolite, benzoylecgonine, were very low compared with corresponding plasma cocaine levels (less than 10%). No significant differences in plasma benzoylecgonine levels were observed among strains at any time point (at 5 min: 1.55 ± 0.14 µg/ml in Wistar, 1.83 ± 0.16 µg/ml in Long-Evans, 1.24 ± 0.29 µg/ml in Fast, and 1.56 ± 0.29 µg/ml in Slow). Benzoylecgonine levels in heart tissue were not detectable at arrhythmia onset in any animals and were very low when animals died (1.46 ± 0.32 µg/g wet weight in Fast versus 1.03 ± 0.31 in Slow, p > .05).

**Effects of Autonomic Blocking Agents on Cocaine Cardiotoxicity.** Figure 5 shows changes in the rate-pressure products in response to cocaine infusion in the absence (vehicle) and presence of autonomic blocking agents. Base-1, 30-min stabilization and before pretreatment with vehicle or autonomic blocking agents; Base-2, 5 min (or 10 min for chlorisondamine) after animals were pretreated with vehicle (1 ml/kg i.v. saline), hexamethonium (20 mg/kg i.v.), chlorisondamine (5 mg/kg i.v.), propranolol (1 mg/kg i.v.), or phentolamine (10 mg/kg i.v.) and before initiation of cocaine infusion (4 mg/kg/min i.v.). Values are mean ± S.E.M. *p < .05 Fast versus Slow strain.
also attenuated peak rate-pressure products during cocaine infusion and completely abolished the difference in peak rate-pressure products between Slow and Fast strains. Pretreatment of animals with phentolamine markedly delayed cocaine-elicited peak hemodynamic responses (~2.5 min versus ~0.5 min in vehicle- and all other agent-pretreated groups) in all animals.

Table 2 presents the effect of different autonomic blocking agents on the cocaine threshold for production of AV conduction block and cardiac arrest. Hexamethonium, chlorisondamine, and propranolol did not significantly modify cocaine doses required to produce AV conduction block and cardiac arrest in any strain, nor did they alter the differences in cocaine doses for the two endpoints between Fast and Slow rats. The nonselective alpha adrenergic receptor blocker phentolamine, on the other hand, not only increased cocaine doses required to produce AV conduction block and cardiac arrest in all strains but also abolished the differences in cocaine doses for these two endpoints between Fast and Slow rats. None of the autonomic blocking agents tested altered arrhythmia patterns induced by cocaine. AV conduction block was still the predominant arrhythmia pattern.

The effect of autonomic blocking agents on plasma cocaine concentrations is shown in Fig. 6. Except for propranolol, all of the autonomic blocking agents attenuated the difference in plasma cocaine concentrations between Slow and the other strains. Plasma cocaine concentrations were markedly reduced in all animals with phentolamine pretreatment and stayed relatively flat from 1.0 through 7.5 min. Differences between Fast and Slow strains for plasma cocaine concentrations at the onset of AV conduction block were abolished by pretreatment with all of the blocking agents.

**Discussion**

Results of our studies provide the first evidence of genetically determined differential sensitivity to cocaine-induced cardiac arrhythmias and arrest in animals. In this model, two genetically selected strains of rats differ markedly in the cocaine doses for these two endpoints between Fast and Slow rats. None of the autonomic blocking agents tested altered arrhythmia patterns induced by cocaine. AV conduction block was still the predominant arrhythmia pattern.

The toxic doses of cocaine that produced arrhythmias and cardiac arrest in our studies are similar to those from other studies with rats under similar experimental conditions (the LD<sub>100</sub> for rats has been reported to be about 35–75 mg/kg) (Smart and Anglin, 1987; Morishima et al., 1993). The lethal (cardiac arrest) dose for Slow rats is slightly below the lower range (32 ± 2 mg/kg), whereas the lethal dose for Fast rats is in the higher range (62 ± 6 mg/kg). The toxic dose is dependent on a number of factors, including route and rate of injection, as well as physiological state of the animals. Anesthesia, mechanical ventilation, and oxygenation all play a role in the prolongation of survival time. However, anesthesia used in our experiments does not contribute to the differential sensitivity because there was no difference in anesthetic (halothane) requirement in the two strains of rats (the MAC for halothane was similar between Fast and Slow rats).

In humans, doses and blood concentrations of cocaine at toxic endpoints are generally lower and vary more than in experimental animals (Smart and Anglin, 1987). As we mentioned earlier, the lethal dose in human has been reported to be 1.2 g, but severe toxicity has been reported with a dose as low as 20 mg (Estroff and Gold, 1986). Also, blood levels of cocaine in victims who died of i.v. cocaine use vary greatly among individuals (from 0.11–75 mg/dl) (Smart and Anglin, 1987). Therefore, results from animal studies on cocaine may not apply to human situations. However, evidence from both our rat studies and from human studies by others (Estroff and Gold, 1986; Smart and Anglin, 1987) shows a similar picture that genetic predisposition plays a very important role in determining sensitivity to the cardio toxicity of cocaine. Elucidation of the mechanisms for the differential sensitivity to cocaine-induced fatal cardiac arrhythmias between these two strains of rats may unveil the nature of genetic hypersensitivity to cocaine-induced sudden death in humans.

Cocaine affects the heart by two mechanisms. First, it accentuates the actions of the sympathetic nervous system by inhibiting the reuptake of norepinephrine and dopamine at the presynaptic level in both the central and peripheral nervous systems (Knuepfer and Branch, 1992; Tella et al., 1992; de Jong, 1994). Second, through its local anesthetic effect (Watt and Pruitt, 1964; de Jong, 1994), cocaine blocks sodium channels (Crumb and Clarkson, 1990; de Jong, 1994), resulting in a slowing of conduction velocity as manifested on the

**TABLE 2**

<table>
<thead>
<tr>
<th></th>
<th>Vehicle (1 ml/kg)</th>
<th>Hexamethonium (20 mg/kg)</th>
<th>Chlorisondamine (5 mg/kg)</th>
<th>Propranolol (1 mg/kg)</th>
<th>Phentolamine (10 mg/kg)</th>
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<tbody>
<tr>
<td><strong>AV block dose</strong></td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>Wistar</td>
<td>22 ± 2</td>
<td>23 ± 3</td>
<td>19 ± 3</td>
<td>28 ± 6</td>
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<tr>
<td>Long-Evans</td>
<td>23 ± 2</td>
<td>26 ± 5</td>
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<td>15 ± 1***</td>
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<td>39 ± 6***</td>
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<td><strong>Cardiac arrest</strong></td>
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<td>33 ± 3***</td>
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<td>28 ± 2***</td>
<td>81 ± 4***</td>
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</table>

Fast and Slow refer to genetically fast- and slow-amphigdala kindling rats, respectively. Wistar and Long-Evans were control strains.

Animals were pretreated with vehicle (saline), hexamethonium, chlorisondamine, propranolol, or phentolamine i.v. 5 min or 10 min (chlorisondamine) before the initiation of cocaine infusion (4 mg/kg/min i.v.).

Values (mg/kg) are mean ± S.E.M.

*<i>p < .05 versus Wistar, **p < .05 versus Long-Evans, ***p < .05 versus Fast, ****p < .05 versus vehicle-pretreated groups of the same strain.
ECG by a prolongation of the P-R and QRS intervals (Watt and Pruitt, 1964; Hale et al., 1989; Kabas et al., 1990). Both ventricular arrhythmias and conduction block occur in humans and are the major causes for cocaine-induced sudden death (Young and Glauber, 1947; Benchimal et al., 1978; Nanji and Filipenko, 1984; Isner et al., 1986; de Jong, 1994). In anesthetized animals, however, conduction block is the predominant arrhythmia pattern induced by cocaine and other local anesthetics (Watt and Pruitt, 1964; Heavenr et al., 1995). The mechanisms for cocaine-induced ventricular fibrillation and AV block are not fully understood. Generation of ventricular fibrillation is attributed to a combined action of cocaine on the autonomic nervous system and ion channels. Conduction block induced by cocaine is primarily related to its local anesthetic property (ion channel blockade) (de Jong, 1994).

The fact that Slow rats have significantly higher peak rate-pressure products than Fast rats during cocaine infusion suggests a greater increase in sympathetic activity induced by cocaine in the Slow strain. The difference in the peak hemodynamic responses to cocaine exposure as well as the rate-pressure product dynamics during cocaine infusion between Slow and Fast rats was abolished by blockade of sympathetic ganglia and alpha adrenergic receptors and partially altered by beta adrenergic blockade. However, only the alpha adrenergic receptor blocker phentolamine abolished the difference in cocaine doses required to produced arrhythmias and cardiac arrest between Fast and Slow strains. Neither ganglionic nor beta adrenergic receptor blockade significantly altered these differences. In addition to its primary alpha adrenergic blocking property, phentolamine has direct antiarrhythmic activity through its electrophysiological effects, similar to those observed with class I agents such as quinidine (Rosen et al., 1971). Phentolamine reduces automaticity and conduction velocity and therefore prevents ventricular arrhythmias (Rosen et al., 1971). In this study, phentolamine prevented (rather than potentiated) cocaine-induced AV conduction block. Apparently, prevention of cocaine-induced AV conduction block with phentolamine is independent of direct electrophysiological actions of phentolamine and is more likely associated with its alpha adrenergic blocking property. We therefore concluded that the alpha adrenergic receptor system is significantly involved in the differential sensitivity to the cardiotoxic effects of cocaine between Slow and Fast rats. However, it is not clear which subtype or subtypes and which related component or components are the key element or elements that determine the differential sensitivity.

Another striking difference between Fast and Slow rats is that plasma cocaine concentration in Slow rats increased much faster than in Fast rats during constant i.v. cocaine infusion at any dose. This higher plasma concentration for a given dose indicates that there was a smaller volume of distribution ($V_d = \text{dose/plasma concentration}$) for cocaine in Slow rats, which could be the immediate and direct cause for the earlier onset of AV conduction block or hypersensitive nature in this strain. The mechanisms for the smaller volume of distribution in the Slow strain versus the Fast strain are not known. The pharmacokinetic difference may be determined by a difference in the alpha adrenergic receptors because alpha adrenergic blockade abolished the difference in hemodynamic responses and plasma cocaine concentrations and, at the same time, abolished the difference in cocaine arrhythmia thresholds between Fast and Slow strains. However, the difference in volume of distribution may not be the sole reason because the differences in sensitivity to the car-

![Fig. 6. Plasma cocaine concentrations versus i.v. cocaine infusion time in the absence (vehicle) and presence of four autonomic blocking agents. The inserted figures show plasma cocaine concentrations at the onset of AV conduction block. Values are mean ± S.E.M. *p < .05 Fast versus Slow strains.](image-url)
dioxicity of cocaine between strains were not significantly altered by the two ganglionic blockers that abolished the pharmacokinetic differences. In addition, plasma and heart cocaine concentrations at arrhythmia onset were lower (~1.5 times) in Slow rats than in Fast rats, indicating that the heart of Slow rats is more sensitive to cocaine-induced arrhythmogenesis.

Examination of our data eliminates other possible contributing factors for the differential sensitivity to the cardioxicity of cocaine among strains. For example, preexisting hypertension does not appear to contribute to the differences in the sensitivity because the Slow and Fast rats are equally hypertensive. Also, hypotension is not responsible for the earlier onset of arrhythmias in the Slow strain because arterial blood pressure at arrhythmia onset was not lower in the Slow than in the other strains. In addition, seizures do not contribute to the increased sensitivity to the cardiotoxic effects of cocaine in the Slow rats because cocaine seizure thresholds were similar between Slow and the other strains. The relationship between seizures and arrhythmias is unclear. The fact that the differential sensitivity to the arrhythmogenic effects of cocaine exists in two strains of rats that are genetically selected by phenotypic differences in kindling suggests a possible link between kindling seizures and cocaine arrhythmogenesis.

In conclusion, the most significant contribution of the present study is to identify the first animal model demonstrating genetically determined differential sensitivity (hypersensitivity versus resistant) to cocaine-induced arrhythmias. There appears to be a significant involvement of the alpha adrenergic receptor system in the differential sensitivity. This study is an initial step toward uncovering the underlying mechanisms of predisposition to the arrhythmogenic action of cocaine and other drugs in humans.

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References


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