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Inhibition of centrally induced ventricular arrhythmias by rilmenidine and idazoxan in rabbits

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Abstract In a model of ventricular arrhythmias of central origin, we investigated the effects of rilmenidine, an oxazoline with antihypertensive properties, and idazoxan, an imidazoline that is an antagonist of the hypotensive effects of rilmenidine.

Bicuculline, a $GABA_A$ receptor antagonist, was administered intracisternally (i.c.) to produce arrhythmias in pentobarbitone anaesthetised rabbits; 10 μg/kg bicuculline i.c. induced polymorphic ventricular ectopic beats and ventricular tachycardia while blood pressure increased by about 50–60% and sinusal heart rate decreased by about 20%. Rilmenidine, either administered intravenously (0.01, 0.1, 1 mg/kg i.v.) or i.c. (3, 10, 30 μg/kg) dose-dependently prevented the occurrence of bicuculline-induced arrhythmias while, because of a lower base-line, the blood pressure values reached were less as compared to controls. Idazoxan administered i.v. (3, 10 mg/kg) had a similar action. Idazoxan i.c. (15 μg/kg) had no significant antiarrhythmic effect but antagonized in part the haemodynamic and antiarrhythmic effects of rilmenidine (1 mg/kg i.v.; 30 μg/kg i.c.).

It is suggested that the antiarrhythmic effects observed with rilmenidine are mainly mediated by blunting the bicuculline-induced increase in the sympathetic nervous output to the heart and the vascular beds. These effects of rilmenidine are likely to originate both from the central and peripheral nervous system. The antiarrhythmic effects of idazoxan i.v. might be related to a blocking action on alpha$_2$-adrenoceptors at the level of the coronary arteries and other vascular beds.

Key words Alpha-adrenergic agents · Arrhythmia · Bicuculline · Idazoxan · Imidazoline compounds · Rilmenidine

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Introduction

Several lines of experimental and clinical evidence suggest that centrally acting antihypertensive drugs bearing an imidazoline or a related chemical structure, such as clonidine or rilmenidine, can prevent the occurrence of ventricular arrhythmias (Thomas and Stephen 1991; Hayashi et al. 1993; Mammoto et al. 1995). These drugs decrease the sympathetic nervous output to the heart and vascular beds, and facilitate the basal parasympathetic tone and the parasympathetic component of the baroreflex (Korner et al. 1974; Mc Kaigue and Harron 1992). These effects are commonly believed to be due mainly to an action on alpha$_2$-adrenergic receptors and/or on receptors specifically sensitive to the imidazoline structure, in the medulla oblongata (Feldman et al. 1990; Gomez et al. 1991; Ernsberger et al. 1995; Guyenet et al. 1995). In addition, drugs with alpha$_2$-adrenoceptor antagonist activity, such as phentolamine, yohimbine or idazoxan, that can antagonize the hypotensive and antiarrhythmic effects of centrally acting antihypertensive imidazolines or related compounds, were also shown to have antiarrhythmic properties (Gould et al. 1975; DiMicco et al. 1977; Segal et al. 1981, 1984; Bernauer 1990; Feldman et al. 1990; Ernsberger et al. 1995; Roegel et al. 1996).

In the present study, we investigated the effects of rilmenidine and idazoxan in a model of ventricular arrhythmias provoked by central administration of bicuculline in rabbits. In such a model, central and peripheral autonomic pathways are involved. The blockade of central nervous system $GABA_A$ receptors in particular increases the sympathetic output to the heart and the vascular beds (DiMicco et al. 1977, Segal et al. 1981, 1984). Rilmenidine, an oxazoline closely related to the imidazoline substances, has been reported to prevent the occurrence of arrhythmias even at low non-hypotensive doses (Mammoto et al. 1995). Idazoxan is an antagonist of the cardiovascular effects of rilmenidine that bears an imidazoline structure and has also been reported to have antiarrhythmic effects (Feldman et al. 1990; Ernsberger et al. 1995; Roegel et al. 1996). The effects of sodium nitroprusside, a peripheral vasodilator, were

also tested to evaluate the possible role of vasodilatation in antiarrhythmic effects of the tested drugs.

Methods

Surgical preparation. Normotensive male rabbits (Zika strain) weighing from 2.4 to 3.5 kg were used. The animals were anaesthetized with sodium pentobarbital (70 to 90 mg/kg) injected through the marginal vein of the ear and after tracheotomy were ventilated with room air (stroke volume of 12 ml/kg; rate of 18/min).

The right femoral vein was cannulated for drug administration and the right femoral artery for blood pressure (BP) monitoring (Statham P23 XL transducer, Gould 20–4615–52 pressure processor amplifier and BS-272 penrecorder – Gould Electronique, Longjumeau, France). Systolic (SAP) and diastolic (DAP) arterial pressures were obtained directly from recordings. Mean arterial pressure (MAP) was calculated as diastolic pressure plus one third of the pulse pressure. Heart rate (HR) was counted from the blood pressure waves by rapid running of the pressure recording. At each heart rate measurement, we checked that the rhythm was sinusal.

Subsequently the animals were immobilized with pancuronium bromide (0.2 mg/kg i.v.). The ventilation parameters were adjusted to maintain normal P_aCO_2, P_aO_2 and pH. Body temperature was maintained with a heated blanket at 38 ±0.5°C (Harvard homeothermic blanket – Harvard apparatus, Edenbridge, UK).

The electrocardiogram (ECG) was continuously recorded from standard leads (stainless-steel needles; derivations D1, D2, D3, aVR, aVL, aVF; 6-channel ECG Hellige EK 512E – Hellige, Freiburg im Breisgau, Germany).

In order to perform the intracisternal injections, the head of the animal was placed in a stereotaxic frame (Unimécanique, Epinay/Seine, France). A surgical approach was performed to expose the atlanto-occipital membrane. Drugs were injected in the cisterna magna under visual control. Normal saline solution (control group) or drugs were injected in a constant volume of 50 µl/kg.

Experimental protocol. Bicuculline (10 µg/kg i.c.) was injected 10 min after administration of the drug to be tested (idazoxan or rilmenidine) or of the vehicle. This dose of bicuculline was the lowest dose that regularly generated ventricular arrhythmias; the same amount of bicuculline given either intravenously or intracerebroventricularly failed to induce arrhythmias. Immobilization was complemented by an additional injection of pancuronium bromide (0.1 mg/kg i.v.) 5 min before bicuculline injection. Cardiovascular parameters (arterial pressure, ECG) were recorded for 30 min after bicuculline administration. The area under the curve (AUC) of the mean arterial pressure, of the heart rate and of the rate-pressure product, an index of the myocardial oxygen demand, were also calculated for the same period.

Evaluation of arrhythmias. Arrhythmias were evaluated according to the Lambeth convention (Walker et al. 1988). The incidence, occurrence (number of episodes) and duration (sec) of the following arrhythmias were recorded: ventricular ectopic beats (VEB), ventricular tachycardia (VT) and ventricular fibrillation (VF). The number of ventricular ectopic beats (VEB) included single ectopic beats (VEBi) as well as those included in VT. VT was regarded as a run of 5 or more ectopics beats. VF was regarded as ineffective ectopic beats; blood pressure collapsed (0–15 mmHg) and no pulse pressure remained. VF as well as the episodes of atrioventricular conduction defects or atrial tachyarrhythmias were recorded but not included in the analysis since they occurred very rarely.

Statistical analyses. All data are expressed as the mean ± SEM. For the gaussian-distributed variables, a one-way analysis of variance was first performed to test for any differences between the mean values of all groups; $P<0.05$ was considered significant. If a difference was established, a Scheffé's test was used as a post-hoc test to localize the significant difference. For all non-gaussian-distributed variables, a square root transform or a natural log transform was performed in order to enable the use of the same parametric tests. If the transformation was ineffective in this respect, a Kruskal-Wallis test was ultimately carried out. If a difference was established with this non-parametric test, the groups of treated animals were compared with each other using a Mann-Whitney test to localize the significant difference; a $P<0.05$ was considered significant. Linear regression was used to analyse the relationship between quantitative variables. All calculations were made by computer-assisted analyses with a commercially available statistical package (BMDP Statistical Software, Cork, Ireland).

Drugs used. The following drugs were used: sodium pentobarbitone (Pentobarbital sodique, Sanofi santé animale, Libourne, France), pancuronium bromide (Organon Teknika, Fresnes, France), bicuculline methiodide (Sigma Chemical Co., St.Louis, Mo., USA), idazoxan HCl (Research Biochemicals Inc., Natick, Mass., USA), rilmenidine (Servier, Neuilly-sur-Seine, France), sodium nitroprusside (Roche, Neuilly-sur-Seine, France). Control animals received an equal volume (0.1 ml/kg i.v.; 50 µl/kg i.c.) of saline solution (0.9% NaCl wt/v dissolved in distilled water). The infusion rate of sodium nitroprusside was adjusted to decrease the mean basal arterial pressure to a similar extent as rilmenidine 1 mg/kg i.v..

Results

Haemodynamic response and arrhythmias induced by intracisternal administration of bicuculline

Intracisternal administration of 10 µg/kg bicuculline increased blood pressure by about 50–60% while heart rate decreased by about 20%. Maximum haemodynamic effects were reached during the first 10 min after bicuculline administration (Tables 1 and 3). Subsequently, blood pressure and heart rate returned to base-line values that were reached about 30 to 40 min after bicuculline administration (result not shown).

Ventricular arrhythmias i.e. VEB and ventricular tachycardia were observed in each animal receiving 10 µg/kg bicuculline intracisternally (Tables 2 and 4). These arrhythmias started within 5 min after bicuculline administration and usually lasted 10 to 15 min. Only one rabbit developed ventricular fibrillation (one spontaneously reversible episode). None of the animals died during the 30 min period after bicuculline i.c. administration.

Effects of intravenously administered rilmenidine on the haemodynamic response and arrhythmias induced by bicuculline

Rilmenidine (before bicuculline injection) dose-dependently (0.01, 0.1 and 1 mg/kg) decreased basal blood pressure and heart rate, the effects reaching statistical significance at the 1 mg/kg dose (Table 1). During the 30 min after bicuculline administration, the AUC of the blood pressure was also dose-dependently decreased by rilmenidine. The AUC of the heart rate, on the other hand, only decreased in the animals treated with 1 mg/kg rilmenidine.

Intravenously administered rilmenidine decreased VEBt, VEBi, VTe and VTt, the highest dose being the most effective (Table 2).

Table 1 Effects of i.v. administered idazoxan, rilmenidine, saline and sodium nitroprusside on haemodynamic parameters before and after intracisternal administration of 10 μg/kg bicuculline

		Before drug injection	After drug injection, before bicuculline administration	10 min after bicuculline	AUC 0–30min
Saline iv	MAP	96±2	93±2	133±3	730±12
$n = 15$	HR	297±4	298±5	234±17	1495±84
Rilmenidine					
(0.01 mg/kg iv)	MAP	98±5	98±5	142±3	678±33
$n = 7$	HR	291±9	293±10	261±22	1581±109
Rilmenidine					
(0.1 mg/kg iv)	MAP	101±3	84±7	121±5	651±29 *
$n = 6$	HR	295±4	263±12	246±15	1571±76
Rilmenidine					
(1 mg/kg iv)	MAP	99±3	68±4 *	106±9	597±40 *
$n = 5$	HR	272±7	216±9 *	172±15	1152±16 *
Idazoxan					
(0.3 mg/kg iv)	MAP	92±4	92±6	130±7	727±46
$n = 5$	HR	272±5	262±7 *	277±7	1636±52
Idazoxan					
(3 mg/kg iv)	MAP	92±4	90±2	131±4	699±15
$n = 5$	HR	266±8	236±8 *	251±19	1535±87
Idazoxan					
(10 mg/kg iv)	MAP	93±3	84±3	118±6	653±24 *
$n = 5$	HR	282±11	244±13 *	228±14	1429±66
Na nitroprusside	MAP	99±2	69±4 *	116±5	626±27 *
$n = 5$	HR	300±18	306±9	288±19	1831±72 *

HR, heart rate in beats per min (bpm); MAP, mean arterial pressure in mmHg; AUC, area under the curve during the 30 min after bicuculline injection (expressed in min·mmHg·5^{-1} for the MAP AUC and in min·bpm·5^{-1} for the HR AUC); * indicates a significant difference from the corresponding value in the group of rabbits injected with saline (NaCl 0,9% i.v.). Results are expressed as the mean ±SEM

Effects of intracisternally administered rilmenidine on the haemodynamic response and arrhythmias induced by bicuculline

Intracisternal administered rilmenidine dose-dependently (3, 10 and 30 μg/kg) decreased the basal blood pressure and heart rate (Table 3). During the 30 min after i.c. bicuculline administration, the AUC of the blood pressure and of the heart rate were also decreased by rilmenidine; this decrease appeared dose-dependent for blood pressure while it was rather similar for heart rate at the three doses used (Table 3).

Intracisternally administered rilmenidine dose-dependently decreased VEBi (Table 4). VEBt, VTe and VTt also decreased but not significantly, probably due to high variation.

Effects of intravenously administered idazoxan on haemodynamic response and arrhythmias induced by bicuculline

The effects of 0.3, 3 and 10 mg/kg intravenously administered idazoxan were studied (Table 1). Basal arterial pressure remained stable up to the dose of 3 mg/kg; blood pressure tended to decrease with the 10 mg/kg dose. Basal heart rate was decreased, the maximal bradycardia being observed with the 3 mg/kg dose. In the 30 min following the i.c. bicuculline administration, the AUC of the blood pressure was not statistically different in animals treated with 0.3 mg/kg and 3 mg/kg idazoxan while it was decreased in animals treated with 10 mg/kg idazoxan. The AUC of the heart rate was not significantly affected. Nevertheless, heart rate changes following bicuculline administration appeared different, tachycardia being substituted for bradycardia in animals treated with 0.3 mg/kg and 3 mg/kg idazoxan.

No significant increase of VEBt, VEBi, VTe and VTt was observed with the 0.3 mg/kg dose of idazoxan (Table 2). At the 3 mg/kg and 10 mg/kg doses, idazoxan reduced the bicuculline-induced arrhythmias.

Effects of intracisternal idazoxan on the haemodynamic response and arrhythmias induced by bicuculline

Basal blood pressure and heart rate of rabbits treated with idazoxan (15 μg/kg) were not significantly different from

Table 2 Effects of intravenously administered saline, rilmenidine, idazoxan and sodium nitroprusside on the development of arrhythmias after intracisternal injection of bicuculline

Treatment	n	RPP	VEB_t	VEB_i	VT_n	VT_e	VT_t
Saline iv	15(0)	181±10	1955±326	1228±247	11	44±13	142±55
Rilmenidine (0.01 mg/kg iv)	7(0)	182±13	595±155 *	207±76 *	5	9±5	100±37
Rilmenidine (0.1 mg/kg iv)	6(0)	171±12	490±335 *	175±110 *	2	9±6	72±55
Rilmenidine (1 mg/kg iv)	5(0)	114±6 *	13±13 *	9±9 *	1	1±1 *	1±1 *
Idazoxan (0.3 mg/kg iv)	5(0)	202±16	2980±850	1962±211	5	102±71	188±150
Idazoxan (3 mg/kg iv)	5(0)	179±15	488±488 *	349±193	1	15±15	74±74
Idazoxan (10 mg/kg iv)	5(0)	155±9	157±157 *	157±157 *	0	0±0 *	0±0 *
Na nitroprusside (15 to 17 µg/kg min iv)	5(0)	190±9	350±293 *	352±295	0	0±0 *	0±0 *

n, number of animals; (), number of deaths; RPP, integral of the rate pressure product during the 30 min after bicuculline administration ($mmHg \times bpm \times min \times 5^{-1} \times 10^3$); VEB_t, total number of ventricular ectopic beats; VEB_i, isolated ventricular ectopic beats; VT_n, number of animals which developed ventricular tachycardia; VT_e, number of episodes of ventricular tachycardia; VT_t, duration of ventricular tachycardia (sec); * indicates a significant difference from the corresponding value in the group of rabbits injected with saline (NaCl 0,9% i.v.). Results are expressed as the mean ±SEM

Table 3 Effects of intracisternally administered idazoxan, rilmenidine and saline on haemodynamic parameters before and after intracisternal injection of 10 µg/kg bicuculline

		Before drug injection	After drug injection, before bicuculline injection	10 min after bicuculline	AUC 0–30min
Saline ic	MAP	93±4	95±3	138±4	755±16
n = 11	HR	289±8	287±8	238±18	1515±71
Rilmenidine (3 µg/kg ic)	MAP	93±3	74±3 *	130±7	707±34
n = 5	HR	278±4	266±6	201±13	1305±77
Rilmenidine (10 µg/kg ic)	MAP	93±5	56±4 *	107±9	615±43 *
n = 5	HR	286±2	242±9 *	220±17	1299±89
Rilmenidine (30 µg/kg ic)	MAP	103±6	53±1 *	102±8	551±35 *
n = 5	HR	304±13	239±10 *	204±16	1267±91
Idazoxan (15 µg/kg ic)	MAP	95±4	92±5	142±9	740±49
n = 5	HR	278±12	262±8	248±17	1543±59

HR, heart rate in beats per min (bpm); MAP, mean arterial pressure in mmHg; AUC, area under the curve during the 30 min after bicuculline injection (expressed in $min \times mmHg \times 5^{-1}$ for the MAP AUC and in $min \times bpm \times 5^{-1}$ for the HR AUC); * indicates a significant difference from the corresponding value in the saline treated group. Results are expressed as the mean ±SEM

vehicle-treated controls (Table 3). During the 30 min after i.c. bicuculline administration, the AUC of the blood pressure and of the heart rate were not significantly affected by idazoxan i.c. either (Table 3).

Intracisternally administered idazoxan (15 µg/kg) had no significant effect on arrhythmias (Table 4). One animal developed ventricular fibrillation (one spontaneously reversible episode of VF).

Effects of intracisternal idazoxan on haemodynamic and antiarrhythmic effects of rilmenidine

We studied the effects of intracisternal administration of idazoxan (15 µg/kg) on the effects of rilmenidine 1 mg/kg i.v. and 30 µg/kg i.c., doses with equipotent antiarrhythmic potencies. Idazoxan was injected just before rilmenidine.

The effects of rilmenidine on basal blood pressure and on the AUC of the blood pressure after bicuculline tended to be or were slightly attenuated by idazoxan i.c (Table 5, compared with corresponding values in Tables 1 and 3).

Table 4 Effects of intracisternally administered saline, rilmenidine and idazoxan on the development of arrhythmias after intracisternal injection of bicuculline

Treatment	*n*	RPP	VEB_t	VEB_i	VT_n	VT_e	VT_t
Saline ic	11 (0)	191±12	1704±460	1105±230	5	9±6	144±110
Rilmenidine (3 µg/kg ic)	5 (1)	154±1	1156±844	211±176 *	2	55±52	195±146
Rilmenidine (10 µg/kg ic)	5 (0)	131±9 *	621±532	151±92 *	1	5±5	101±101
Rilmenidine (30 µg/kg ic)	5 (0)	113±4 *	1±1	1±1 *	0	0±0	0±0
Idazoxan (15 µg/kg ic)	5 (1)	191±17	2161±1069	569±325	3	85±50	311±148

n, number of animals; (), number of deaths; RPP, integral of the rate pressure product during the 30 min after bicuculline administration ($mmHg \times bpm \times min \times 5^{-1} \times 10^3$); VEB_t, total number of ventricular ectopic beats; VEB_i, isolated ventricular ectopic beats; VT_n, number of animals which developed ventricular tachycardia; VT_e, number of episodes of ventricular tachycardia; VT_t, duration of ventricular tachycardia (sec); * indicates a significant difference from the corresponding value in the group of rabbits injected with saline (NaCl 0,9% i.c.). Results are expressed as the mean ±SEM

Table 5 Effects of idazoxan and rilmenidine administered simultaneously on haemodynamic parameters before and after intracisternal injection of 10 µg/kg bicuculline

		Before drug injection	After drug injection, before bicuculline injection	10 min after bicuculline	AUC 0–30min
Idazoxan (15 µg/kg ic) Rilmenidine (1 mg/kg iv)	MAP	88±4	82±4 *°	123±6	644±13°
	HR	288±12	236±9	290±17	1593±107°
Idazoxan (15 µg/kg ic) Rilmenidine (30 µg/kg ic)	MAP	96±4	86±3°	121±5	669±29°
	HR	296±8	272±9°	215±17	1272±9 *

HR, heart rate in beats per min (bpm); MAP, mean arterial pressure in mmHg; AUC, area under the curve during the 30 min after bicuculline injection (expressed in $min \times mmHg \times 5^{-1}$ for the MAP AUC and in $min \times bpm \times 5^{-1}$ for the HR AUC); * and ° indicate a significant difference from the corresponding value in the groups treated with idazoxan and rilmenidine alone, respectively (see Tables 1 and 3). Results are expressed as the mean ±SEM

Rilmenidine's influence on basal heart rate was not significantly altered by idazoxan i.c. In contrast, pretreatment with idazoxan i.c. antagonized the effects of rilmenidine i.v. on the AUC of the heart rate after bicuculline. Changes in heart rate after bicuculline appeared even totally different in animals pretreated with idazoxan i.c. and rilmenidine i.v, tachycardia occurring instead of bradycardia (Tables 5, compared to corresponding values in Table 1). The effects of rilmenidine i.c. on the AUC of the heart rate after bicuculline, were not significantly changed by pretreatment with idazoxan i.c.

The antiarrhythmic effects of rilmenidine tended to be inhibited in rabbits treated simultaneously with idazoxan i.c. (Table 6, compared to corresponding values in Tables 2 and 4); compared to animals treated with idazoxan alone, only VEB_t and VT_t were significantly decreased.

Effects of intravenously administered sodium nitroprusside on the haemodynamic response and arrhythmias induced by bicuculline

An infusion rate of 15 to 17 µg/kg/min sodium nitroprusside was necessary to decrease the mean basal arterial pressure to a similar level as observed with rilmenidine 1 mg/kg i.v. (Table 1).

Basal arterial pressure decreased (MAP: –30±3 mmHg) while basal heart rate remained stable (Table 1). In the 30 min of bicuculline administration, the AUC of the blood pressure was decreased compared with the control animals; in contrast, the AUC of the heart rate increased.

VEB_t, VEB_i, VT_e and VT_t decreased compared with the control group (Table 2).

Table 6 Effects of idazoxan and rilmenidine administered simultaneously on the development of arrhythmias after intracisternal injection of bicuculline

Treatment	*n*	RPP	VEB_t	VEB_i	VT_n	VT_e	VT_t
Idazoxan (15 µg/kg ic) Rilmenidine (1 mg/kg iv)	5(0)	172±11	396±345 *	73±46	1	7±7	69±69 *
Idazoxan (15 µg/kg ic) Rilmenidine (30 µg/kg ic)	5(0)	142±13 *°	150±75 *	146±75	2	1±1	1±1 *

n, number of animals; (), number of deaths; RPP, integral of the rate pressure product during the 30 min after bicuculline administration ($mmHg \times bpm \times min \times 5^{-1} \times 10^3$); VEB_t, total number of ventricular ectopic beats; VEB_i, isolated ventricular ectopic beats; VT_n, number of animals which developed ventricular tachycardia; VT_e, number of episodes of ventricular tachycardia; VT_t, duration of ventricular tachycardia (sec); * and ° indicate a significant difference from the corresponding value in the group treated with idazoxan and rilmenidine alone, respectively (Tables 2 and 4). Results are expressed as the mean ±SEM

Discussion

In this study, the effects of rilmenidine and idazoxan were tested in a model of ventricular arrhythmias provoked by i.c. injection of bicuculline in rabbits. A similar model has already been used to induce cardiac ischemia and arrhythmias (DiMicco et al. 1977, Segal et al. 1981, 1984). It was shown that blockade of central nervous system $GABA_A$ receptors increases the sympathetic tone to the heart and the vascular beds. Coronary constriction occurred as indicated by an increase in coronary vascular resistance followed by ST segment elevation. Cardiac load also increased. Therefore, arrhythmias observed under such conditions are likely to be due, at least in part, to ischemia.

The intracisternal route was used to administer bicuculline in order to reach the medullary structures involved in the control of the sympathetic and parasympathetic nervous output to the heart and vascular beds (DiMicco et al. 1977; Chalmers and Pilowski 1991; Van Giersbergen et al. 1992; Reis et al. 1994). GABA is a physiological inhibitory neurotransmitter in various medullary structures involved in baroreflex function, and high parasympathetic tone can be involved in occurrence of arrhythmias, especially when the sympathetic activity is high (DiMicco et al. 1977; Skinner 1993). A relatively low dose of bicuculline (10 µg/kg), which was the threshold dose to reproducibly induce arrhythmias, was used. The baroreflex, as indicated by bradycardia following blood pressure increase, appeared to be efficient under these conditions (Tables 1 and 3). The occurrence of arrhythmia seemed to be linked to the respiration cycle. The parasympathetic system might thus be involved, associated with the sympathetic output increase, in the mechanisms of arrhythmia in our model (Japundzic et al. 1990).

Under our experimental conditions, there was a strong positive link between the antiarrhythmic effects observed with rilmenidine, idazoxan and sodium nitroprusside and the AUC of the blood pressure in the 30 min period following the injection of bicuculline; a weaker relationship was observed between antiarrhythmic effects and basal blood pressure (before bicuculline administration). In contrast, when considering the AUC of the heart rate and of the rate-pressure product during the same period, the linking with antiarrhythmic effects was not significant in animals treated with rilmenidine or idazoxan, and negative in animals treated with nitroprusside. The baroreflex function, as estimated from heart rate change (i.e. bradycardic response) following blood pressure increase caused by bicuculline, was similar to controls or decreased (Tables 1 and 3). Therefore, the antiarrhythmic effects of rilmenidine, idazoxan and sodium nitroprusside are likely to have resulted primarily from blunting the bicuculline-induced increase in the sympathetic nervous output to the cardiovascular system, and/or by blocking and/or attenuating its consequences at the level of the heart and/or of the vascular beds.

The antiarrhythmic effects of rilmenidine were probably mainly mediated by blunting bicuculline-induced increase in the sympathetic nervous output. Rilmenidine, administered either i.v. or i.c., dose-dependently decreased the AUC of the blood pressure after bicuculline, and inhibited the occurrence of arrhythmias. The doses of rilmenidine i.c. required to evoke such effects, were about 30 fold lower than the i.v. active doses. Moreover, the effects of rilmenidine on resting blood pressure, the AUC of the blood pressure and the occurrence of arrhythmias following bicuculline administration appeared to be prevented by idazoxan i.c.. The rather high dose of idazoxan that was used in this series of experiments is assumed to antagonize the central hypotensive and bradycardic effects of rilmenidine (Feldman et al. 1990).

The influence of idazoxan i.c. on the effects of rilmenidine, injected either i.c. or i.v., were tested in order to discriminate between central and peripheral effects of rilmenidine, when administered i.v.. The doses of rilmenidine (30 µg/kg i.c., 1 mg/kg i.v.) were selected because they induced similar antiarrhythmic effects. Since the decrease of the AUC of the heart rate observed after the administration of bicuculline i.c. in animals pretreated with 1 mg/kg rilmenidine was completely antagonized by the simultaneous administration of idazoxan i.c., this effect was prob-

ably mainly of central origin in rabbits treated with rilmenidine i.v. alone. As idazoxan antagonized the decrease in the AUC of the heart rate after 15 μg/kg i.c. bicuculline in animals treated with rilmenidine i.v. but not in animals treated with rilmenidine i.c., the "central" concentration of rilmenidine attained after rilmenidine i.v. may be assumed to have been less than after rilmenidine i.c.. A similar anti-arrhythmic effect was nevertheless observed after both pre-treatments. This observation suggests the involvement of peripheral effects of rilmenidine, when administered i.v., in its antiarrhythmic action. An effect on presynaptic alpha$_2$-adrenoceptors might contribute to such peripheral effects as it is assumed to contribute as well to the hypotensive effects of i.v. rilmenidine (Laubie et al. 1985; Koenig-Bérard et al. 1988; Szabo et al. 1993; Urban et al. 1995, Van Zwieten et al. 1986).

The antiarrhythmic effects of idazoxan were probably mainly related to a blockade of some effects mediated by the bicuculline-induced increase in sympathetic nervous output, at the level of the heart and vascular beds. When administered i.v. at the doses of 3 mg/kg and 10 mg/kg, idazoxan decreased the AUC of the blood pressure after bicuculline i.c. and inhibited the occurrence of arrhythmias. Since the i.c. administration of idazoxan (15–150 μg/kg) had no anti-arrhythmic effects, the anti-arrhythmic effects of systemically delivered idazoxan are unlikely to originate in the central nervous system even though idazoxan passes the blood-brain barrier and acts on central receptors after i.v. injection (Hannah et al. 1983; Freedman and Aghajanian 1984). The anti-arrhythmic effects and/or antiischaemic effects of idazoxan and other drugs with alpha$_2$-adrenoceptor antagonist activity have been suggested to be mediated through the blockade of peripheral postsynaptic alpha-adrenergic receptors in the heart and blood vessels (Segal et al. 1981, 1984; Seitelberger et al. 1991; Baumgart and Heusch 1995; Roegel et al. 1996). Stimulation of alpha$_2$-adrenergic receptors also induces coronary constriction during stress and cardiac ischemia (Seitelberger et al. 1991; Baumgart and Heusch 1995). So, idazoxan might prevent the bicuculline-induced vasoconstriction by blocking postsynaptic alpha$_2$-adrenergic receptors, in particular in the coronary arteries.

Presynaptic alpha$_2$-adrenoceptors are assumed to participate in the increased sympathetic activity induced by idazoxan. Therefore, the inconstant pro-arrhythmic effects observed in rabbits treated with 300 μg/kg idazoxan i.v., that were rather comparable to those induced by idazoxan i.c., might be explained by a higher pre/postsynaptic selectivity of idazoxan at this particular dose (Heyndrickx et al. 1984; Dabiré 1986).

Although the antiarrhythmic effects we observed with rilmenidine and idazoxan are likely to have resulted from inhibitory effects on the sympathetic nervous output to the vascular beds, the involvement of effects on cardiac sympathetic and parasympathetic tone cannot be excluded. The discrepancies between the antiarrhythmic effects of 0.1 mg/kg i.v. rilmenidine and 10 mg/kg i.v. idazoxan, treatments that are equally effective in blunting the blood pressure AUC after bicuculline, might be explained by effects on cardiac sympathetic and parasympathetic tone (Tables 1 and 2). The effects of rilmenidine on heart rate were mainly of central origin as they were practically maximal already after the lowest dose of rilmenidine i.c. (3 μg/kg) but only after the highest dose of rilmenidine i.v. (10 mg/kg)(Tables 1 and 3). The decrease of the AUC of the heart-rate observed, after the administration of bicuculline i.c., in animals pretreated with 1 mg/kg rilmenidine was completely antagonized by simultaneous administration of 15 μg/kg idazoxan i.c. (Tables 1 and 5). In contrast, the effects on heart rate induced by idazoxan probably originated mainly in the periphery as i.c. administration of 15 μg/kg (Table 3) or of higher doses (data not shown) appeared ineffective in this respect.

In conclusion, our data suggest that both rilmenidine and idazoxan can prevent ventricular arrhythmias of central origin. It is suggested that the antiarrhythmic effects observed with rilmenidine are mainly mediated by blunting the bicuculline-induced increase in sympathetic nervous output to the heart and the vascular beds. These effects of rilmenidine are likely to originate both in the central and the peripheral nervous system. On the other hand, the antiarrhythmic effects of idazoxan are probably mainly related to a blockade, at the level of the heart and vascular beds, of the bicuculline-induced increased sympathetic nervous output; these effects are believed to mainly result from an action on alpha$_2$-adrenoceptors.

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References

Baumgart D, Heusch G (1995) Neuronal control of coronary blood flow. Basic Res Cardiol 90:142–159

Bernauer W (1990) Antiarrhythmic and antinecrotic effects of yohimbine stereoisomers in rats during coronary occlusion and reperfusion. Basic Res Cardiol 85:132–141

Chalmers J, Pilowsky P (1991) Brainstem and bulbospinal neurotransmitter systems in the control of blood pressure. J Hypertens 9:675–694

Dabiré H (1986) Idazoxan: a novel pharmacological tool for the study of alpha$_2$-adrenoceptors. J Pharmacol (Paris) 17:113–118

DiMicco JA, Hamilton BL, Gillis RA (1977) Central nervous system sites involved in the cardiovascular effects of picrotoxin. J Pharmacol Exp Ther 203:64–71

Ernsberger P, Collins LA, Graves ME, Dreshaj IA, Haxhiu M (1995) Imidazoline I_1 receptors in the ventrolateral medulla and their role in cardiorespiratory control. In: Trouth CO, Millis RM, Kiwull-Schöne HF, Schläfke ME (eds) Ventral brainstem mechanisms and control of respiration and blood pressure. Marcel Dekker, New York, pp 319–358

Feldman J, Tibiriça E, Bricca G, Dontenwill M., Belcourt A, Bousquet P (1990) Evidence for the involvement of imidazoline receptors in the central hypotensive effect of rilmenidine in the rabbit. Br J Pharmacol 100: 600–604

Freedman JE, Aghajanian GK (1984) Idazoxan (RX 781094) selectively antagonizes alpha$_2$-adrenoceptors on rat central neurons. Eur J Pharmacol 105:265–272

Gomez RE, Ernsberger P, Feinland G, Reis DJ (1991) Rilmenidine lowers arterial pressure via imidazole receptors in brainstem C1 area. Eur J Pharmacol 195:181–191

Gould L, Reddy CVR, Weinstein T, Gomprecht RF (1975) Antiarrhythmic prophylaxis with phentolamine in acute myocardial infarction. J Clin Pharmacol 15:191–197

Guyenet PG, Lynch KR, Rosin DL, Stornetta RL, Allen AM (1995) Alpha$_2$-adrenergic receptors rather than imidazoline binding sites mediate the sympatholytic effect of clonidine in the rostral ventrolateral medulla. In: Trouth CO, Millis RM, Kiwull-Schöne HF, Schläfke ME (eds) Ventral brainstem mechanisms and control of respiration and blood pressure. Marcel Dekker, New York, pp 281–303

Hannah JAM, Hamilton CA, Reid JL (1983) RX781094, a new potent alpha$_2$ adrenoceptor antagonist: in vivo and in vitro studies in the rabbit. Naunyn-Schmiedeberg's Arch Pharmacol 322:221–227

Hayashi Y, Kamibayashi T, Maze M, Yamatodani A, Sumikawa K, Kuro M, Yoshiya I (1993) Role of imidazoline-preferring receptors in the genesis of epinephrine-induced arrhythmias in halothane-anesthetized dogs. Anesthesiology 78:524–530

Heyndrickx GR, Vilaine JP, Moerman EJ, Leusen I (1984) Role of prejunctional alpha$_2$-adrenergic receptors in the regulation of myocardial performance during exercice in conscious dogs. Circ Res 54:683–693

Japundzic N, Grichois ML, Zitoun P, Laude D, Elghozi JL (1990) Spectral analysis of blood pressure and heart rate in conscious rats: effects of autonomic blockers. J Auton Nerv Syst 30:91–100

Koenig-Berard E, Tierney C, Beau C, Delbarre G, Lhoste F, Labrid C (1988) Cardiovascular and central nervous system effects of rilmenidine in rats. Am J Cardiol 61:22D-31D

Korner PI, Oliver JR, Sleight P, Chalmers JP, Robinson JS (1974) Effects of clonidine on the baroreceptor-heart rate reflex and on single aortic baroreceptor fibre discharge. Eur J Pharmacol 28:189–198

Laubie M, Poignant JC, Scuvee-Moreau J, Dresse A, Schmitt H (1985) Pharmacological properties of (N-dicyclopropyl-methylamino-2-oxazoline (S3341)), an alpha$_2$-adrenoceptor agonist. J Pharmacol (Paris) 16: 259–278

Mac Kaigue JP, Harron DWG (1992) The effects of rilmenidine on tests of autonomic function in humans. Clin Pharmacol Ther 52:511–517

Mammoto T, Kamibayashi T, Hayashi Y, Takada K, Yamatodani A, Yoshiya I (1995) Rilmenidine prevents epinephrine induced arrhythmias in halothane anaesthetized dogs. J Cardiovasc Pharmacol 26 [Suppl 2]:S40-S43

Reis DJ, Golanov EV, Ruggiero DA, Sun MK (1994) Sympatho-excitatory neurons of the rostral ventrolateral medulla are oxygen sensors and essential elements in the tonic and reflex control of the systemic and cerebral circulations. J Hypertens 12 [Suppl 10]:S159–S180

Roegel JC, De Jong W, Monassier L, Feldman J, Bousquet P (1996) Comparative effects of idazoxan, prazosin and yohimbine on coronary ligation induced arrhythmias in spontaneously hypertensive rats. J Cardiovasc Pharmacol 27:226–234

Segal SA, Pearle DL, Gillis RA (1981) Coronary spasm produced by picrotoxin in cats. Eur J Pharmacol 76:447–451

Segal SA, Thomas J, Gillis RA (1984) Blockade of central nervous system GABAergic tone causes sympathetic-mediated increases in coronary vascular resistance in cats. Circ Res 55:404–415

Seitelberger R, Guth BD, Heusch G, Ross J (1991) Alpha$_2$-adrenergic coronary constriction in ischemic myocardium during exercise. In: Heusch G, Ross J (eds) Adrenergic mechanisms in myocardial ischemia. Steinkopff, Darmstadt, pp 207–217

Skinner JE (1993) Neurocardiology: brain mechanisms underlying fatal cardiac arrhythmias. Neurologic Clinics 11:325–351

Szabo B, Urban R, Starke K (1993) Sympatho-inhibition by rilmenidine in conscious rabbits; involvement of alpha$_2$-adrenoceptors. Naunyn-Schmiedeberg's Arch Pharmacol 348:593–600

Thomas GP, Stephen PM (1991) Protective action of clonidine against the arrhythmogenic and lethal effects of ouabain in guinea pigs. Br J Pharmacol 104:995–999

Urban R, Szabo B, Starke K (1995) Involvement of peripheral presynaptic inhibition in the reduction of sympathetic tone by moxonidine, rilmenidine and UK 14304. Eur J Pharmacol 282:29–37

Van Giersbergen PLM, Palkovits M, De Jong W (1992) Involvement of neurotransmitters in the nucleus tractus solitarii in cardiovascular regulation. Physiol Rev 72:789–824

Van Zwieten PA, Thoolen MHMC, Jonkman FAM, Willfert B, De Jong A, Timmermans PBMWM (1986) Central and peripheral effects of S3341 [(N-dicyclopropyl-methylamino-2-oxazoline)] in animal models. Arch Int Pharmacodyn Ther 279:130–149

Walker MJA, Curtis MJ, Hearse DJ, Campbell RWF, Janse MJ, Yellon DM, Cobbe SM, Coker SJ, Harness JB, Harron DWG, Higgins AJ, Julian DG, Lab MJ, Manning AS, Northover BJ, Parratt JR, Riemersma RA, Riva E, Russell DC, Sheridan DJ, Winslow E, Woodward B (1988) The Lambeth Conventions: guidelines for the study of arrhythmias in ischemia, infarction, and reperfusion. Cardiovasc Res 22:447–455