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Original article

Synthesis and antihypertensive effects of new methylthiomorpholinphenol derivatives

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Abstract

We present in this work the synthesis and cardiovascular effects of new methylthiomorpholine compounds and they were compared with cardiovascular drugs such as captopril, losartan and omapatrilat.
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Keywords: Cardiovascular effects; Thiomorpholin; Methylthiomorpholinphenols

1. Introduction

In the last twenty years, cardiovascular diseases have become the world's leading cause of death [1]. In this setting, well-formulated health promotion programs may play an important role, as they are thought to reduce chronic disease-related morbidity and mortality, as well as health care costs [2]. It is known that such health programs may be effectively delivered at the workplace, in addition to improve absenteeism and work safety [3–6]. Under most real-world conditions, it is difficult and costly for worksite health promotion programs to follow strictly controlled research designs over an extended period of time, impeding the evaluation and determination of the most effective programmatic structure to achieve health improvements. On the other hand, it is estimated that about one million patients are hospitalized for acute coronary events each year in the United States, and it is well known that low socioeconomic status is associated with increased risk of cardiovascular disease in both men and women, and in different ethnic groups [7–9]. Risk factors for cardiovascular disease are prevalent in men and women [10]. In young adults, education level is inversely associated with 5-year weight gain [11] and 10-year incidence of high blood pressure [12], while financial hardship is associated with 10-year incidence of hypertension [13].

The dihydropyridine calcium channel blocker compounds such as nifedipine and isradipine were originally developed for the treatment of hypertension [14,15], and today there are many compounds used as antihypertensives [16]. In 1979, a research group in the People's Republic of China noted, while examining the antimalarial properties of derivatives of febrifugine, that one compound in clinical trials, changrolin, Fig. 1, was effective as an arrhythmic agent. In 1983, Stout and his research group of the American Hospital Supply Corporation, McGaw Park, Illinois, studied the structure of changrolin for its dissimilarity with currently marketed antiarrhythmics [17–20]; there are also other recent studies about the biological structure–activity relationships [21,22]. And so, we take the phenol and methylpyrrolidine rings as a structural

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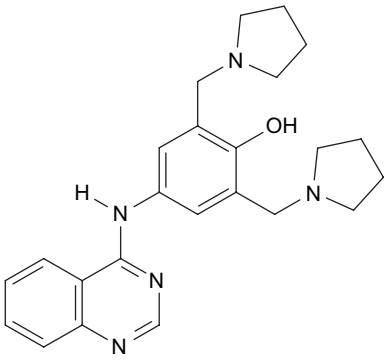


Fig. 1. Changrolin.

requirement to show cardiovascular effects and we change the pyrrolidine rings to methylthiomorpholin rings. In our experience, methylmorpholinphenol and methylpiperidinylphenol derivatives show cardiovascular effects [23] and, in the literature, there is only one report about the cardiovascular effects of methylmorpholinphenols [24], and only two reports about the biological activity of thiomorpholinphenols, which have been reported with antimycobacterial activity [25] and against *Candida* [26]. We now report, as part of the Drug Design in Medicinal Chemistry Program of the UNAM, new methylthiomorpholinphenol compounds with cardiovascular effects, considering that the development of new antihypertensive drugs is justified as there is a need to search for medicines that promote a decrease in blood pressure, such as monotherapy, to achieve a good protection for most hypertensive patients and a reduction in adverse reactions.

2. Experimental

2.1. Chemistry

2.1.1. General methodology of synthesis

Methylthiomorpholinphenol compounds were prepared from phenol derivatives (1 eq.), thiomorpholine (2.1 eq.) and formaldehyde (37%) (2 eq.). They were mixed in a round flask fitted with a condenser. The mixture was irradiated with infrared light using a medicinal infrared lamp (250 W) under solvent-free conditions [27]. The reaction was monitored with TLC using a gradient solvent system (*n*-hexane/ethylacetate) until the reaction was complete. The products were purified by recrystallization and by using silica gel column chromatography using gradient solvents (*n*-hexane/ethylacetate). The temperature of the reaction mixture is in the range of 120 °C–180 °C. Summary of reaction is described in Table 1.

2.1.2. Spectroscopy

*2.1.2.1. 4-Chloro-2-(thiomorpholin-4-ylmethyl)phenol – compound **2** (LQM301).* IR ($CHCl_3$ film) cm^{-1} 3502 (O–H), 3010 (C_{sp^2}–H Ar), 2985 (C_{sp^3}–H). ^1H NMR ($CDCl_3$) δ: 10.56 (1H, s, OH), 7.11 (1H, dd, $J = 8.7$ Hz, 2.7 Hz), 6.94 (1H, d, $J = 2.7$ Hz), 6.74 (1H, d, $J = 8.7$ Hz), 3.65 (2H, s, Ar-CH_2), 2.81 (4H, m, –S–CH_2–), 2.71 (4H, m, –N–CH_2–). ^{13}C NMR ($CDCl_3$) δ: 156 (C), 128.56 (CH), 128.31 (CH), 123.61 (C), 122.11 (C), 117.37 (CH), 61.63 (Ar-CH_2), 54.27 (–S–CH_2–), 27.73 (–N–CH_2–). FAB-MS *m/z* (rel%) (M + 1) 244 (100%), 215, 180, 154.

*2.1.2.2. 4-tert-Butyl-2-(thiomorpholin-4-ylmethyl)phenol – compound **3** (LQM302).* IR ($CHCl_3$ film) cm^{-1} 3456 (O–H), 3197 (C_{sp^2}–H Ar), 2886 (C_{sp^3}–H). ^1H NMR ($CDCl_3$) δ: 10.33 (1H, s, OH), 7.18 (1H, dd, $J = 8.4$ Hz, 2.7 Hz), 6.94 (1H, d, $J = 2.7$ Hz), 6.74 (1H, d, $J = 8.4$ Hz), 3.70 (2H, s, Ar-CH_2), 2.82 (4H, m, –S–CH_2–), 2.71 (4H, m, –N–CH_2–), 1.27 (9H, CH_3). ^{13}C NMR ($CDCl_3$) δ: 155 (C), 141.8 (C), 125.60 (CH), 125.49 (CH), 119.77 (C), 115.47 (CH), 62.51 (Ar-CH_2), 54.36 (–N–CH_2–), 33.84 (C), 31.48 (CH3), 27.79 (–S–CH_2–). FAB-MS *m/z* (M + 1) 266 (80%), 265 (100%), 163 (45%).

*2.1.2.3. 4-tert-Butyl-2,6-bis(thiomorpholin-4-ylmethyl)phenol – compound **4** (LQM303).* IR ($CHCl_3$ film) cm^{-1} 3403 (O–H), 3089 (C_{sp^2}–H Ar), 2986 (C_{sp^3}–H). ^1H NMR ($CDCl_3$) δ: 10.69 (1H, s, OH), 7.09 (2H, s,), 3.71 (4H, s, Ar-CH_2), 2.86 (8H, m, –S–CH_2–), 2.76 (8H, m, –N–CH_2–), 1.27 (9H, CH_3). ^{13}C NMR ($CDCl_3$) δ: 153.6 (C), 141.14 (C), 125.79 (CH), 121.22 (C), 58.81 (Ar-CH_2), 54.42 (–N–CH_2–), 33.78 (C), 31.47 (CH_3), 27.74 (–S–CH_2–). FAB-MS *m/z* (M + 1) 381 (35%), 278 (100%), 175 (50%).

*2.1.2.4. 4,6-Bis(thiomorpholin-4-ylmethyl)1,2,3-benzenetriol – compound **5** (LQM304).* IR ($CHCl_3$ film) cm^{-1} 3429 (O–H), 3065 (C_{sp^2}–H Ar), 2872 (C_{sp^3}–H). ^1H NMR ($CDCl_3$) δ: 8.401 (3H, s, OH), 6.20 (1H, s), 3.61 (4H, s, Ar-CH_2), 2.81 (8H, m, –S–CH_2–), 2.72 (8H, m, –N–CH_2–). ^{13}C NMR ($CDCl_3$) δ: 144.8 (C), 132.51 (C), 118.42 (CH), 111.74 (C), 61.63 (Ar-CH_2), 54.22 (–N–CH_2–), 27.81 (–S–CH_2–). FAB-MS *m/z* (M + 1) 357 (10%), 254 (100%), 102 (91%).

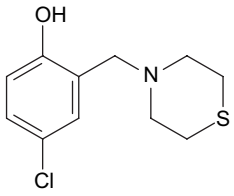
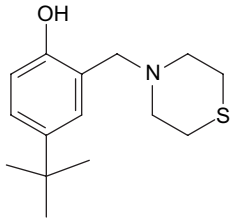
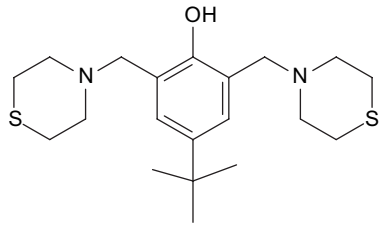
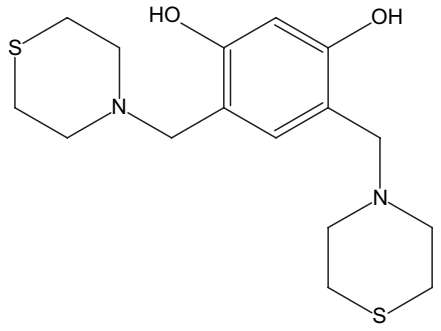
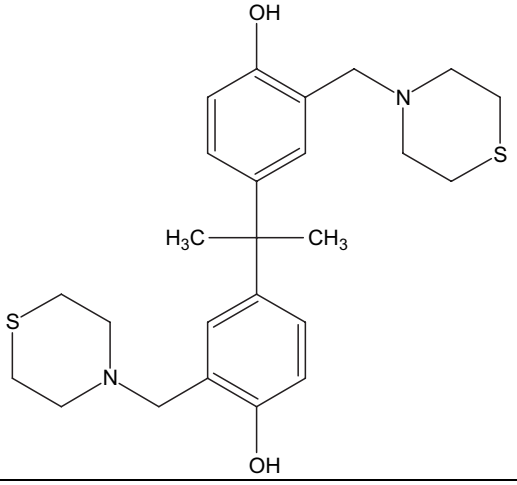
*2.1.2.5. 4-[1-(4-Hydroxy-3-(thiomorpholin-4-ylmethyl)phenyl)-1-methylethyl]-2-(thiomorpholin-4-ylmethyl)phenol – compound **6** (LQM305).* IR ($CHCl_3$ film) cm^{-1} 3473 (O–H), 3034 (C_{sp^2}–H Ar), 2895 (C_{sp^3}–H). ^1H NMR ($CDCl_3$) δ:10.42 (2H, s, OH), 7.0 (1H, dd, $J = 2.4$ Hz, 8.4 Hz), 6.79 (1H, d, $J = 2.4$ Hz), 6.70 (1H, d, $J = 8.4$ Hz), 3.64 (4H, s, Ar-CH_2), 2.80 (8H, m, –S–CH_2–), 2.74 (8H, m, –N–CH_2–), 1.57 (3H, s, CH_3). ^{13}C NMR ($CDCl_3$) δ: 155.09 (C), 141.70 (C), 127.06 (CH), 126.85 (CH), 119.78 (C), 115.30 (CH), 62.48 (Ar-CH_2), 54.35 (–N–CH_2–), 41.38 (C), 31.07 (CH_3), 27.83 (–S–CH_2–). FAB-MS *m/z* (M + 1) 459 (40%), 238 (100%), 237 (32%).

3. Biological activity

3.1. Materials and methods

We compared methylthiomorpholinphenol derivatives with captopril (angiotensin-converting enzyme, ACE), losartan (AT_1 receptor antagonist) and omapatrilat (neutral

Table 1
Experimental data of compounds **2**–**6**

Compound	Structure	MP (°C)	Yield (%)	Reaction time (min)
2 (LQM301)		127–129	60	7
3 (LQM302)		85–87	70	15
4 (LQM303)		95–97	80	20
5 (LQM304)		193–195	87	11
6 (LQM305)		163–165	50	11

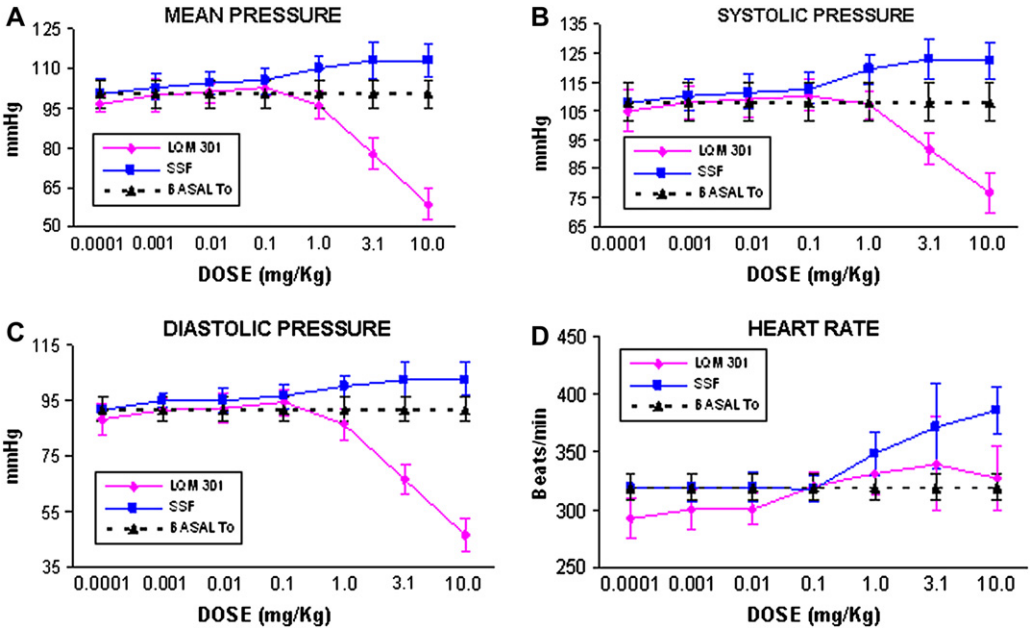


Fig. 2. Dose–effect curve of compound LQM301 in anesthetized male Wistar rat. The hypotensive effect of compound LQM301 was significant at doses of 1.0, 3.1 and 10.0 mg/kg. This compound significantly decreased heart rate at doses of 0.0001, 0.001 and 0.01 mg/kg.

endopeptidase inhibitor and ACE). The results of this work are shown with a dose–effect curve for each compound. An invasive arterial pressure model was used to determine the compounds' hypotensive effect.

3.1.1. Reagents

Captopril SIGMA, omapatrilat and losartan were provided by Bristol-Meyers-Squibb, and Merck Sharp & Dome Laboratories, respectively. All the compounds were synthesized in Laboratorio de Quimica Medicinal, FESC-UNAM. We used 12 weeks old male Wistar normotensive and hypertensive rats, weighing between 280 and 320 g. The rats were maintained on a 12 h light/12 h dark cycle and had free access to standard rat chow and drinking water. The experimental procedures were approved by the Institutional Animal Care and Use Committee of our institution.

3.2. Determination of hypotensive effect

In order to establish the biological effect of methylthiomorpholin derivatives, we chose five compounds that were labeled LQM301–LQM305 and solubilized using a mixture of 0.1 mL

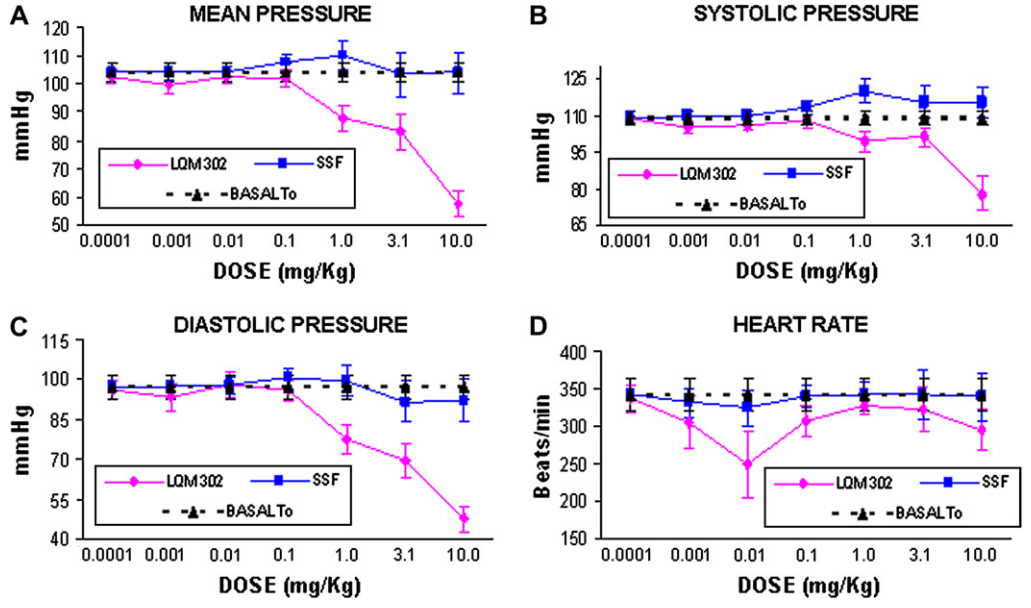


Fig. 3. Dose–effect curve of compound LQM302 in anesthetized male Wistar rat. The hypotensive effect of compound LQM302 turned out to be significant at doses of 0.001, 0.1, 1.0, 3.1 and 10.0 mg/kg. This compound significantly decreased heart rate at doses of 0.01, 0.1, 1.0, and 10.0 mg/kg.

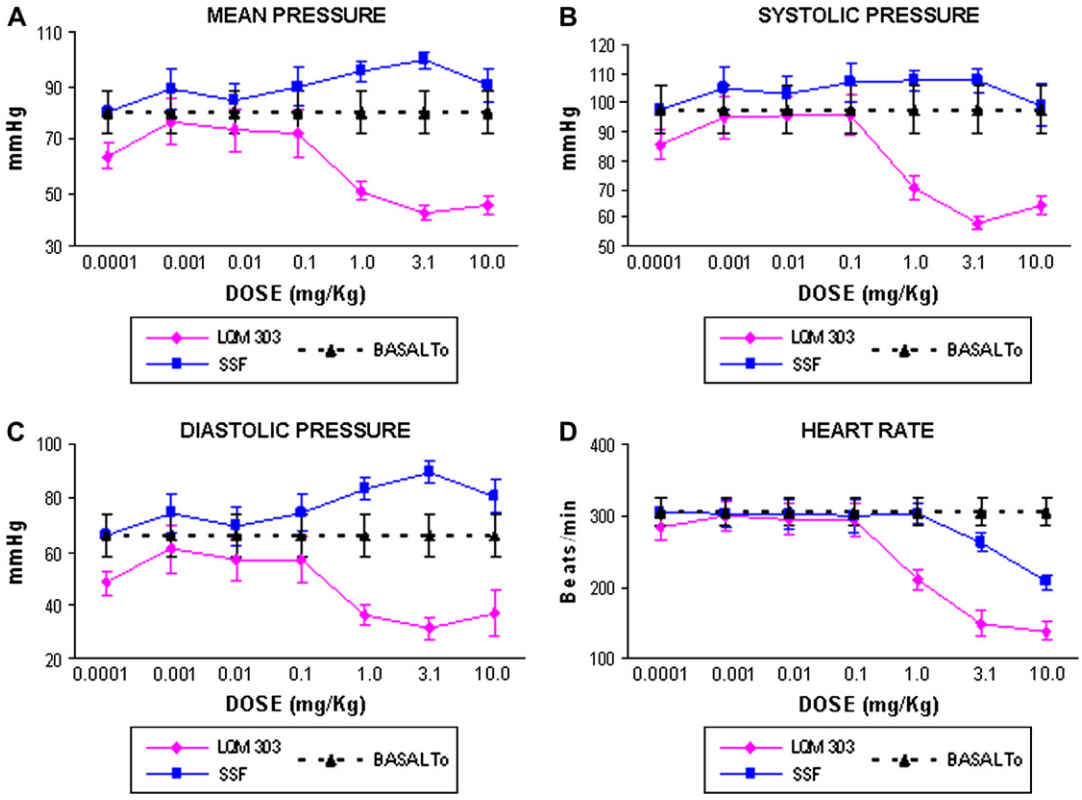


Fig. 4. Dose–effect curve of compound LQM303 in anesthetized male Wistar rat. The hypotensive effect of compound LQM303 was significant at all doses. This compound significantly decreased heart rate at doses of 1.0, 3.1 and 10.0 mg/kg.

of chlorhydric acid solution (0.000592 M) and 0.9 mL physiological saline solution, obtaining a final volume of 1.0 mL.

Subsequently, the rats were anesthetized with sodium pentobarbital (45 mg/kg, ip), a tracheotomy was performed, the right carotid artery was dissected and cannulated with a catheter (PE50) connected to a pressure transducer which was connected to a Digi-Med Blood Pressure Analyzer to monitor systolic (SAP), diastolic (DAP) and mean arterial pressure

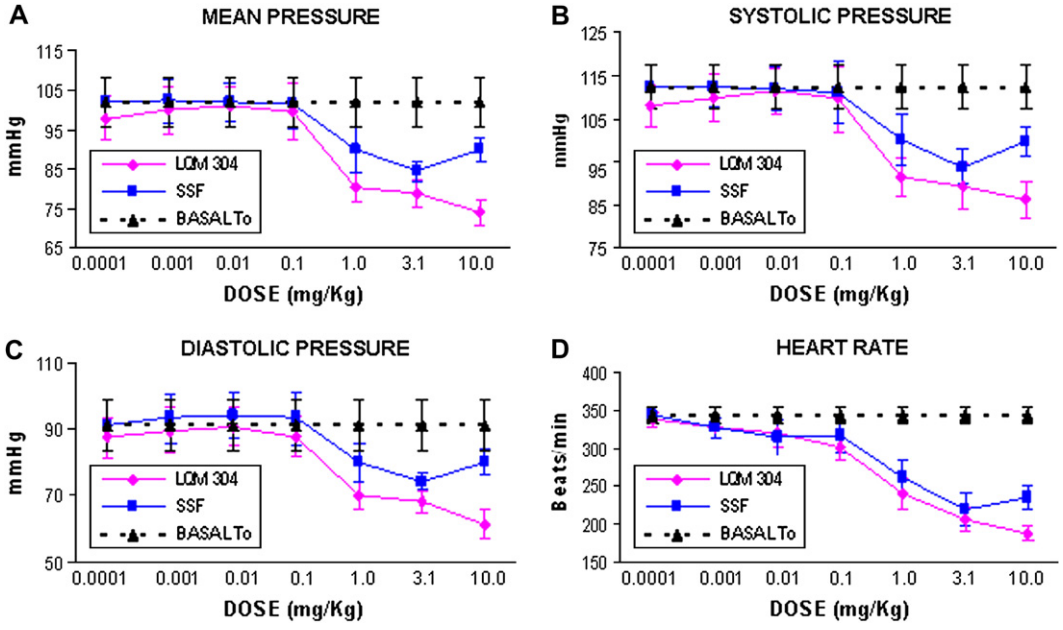


Fig. 5. Dose–effect curve of compound LQM304 in anesthetized male Wistar rat. The hypotensive effect of compound LQM304 decreased significantly at doses of 1.0, 3.1 and 10.0 mg/kg. This compound significantly decreased heart rate at doses of 0.01, 0.1, 1.0, 3.1 and 10.0 mg/kg.

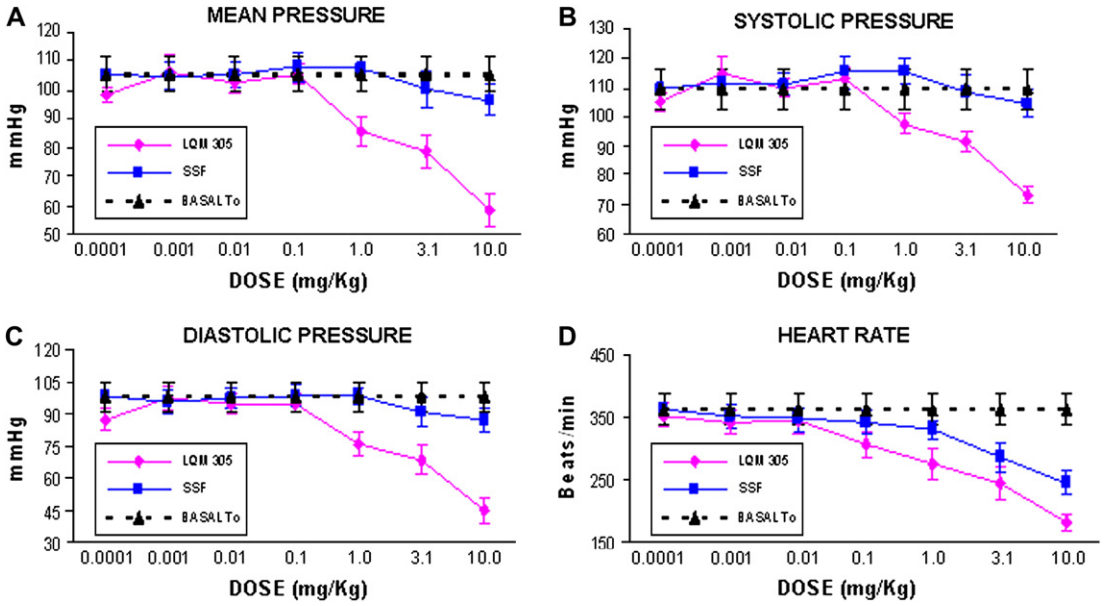


Fig. 6. Dose–effect curve of compound LQM305 in anesthetized male Wistar rat. The hypotensive effect of compound LQM305 was significant at doses of 0.0001, 1.0, 3.1 and 10.0 mg/kg. This compound significantly decreased heart rate at doses of 0.01, 0.1, 1.0, 3.1 and 10.0 mg/kg.

(MAP), and heart rate. Registers were obtained using the program DMSI-2000_1.

In the first stage, the biological activity of the compounds was established using anesthetized normotensive rats with the objective of establishing in a quick and practical manner the biological effect of captopril, losartan, omapatrilat and the LQM series compounds; in the second experimental stage, the conscious spontaneous hypertensive rat model was applied to

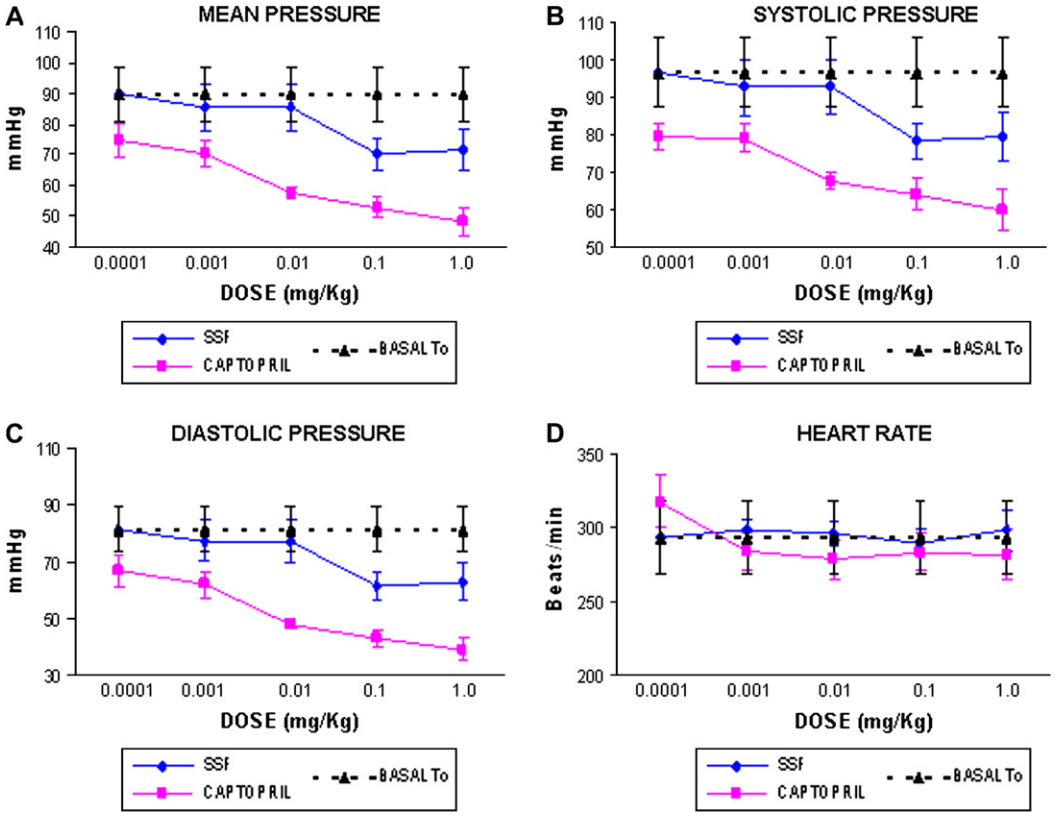


Fig. 7. Dose–effect curve of captopril (angiotensin-converting enzyme inhibitor) in anesthetized male Wistar rat. The hypotensive effect of the captopril was significant at all doses. Its effect on heart rate was not significant.

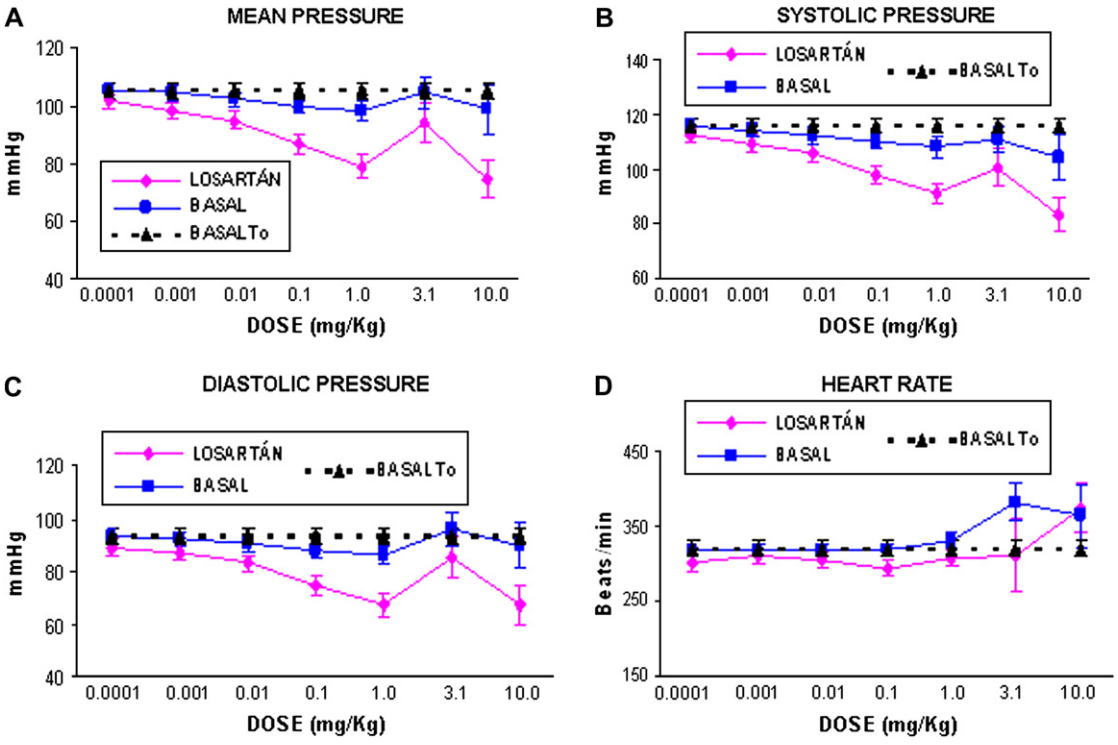


Fig. 8. Dose–effect curve of losartan (AT1 receptor antagonist) in anesthetized male Wistar rat. The hypotensive effect of the Losartan was significant at all doses. Its effect on heart rate was not significant.

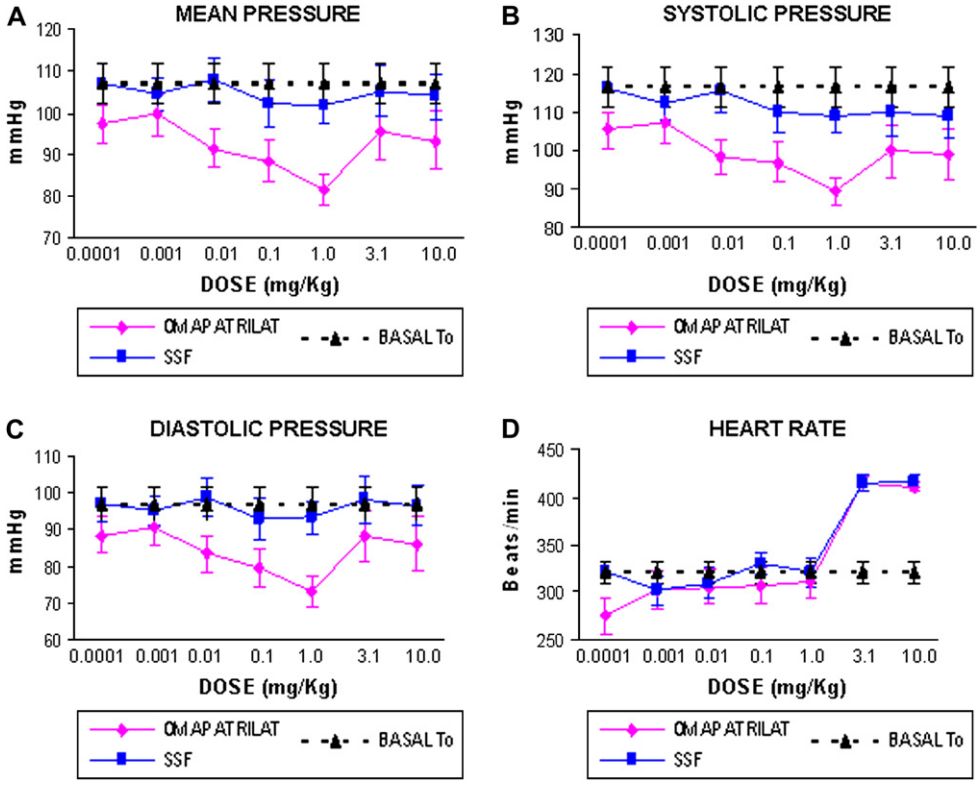


Fig. 9. Dose–effect curve of the reference drug omapatrilat (neutral endopeptidase inhibitor) in anesthetized male Wistar rat. The hypotensive effect of omapatrilat was significant at all doses. The effect on heart rate was not significant.

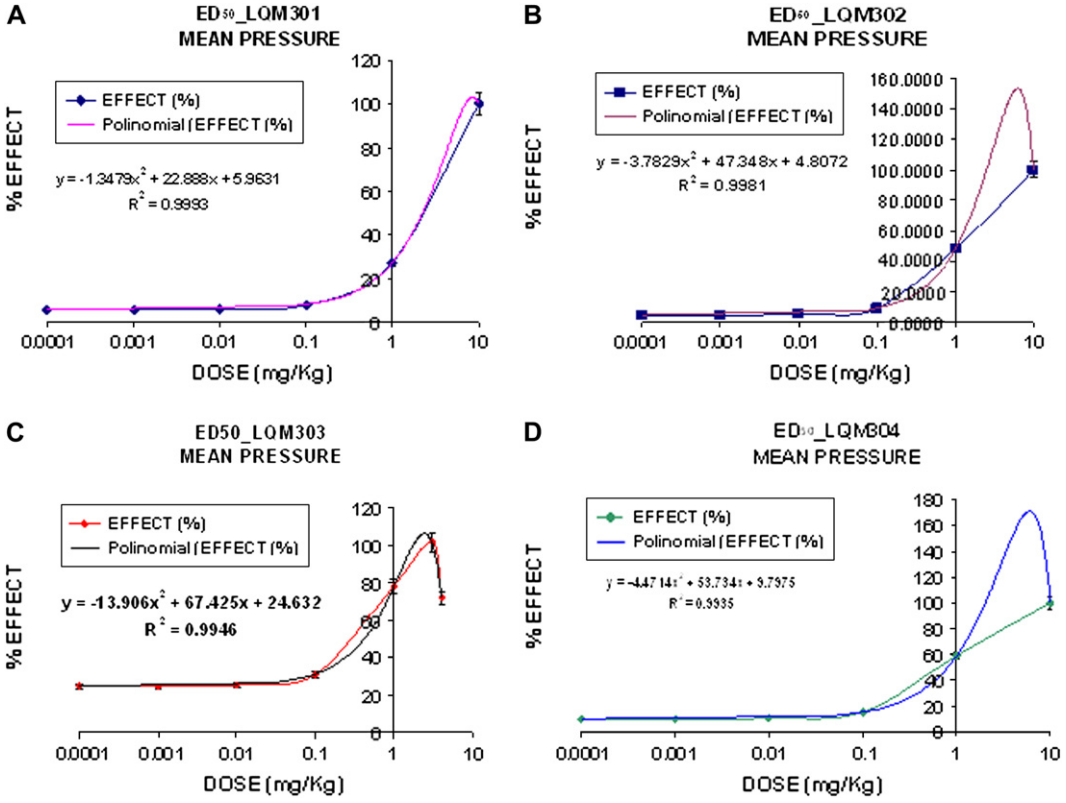


Fig. 10. Dose–effect curves (A–D) of compounds LQM301, LQM302, LQM303 and LQM304 in normotensive anesthetized rat. Each graph shows the mathematical model with its ED_{50} calculated.

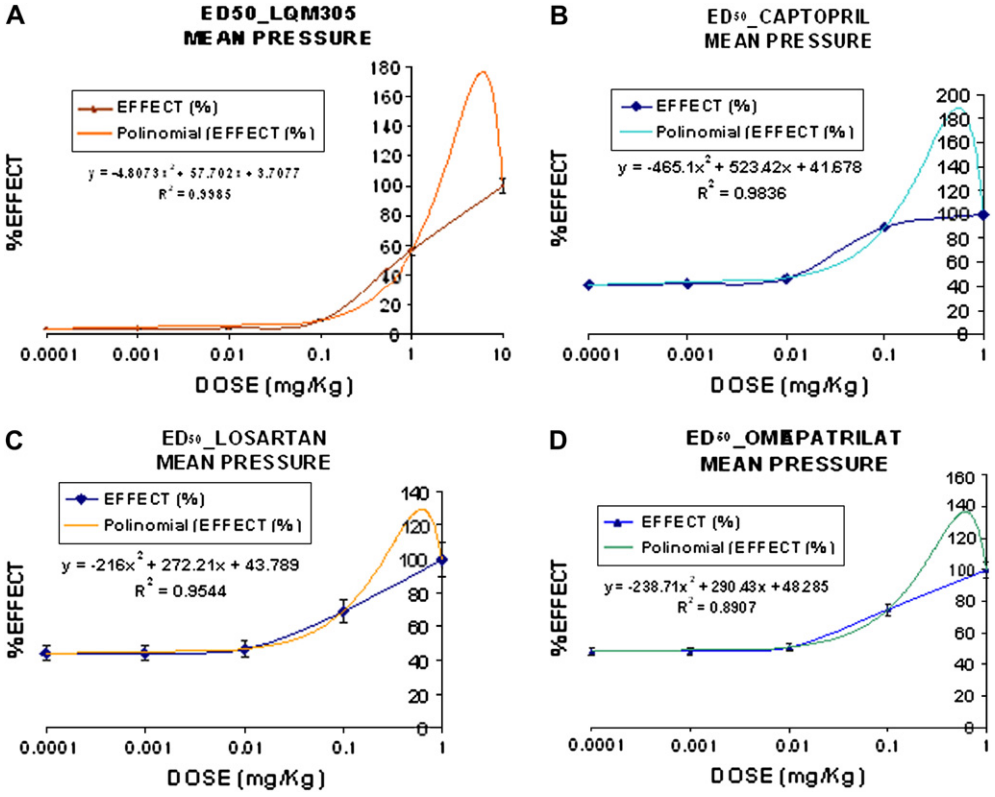


Fig. 11. Dose–effect curves (A–D) of compounds LQM305, captopril, losartan and omapatrilat in normotensive anesthetized rat. Each graph shows the mathematical model with its ED_{50} calculated.

Table 2
Mean effective dose of synthesized compounds and positive controls

Compound	LQM301	LQM302	LQM303	LQM304	LQM305	Captopril	Losartan	Omapatrilat
ED_{50} (mg/kg)	2.2122	1.0410	0.4111	0.3631	1.4091	0.00062	0.02815	0.0058

better evaluate hypotension effect, recording systolic and diastolic arterial pressure and heart rate.

The biological activity of each compound (LQM301–LQM305) was determined by means of a dose–effect curve (0.0001, 0.001, 0.01, 0.1, 1.0, 3.1 and 10 mg/kg IV, $n = 5$). Each compound was administered through the femoral vein. The captopril, omapatrilat and losartan dose–effect curves were determined (0.0001, 0.001, 0.01, 0.1 and 1.0 mg/kg IV, $n = 5$) with the purpose of comparing mean effective dose (ED_{50}) with experimental compounds. The doses were administered every five minutes allowing for the hypotensive effect to take place.

4. Statistical analysis

The data were analyzed using Excel and Statistica Software, the values appear as average ($n = 5$) ± SEM. The comparison of the results of both curves was conducted by Student's *t*-test and ANOVA $^*P < 0.05$.

5. Results and discussion

First step: The arterial pressure model was used to determine the hypotensive effect of all compounds. The results are shown in Figs. 2–9. The values are plotted as follows: A: mean arterial pressure (MAP) vs. dose; B: systolic arterial pressure (SAP) vs. dose; C: diastolic arterial pressure (DAP) vs. dose.

The black dotted line is the extension of the initial value of the arterial pressure (Basal t_0); the blue line[1] indicates pressure values before the addition of each dose (Basal); and the pink line indicates pressure values after compound addition (effect). For the case of losartan effect, in Fig. 8 we can observe its hypotensive effect, because there is a difference between the basal pressure and the final pressure after the administration of the drug of reference. The change in mean arterial pressure after administering 10.0 mg/kg of losartan is 41.31 mmHg; for the systolic arterial pressure, it is 43.62 mmHg; for the diastolic arterial pressure it is 45.64 mmHg, therefore, losartan does exhibit hypotensive effect in the experiment with the anesthetized rat model.

Dose–effect curves were used to calculate the ED_{50} of compounds LQM301–LQM305, and captopril, losartan and omapatrilat. The results are shown in Figs. 10 and 11. In these figures, we can also observe the mathematical models used for the ED_{50} calculations.

[1] For interpretation of the references to colour in text, the reader is referred to the web version of this article.

LQM301, LQM302, LQM304 and LQM305 compounds showed a gradual effect on systolic, diastolic and mean arterial pressure, starting at 0.1 mg/kg (Figs. 2–6), whereas captopril, losartan and omapatrilat, showed the same effect at 0.001 mg/kg. Regarding heart rate, LQM302–LQM305, reduced it gradually and significantly, whereas captopril, losartan and omapatrilat did not show a significant effect on this variable.

Table 2 shows ED_{50} in mg/kg of LQM301–LQM305, and the three reference drugs. Captopril showed the lowest ED_{50} among all the compounds. On the other hand, it was determined that captopril's ED_{50} is three times lower than LQM303, LQM304 and four times lower than LQM301, LQM302 and LQM305.

Figs. 12–15 show efficacy and potency on systolic, diastolic and mean arterial pressure and on heart rate of all tested compounds as well as captopril, losartan and omapatrilat. Captopril was the most potent and effective, LQM303 exhibited some similarity with captopril in diastolic and mean arterial pressure, but they differed in systolic arterial pressure and potency, as shown in Figs. 13 and 14. LQM301, LQM302, LQM304 and LQM305 exhibited similar effects to losartan and omapatrilat. This observation was made based only on the behavioral tendencies of the curves representing the dose–effect % relationship, Figs. 13 and 14.

On the other hand, captopril, losartan and omapatrilat did not have a significant effect on heart rate, whereas LQM303–LQM305 induced a significant decrease in heart

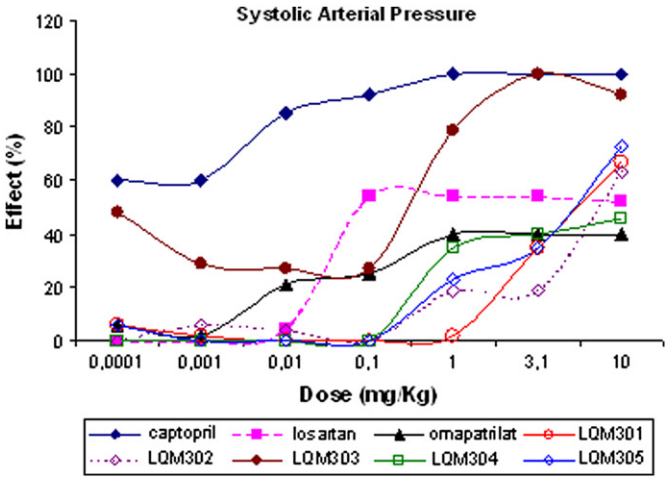


Fig. 12. Dose–effect (%) curves of five thiomorpholine compounds, captopril, losartan and omapatrilat (antihypertensive drugs). The graph shows their efficacy and potency on systolic arterial pressure. The captopril was more effective in reducing systolic arterial pressure than LQM303 > losartan > omapatrilat = LQM304 > LQM305 = LQM302 > LQM301 at 1 mg/kg.

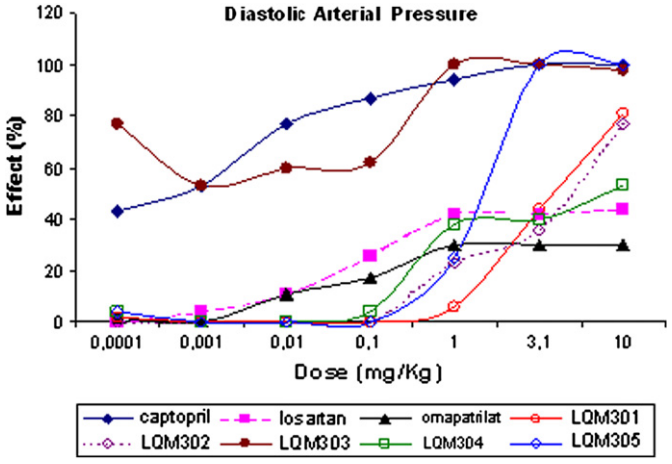


Fig. 13. Dose–effect (%) curves of five thiomorpholine compounds, captopril, losartan and omapatrilat (antihypertensive drugs). The graph shows their efficacy and potency on diastolic arterial pressure. Captopril was more effective in reducing diastolic arterial pressure than LQM303 > losartan = LQM304 > omapatrilat > LQM302 > LQM305 > LQM301 at 1 mg/kg.

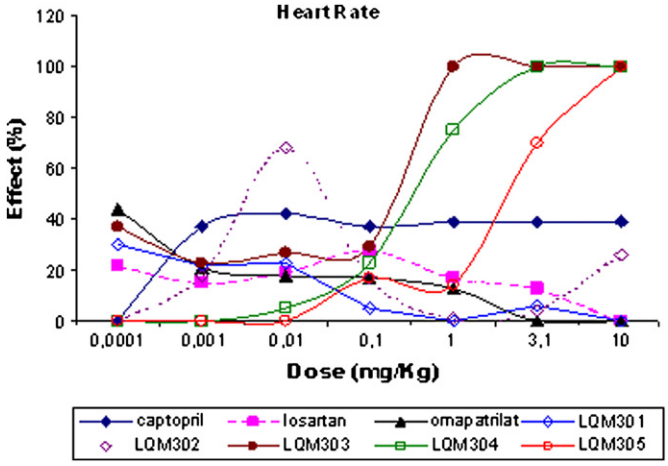


Fig. 15. Dose–effect (%) curves of five thiomorpholine compounds, captopril, losartan and omapatrilat (antihypertensive drugs). The graph shows their efficacy and potency on heart rate. LQM303 was more effectivein reducing heart rate than LQM304 > captopril > losartan = LQM305 = omapatrilat > LQM301 = LQM302 at 1 mg/kg.

rate, this may occur because LQM compounds have a depressing effect on the Sino auricular node, which controls the cardiac rhythm, therefore decreasing heart rate.

Second step: In the second experimental stage, the conscious hypertensive rat model was used to better evaluate hypotensive effect, recording systolic and diastolic arterial pressure, and heart rate.

The administration of the reference antihypertensives and the compounds of the experimental series was in a dose of 1.0 mg/kg orally. The experimental results show a decrease in systolic and diastolic pressures and heart rate.

Table 3 shows the results obtained in the maximum decrease of systolic and diastolic pressure and heart rate for each compound of the experimental series and of the reference antihypertensives during the 2.0 h that the experiment lasted, for the spontaneous hypertensive rat experimental model. Figs. 16–22 show the graphs of the tendencies in these experiments.

Based on the findings of the experiment with the conscious spontaneous hypertensive rat model, where the change in systolic arterial pressure was of 82.3 mmHg, 80 min after administering 1.0 mg/kg of losartan, and change in diastolic pressure was of 66.35 mmHg, also 80 min after administering losartan, it was confirmed that losartan exhibits the hypotensive effect that has been known until now, as can be observed in Table 3.

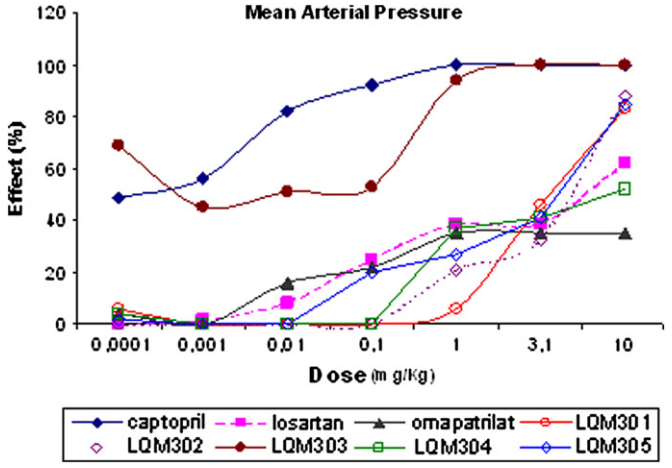


Fig. 14. Dose–effect (%) curves of five thiomorpholine compounds, captopril, losartan and omapatrilat (antihypertensive drugs). The graph shows their efficacy and potency on mean arterial pressure. Captopril and LQM303 were more effective in reducing mean arterial pressure than losartan = omapatrilat = LQM304 > LQM305 > LQM302 > LQM301 at 1 mg/kg.

6. Conclusion

The synthesis of the experimental LQM compounds is very simple, and they are easy to purify. The results obtained suggest that LQM compounds have a moderate hypotensive effect and could be used for chronic patients.

In this research, some of these compounds were compared with captopril (ACE inhibitors), losartan (antagonist of AT1

Table 3
The results of maximum decrease in systolic pressure, diastolic pressure, and heart rate, of the compounds that form the experimental series and of the antihypertensive compounds used as reference

Compound	ΔP_{max} systolic/ time (mmHg/min)	ΔP_{max} diastolic/ time (mmHg/min)	Δ_{max} heart rate/ time (beat/min/min)
LQM301	73.9/100	46.7/100	72.2/110
LQM302	71.47/100	31.8/60	147.88/100
LQM303	95.755/90	48.225/90	160.448/100
LQM304	45.6/60	29.2/60	92.5/120
LQM305	28.086/60	12.5875/60	106.566/120
CAPTOPRIL	42.0/50	36.45/100	52.04/70
LOSARTAN	82.3/80	66.35/80	38.37/80

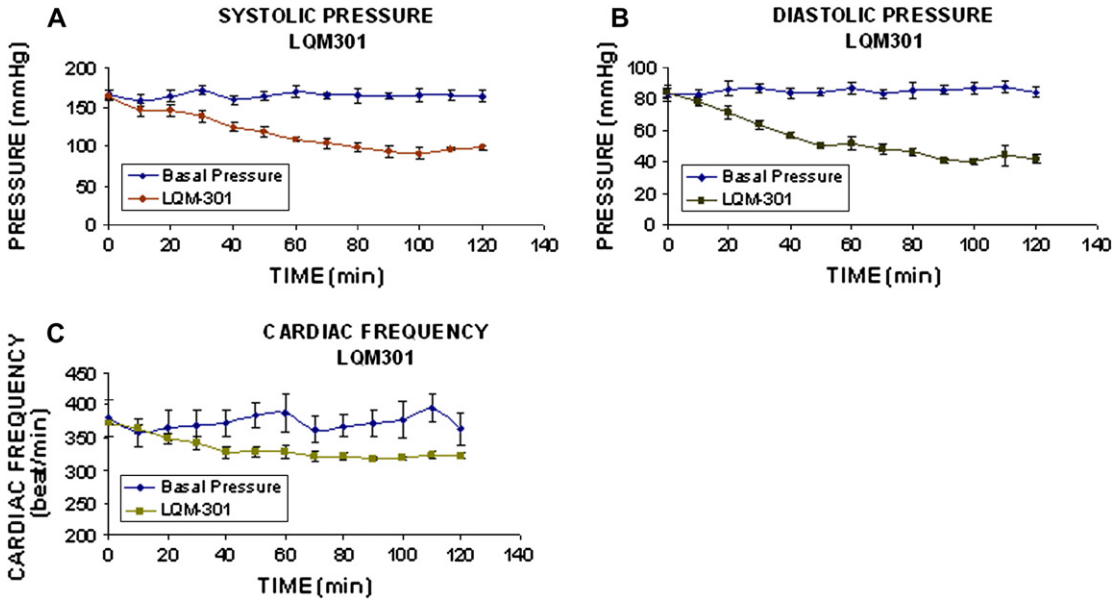


Fig. 16. Curve effect vs. time of compound LQM301 (A–C). Oral administration: 1.0 mg/kg dose. Spontaneous hypertensive rat. Each graph shows experimental conditions and maximum decrease. (A) Variation of systolic pressure vs. time. The maximum decrease was 73.9 mmHg, 100 min after administration. (B) Variation of diastolic pressure vs. time. The maximum decrease was 46.7 mmHg. (C) Variation of cardiac frequency vs. time. The maximum decrease was 72.2 beats/min.

receptors) and omapatrilat (inhibitor of neutral endopeptidase and ACE) in normotensive and hypertensive rat. The results obtained in the dose–effect curves in the arterial pressure model show two important candidate compounds (LQM303 and LQM304), since these have a lower ED_{50} than the other synthesized compounds. Also, the dose–effect curve of LQM303 shows a similarity in the tendency to decrease systolic, diastolic and mean arterial pressure. Therefore, it would be important to analyze the influence of the functional groups of this molecule (LQM303) in order to determine which of

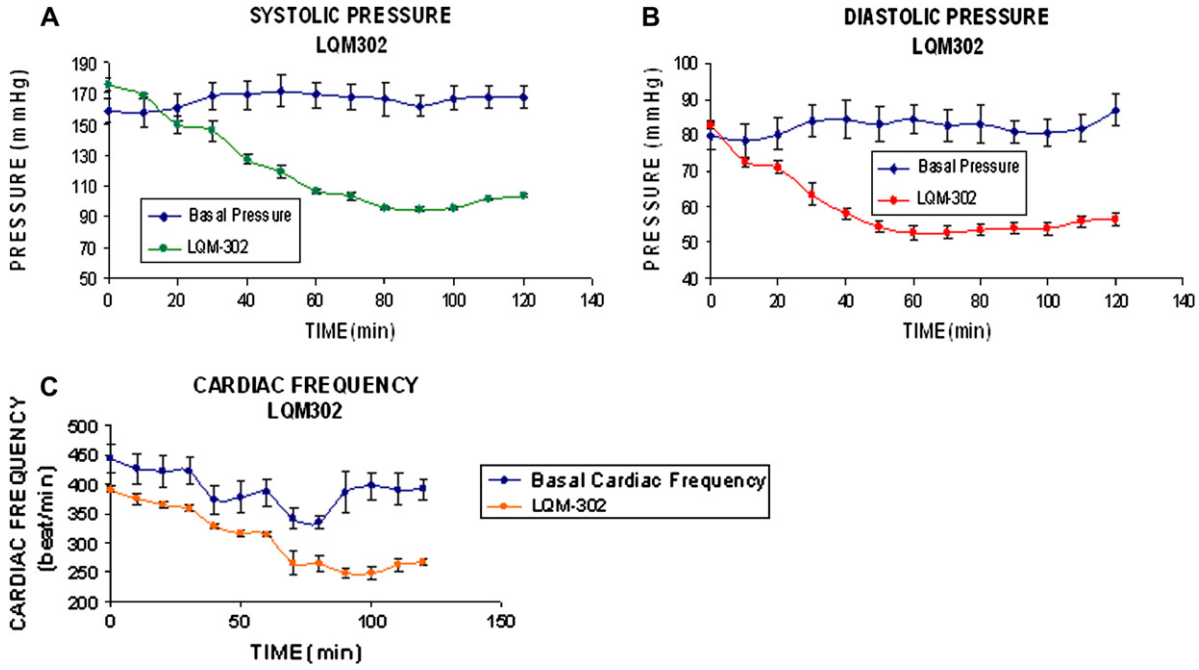


Fig. 17. Curve effect vs. time of compound LQM302 (A–C). Oral administration, 1.0 mg/kg dose. Spontaneous hypertensive rat. Each graph shows experimental conditions and maximum decrease. (A) Variation of systolic pressure vs. time. The maximum decrease was 71.4 mmHg, 100 min after administration. (B) Variation of diastolic pressure vs. time. The maximum decrease was 31.8 mmHg, 60 min after administration. (C) Variation of heart rate vs. time. The maximum decrease was 147.88 beats/min, 100 min after administration.

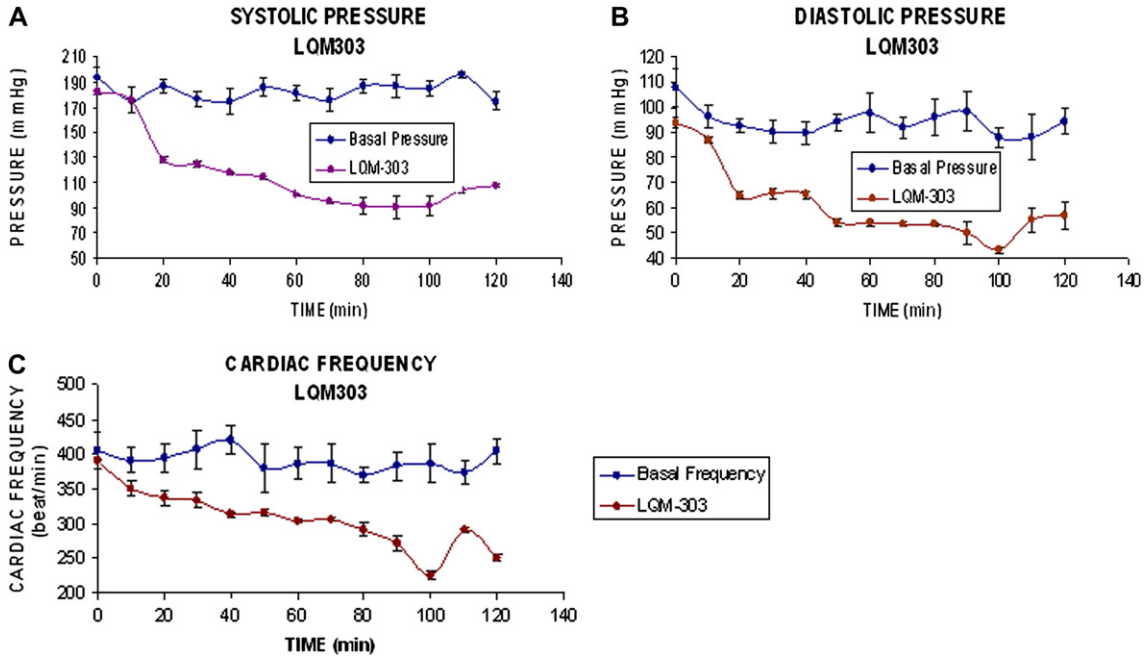


Fig. 18. Curve effect vs. time of compound LQM303 (A–C). Oral administration, 1.0 mg/kg dose. Spontaneous hypertensive rat. Each graph shows experimental conditions and maximum decrease. (A) Variation of systolic pressure vs. time. The maximum decrease was 95.755 mmHg, 90 min after administration. (B) Variation of diastolic pressure vs. time. The maximum decrease was 48.225 mmHg, 90 min after administration. (C) Variation of heart rate vs. time. The maximum decrease was 160.448 beats/min, 100 min after administration.

them is involved in the pharmacological activity. In addition, we determined that some compounds exist that not only reduce arterial pressure but also reduce heart rate. This could be relevant because, in these compounds, we can find the groups that influence the decrease in cardiac rhythm and then test them in a cardiac arrhythmia model. On account of their characteristics, they are prime candidates to exert possible anti-arrhythmic activity.

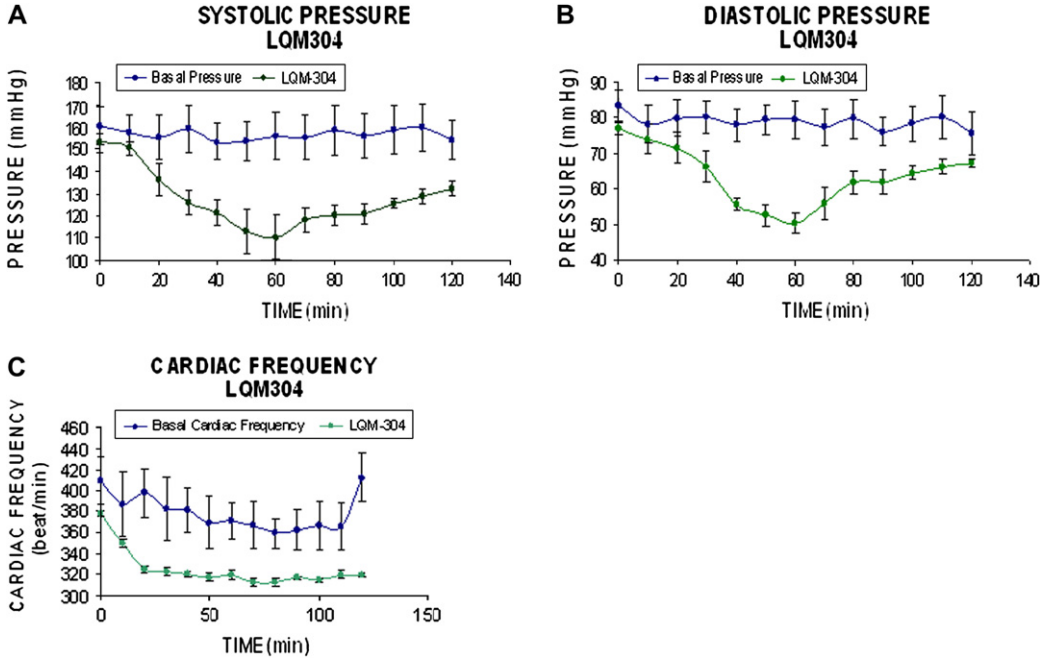


Fig. 19. Curve effect vs. time of compound LQM304 (A–C). Oral administration, 1.0 mg/kg dose. Spontaneous hypertensive rat. Each graph shows experimental conditions and maximum decrease. (A) Variation of systolic pressure vs. time. The maximum decrease was 45.6 mmHg, 60 min after administration. (B) Variation of diastolic pressure vs. time. The maximum decrease was 29.2 mmHg, 60 min after administration. (C) Variation of heart rate vs. time. The maximum decrease was 92.5 beats/min, 120 min after administration.

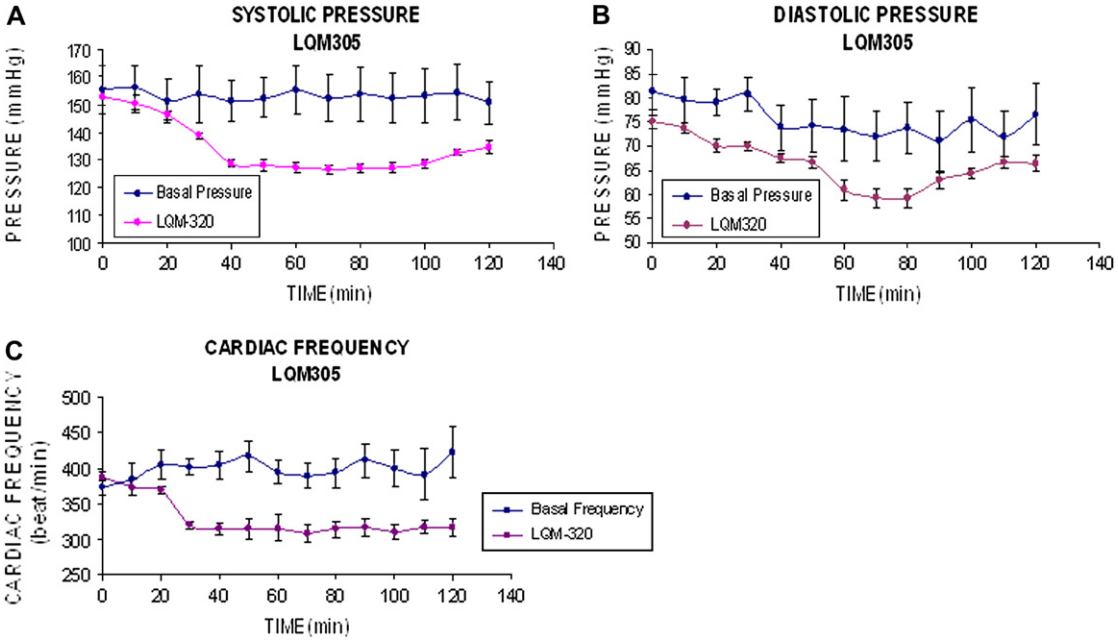


Fig. 20. Curve effect vs. time of compound LQM305 (A–C). Oral administration, 1.0 mg/kg dose. Spontaneous hypertensive rat. Each graph shows experimental conditions and maximum decrease. (A) Variation of systolic pressure vs. time. The maximum decrease was 28.086 mmHg, 60 min after administration. (B) Variation of diastolic pressure vs. time. The maximum decrease was 12.558 mmHg, 60 min after administration. (C) Variation of heart rate vs. time. The maximum decrease was 160.566 beats/min, 120 min after administration.

Considering the ED_{50} of each tested compound, the scale of effective pharmacological effect would be captopril > omapatrilat > losartan > LQM304 > LQM303 > LQM302 > LQM305 > LQM301.

The hypotensive effect of omapatrilat, losartan and captopril showed a gradual relationship of hypotensive effect vs. dose (mg/kg IV) in this model; whereas LQM compounds decrease blood pressure between 5 and 10 mmHg at a dose of

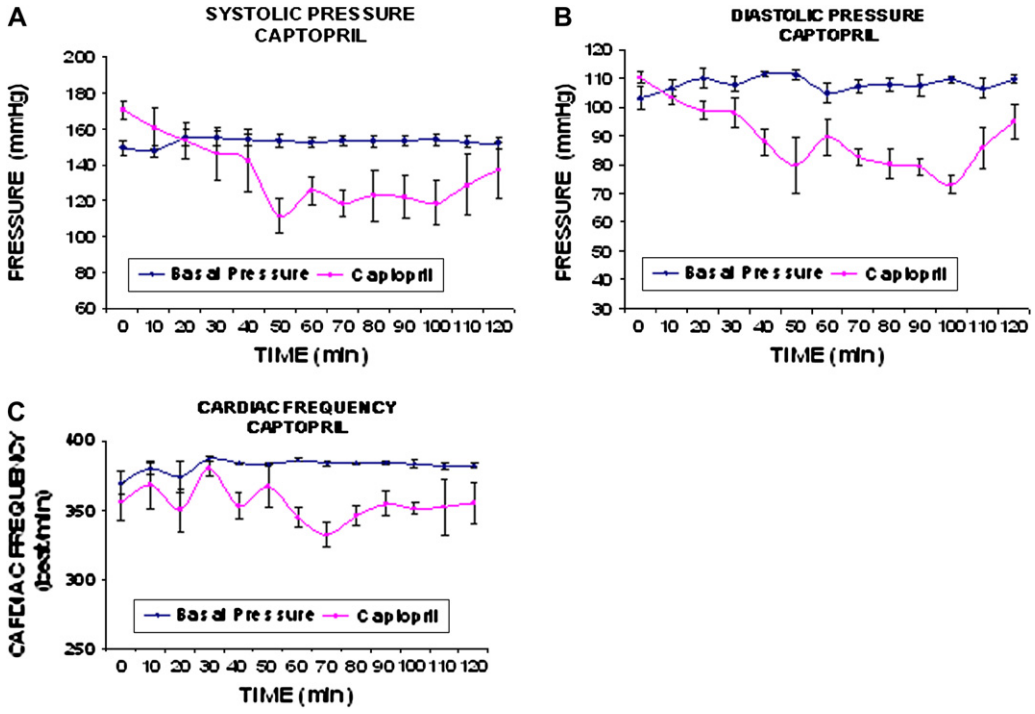


Fig. 21. Curve effect vs. time of compound captopril (A–C). Oral administration, 1.0 mg/kg dose. Spontaneous hypertensive rat. Each graph shows experimental conditions and maximum decrease. (A) Variation of systolic pressure vs. time. The maximum decrease was 42.0 mmHg, 50 min after administration. (B) Variation of diastolic pressure vs. time. The maximum decrease was 36.45 mmHg, 100 min after administration. (C) Variation of cardiac frequency vs. time. The maximum decrease was 52.04 beats/min, 70 min after administration.

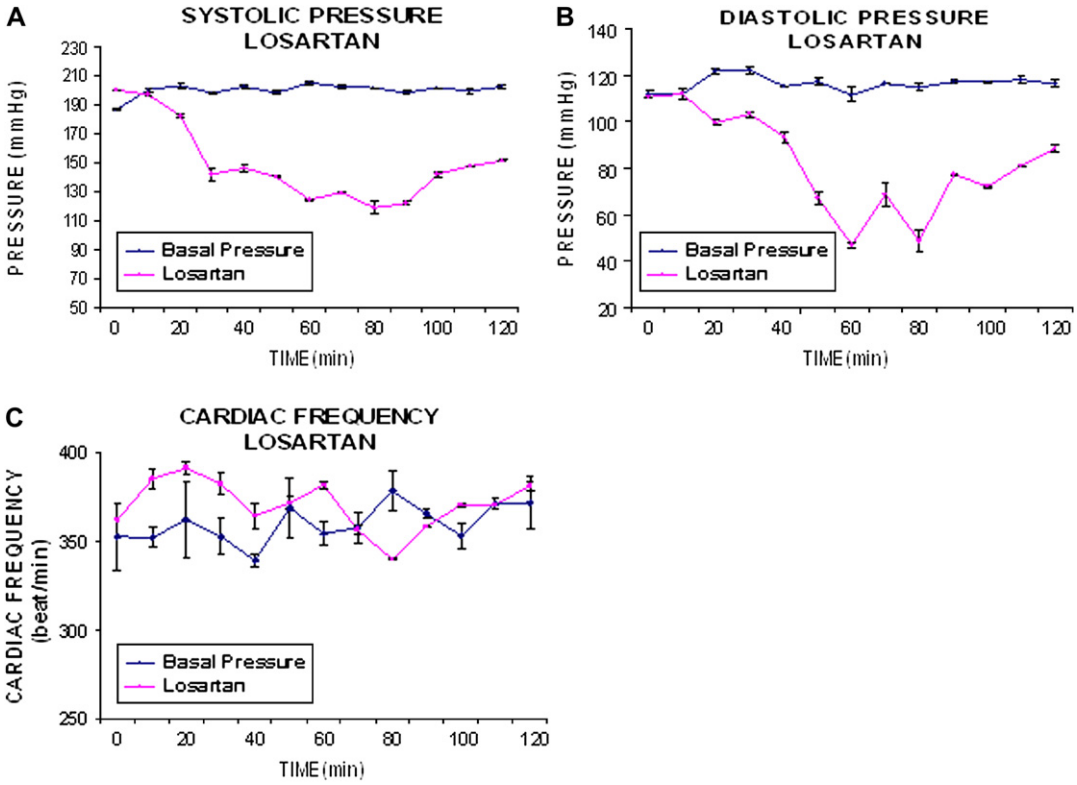


Fig. 22. Curve effect vs. time of compound losartan (A–C). Oral administration, 1.0 mg/kg dose. Spontaneous hypertensive rat. Each graph shows experimental conditions and maximum decrease. (A) Variation of systolic pressure vs. time. The maximum decrease was 82.3 mmHg, 80 min after administration. (B) Variation of diastolic pressure vs. time. The maximum decrease was 66.35 mmHg, 80 min after administration. (C) Variation of cardiac frequency vs. time. The maximum decrease was 38.37 beats/min, 80 min after administration.

0.001, 0.01 and 0.01 (mg/kg IV) after the hypotensive effect becomes gradual, so LQM compounds do not reduce blood pressure in a sudden manner as in the case of vasodilatations and β-adrenergic blockers, angiotensin-converting enzyme inhibitors (ACE), receptors AT1 antagonists, and neutral endopeptidase inhibitors.

Finally, we can say that the development of new antihypertensive drugs is justified because it is necessary to search for drugs that are able to reduce blood pressure, like monotherapy, in order to achieve good protection for the majority of hypertensive patients and a reduction of adverse reactions.

As shown in Table 2, the compound that exhibited the highest antihypertensive activity in the conscious spontaneous hypertensive rat model was LQM303, confirming what has been observed in the anesthetized rat model.

The magnitude of the effect of the compounds on systolic pressure in decreasing order was LQM303 > losartan > LQM301 > LQM302 > LQM304 > captopril > LQM305.

For diastolic pressure, the magnitude of the effect in decreasing order was losartan > LQM303 > LQM301 > captopril > LQM302 > LQM304 > LQM305.

And for heart rate, it was: LQM303 > LQM302 > LQM305 > LQM304 > LQM301 > captopril > losartan.

Finally, we observed that of the studied compounds, LQM303 exhibits the best decreasing effect in both systolic and diastolic pressure; it also exhibits the best heart rate-decreasing effect.

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