# Hypotensive effect of alpha-lipoic acid after a single administration in rats

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## Abstract

**Objective:** The effect of alpha-lipoic acid on blood pressure was investigated many times in chronic studies, but there are no studies on the effect of this compound after a single administration. Alpha-lipoic acid is a drug used in diabetic neuropathy, often in obese patients, to treat hypertension. Therefore, knowledge of the potential antihypertensive effect of alpha-lipoic acid even after a single dose and possibly too much pressure reduction is interesting and useful.

**Methods:** The mechanism of the hypotensive effect of alpha-lipoic acid was examined in normotensive rats *in vivo* after a single intraperitoneal administration, blood pressure in the left carotid artery of the rats was measured prior to the administration of the compounds (alpha-lipoic acid and/or glibenclamide) and 80 min thereafter.

**Results:** Alpha-lipoic acid at a dosage of 50 mg/kg b.w. i.p. significantly decreased the blood pressure from the 50$^{th}$ min after drug administration. This cardiovascular effect of this compound was reversed by glibenclamide, a selective $K_{ATP}$ blocker. Glibenclamide alone at this dose did not significantly affect the blood pressure. Statistical significance was evaluated using two-way ANOVA.

**Conclusion:** This suggests that alpha-lipoic acid affects ATP-dependent potassium channels. It is possible that this is an indirect effect of hydrogen sulfide because alpha-lipoic acid can increase its concentration. The results obtained in this study are very important because the patients taking alpha-lipoic acid may be treated for co-existing hypertension. Therefore, the possibility of blood pressure lowering by alpha-lipoic acid should be taken into account, although it does not lead to excessive orthostatic hypotension. *(Anatol J Cardiol 2016; 16: 306-9)*

**Keywords:** alpha-lipoic acid, blood pressure, hydrogen sulfide, $K_{ATP}$ channels

## Introduction

Alpha-lipoic acid has gained considerable interest as an antioxidant. It is readily absorbed from the diet, transported to tissues, and taken up by cells, where a large proportion of it is rapidly converted to its reduced form, dihydrolipoic acid (1). It cooperates with rhodanese in the sulfane sulfur metabolism and hydrogen sulfide ($H_2S$) release. Sulfane sulfur is an essential storage form of $H_2S$ in the body (2, 3). Therefore, some of the effects observed after alpha-lipoic acid administration can be associated with the action of $H_2S$.

In the cardiovascular system, endogenous $H_2S$ has important physiological effects including vascular tone regulation (4). $H_2S$ induces hypotension *in vivo* (5) and vasodilatation *in vitro* (6) by opening $K_{ATP}$ channels in vascular smooth muscle cells (5-7). $H_2S$ is endogenously produced by the heart tissue as a physiological regulator of cardiac function (8). Glibenclamide, a $K_{ATP}$ channel antagonist, attenuates the hypotensive effect of $H_2S$ *in vivo* and vasodilatation *in vitro* (3, 7).

The effect of alpha-lipoic acid on blood pressure was investigated many times in chronic studies (8-12), but there are no studies on the effect of this compound after a single administration. Alpha-lipoic acid is a drug used in diabetic neuropathy and diabetes, which occurs frequently together with hypertension, especially in obese patients, to treat hypertension. Therefore, knowledge of the potential antihypertensive effect of alpha-lipoic acid even after a single dose and possibly too much pressure reduction is interesting and useful.

## Methods

The mechanism of the hypotensive effect of alpha-lipoic acid was examined in normotensive rats *in vivo* after a single intraperitoneal administration.

The experiments were conducted on male Wistar rats (weight, 180–250 g). The animals were housed in plastic cages in a room at a constant temperature of 20±2°C with a natural light–dark cycle. They had free access to a standard pellet diet and

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water. Groups consisted of the following number of animals: group 1, n=7; group 2, n=7; group 3, n=6; group 4, n=8.

The rats in the groups were treated as follows:

Group 1: alpha-lipoic acid at a dosage of 50 mg/kg b.w., i.p., suspended in 1% Tween 80.

Group 2: glibenclamide at a dosage of 50 mg/kg b.w., i.p., suspended in 1% Tween 80.

Group 3: glibenclamide at a dosage of 50 mg/kg b.w., i.p. 30 min before alpha-lipoic acid at a dosage of 50 mg/kg b.w., i.p.

Group 4: 0.25 mL of 1% Tween 80-control group.

All experiments were conducted according to the guidelines of the Animal Use and Care Committee of the Jagiellonian University (No.: 96/2011, date: 09.21.2011). The entire surgery was performed under thiopental anesthesia, and all efforts were made to minimize suffering.

The rats were anesthetized with thiopental (70 mg/kg) by an intraperitoneal injection. The left carotid artery was cannulated with a polyethylene tubing filled with heparin solution in saline to facilitate pressure measurements using the Datamax apparatus (Columbus Instruments, Ohio). Blood pressure was measured prior to the administration of the compounds (time 0 min-control pressure) and 80 min thereafter.

### Statistical analysis

The results of the blood pressure measurements are presented as the means±S.E.M., and statistical significance of the differences was evaluated using a two-way ANOVA, Bonferroni's multiple comparison test (four groups and ten factors). The differences were considered statistically significant when $p<0.05$.

## Results

Alpha-lipoic acid intraperitoneally administered at a dosage of 50 mg/kg of b.w significantly lowered blood pressure 50 min after the administration to rats [systolic $F(10,278)=2.828$, $p=0.0023$; diastolic $F(10.268)=3.689$, $p=0.0001$]. All animals survived the experience. There were no complications with the surgery. Glibenclamide (50 mg/kg) intraperitoneally administered 30 min before alpha-lipoic acid inhibited its in all the time measuring points, there was no statistically significant difference between the pressure indicated between the glibenclamide-treated groups and the control group ($p>0.05$). The results are shown in Figure 1. Table 1 shows the percentage of pressure drops in each group.

## Discussion

In this study we demonstrated a significant effect of alpha-lipoic acid at a dosage of 50 mg/kg of b.w. on blood pressure in rats after one intraperitoneal administration. Glibenclamide, a $K_{ATP}$ channel antagonist, attenuated this effect.

There are many papers in literature describing the impact of the chronic administration of alpha-lipoic acid on blood pressure pointing to its hypotensive effect after repeated administration or prevention of the development of hypertension (8-12). However, there is no described effect of alpha-lipoic acid on blood pressure after a single administration. In the present study, alpha-lipoic acid was intraperitoneally administered to rats at a dosage of 50 mg/kg of b.w. It was demonstrated that this dosage significantly reduced blood pressure, but the effect was visible beginning from the 50$^{th}$ minute after the administration. Typically, the hypotensive effect after intraperitoneal treatment with a blood pressure-lowering compound is faster. There may be various reasons explaining why the hypotensive effect was not manifested until 50$^{th}$ minute after the administration, including poor absorption of the compound after the intraperitoneal administration. Alpha-lipoic acid may be subject to changes or metabolized before its activity may be expressed or it may induce the formation of blood pressure reducing agents.

We also performed studies on the impact of the co-administration of glibenclamide, an ATP-dependent potassium channel inhibitor because it is known that the $K_{ATP}$ channel opening is involved in the integrated vasodilator response to $H_2S$ *in vivo* in rats and that this effect is abrogated by the glibenclamide-

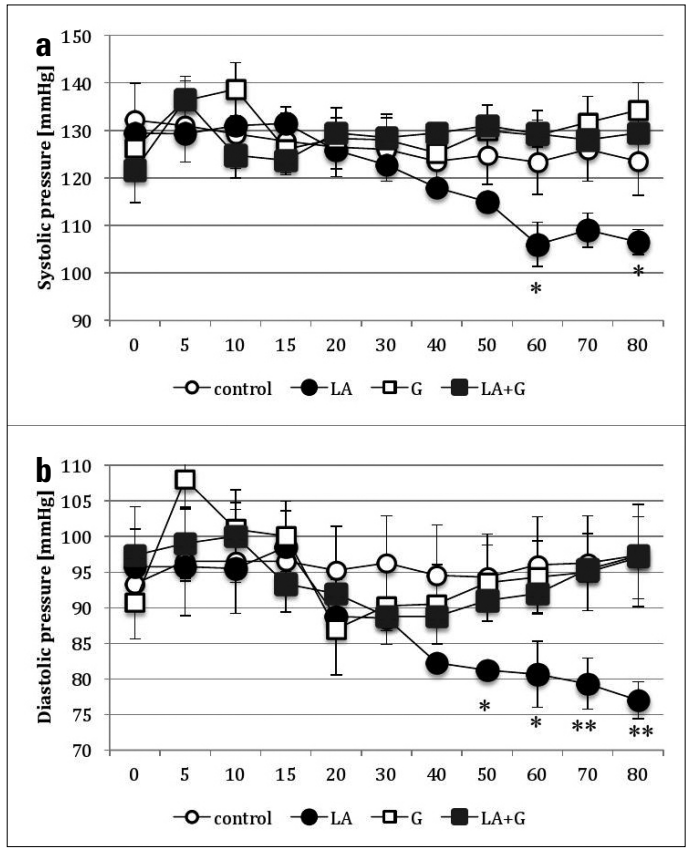


**Figure 1. a, b. The effect of alpha-lipoic acid, glibenclamide, and both compounds on blood pressure after a single administration**

The data represent systolic (a) and diastolic (b) blood pressure, means±S.E.M., after the administration of alpha-lipoic acid (50 mg/kg b.w., intraperitoneal injection)-LA group, or glibenclamide (50 mg/kg b.w., intraperitoneal injection)–G group, or alpha-lipoic acid (50 mg/kg b.w., intraperitoneal injection) plus glibenclamide (50 mg/kg b.w., intraperitoneal injection)-LA+G group, or 1% Tween 80 (0.25 mL, intraperitoneal injection)-control group; *significant LA group vs. control group in each time point: *$P<0.05$, **$P<0.01$ (two-way ANOVA, Bonferroni's multiple comparison test)

**Table 1. The percentage of changes in systolic blood pressure**

| Group\time | 5 | 10 | 15 | 20 | 30 | 40 | 50 | 60 | 70 | 80 |
|---|---|---|---|---|---|---|---|---|---|---|
| Control | -0.9 | -2.19 | -3.33 | -4.31 | -4.69 | -6.58 | -5.6 | -6.73 | -4.69 | -6.58 |
| LA | -0.15 | 1.16 | 1.54 | -2.86 | -5.17 | -8.88 | -11.2 | -18.2 | -15.8 | -17.8 |
| G | 7.9 | 9.82 | -0.24 | 1.58 | 1.35 | -0.79 | 2.93 | 2.14 | 4.28 | 6.33 |
| LA+G | 12.16 | 2.54 | 1.64 | 6.41 | 5.59 | 6.41 | 7.64 | 6.25 | 5.18 | 6.41 |

Groups: control-0.25 mL of 1% Tween 80; G-glibenclamide at a dose of 50 mg/kg b.w., i.p., suspended in 1% Tween 80; LA–alpha-lipoic acid at a dose of 50 mg/kg b.w., i.p., suspended in 1% Tween 80; LA+G - glibenclamide at a dose of 50 mg/kg b.w., i.p. 30 min before alpha-lipoic acid at a dose of 50 mg/kg b.w., i.p.; Time after administration [min]

induced blockade of the $K_{ATP}$ channel (13).

Other studies have shown that glibenclamide abolishes the decongestant action of alpha-lipoic acid, suggesting that ATP-dependent potassium channels are involved in this action (14, 15) and that this effect could be associated with the formation of $H_2S$.

There are reports indicating that the anti-inflammatory effect of alpha-lipoic acid may be associated with increased sulfane sulfur levels and $H_2S$ release (16). Additionally, other thiols such as L-cysteine or glutathione may be sulfane sulfur donors (17, 18) and may increase the levels of $H_2S$ (17). It has been demonstrated that $H_2S$ is released from bound sulfur in the presence of physiological concentrations of glutathione and cysteine under slightly alkaline conditions. Extracellular potassium increases the intracellular pH to the level that supports $H_2S$ release (17).

Therapy for cardiovascular system diseases utilizes the cardioprotective properties of compounds such as nicorandil (coronary heart disease), narcotic analgesics (post-myocardial infarction medication, cardioprotective effect), and adenosine (arrhythmias) for opening ATP-dependent potassium channels. Nicorandil opens $K_{ATP}$ channels, thereby dilating peripheral resistance and coronary arterioles. It may cause a decrease in preload and blood pressure. Therefore, the results are very interesting, suggesting that alpha-lipoic acid, a drug used in diabetic neuropathy, may have cardioprotective properties related to this mechanism.

## Study limitations

The most important limitation of our study was the lack of detailed studies at the cellular level.

Another very important limitation of our study was the duration of the experiment.

## Conclusion

This study suggests that the observed effect of alpha-lipoic acid on blood pressure is associated with increased levels of sulfane sulfur, opening of ATP-dependent potassium channels, and release of $H_2S$.

The results obtained in this study are very important because the patients taking alpha-lipoic acid may be treated for co-existing hypertension. Therefore, they must take into account the possibility of the blood pressure-lowering effect of alpha-lipoic acid that does not lead to excessive orthostatic hypotension.

**Conflict of interest:** None declared.

**Peer-review:** Externally peer-reviewed.

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## References

1. Dinçer Y, Telci A, Kayalı R, Yılmaz İA, Çakatay U, Akçay T. Effect of lipoic acid on lipid peroxidation and anti-oxidant enzyme activities in diabetic rats. Clin Exp Pharmacol Physiol 2002; 29: 281-4.
2. Bilska A, Dudek M, Iciek M, Kwiecień I, Sokolowska-Jeżewicz M, Filipek B, et al. Biological actions of lipoic acid associated with sulfane sulfur metabolism. Pharmacol Rep 2008; 60: 225-32.
3. Elsey DJ, Fowkers RC, Baxter GF. Regulation of cardiovascular cell function by hydrogen sulfide (H2S). Cell Biochem Funct 2010; 28: 95-106.
4. Wang XB, Jin HF, Tang CS, Du JB. Significance of endogenous suphur-containing gases in the cardiovascular system. Clin Exp Pharmacol Physiol 2010; 37: 745-52. [Crossref]
5. Beltowski J. Hydrogen sulfide as a biologically active mediator in the cardiovascular system. Postepy Hig Med Dosw 2004; 58: 285-91.
6. Al-Magableh MR, Hart JL. Mechanism of vasorelaxation and role of endogenous hydrogen sulfide production in mouse aorta. Naunyn Schmiedebergs Arch Pharmacol 2011; 383: 403-13.
7. Tang G, Wu L, Wang R. Interaction of hydrogen sulfide with ion channels. Clin Exp Pharmacol Physiol 2010; 37: 753-63. [Crossref]
8. Vasdev S, Gill V, Parai S, Gadag V. Dietary lipoic acid supplementation attenuates hypertension in Dahl salt sensitive rats. Mol Cell Biochem 2005; 275: 135-41. [Crossref]
9. Takaoka M, Kobayashi Y, Yuba M, Ohkita M, Matsumura Y. Effects of α-lipoic acid on deoxycorticosterone acetate–salt-induced hypertension in rats. Eur J Pharmacol 2001; 424: 121-9. [Crossref]
10. Midaoui AEL, de Champlain J. Prevention of hypertension, insulin resistance, and oxidative stress by α-lipoic acid. Hypertension

2002; 39: 303-7. [Crossref]
11. Thirunavukkarasu V, Nandhini AT, Anuradha CV. Lipoic acid attenuates hypertension and improves insulin sensitivity, kallikrein activity and nitrite levels in high fructose-fed rats. J Comp Physiol B 2004; 174: 587-92. [Crossref]
12. Ergür BU, Mıcılı SÇ, Yılmaz O, Akokay P. The effects of α-lipoic acid on aortic injury and hypertension in the rat remnant kidney (5/6 nephrectomy) model. Anatol J Cardiol 2015; 15: 000-000 [doi: 10.5152/akd.2014.5483]. [Crossref]
13. Zhao W, Zhang J, Lu Y, Wang R. The vasorelaxant effect of H2S as a novel endogenous gaseous KATP channel opener. EMBO J 2001; 20: 6008-16.
14. Dudek M, Bilska-Wilkosz A, Knutelska J, Mogilski S, Bednarski M, Zygmunt M, et al. Are anti-inflammatory properties of lipoic acid associated with the formation of hydrogen sulfide? Pharmacol Rep 2013; 65: 1018-24. [Crossref]
15. Dudek M, Knutelska J, Bednarski M, Nowiński L, Zygmunt M, Bilska-Wilkosz A, et al. Alpha lipoic acid protects the heart against myocardial post ischemia-reperfusion arrhythmias via KATP channel activation in isolated rat hearts. Pharmacol Rep 2014; 66: 499-504.
16. Zygmunt M, Dudek M, Bilska-Wilkosz A, Bednarski M, Mogilski S, Knutelska J, et al. Anti-inflammatory activity of lipoic acid in mice peritonitis model. Acta Pol Pharm 2013; 70: 899-904.
17. Ishigami M, Hiraki K, Umemura K, Ogasawara Y, Ishii K, Kimura H. A Source of Hydrogen sulfide and a mechanism of its release in the brain. Antioxid Redox Signal 2009; 11: 205-14. [Crossref]
18. Iciek M, Wlodek L. Biosynthesis and biological properties of compounds containing highly reactive, reduced sulfane sulfur. Pol J Pharmacol 2001; 53: 215-25.


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