بسم الله الرحمن الرحيم

قالوا سبحانك لا علم لنا
إلا ما علمتنا
إنك أنت العليم الحكيم

صدق الله العظيم

سورة البقرة الآية ٣٢

ALPHA-LIPOIC ACID AND AMLODIPINE AMELIORATE MYOCARDIAL INFARCTION INDUCED BY ISOPROTERENOL IN RATS

By

Nayira A. Abdel Baky , Nouf M. Al- Rasheed, Nawal M. Al-Rasheed, Iman Y. Zaghloul , and Mahasen A. Radwan

Pharmaology Department, College of Pharmacy (Girls), KSU

Introduction

- **In the 21st century, there is an ongoing effort to integrate complementary and alternative medications into the practice of conventional medicine for the treatment of serious cardiovascular disorders as ischemic heart diseases and specifically myocardial infarction.**

- **Epidemiological studies indicate that ischemic heart diseases, especially myocardial infarction, will constitute the major disease-burden worldwide in the year 2020. In fact, myocardial infarction is considered the most leading cause of death worldwide even with the huge improvement in clinical care, and the wide use of various health innovations**

- Alpha-lipoic acid has generated considerable clinical interest as a thiol-replenishing and redox-modulating agent.

- Biologically, alpha-lipoic acid functions as a coenzyme in multi-enzyme complexes that catalyse the oxidative decarboxylation of alpha-keto acids. It has been identified as a key antioxidant found naturally in our diets, but it appears to have an enhanced function when given as a supplement.

- This antioxidant can scavenge a number of free radicals in hydrophilic and lipophilic environments. In addition, alpha-lipoic acid was found to be capable of regenerating many endogenous antioxidants in the body.

- Moreover, alpha-lipoic acid is endowed with other beneficial activities, as in the prevention and treatment of oxidative stress in a number of models or clinical conditions including ischemia reperfusion injury , diabetes, HIV infection, and neurodegenerative diseases

- Calcium channel antagonists have been extensively used in the treatment of ischemic heart disease because of their ability to trigger coronary vasodilatation. They have been shown to exert beneficial myocardial effects via their inhibition of the slow inward Ca^{2+} current through cardiac L-type Ca^{2+} channels. This effect has been thought to reduce the activation of intramyocardial Ca^{2+} - dependent enzymes , which are known to contribute to cardiac dysfunction and fibrotic processes. This is in addition to the negative inotropic and/or chronotropic energy-saving effects of these compounds.

- **Amlodipine** is a long-acting dihydropyridine Ca^{2+} antagonist that is effective in treatment of hypertension. However, research on the non-hypotensive effects of amlodipine has attracted more attention in recent years, especially due to its potent antioxidant activity arising from distinct biophysical interactions with the membrane lipid bilayer. This mechanism may contribute to its cytoprotective action in cardiovascular diseases.

Aim of The Work

- The pathophysiological effects of myocardial infarction is the imbalance between the generation of reactive oxygen species due to ischemia and the antioxidant defense system of the heart tissue.

- **Protection against myocardial infarction may be related to the maintenance of the antioxidant status of the heart.**

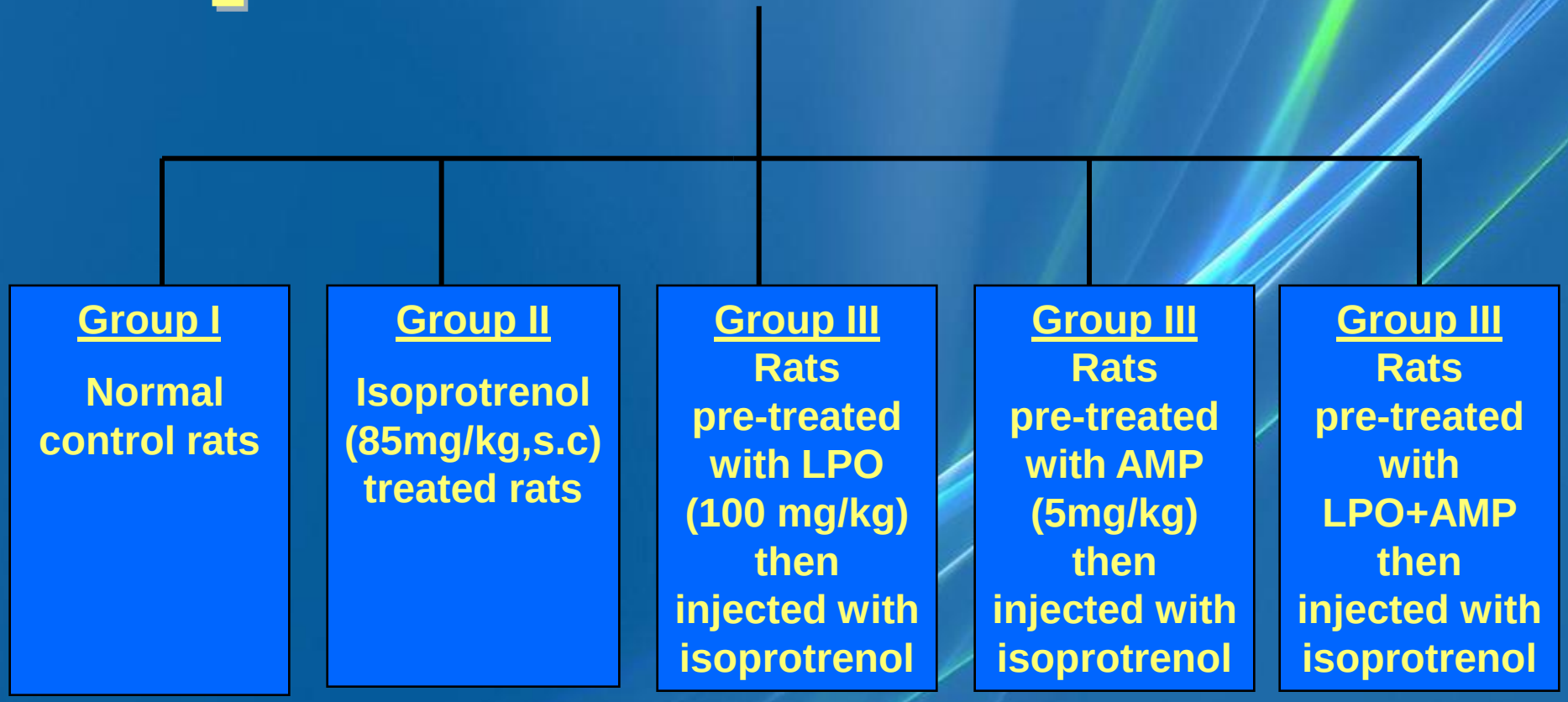
- **Alpha-lipoic acid is a new food supplement with antioxidant properties.**

- **Amlodipine (AML) is an arteriodilator antihypertensive drug with antioxidant properties.**

Our Aim was to investigate whether the combination of alpha-lipoic acid and amlodipine offers a protective synergism against myocardial infarction in a well-characterised experimental model; the induction of myocardial infarction by isoproterenol in rats.

Materials and Methods

Experimental Protocol



Parameters Measuered

- *Survival and general toxicity.*
- **Electrocardiograph parameters.**
- **Myocardial injury marker enzymes.**
- **Oxidative stress biomarkers.**
- **Histopathological examination**

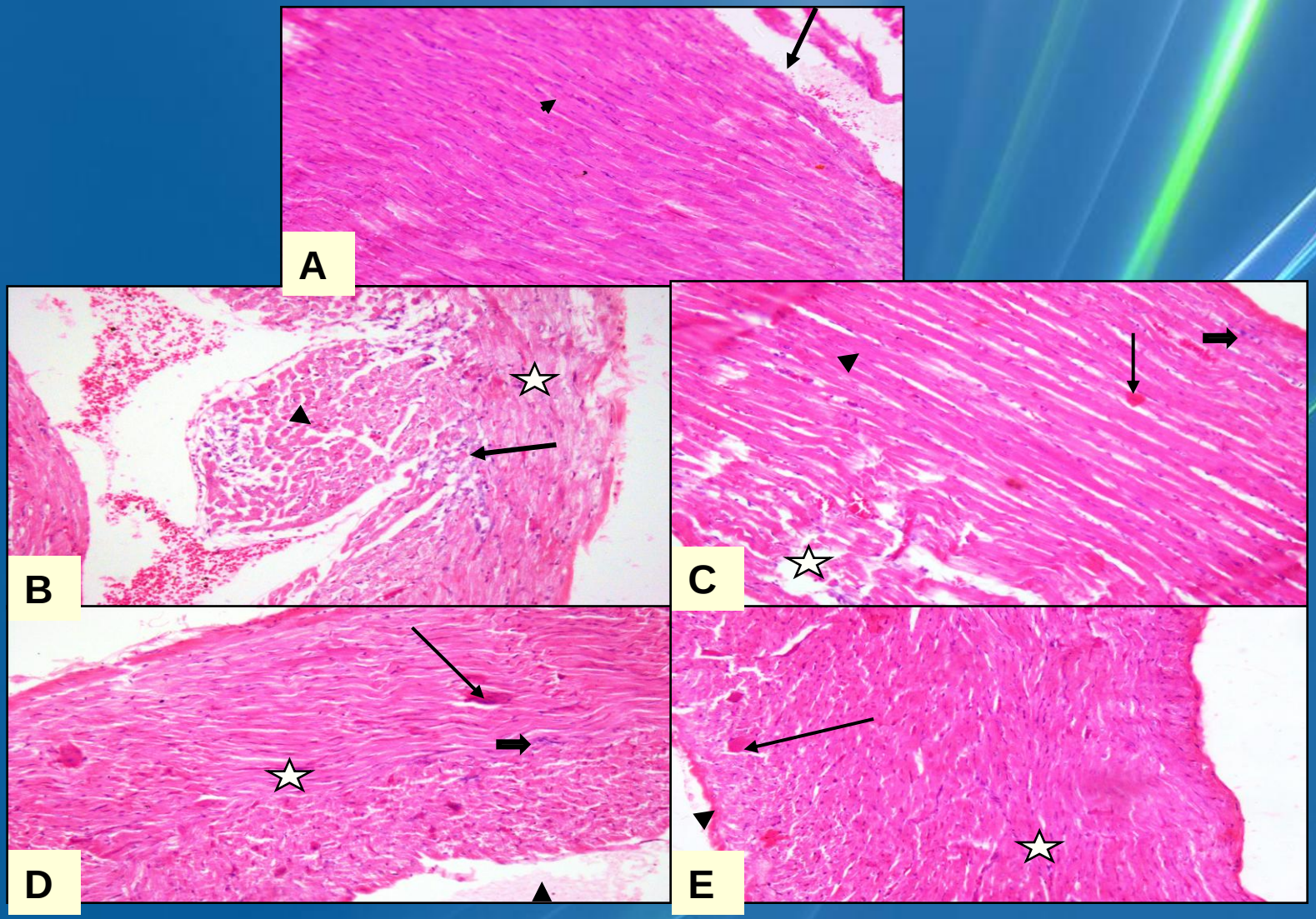
Results

Effect of α-Lipoic acid (LPO), amilodipine (AML) and their combination on body weight and mortality % of infarcted rats.

Treatment	Body weight (g)		Heart weight (g)	Mortality %
	Initial	Final		
CONT	200.2 ± 2.46	212.4 ± 1.91	0.834 ± 0.061	0
ISO	203.4 ± 1.81	210.0 ± 1.65	1.12 ± 0.018[a]	40
LPO+ISO	200.6 ± 2.09	214.5 ± 1.84	1.072 ±0.025[b]	20
AML+ISO	202.2 ± 2.75	206. 3± 1.17	0-966 ± 0.036	0
COMB+ISO	209.0 ± 3.96	217.0 ± 2.06	0.940 ± 0.033 *	0

*Values are means of (6-10) data points ± S.E.M. aP<0.001, bP<0.01 compared to the control group, *P<0.05 compared to the ISO-treated group, respectively, using ANOVA followed by Bonferroni as a post-ANOVA test.*

Figure 4. Light photomicrographs of rat heart tissue stained by H&E and magnified X200.

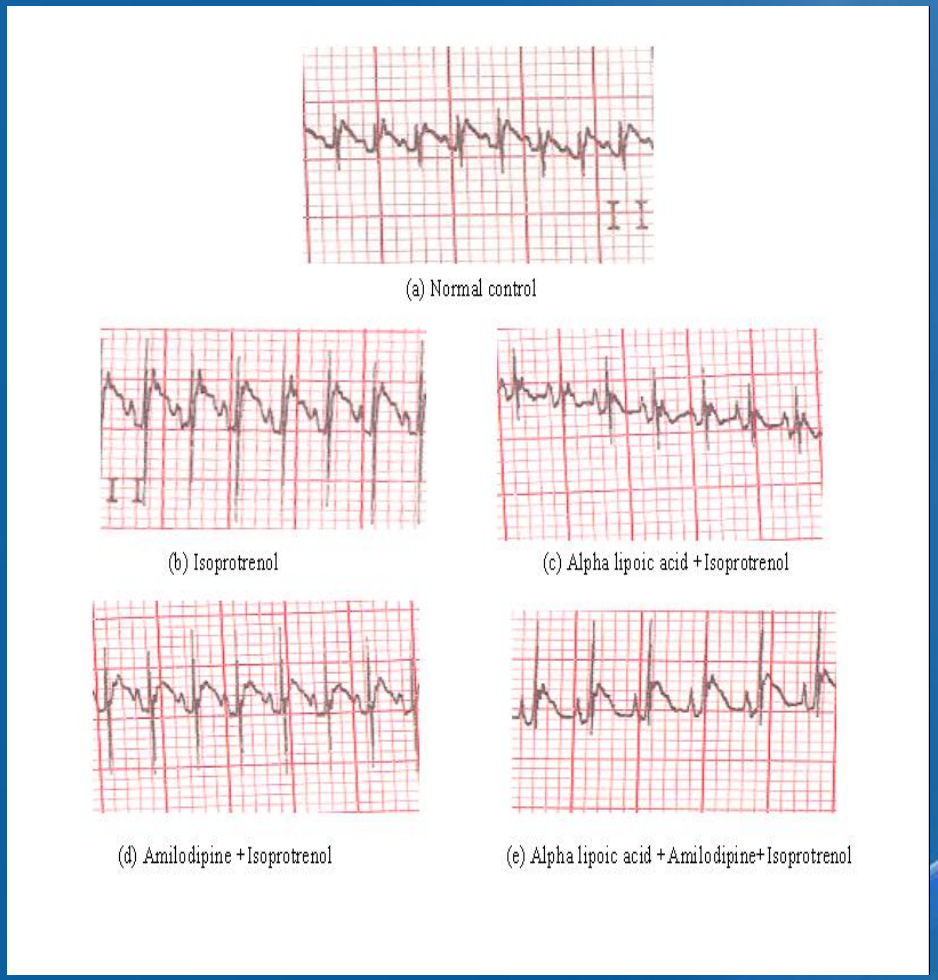


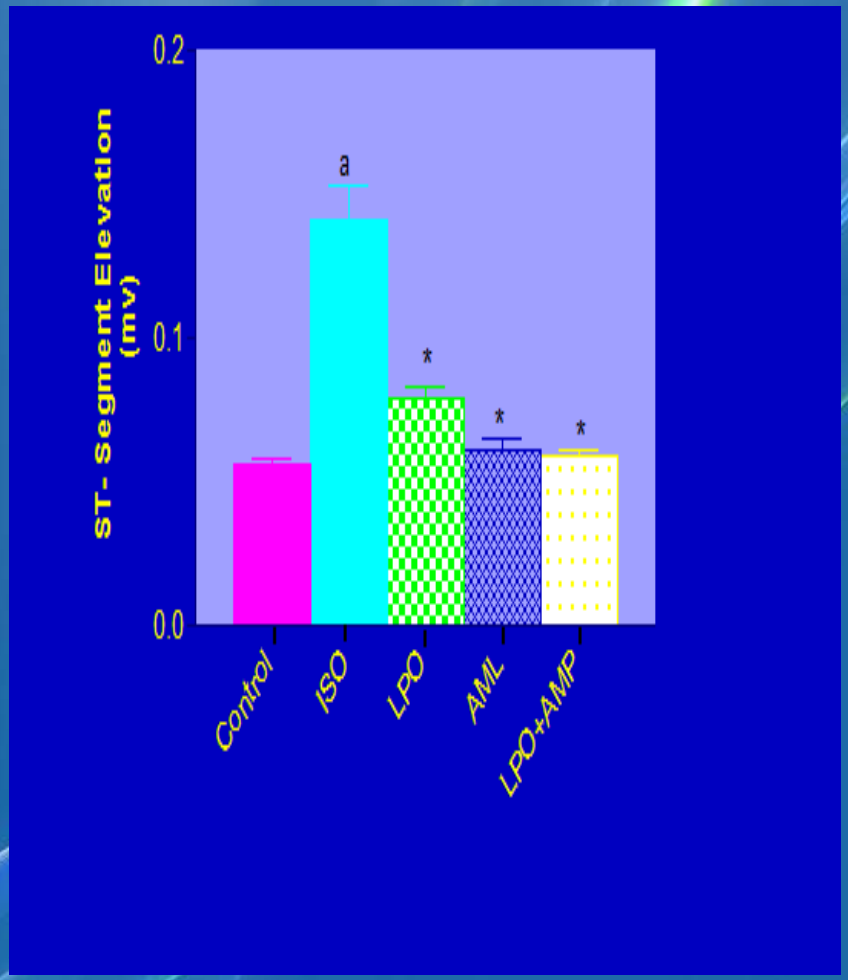
Effects of α-Lipoic acid (LPO), amilodipine (AML) and their combination on cardiac marker enzymes

Animal Groups	LDH	CPK-MB	ALT	AST
CONT	220.7 ± 10.5	209.3 ± 15.04	42.88 ± 1.93	93.20 ± 2.21
ISO	400.4 ± 20.3[a]	430.4 ± 6.27[a]	75.52 ± 1.22[a]	243.75 ± 15.87 [a]
LIP+ISO	273.5 ± 13.2 [*]	280.8 ± 10.77[b *#]	58.20 ± 1.01 [a*]	149.5 ± 1.34 [b*]
AML+ISO	257.7 ± 19.4 [*]	254.6 ± 6.2 [c*]	51.83 ± 2.75 [c*]	148.1 ± 9.88 [b*]
COMB+ISO	219.4 ± 17.1 [*]	232.2 ± 5.07 [*]	53.71 ± 1.39 [b*]	148.6 ± 5.10 [b*]

Values are means of (6-10) data points ± S.E.M. CPK-MB, LDH, AST, and ALT concentrations are expressed in U/l, *aP<0.0* compared to the control group, b*P<0.001* compared to the isoprotrenol treated group, and c*P <0.05* compared to the combination treated group, respectively, using ANOVA followed by Bonferroni as a post-ANOVA test.

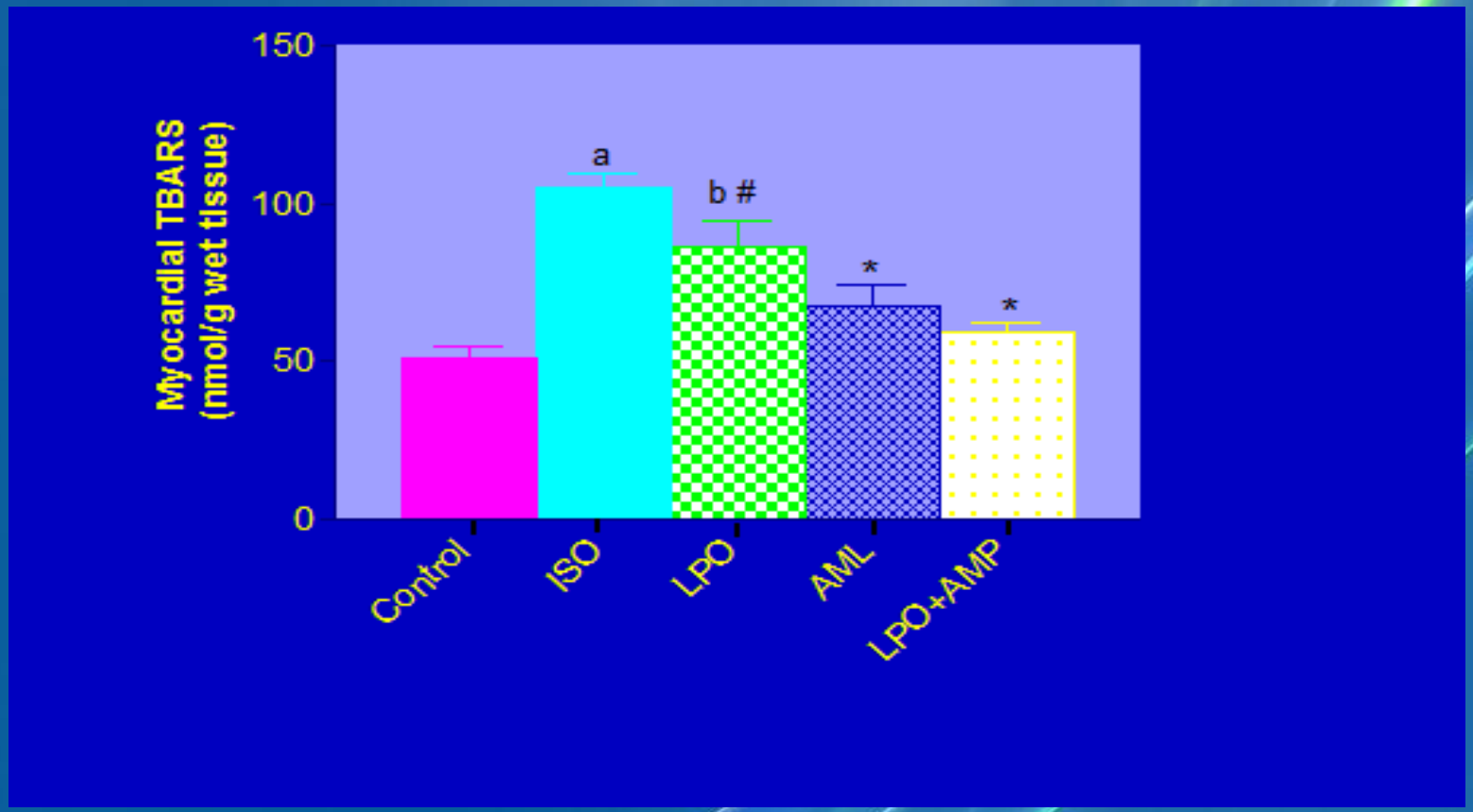
Effect of α-Lipoic acid (LPO), amilodipine (AML) and their combination on ECG-patterns of ISO-induced myocardial infarction in rats





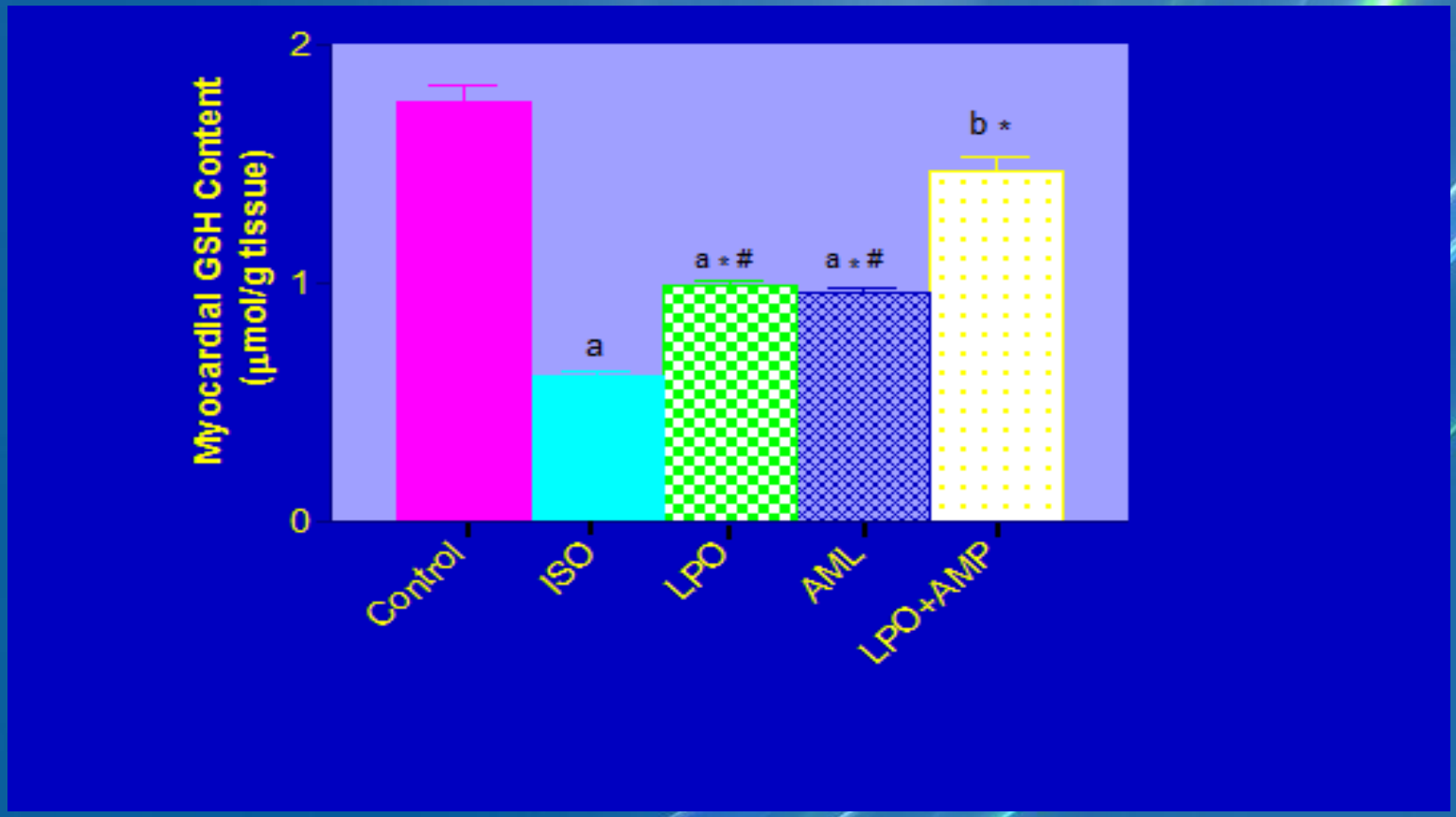
Data are presented as means ±S.E.M. (n= 6-10). aP<0.001 compared to the control group, *P<0.001 compared to the isoprotrenol-treated group, respectively, using ANOVA followed by Bonferroni as a post-ANOVA test

Effect of α-Lipoic acid (LPO), amilodipine (AML) and their combination on myocardial lipid peroxidation.



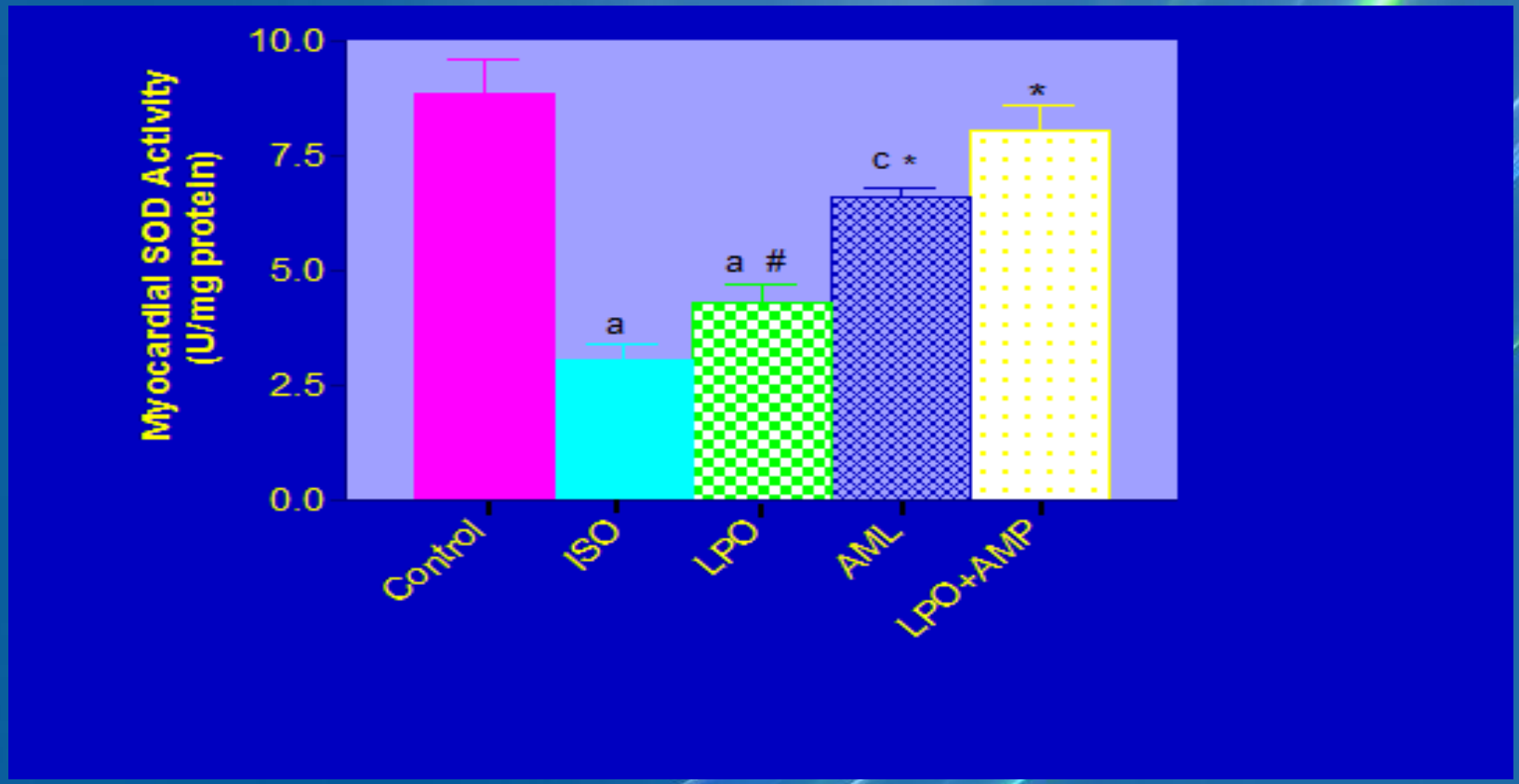
Data are presented as means ± S.E.M. (*n*=6-10). *aP<0.001, bP*<0.01 compared to the control group, **P<0.001* compared to the isoprotrenol-treated group, and #*P<0.05* compared to the combination-treated group, respectively, using ANOVA followed by Bonferroni as a post-ANOVA test.

Effect of α-Lipoic acid (LPO), amilodipine (AML) and their combination on myocardial glutathione content.



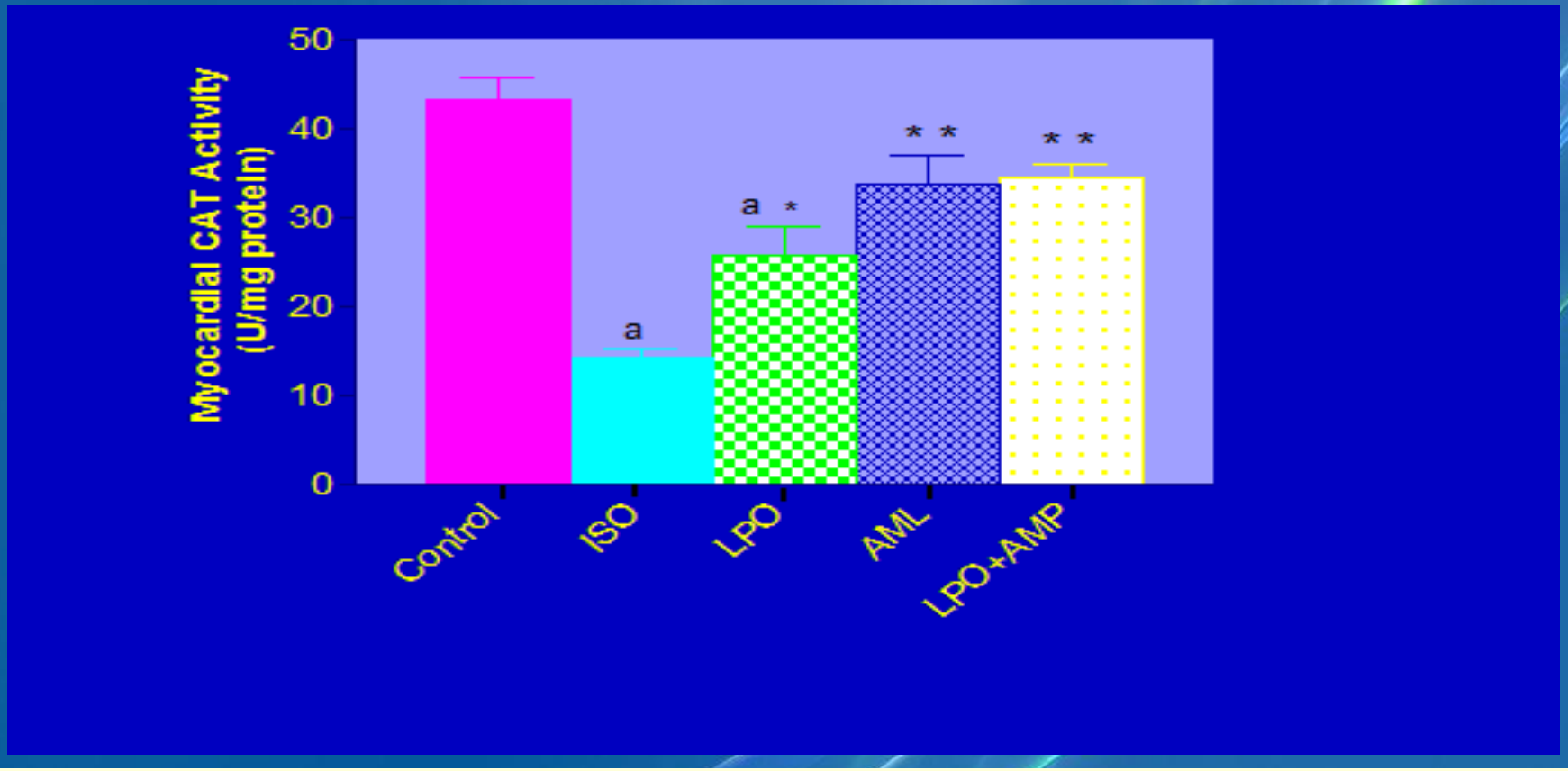
Data are presented as means ± S.E.M. (n=6-10). *aP<0.001, bP*<0.01 compared to the control group, **P<0.001* compared to the isoprotrenol-treated group, and #*P<0.001* compared to the combination-treated group, respectively, using ANOVA followed by Bonferroni as a post-ANOVA test.

Effect of α-Lipoic acid (LPO), amilodipine (AML) and their combination on myocardial superoxide dismutase (SOD) activity.



Data are presented as means ± S.E.M. (n=6-10). *aP<0.001, bP*<0.01, c*P*<0.05 compared to the control group, **P<0.001* compared to the isoprotrenol-treated group, and #*P<0.001* compared to the combination-treated group, respectively, using ANOVA followed by Bonferroni as a post-ANOVA test.

Effect of α-Lipoic acid (LPO), amilodipine (AML) and their combination on myocardial catalase (CAT) activity.



Data are presented as means ± S.E.M. (*n*=6-10). *aP<0.001* compared to the control group, and **P<0.05* ***P<0.001* compared to the isoprotrenol-treated group, respectively, using ANOVA followed by Bonferroni as a post-ANOVA test.

Conclusion

- Subcutaneous injection of isoproterenol to male Wistar rats induced marked alterations in ECG-patterns, and significanty increased the activities of serum myocardial injury marker enzymes.

- Moreover, the level of myocardial lipide peroxides was significantly increased, while the level of myocardial reduced glutathione and the activities of antiperoxidative enzymes were significantly reduced.

- Daily pre-treatment with either alpha-lipoic acid or amlodipine for respective periods of 15 or 7 days altered almost all of the above mentioned myocardial changes induced by isoproterenol.

- Meanwhile, the combination of alpha-lipoic acid and amlodipine synergisticlly reduced all isoproterenol-induced elevations in diagnostic marker enzyme levels and almost restored normal ECG-patterns in experimental animals.

- In addition, this combination exerted a synergistic antioxidant effect by blocking the induction of lipid peroxidation, and prevented isoproterenol-induced alterations in the level of reduced glutathione and the activities of antiperoxidative enzymes.

- Histopathological observations were found to support these biochemical findings.

- As far as we are aware, the present study demonstrates for the first time that LPO ameliorates ISO–induced myocardial infarction in rats and that co–pretreatment with AML significantly potentiated this cardioprotective effect.
- The overall cardioprotective effect of LPO is probably related to its direct quenching of oxidative free radicals and to its ability to maintain the activities of free radical scavenging enzymes and the level of GSH, which protect myocardial membranes against oxidative damage by decreasing lipid peroxidation.
- These observations highlight that LPO is a promising supplement for improving defence mechanisms in the heart against oxidative stress caused by myocardial infarction.


Thank You