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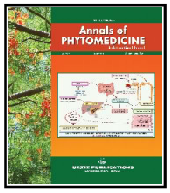

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Ameliorative effect of quercetin in lead-induced toxicity on pulmonary artery of goat (*Capra hircus*)

Gaurang Sharma, Pratishtha Sharma◆, Ashok Gaur and Shourbh Bithu

Department of Veterinary Pharmacology and Toxicology, College of Veterinary and Animal Science Rajasthan, University of Veterinary and Animal Sciences, Bikaner-334001, Rajasthan, India

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Abstract

There is no level of blood lead (Pb) which can be regarded as safe for animals or humans; deleterious effects of Pb in the form of endothelial dysfunction, hypertension, cardiovascular dysregulation, and autonomic dysfunction have largely been documented. Lead, being a highly toxic heavy metal, affects nearly most of the organs and systems within an organism's body. Previous studies suggested quercetin, a normal dietary flavonoid, has a remarkable relaxing effect on smooth muscles of lung tissue. The smooth muscle rings, both untreated (control) and treated with Pb acetate were subjected to 5-HT and PE (contractile agents) and ACh and SNP (relaxing agents) both with or without the presence of quercetin in increasing doses. If, the Pb acetate is not present, the mean EC_{50} values related to 5-HT and PE were 7.359×10^{-6} M and 5.953×10^{-6} M, respectively. In the presence of Pb acetate in solution, these values decreased to 9.140×10^{-7} M and 3.164×10^{-6} M, respectively, indicating a shift towards the left in the curve. The mean values of IC_{50} for ACh and SNP were 2.445×10^{-8} M and 2.764×10^{-6} M, respectively, in the absence of Pb acetate. In the presence of Pb acetate in solution, these values increased to 5.258×10^{-7} M and 2.800×10^{-6} M, respectively, indicating a right shift in the curves. This shows the increase in the contractile nature of tissue due to Pb. In quercetin-containing solution, the values of mean EC_{50} for 5-HT and PE were increased to 1.631×10^{-6} M and 1.167×10^{-5} M, respectively indicating a right shift in the curve. Likewise, IC_{50} values were decreased to 4.730×10^{-8} M and 9.993×10^{-7} M, respectively, for ACh and SNP indicating a shift in the curve towards the left. This suggests quercetin has an ameliorative effect against the toxic effect of Pb on the pulmonary artery of goats by vasorelaxation.

1. Introduction

Lead (Pb) is an important metal toxicant and no exposure to Pb can be regarded as safe because it tends to persist inside the body, specially in the brain, and bones, for a long time and in the ecosystem as well. Lead exists in various forms, including its metallic state, divalent or tetravalent cations, and organic compounds. The predominant environmental form is divalent Pb, whereas inorganic tetravalent Pb compounds do not occur naturally. Notably, organo-lead complexes, predominantly composed of tetravalent Pb, include well-known examples such as the gasoline additive tetraethyl Pb which is now banned in many countries. Various steps have been taken in the developed world towards limiting Pb exposure with the use of lead-free fuel, lead-free paints, lead-free ammunition, and many other products but its erstwhile use in developed countries, metallurgical activities, and poor regulations in developing countries continue to render Pb one of the most significant environmental pollutants worldwide (Thompson, 2018; Ufelle and Barchowsky, 2019). Old farm buildings with chipping and alligatoring lead paint, old and used lead batteries lying unattended in farmland, licking vices of animals, cheap yet very effective lead paints, rearing of animals near metallurgical operations, battery recycling facilities, lead ammunition contaminated land, *etc.*, all these and other factors contribute to Pb poisoning which is still prevalent among livestock animals in all parts of India including Rajasthan. Blood Pb toxic levels are reported in wild animals in captivity (Bates, 2017; Gupta and Bakre, 2012; Gupta, 2013). Several epidemiological studies conducted and various meta-analyses have shown that blood Pb more than 10 µg/100 ml causes cardiovascular diseases, increased blood pressure, and chronic pulmonary disease, in both animal species and in humans (ATSDR, 2020; Cheng *et al*., 2001; Marques *et al*., 2001) as well. In its divalent or tetravalent metallic cation forms within the body, Pb exhibits a higher affinity for endogenous protein binding sites compared to calcium and zinc ions. This heightened affinity is attributed to the larger ionic radius and greater electronegativity of Pb (Garza *et al*., 2006; Halmo and Nappe, 2021; Mitra *et al*., 2017). Several oxidative stress-generated deleterious effects, physiological dysfunctions, and biochemical alterations in laboratory animals and humans are associated with Pb exposure (Courtois *et al*., 2003; Halmo and Nappe, 2021; Mitra *et al*., 2017). The respiratory tract becomes a remarkable route of Pb exposure from atmospheric pollution and a target as well. Lead inhibits the relaxation of muscles mainly smooth muscles in response to vasodilators as well as bronchodilators (Gupta and Fahim, 2007; Zhang *et al*., 2005). Smooth muscle tissues of the airway, GIT, and vascular system have shown contraction *in vitro* upon the revelation of Pb (Sopi *et al*., 2009; Valencia-Hernandez *et al*., 2001; Zhang *et al*., 2005). Non-adrenergic and non-cholinergic

Corresponding author: Dr. Pratishtha Sharma
Department of Veterinary Pharmacology and Toxicology, College of Veterinary and Animal Science, Rajasthan University of Veterinary and Animal Sciences, Bikaner-334001, Rajasthan, India
E-mail: drpratishthasharma@gmail.com
Tel.: +91-9251415775

(NANC) relaxation of the gastric fundus of rats is hindered due to persistent Pb exposure, potentially by influencing the release of nitric oxide (NO) interacting with NANC nerves along with intracellular signaling mechanisms. (Santos *et al*., 2006). Exposure to Pb induces hypertension and impairs endothelial function. At low-level exposure for short term, Pb induces alterations in pulmonary hemodynamics and elevates oxidative stress that trigger heightened hyperpolarization in the pulmonary vasculature *via* Kv and SKCa channels, potentially serving a response in declining nitric oxide sensitivity (Covre *et al*., 2016). Lead interferes with endogenous nitric oxide generation and subsequent cGMP-dependent vasorelaxation, possibly through Pb-induced ROS generation. Lead mimics calcium in activating renin secretion in the renin-angiotensin-aldosterone system, it also elevates blood pressure. Lead alters calcium-activated contraction and functioning of smooth muscle cells, leading to increased contractility by reducing Na^+/K^+-ATPase pump activity and by stimulating Na^+/Ca^{+2} exchange pump activity. Additionally, Pb tends to elevate the secretion of vasoconstrictive ligands, such as endothelin and thromboxane (ATDSR, 2020).

Quercetin (QUE) is an antioxidant flavonoid phytochemical that is chemically pentahydroxyflavone (NCBI, 2022). Many common foods, *viz.*, red onions, radish leaves, fennel, lovage, broccoli, berries, *etc*., are rich in QUE (Bhagwat and Haytowitz, 2016). QUE has been found beneficial in ailments like diabetes (Ain *et al*., 2022; Alam *et al*., 2014; Hamilton *et al*., 2018; Mahesh and Menon, 2004; Srinivasan *et al*., 2018; Vessal *et al*., 2003; Yang and Kang, 2018; Youl *et al*., 2010), aging (Lu *et al*., 2006), pulmonary as well as cardiovascular diseases, osteoporosis (Boots *et al*., 2008) as well as hepatotoxicity (Yadav *et al*., 2021), and nephrotoxicity (Kilaru *et al*., 2022). QUE is protective against Pb-induced injuries to the liver (Liu *et al*., 2010a; Liu *et al*., 2013) and kidneys (Liu *et al.,* 2010b; Liu *et al*., 2012). Orally administering a daily combination of quercetin (QUE) and curcumin to rats showed alleviating effects on cadmium-induced variations observed in both the cortex of the brain and the heart. (Makwana *et al*., 2021). Quercetin (QUE) notably decreased hydrogen peroxide (H_2O_2), reactive oxygen species (ROS) levels, and malondialdehyde (MDA), concurrently reducing the proportion of glutathione (GSH) to oxidized glutathione (GSSG), in the rats' excretory cells and liver with Pb treatment. (Samarghandian *et al*., 2013). Quercetin demonstrated protective properties against Pb toxicity in Japanese quails by decreasing oxidative stress and suppressing proteins such as Caspase 3 and Caspase 9 proteins (Arslan *et al*., 2022). The administration of QUE partially suppressed the activation of nuclear factor kappa B(NF-κB) and mitogen-activated protein kinases (MAPKs), thereby mitigating Pb-induced histopathological changes in rat kidneys, as well as oxidative damage, through inhibition of ROS generation in the kidney cells (Liu *et al*., 2012). QUE also significantly restored the functioning of catalase enzyme (CAT), glutathione peroxidase enzyme (GPx), and superoxide dismutase (SOD). Furthermore, QUE decreased oxidative DNA damage and apoptotic changes in the kidney and liver of rats exposed to Pb. Certain studies indicate that the endothelium-dependent vasorelaxant impact of QUE is associated with nitric oxide (Ajay *et al*., 2003; Chan *et al*., 2000; Chen and Pace-Asciak, 1996; Khoo *et al*., 2010) while some others report it as endothelium-independent (Duarte *et al*., 1993; Perez-Vizcaino *et al*., 2002) including another report which suggests that its action involves the activation of L-type vascular calcium channels (Saponara *et al*., 2002).

The medicinal and therapeutic attributes of QUE, coupled with its low toxicological profile, position it as a highly promising agent in addressing heavy metal toxicity. Quercetin exhibits Pb-chelating properties by establishing coordination bonds with ionic Pb via its ortho-phenolic groups situated on the quercetin B ring. This interaction aids in alleviating the adverse effects of Pb exposure (Bravo and Anacona, 2001). Chelation therapy proves highly beneficial in addressing Pb toxicity, facilitating the removal of Pb from various compartments of the body, and promoting its excretion through urine. Prevention is considered the best approach and numerous naturally occurring antioxidants like QUE have been identified for their potential in preventing and treating oxidative stress and Pb-induced toxicity (Flora *et al*., 2012). Studies also suggested that the QUE and melatonin liposomes have antioxidant properties and may be helpful in the cure of pulmonary diseases (Yeligar *et al*., 2021).

Small ruminants especially goats are reared primarily on range feeding. The curious nature of goats and their constant desire to explore and investigate anything unfamiliar adds to their easy exposure to pollutants and sources of toxicity. Existing data from other researchers on the detrimental impacts of Pb on smooth muscle tissues in laboratory animals, coupled with the antioxidant properties of QUE, provided the basis for our current investigation. We aimed to assess the *in vitro* impact of Pb on goat pulmonary artery smooth muscle and explore any potential mitigation through the concurrent administration of QUE.

2. Materials and Methods

2.1 Chemicals

Pure chemicals like 5-hydroxytryptamine (5-HT), phenylephrine (PE), acetylcholine (ACh), and sodium nitroprusside (SNP) from Sigma-Aldrich, USA; quercetin (QUE) from Himedia India were procured, respectively. Stock solutions of 5-HT, PE, ACh, SNP, and Pb acetate were prepared in sterile triple glass distilled water. Fresh dilutions of the necessary concentrations for each chemical were prepared in Modified Kreb's-Henseleit solution (MKHS) on the day of usage. The stock solution of QUE (10 mM strength) was prepared by dissolving the same with a minimum volume of N/10 sodium hydroxide, followed by the addition of sterile triple glass distilled water to render it up to the required volume while adjusting pH to 7.4 with a minimum quantity of N/10 hydrochloric acid. Working solutions of QUE were prepared afresh and used the same day. Analytical-grade chemicals and reagents were used for the present study.

2.2 Tissue preparations

The whole lung lobes of freshly slaughtered healthy goats (20-25 kg) were obtained from a local abattoir. These tissue samples were transferred immediately to glass jars having cold (4-6°C) oxygenated MKHS with pH 7.4 having (in mM): NaCl (118), $NaHCO_3$ (25), KCl (4.8), KH_2PO_4 (1.2), $MgSO_4$ (1.2), $CaCl_2$ (1.2) and anhydrous dextrose (11.1) and transported under the cold chain to reach the laboratory within 15-20 min. Pulmonary arteries were meticulously dissected under a stereo-microscope, removing any adhering connective tissue and fascia. Subsequently, annular ring segments measuring 2-3 mm in length were cut from these arteries. Then, these annular arterial rings were utilized for isometric contraction studies.

2.3 Isometric contraction recording

Freshly dissected arterial rings were mounted in a tissue bath tube with the help of two 'L' shaped hooks, made of 32-gauge stainless steel wire, passing through its lumen. The mounted rings in the tissue organ bath of 20 ml capacity were then allowed to equilibrate under a resting tension of 1g while immersed in a thermostatically controlled MKHS maintained consistent at 37.0 ± 0.5°C and aerated continuously with 95% O_2 level and 5% CO_2 (carbogen). The arterial ring was kept on equilibration for two hours before recording the muscle tension. Throughout this period, MKHS in the tissue bath was changed every 15 min. An increase or decrease in real-time tension was sensed by a highly sensitive isometric force transducer (Force transducer Model: ML 4856 /26T, ADInstruments Pty Ltd., Australia) connected with a compatible amplifier (Power Lab 26T Series Unit, ADInstruments Pty Ltd., Australia) and recorded using Lab chart® v8.1.10 computer software (Power Lab, Australia).

After the completion of the equilibration period, the arterial rings were exposed to an isotonic 80 mM potassium depolarizing solution (KDS) to assess the viability of the tissue. KDS was prepared by substituting NaCl in MKHS with an equimolar quantity of KCl. When contractile responses plateaued, the rings were given washings *in situ* with two or three changes of MKHS. The arterial rings other than the control rings were then exposed to Pb acetate, by bathing in 0.5mM concentration of Pb acetate chloride in MKHS for 30 min. Subsequently, contractile (5-HT, phenylephrine) and relaxing agents (acetylcholine, and sodium nitroprusside) were added separately later to get cumulative dose-response curves (DRCs). Then QUE was further added to the Pb acetate-exposed arterial rings, and then again to those as mentioned above contractile as well as relaxing agents were added to obtain cumulative DRCs. Their respective half-maximal effective concentration (EC_{50}) values or half-maximal inhibitory concentration (IC_{50}) values, and negative logarithm to base 10 of the EC_{50} or IC_{50}, the pD_2 estimates were determined by GraphPad Prism ® software version 8.02.

2.4 Experimental protocol

All experiments were conducted on pulmonary arterial rings treated with control, Pb, and Pb combined with QUE. The contraction response was evaluated by adding increasing concentrations cumulatively (10^{-9} M to 3×10^{-4} M) of PE and 5-HT, respectively. The DRCs were studied for: (1) the control untreated arterial rings; (2) the Pb-treated arterial rings; and (3) the Pb plus quercetin-treated arterial rings. The relaxation response was assessed by adding cumulative concentration (10^{-9} M to 3×10^{-4} M) of ACh and SNP, respectively, on the arterial rings precontracted with 5-HT. Concentration-relaxation curves were also constructed for the following groups: (1) untreated arterial rings serving as controls, (2) arterial rings treated with Pb, and (3) arterial rings treated with both Pb and QUE.

2.5 Statistical analysis

The study evaluated the effects of both contractile and relaxing agents on pulmonary arterial rings from goats by assessing the percentage of contraction and relaxation responses, respectively. Graphical representations of mean values along with their standard error (SEM) were created using Microsoft Excel, and calculations were conducted using GraphPad Prism 5 software, version 8.0.2, to determine the concentration at which 50% of the maximum effect occurred (EC_{50} for agonists and IC_{50} for antagonists) for each agent. Geometric means of EC_{50} and IC_{50} values, along with their respective 95% confidence intervals, were provided. Group differences were analyzed using a two-way analysis of variance (ANOVA) followed by Bonferroni multiple comparison tests, performed with GraphPad Prism software, version 8.0.2. The EC_{50} and IC_{50} values are presented as geometric means with their respective 95% confidence limits. Significance was established as $p<0.05$.

3. Results

3.1 Effect of spasmogens and vasorelaxants

Per cent contractile responses of 5-HT and PE (Figure 1) in equilibrated pulmonary arterial rings were plotted with a 0.5 log molar dose difference (from 10^{-9} M to 3×10^{-5} M) to obtain a dose-response curve (DRC). Half maximal effective concentration (EC_{50}) value and affinity (pD_2) were deduced and presented in Table 1. In the 5-HT (10 µM) pre-contracted pulmonary arterial rings, per cent relaxation by ACh and SNP (Figure 1) were plotted with 0.5 log molar dose difference (from 10^{-9} M to 3×10^{-5} M) to obtain DRC. Half-maximal inhibitory concentration (IC_{50}) and pD_2 value were deduced and shown in Table 1. At similar concentrations, ACh *in vitro* caused greater relaxation in pulmonary vasculature than SNP.

3.2 Effect of lead

After incubating pulmonary arterial rings with Pb acetate for 20 min, increased contractions were observed in response to 5-HT and PE compared to untreated rings (Figure 1). Table 1 presents the EC_{50} and pD_2 values. A notable decrease ($p<0.05$) in mean EC_{50} and a leftward shift in the dose-response curve were observed for 5-HT, accompanied by a significant increase ($p<0.05$) in pD_2. However, for PE, there was a non-significant decrease in mean EC_{50} with a leftward shift in the dose-response curve. In contrast, the mean IC_{50} value for the relaxant response of ACh and SNP significantly increased ($p<0.05$) in Pb-treated arterial rings compared to untreated ones, resulting in decreased contractions, decreased pD2, and a rightward shift in the dose-response curve (DRC).

3.3 Effect of quercetin on lead-treated pulmonary vasculature

The addition of QUE to the Pb-treated tissue resulted in a decrease in contraction caused by contractile agents, *viz.*, 5-HT and PE and right shift of DRCs. Looking at EC_{50} and pD_2 values of 5-HT and PE in Table 1 suggests a vasorelaxant effect of QUE in Pb-treated arterial rings. Similarly, the presence of QUE in addition to Pb-treated rings significantly enhanced relaxation by ACh and SNP (Figure 1) than absence in Pb-treated rings, as visible at various concentrations with a left shift of curves between per cent relaxation and concentration drawn between the two treatments. There was a significant ($p<0.05$) decrease in mean IC_{50} for the response of ACh in the presence of QUE in Pb-treated arterial rings than in its absence. Similarly, we observed a significant ($p<0.05$) decrease in mean IC_{50} for relaxation caused by SNP in the presence of QUE in Pb-treated arterial rings than its absence in tissue (Table 1).

Table 1: Comparative EC_{50}/IC_{50} and pD_2 values for different agents in the absence or presence of quercetin (10 µM), in lead-treated pulmonary arterial rings of goats

S.No.	Treatment groups (dose from 10^{-9} M to 3×10^{-5} M)	EC_{50} / IC_{50} values of the agent with a 95 per cent confidence limit range	pD_2 estimate
1.	5-HT in the absence of lead	$(7.359 \pm 1.19) \times 10^{-6}$ M	5.13 ± 0.08
2.	5-HT in the presence of lead	$(9.140 \pm 0.33) \times 10^{-7}$ M$^{\#}$	6.03 ± 0.01*
3.	5-HT in the presence of lead with quercetin	$(1.631 \pm 0.68) \times 10^{-6}$ M$^{\$}$	5.78 ± 0.28
4.	PE in the absence of lead	$(5.953 \pm 1.16) \times 10^{-6}$ M	5.22 ± 0.12
5.	PE in the presence of lead	$(3.164 \pm 1.09) \times 10^{-6}$ M	5.49 ± 0.11
6.	PE in the presence of lead with quercetin	$(1.167 \pm 0.40) \times 10^{-5}$ M	4.93 ± 0.19
7.	ACh in the absence of lead	$(2.445 \pm 4.00) \times 10^{-8}$ M	7.61 ± 0.60
8.	ACh in the presence of lead	$(5.258 \pm 2.03) \times 10^{-7}$ M*	6.27 ± 0.12
9.	ACh in the presence of lead with quercetin	$(4.730 \pm 0.75) \times 10^{-8}$ M$^{\#}$	7.32 ± 0.69
10.	SNP in the absence of lead	$(2.764 \pm 0.46) \times 10^{-6}$ M	5.56 ± 0.07
11.	SNP in the presence of lead	$(2.800 \pm 0.49) \times 10^{-6}$ M*	5.55 ± 0.09
12.	SNP in the presence of lead with quercetin	$(9.993 \pm 6.37) \times 10^{-7}$ M$^{\#\$}$	6.01 ± 0.40

Given values are presented as Mean ± SEM. EC_{50} /IC_{50} data underwent analysis through two-way ANOVA, followed by Bonferroni's multiple comparisons post-hoc test. For the pD_2 estimate, analysis was conducted via one-way ANOVA, and this one-way ANOVA followed by Tukey's multiple comparisons post-hoc test.Top of Form*p<0.05 Agent in the absence of lead vs Agent in the presence of lead, $^{\#}p$<0.05 Agent in the presence of lead vs Agent in the presence of lead acetate with quercetin, $^{\$}p$<0.05 Agent in absence of lead vs Agent in presence of lead with quercetin.

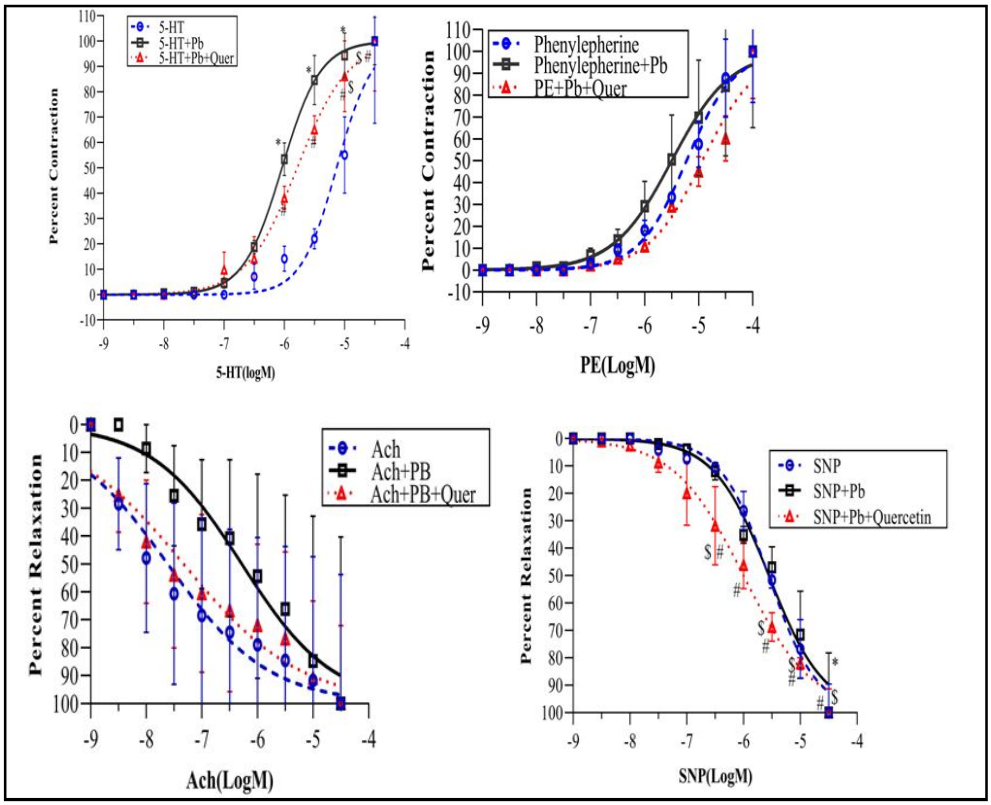

Figure 1: Comparative Per cent contractile/Relaxation response curves (mean ± SEM) of exponentially ascending concentrations in lead-treated pulmonary arterial rings of goats in the presence and absence of quercetin for different agents. Vertical error bars over scatter plot points represent SEM. Data underwent analysis using two-way ANOVA, followed by Bonferroni's multiple comparisons post-hoc test. [*p<0.05 for normal vs Normal + Pb, #p<0.05 in Normal vs Normal + Pb + Quer, $$p$<0.05 in Normal+ Pb + Quer vs Normal + Pb.]

4. Discussion

Numerous studies have demonstrated the relationship between exposure to Pb and the development of hypertension condition in both humans and animals. (Sharp *et al*., 1987; Sharp *et al*., 1988). Experimental acute exposure of Pb in rats intravenously gives a significant increase in systolic arterial pressure after 60 min (Simões *et al*., 2011).

In rats, subjecting them to experimental low-dose Pb exposure for a short duration results in damage to the pulmonary vasculature. This observed harm correlates with heightened oxidative stress and a specific endothelial modulation affecting the tone of the resilient pulmonary artery (Covre *et al*., 2016).

In addition to its chief toxic effect, several previous tissue bath studies report the contractile effect of Pb on several smooth muscles of various laboratory animal models (Sopi *et al*., 2009; Valencia *et al*., 2001). A study on rat aorta vascular smooth muscle reports that Pb acts like calcium in its contractile mechanism (Valencia *et al*., 2001). Pb induced dose-dependent contraction in dog tracheal smooth muscle and potentiated the effect of bronchoconstrictors like bradykinin (Sopi *et al*., 2009). Pb aerosol exposure increased hyperreactivity in guinea pig tracheal smooth muscle to ovalbumin (Boskabaddy and Farkhondeh, 2013). Also, Pb dose-dependently increases contraction in rat thoracic aorta (Karimi *et al*., 2002; Simões *et al*., 2011; Valencia- Hernandez *et al*., 2001) and reduced nitric oxide production or its availability at biophase were suggested as its cause (Simões *et al*., 2011). Such findings were also reported this effect in rabbit thoracic aorta (Valencia- Hernandez *et al*., 2001), and rat tail artery (Silveira *et al*., 2010). The present study finds a clear vasoconstrictive effect of Pb incubation *in vitro* on goat pulmonary arterial rings. Changes were noticed in the nitric oxide (NO) and cyclooxygenase pathways in the aortic rings of rats exposed to Pb, suggesting a possible contribution of vasoconstrictors derived from the endothelium (Karimi *et al*., 2002).

Quercetin, an extensively studied flavonol (Truzzi *et al*., 2021), is a common dietary component found in numerous fruits. It is known to possess various pharmacological effects, including a smooth muscle relaxant effect. QUE demonstrated a notable vasorelaxant effect on endothelium-intact isolated cerebral basilar artery rings pre-contracted with 60 mM KCl (Yuan *et al*., 2018). It's antioxidant and metal-chelating abilities stem from the numerous hydroxyl groups in its chemical composition and the presence of conjugated electrons (Flora *et al*., 2012).

In the present study, it was observed that Pb acetate increased the affinity of 5-HT and PE by 0.90 and 0.27 log units, respectively as compared to that of normal pulmonary arterial rings of goats, suggesting that Pb increased the contractile nature of rings. Our results corroborate the findings of other investigators (Inneh and Ebeigbe, 2016; Zhang *et al*., 2005), as per their findings, it was reported that Pb has the potential to directly enhance the contraction of rat aortal rings to 5-HT and rabbit aortic rings to PE, respectively. The enhanced contraction caused by Pb is endothelium-dependent. The heightened production of $O_2^{\bullet -}$ induced by Pb could potentially correlate with the amplified contractile reaction to 5-HT. (Zhang *et al*., 2005). When the Pb acetate-treated rings are incubated in QUE, there is a decrease in the affinity of 5-HT by 0.25 log and PE by 0.56 log units as compared to Pb-treated pulmonary arterial rings of goat, suggesting a vasorelaxant effect of QUE. Reduction in 5-HT and PE-induced contractions in QUE-treated arterial rings was also reported by other investigators (Ajay *et al*., 2006; Choi *et al*, 2016; Yuan *et al*., 2018). The affinity of ACh is decreased by 1.34 log units in Pb - treated rings as compared to that of normal pulmonary arterial rings of goats. Lead acetate has a significant inhibitory effect on relaxation responses induced by acetylcholine in both endothelium-intact rabbit aortic rings and rat aortic rings (Inneh and Ebeigbe, 2016; Zhang *et al*., 2009). After treatment with QUE, the affinity of ACh is increased by 1.05 log units in Pb acetate-treated pulmonary arterial rings of goats. Previous studies also suggested the relaxant effect of QUE in rat aortic rings (Ajay *et al*., 2006; Khoo *et al*., 2010; Li *et al*., 2012). Lead acetate decreased the affinity of SNP by 0.006 log units as compared to that of normal pulmonary arterial rings of goats. Relaxation to SNP was reduced in Pb-treated rats compared to control (Marques, 2001). Quercetin treatment increased the affinity of SNP by 0.46 log units. Quercetin enhanced cyclic GMP-dependent relaxations to SNP (Suri *et al*., 2010).

5. Conclusion

In conclusion, Pb shows contraction in the pulmonary artery of goats and potentiates contraction by other endogenous spasmogens. Higher blood Pb levels can exacerbate the progression of hypertension, cardiovascular diseases, and pulmonary arterial hypertension (PAH) and increase the risk of cardiovascular mortality. By vasorelaxation and attenuation of contractile response of Pb and other spasmogens on the pulmonary artery of goats, quercetin shows promise as an alternative to partially alleviate the toxic effect of Pb. It paves the way for further studies and exploration of its therapeutic potential for Pb-induced hypertension.

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Conflict of interest

The authors declare no conflict of interest relevant to this article.

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