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SYSTEMATIC REVIEW ARTICLE

Dose-related Effects of Resveratrol in Different Models of Pulmonary Arterial Hypertension: A Systematic Review

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Abstract: ***Background*:** Pulmonary Arterial Hypertension (PAH) is a severe and progressive disease of pulmonary arterioles. This pathology is characterized by elevation of the pulmonary vascular resistance and pulmonary arterial pressure, leading to right heart failure and death. Studies have demonstrated that resveratrol possesses a protective effect on the mechanisms related to the genesis of the PAH-induced by different models.

***Objective*:** This study aimed to investigate the dose-related effects of resveratrol in different models of pulmonary arterial hypertension.

***Methods*:** To identify eligible papers, we performed a systematic literature search on Scielo, PubMed, and Scholar Google. The research was limited to articles written in English in the last 10 years. We used the following descriptors to search: Pulmonary Arterial Hypertension and Resveratrol, OR Resveratrol, and Animal models of Pulmonary Arterial Hypertension, OR Resveratrol, and *in vitro* models of Pulmonary Arterial Hypertension.

***Results*:** 1724 studies were identified through the descriptors used, fifty-five studies with different models of pulmonary arterial hypertension were selected for the full review, forty-four were excluded after application of exclusion and inclusion criteria, totalizing eleven studies included in this systematic review.

***Conclusion*:** The results showed that resveratrol, at low and high doses, protects in a dose-dependent manner against the development of PAH induced through monocrotaline, normoxia and hypoxia models. In addition to having chemopreventive, anti-inflammatory, antioxidant and anti-proliferative properties. In the case of PAH-related myocardial injury, resveratrol protects cells from apoptosis, thus working as an antiapoptotic agent.

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1. INTRODUCTION

Pulmonary Arterial Hypertension (PAH) is a progressive disease with hemodynamic consequences characterized by an increase in pulmonary vascular resistance, leading to right ventricular overload and right ventricular failure [1-3]. It is a pathological condition common to a variety of etiologies; however, its pathophysiological mechanisms are not well known [4-6].

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The average pulmonary artery pressure is 14±3 mmHg [7], PAH is defined as (mPAP) equal to or exceeding 25 mmHg according to An official American Thoracic Society clinical practice guideline: diagnosis, risk stratification, and management of pulmonary hypertension of sickle cell disease [8]. It can also be observed that the right ventricular function is compromised when there is an acute increase in the pressure in the pulmonary artery with levels greater than 40 mmHg. However, when presented chronically, it can determine the adaptations of right ventricular function up to levels close to 60 mmHg without impairment of function [9-11].

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Epidemiological data of this disease differ in the literature. However, it is possible to highlight values of 15-26 cases per million inhabitants per year in the world, affecting predominantly women with a female to male ratio of 1.7:1, and the onset of symptoms mainly after the second or third decade of life [12, 13]. Highlight also a high prevalence in collagen diseases, with 15% of individuals presenting PAH frequently associated with systemic sclerosis and in the masonic schistosomiasis responsible for one-third of the cases of PAH in Brazil, more commonly reported in patients with the hepatosplenic form [14-17].

PAH has been hemodynamically classified as pre-capillary, post-capillary, or combined pre- and post-capillary since there may be overlaps among these conditions [18]. The first is defined by the concomitant presence of mPAP >20 mmHg, pulmonary arterial wedge pressure (PAWP) ≤15 mmHg, and pulmonary vascular resistance (PVR) ≥3 Wood Units (WU). This group comprises the majority of the etiopathogenic factors of the PAH [19]. The second group is characterized when the pulmonary capillary pressure exceeds 15 mmHg with preserved ejection fraction. This category includes left heart diseases, systolic, or diastolic left ventricular dysfunction, which can lead to heart failure [20]. This latter group may present with isolated post-capillary PAH or combined post-capillary PAH with a pre-capillary component, as indicated by an elevated diastolic pressure gradient and an increased PVR [18]. Because of this, alterations can be observed between the different forms of PAH such as vascular remodeling, including the cellular proliferation of the intima and media, endothelial dysfunction, increased vasoconstriction and activation of inflammatory processes [21-26].

Some mechanisms of the pathophysiology of the PAH are still not fully elucidated. The current therapies used to treat this disease include the use of drugs that act, promoting a decrease in pulmonary artery resistance and inducing vasodilatation. The three different pathways used are selective endothelin receptor antagonist, nitric oxide pathway as a selective inhibitor of phosphodiesterase type 5 and guanylate cyclase stimulant, and prostacyclin pathway by the adenosine monophosphate [21, 27-29].

Like this, the use of in vivo and in vitro models that replicate the developmental processes of this pathology and allow the evaluation of new substances in the treatment of PAH have been performed. Among these substances is the resveratrol, a polyphenol that acts with antioxidant and anti-inflammatory properties, modulator of endothelial function, inhibitor of vascular smooth muscle cell proliferation [30-32], and regulator of the expression of factors responsible for activation, induction or even in the control of genes involved in the vasoconstriction [9, 12, 33].

Thus, this review article aimed to investigate the effects related to the dose of resveratrol in different models of pulmonary arterial hypertension.

2. METHODS

2.1. Systematic Review Strategies

A systematic review has been carried out to summarize the available results of experimental studies. To identify eligible papers, we performed a systematic literature search on Scielo, Pubmed, and Scholar Google. The research was limited to articles written in English in the last 10 years; thus, papers published between April 2009 and April 2019 were included. We use the following descriptors to search: Pulmonary Arterial Hypertension and Resveratrol, OR Resveratrol, and animal models of Pulmonary Arterial Hypertension, OR Resveratrol, and *in vitro* models of Pulmonary Arterial Hypertension. Besides, scanning of reference lists was performed from the retrieved studies to identify any articles that may have been missed from the literature search. The studies included in this review were selected based on the inclusion and exclusion criteria presented in Fig. (**1**).

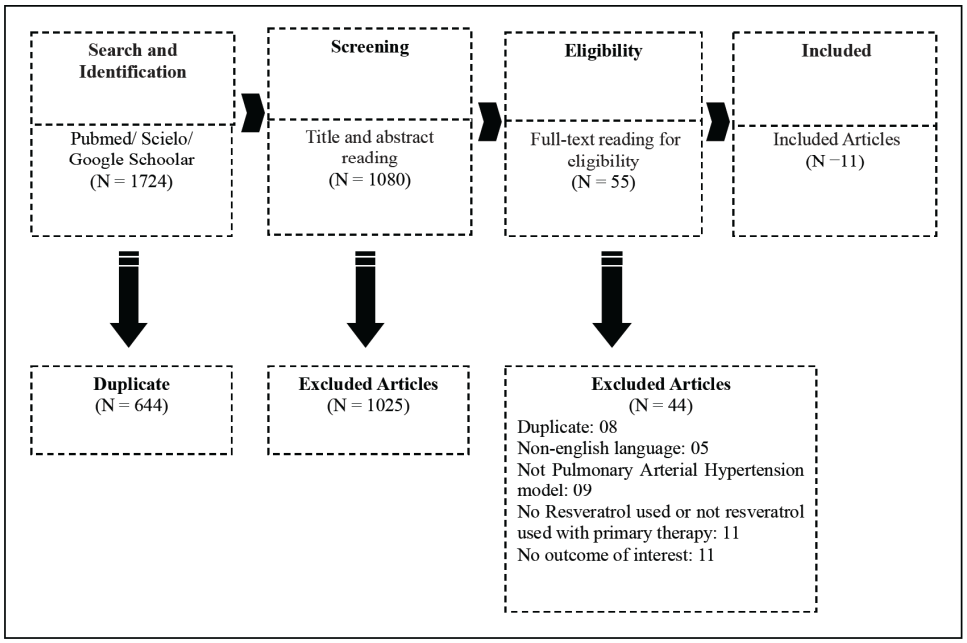

Fig. (1). Flow diagram of search strategy and study selection.

2.2. Study Selection Criteria

To select only the relevant studies, the titles and abstracts of all the citations identified by the bibliographic research were examined. For this purpose, all those studies were included that investigated the specific topic of resveratrol use as primary therapy and aspects related to pulmonary arterial hypertension in studies that involved in vivo and in vitro models.

For characterization aspects of included studies, the following criteria were adopted as the most important: species, strain, sex or cell culture, age, weight, number of control and treated animals, Pulmonary Arterial Hypertension model, Resveratrol dose, drug administration, follow-up in weeks and aim of the study. Then all the data needed for the characterization were organized and presented in tabular form. The data that were not identified in the studies were filled out with n.e (not evaluated).

3. RESULTS

Of the 1724 studies identified through a systematic review of the literature, fifty-five studies with different models of pulmonary arterial hypertension were identified for the full considerations. Eight duplicate articles were excluded, five were not in English, nine were not Pulmonary Arterial Hypertension model, eleven with no use of Resveratrol or not used as primary therapy, and eleven studies did not have the outcome of interest. Total eleven studies were included and characterized as shown in Table **1**. The main results of the studies included in this review are evidenced in Table **2**.

Table 1. Characteristics of studies included in the systematic review.

Author (Year)	Species, Strain, Sex or Cell Culture	Age (Weight)	No. of Control Animals	No. of Treated Animals	PAH Model	Resveratrol Dose	Drug Administ.	Timing of Resveratrol Therapy	Followup (Weeks)	Aim
Csiszar *et al.* [35] (2009)	Rats, Sprague-Dawley, Males.	Adults (300g).	Control: 14 or 21 days; MCT: 14 or 21 days; (n=6, per group).	MCT + RES: 14 and 21 days (n=6, per group).	Monocrotaline- induced PAH (60 mg/kg SC).	25 mg/kg per day	In drinking water.	From day 1 to day 14 or day 21 after MCT injection.	Up to 3 weeks	To investigate the efficacy of resveratrol to present PAH.
Yang *et al.* [29] (2010)	Rats, Sprague-Dawley, Males.	10 weeks old (180-200g).	Control (n=8); MCT (n=12).	MCT + RES: 10 mg/kg MCT + RES: 30 mg/kg (n=8, per group).	Monocrotaline-induced PAH (50 mg/kg SC).	10 or 30 mg⁄kg per day	Intragastric	1 to day 21 after MCT injection.	3 weeks	To investigate the protective effects of resveratrol against right ventricular hypertrophy.
Paffetti, Lucas and Campen [25] (2012)	Rats, Sprague-Dawley, Males.	8-10 weeks old (>300g).	Control Saline or resveratrol; MCT saline or resveratrol (n=8-12, per group)	MCT + RES; MCT + Sildenafil; (n=8-12, per group).	Monocrotaline-induced PAH (50 mg/kg IP).	3 mg/kg per day	In drinking water.	From day 28 to day 42 after MCT injection.	6 weeks	To verify the atrophy pathways in the pulmonary artery after the use of resveratrol.
Paffetti *et al.* [3] (2012)	Rats, Sprague-Dawley, Males.	8-10 weeks old (approximately 300g).	Control; MCT; (n=12, per group).	Saline + RES; MCT + RES; (n=12, per group).	Monocrotaline-induced PAH (50 mg/kg IP).	3 mg/kg per day.	In drinking water.	From day 28 to day 42 after MCT injection.	6 weeks	To assess longitudinal changes in cardiac function, morphology, and perfusion.
Chen *et al.* [30] (2014)	*In vivo:* Rats, Sprague-Dawley, Males. *In vitro:* Human PASMC.	Neonatal 2 days old (n.e).	Vehicle: normoxia or hypoxia; (n= 8–12 per group). Group 0 – Positive control.	Resveratrol: normoxia or hypoxia; (n= 8–12 per group). Group 40 µM Group 80 µM Group 100 µM, in 21% or 1% de O_2.	*In vivo:* Hypobaric hypoxic chamber with barometric pressure maintained at 435 – 5 Torr for 14 days. *In vitro:* Normoxia and hypoxia-induced proliferation in human pulmonary artery smooth muscle cells (incubated in 21% O_2, 5% CO_2, balance N_2 (normoxia) or 1% O_2, 5% CO_2, balance N_2 (hypoxia) for 30 min, 24, 48, or 120hours.	*In vivo:* 100 mg⁄kg per day. *In vitro:* 40, 80, or 100 µM.	Subcutaneous Injections. *In vitro*	From day 1 to day 14 after placed into hypoxic chamber. 30 min, 24, 48, or 120 hours.	2 weeks 1 week	To investigate the effects in the present hypoxia-induced proliferation in human pulmonary artery smooth muscle cells *via* inhibition of arginase II induction.

(Table 1) Contd…

Author (Year)	Species, Strain, Sex or Cell Culture	Age (Weight)	No. of Control Animals	No. of Treated Animals	PAH Model	Resveratrol Dose	Drug Administ.	Timing of Resveratrol Therapy	Followup (Weeks)	Aim
Zhou *et al.* [41] (2015)	Rats, Sprague-Dawley, Males.	Adults (280-300g)	Control; MCT + vehicle; (n=20, per group).	MCT + RES 2,5 mg/kg; MCT + RES 20 mg/kg; (n=20, per group).	Monocrotaline- induced PAH (60 mg/kg SC).	2,5 or 20 mg/kg per day.	Intragastric	From day 1 to day 14 or day 21 after MCT injection.	3 weeks	To examine the effects of resveratrol on cardiac and pulmonary trunk remodeling, and common plasma markers of vascular function.
Wilson *et al.* [26] (2016)	Rats, Sprague-Dawley, Males.	Adults (260±6g).	Control; MCT; (n=12, per group).	MCT + RES (n=12, per group).	Monocrotaline-induced PAH (60 mg/kg SC).	25 mg/kg per day.	Intragastric	From day 1 to day 21 after MCT injection.	3 weeks	To examine the effects of resveratrol on cardiac and pulmonary trunk remodeling, and common plasma markers of vascular function.
Xu *et al.* [18] (2016)	Rats, Sprague-Dawley, Males	Adults (n.e)	Normoxia; Hypoxia; (n=6, per group).	Normoxia + RES; Hypoxia + RES; (n=6, per group).	Normoxia and Hypoxia-induced PAH per 28 days (Normoxia: ambient barometric pressure: 718 mmHg/PO_2 150.6 mmHg; Hypoxia: 380 mmHg/PO_2 79.6 mmHg.	40 mg/kg per day	Intragastric	From day 1 to day 21 after placed into normoxia and hypoxic chamber.	3 weeks	To investigate the effects of resveratrol on HPH development.
Guan *et al.* [39] (2017)	Rats, Sprague-Dawley, Males	50 days old (150-180g)	Positive control (cells + saline + hypoxia).	RES (10, 30 or 100 µmol/l); Resveratrol + LY-294002; Resveratrol + PI3K inhibitor; Resveratrol + IGF-1; Evaluated in 24, 48 and 72hours.	*In vitro:* A hypoxic environment was induced using an autonomous plexiglass chamber supplied with 5% CO_2 and 95% N_2 at 20 ml/min.	Resveratrol (10, 30 and 100 µmol/l).	*In vitro*	24, 48 and 72 hours.	1 week	To investigate the role of resveratrol by examining alterations in expression levels of genes associated with the PI3K/AKT pathways, proliferation, and migration.
Yu *et al.* [31] (2017)	Rats, Wistar, Males.	Adults (200–300g)	Normoxia (n= n.e) Hypoxia (n= n.e).	Hypoxia + RES (n= n.e) Hypoxia + SRT1720 (n= n.e).	*In vivo:* Hypoxia-induced PAH per 21 days (hypoxia involved fractional inspired oxygen of 0.21 and 0.12). *In vitro:* For hypoxic cultivation, an atmosphere of 92% N_2/5% CO_2/3% O_2 was used.	25 mg/kg per day	Intragastric	*In vivo:* From day 1 to day 21 after placed into hypoxic chamber *In vitro:* pulmonary cells were treated at 80% confluence using SRT1720 for 24 or 48 hours.	3 weeks	To investigate the action and potential mechanism of resveratrol on PAH, focusing on the role of SIRT1 (Silent information Regulator 1) in apoptosis of pulmonary artery smooth muscle cells.
Shi *et al.* [28] (2018)	Rats, Sprague-Dawley, Males.	Adults (190–200g)	Control; MCT; (n=8, per group).	MCT and PF543; MCT and PDTC; MCT, PF543 and PDTC; MCT and resveratrol; MCT, resveratrol, and PF543; MCT, resveratrol and PDTC; (n=8, per group).	MCT- induced PAH (60 mg/kg IP).	25 mg/kg per day	Intragastric	From day 1 to day 28 after MCT injection.	4 weeks	To explore the molecular mechanisms underlying SphK1 inducing pulmonary vascular remodeling and RES suppressing PAH.

Note: No: number; PAH: pulmonary arterial hypertension; MCT: monocrotaline; RES: resveratrol; SC: subcutaneous; IP: intraperitoneal; n.e: not evaluated; HPH: hypoxia induced pulmonary hypertension; LY-294002: (2-(4-morpholinyl)-8-phenyl-4H-1-benzopyran-4-one), an inhibitor of phosphoinositide-3 kinase; PI3K: phosphoinositide 3-kinase; IGF-1: insulin-like growth factor 1; AKT: Protein kinase B; SRT1720: activating drug of SIRT1; PF543: the drug that acts as a specific inhibitor of sphingosine kinase 1; PDTC: Pyrrolidine Dithiocarbamate, an anti-oxidant; SphK1: sphingosine kinase 1;

Table 2. Detailed information on effects of the resveratrol *in vitro* and *in vivo* models of pulmonary arterial hypertension.

Author (year)	Cardiovascular Function	Endothelial Function	Inflammatory Markers	Oxidative Markes	Others Variables
Csiszar *et al.* [35] (2009)	Resveratrol treatment from day 1 normalized right ventricular systolic pressure in MCT-injected rats at both the 2- and 3-week periods; Resveratrol treatment prevented MCT-induced RV hypertrophy; Resveratrol treatment normalized medial wall thickness, avoiding the increase in PASMC mass in vessels of MCT-treated rats; The number of PCNA-positive cells was considerably reduced in animals treated with resveratrol.	Increase of small pulmonary arteries relaxation; Improved eNOS expression.	Resveratrol treatment significantly attenuated the mRNA expression of IL-6, IL-1, TNF-α, PDGF α, PDGF β, TGF-β, MCP-1.	Expression of NAD(P)H oxidase subunits were upregulated; Downregulation of NOX-1 and gp91phox gene; Improved eNOS expression.	n.e
Yang *et al.* [29] (2010)	The dose of 30mg/kg showed lower right ventricular hypertrophy, right ventricle mass index, cardiomyocyte length, and cardiomyocyte cross-sectional area among the MCT-induced PAH groups; RV free wall thickness was decreased by 25.26 and 40.67%, in the dose of 10 e 30mg/kg respectively, compared with untreated MCT group; Pulmonary arterial acceleration time showed a significant increase (50.63% and 67.75%, respectively, in the 10 and 30 mg⁄kg) in the resveratrol treated MCT groups; Treatment with 10 and 30 mg⁄kg decreased right ventricular systolic pressure by 26.26% and 37.36%, respectively. There were no significant changes in mean systemic blood pressure among the groups.	n.e	n.e	n.e	After treatment with resveratrol (10 and 30 mg/kg), the morphological structure of the myocardium exhibited less hypertrophy, without fibrosis, and the myofibrilla were lined up; Treatment with resveratrol (10 and 30 mg/kg) significantly improved mitochondrial proliferation, swelling, vacuolization, and medullary sheath-like degeneration, also decreasing sarcoplasmic reticulum and dissolution of the myofilaments, observing still decrease of broken Z-lines and irregular pattern of transverse striations.
Paffetti, Lucas and Campen [25] (2012)	Resveratrol and Sildenafil (used as a clinical reference standard) treatments initiated 28 days after MCT injection caused a significant reduction in right ventricular systolic pressure in MCT-injected rats, with no effect in saline controls; Right ventricular hypertrophy, measured by the RV/LV+S, show decrease only in the resveratrol therapy group; There was no decrease in right ventricular hypertrophy after treatment with resveratrol.	Resveratrol restores acetylcholine-induced relaxation and KCl-induced contractile responses in pulmonary arterial systolic pressure from MCT-pulmonary hypertensive rats.	After day 42 in resveratrol therapy, there were no apparent elevations in circulating interleukins 2, 4, or 6, TNF-α, IFN-γ, MIP-1α, MCP-1, or G-CSF caused by MCT.	Development of MCT-induced pulmonary hypertension is associated with transcriptional down-regulation of atrogin-1, MuRF-1, eNOS and Kv1.5 mRNA expression.	The treatment with resveratrol shows a partial reduction in wall thickness in vessels ranging from 75 to 150μm in diameter.
Paffetti *et al.* [3] (2012)	Resveratrol led to significant attenuation in systolic and mean right ventricular pressures in monocrotaline injected rats; Resveratrol normalized the magnitude of right ventricular contractility (+dP/dtmax) to the level of controls; Resveratrol reversed both right ventricular hypertrophy and pronounced leftward septal deviation in monocrotaline treated rats.	Monocrotaline induced a progressive increase in 99mTc-Annexin binding in lung apoptosis cells compared to saline controls. Resveratrol treatment abrogated this increase in 99mTc-Annexin in monocrotaline-treated rats.	n.e	n.e	n.e
Chen *et al.* [30] (2014)	Hypobaric hypoxia exposure for 14 days increased the right ventricular hypertrophy measured by the RV/(LV+S) ratio. Treatment with 14 days of resveratrol normalized the chronic hypoxia-induced right ventricular hypertrophy in the neonatal rats.	Nitrite levels were measured from the media of normoxic and hypoxic hPASMC treated with resveratrol. Hypoxia significantly decreased nitrite levels, relative to normoxia. However, resveratrol treatment no-showed significant results on nitrite levels.	n.e	n.e	The addition of resveratrol for 48h prevented hypoxia-induced arginase II protein expression in all doses evaluated; Resveratrol did not affect arginase I protein levels in either normoxia or hypoxia; Treatment with resveratrol prevented the hypoxia-induced arginase II mRNA expression after 24h; Resveratrol did not affect arginase I mRNA expression in normoxia or hypoxia; hPASMC proliferation in vitro: hPASMC were evaluated for apoptosis by measuring cleaved caspase 3. There was no difference in cleaved caspase 3 protein expression in normoxia or hypoxia with or without resveratrol treatment; Inhibition of Akt phosphorylation prevents hypoxia-induced arginase II protein expression, result evidenced in all resveratrol doses evaluated.

(Table 2) Contd…

Author (year)	Cardiovascular Function	Endothelial Function	Inflammatory Markers	Oxidative Markes	Others Variables
Zhou *et al.* [41] (2015)	Resveratrol significantly prevented mPAP from increasing in both the 2.5 mg.kg^{-1}d^{-1} group and 20 mg/kg/day groups at both 14 and 21 days; Treatment with resveratrol decreased RVHI in a dose-dependent manner; Resveratrol attenuated the muscularization of intra-acinar arteries; 2.5 mg/kg/day and 20 mg/kg/day of resveratrol attenuated the increase of medial wall thickness.	n.e	n.e	n.e	PDGF-BB (10 ng/mL) treatment decreased expression of p21 but increased cyclin D1 and cyclin E expression in HPASMCs; Flow cytometry analysis showed significant G0/G1 accumulation of hPASMCs with SIRT1 overexpression 12 hours after PDGF-BB treatment; Resveratrol Increases SIRT1 and p21 Expression but Decreases Cyclin D1 Expression in Lungs of MCT-Induced PAH Rats; Resveratrol increased p21 expression but decreased cyclin D1 and cyclin E expressions after PDGF-BB stimulation.
Author (year)	Cardiovascular Function	Endothelial Function	Inflammatory Markers	Oxidative Markers	Others Variables
Wilson *et al.* [26] (2016)	Resveratrol does not alter the weight heart, but the hypertrophy in the right ventricle was adjusted when compare with the control group. Resveratrol treatment was beneficial and reversed total heart surface area to a level observed in control hearts. This reduction in total heart surface area was attributed to a decrease in both total heart muscle area.	n.e	n.e	n.e	Resveratrol was beneficial and significantly reduced tunica media thickness in the pulmonary trunk compared with MCT-treated rats.
Xu *et al.* [18] (2016)	After 28 days of hypoxia exposure, the myocytes of the right ventricle in the hypoxia group were significantly enlarged versus the normoxic groups. The hypoxia exposure also markedly increased RVHI versus the normoxic groups and resveratrol treatment significantly decreased the elevation of RVHI; The chronic hypoxic condition also dramatically increased RVSP versus the normoxic groups, and resveratrol treatment notably decreased the increased RVSP. Similarly, there was no significant difference found in the two normoxic groups.	n.e	Inflammatory factors IL-6, IL-1β, TNF-α, and cytokine VEGF, were all significantly increased in mRNA levels after chronic hypoxia, and all those factors were decreased considerably after resveratrol treatment.	Compared to the normoxic groups, there was a significant elevation of H_2O_2 found in rat lungs after 28 days of hypoxic exposure, and resveratrol treatment significantly reduced its height in a dose-dependent way.	Chronic hypoxia exposure resulted in thickened pulmonary arterial tunica media and accumulated extracellular matrix and resveratrol treatment significantly reduced this process in a dose-dependent way.
Guan *et al.* [39] (2017)	Resveratrol inhibits the development of experimental PAH per hypoxia.	n.e	n.e	n.e	Migration of PASMCs in the resveratrol-treated group was reduced compared with the cells treated with hypoxia, and this effect was dose-dependent, inhibiting hypoxia; Protein expression levels of AKT were increased significantly in the hypoxic group compared with the 10 and 30 μmol/l resveratrol treated groups, and this effect was dose-dependent; Resveratrol inhibits hypoxia-induced proliferation and migration of PASMCs by inhibiting the PI3K/AKT signaling pathway; The protein level of p Akt was significantly suppressed by resveratrol.

(Table 2) Contd…

Author (year)	Cardiovascular Function	Endothelial Function	Inflammatory Markers	Oxidative Markes	Others Variables
Yu *et al.* [31] (2017)	Resveratrol improves right ventricular systolic pressure and mitigate RVHI.	n.e	n.e	n.e	Resveratrol reverses pulmonary vascular remodeling and contributes to the improvement of mitochondrial dysfunction.
Shi *et al.* [28] (2018)	Resveratrol inhibits the development of experimental PAH; The RVSP and right ventricular hypertrophy index decrease in MCT-treated rats after treatment with resveratrol; The combination resveratrol with two agents (resveratrol+PF543 or resveratrol+PDTC) in rats MCT-treated decreased significantly RVSP and RVHI.	n.e	n.e	n.e	The SphK1, cyclin D1, and S1P protein levels were decreased in resveratrol-treated PAH rats, as well as these same variables were decreased after the combination of resveratrol with two agents (resveratrol+PF543 or resveratrol+PDTC).

Note: MCT: monocrotaline; PASMC: artery smooth muscle cells; PCNA: proliferating cell nuclear antigen; eNOS: endothelial nitric oxide synthase; IL: interleukin; TNF-α: Tumor necrosis factor α; PDGF α: α-receptor for platelet-derived growth factor; PDGF β: β-receptor for platelet-derived growth factor; TGF-β: transforming growth factor β; MCP-1: monocyte chemoattractant protein–1; NAD(P)H oxidase: phagocyte nicotinamide adenine dinucleotide phosphate oxidase; NOX-1: hydrogen peroxide; n.e: not evaluated; KCL: potassium chloride; IFN-γ: interferon-γ; MIP-1α: macrophage inflammatory protein-1α; G-CSF: granulocyte colony-stimulating factor; MuRF-1: muscle ring finger 1; Kv1.5: voltage-dependent K+ channel; mRNA: messenger ribonucleic acid; hPASMC: human pulmonary artery smooth muscle cells; mRNA: messenger ribonucleic acid; mPAP: mean pulmonary artery pressure; RVHI: right ventricular hypertrophy index; PDGF-BB: platelet-derived growth factor-BB; SIRT1: silent information regulator 1; PAH: pulmonary arterial hypertension; RVSP: right ventricular systolic pressure; VEGF: vascular endothelial growth factor;; H2O2: hydrogen peroxide; PASMC: pulmonary artery smooth muscle cells; PI3K: phosphoinositide 3-kinase; AKT: Protein kinase B; RVHI: right ventricular hypertrophy index; PF543: drug that act like a specific inhibitor of sphingosine kinase 1; PDTC: pyrrolidine dithiocarbamate, an anti-oxidant; SphK1: sphingosine kinase 1.

4. DISCUSSION

Resveratrol (RES) is a phenolic compound naturally occurring in a variety of plants such as grapes and peanuts [34, 35]. Recent studies showed that RES presents a series of cardioprotective effects, including anti-inflammatory, antioxidant, anti-proliferative properties, as well as anti-atherosclerosis and anti-hypertensive action [5, 30, 36-38]. Furthermore, evidence suggests that RES could attenuate the developed of PAH once this substance acts on pathophysiological mechanisms of the disease such as vascular remodeling, inhibition of proliferation of the intima and media, inhibition of endothelial dysfunction, decreased vasoconstriction and activation of inflammatory processes [5, 30, 39-44]. Thus, this review article aimed to investigate the effects related to the dose of resveratrol in different models of pulmonary arterial hypertension.

The studies presented evidenced significant results on the anti-inflammatory response of RES in myocardial cells as observed at doses of 3 mg/kg, 25 mg/kg, and 40 mg/kg. It is recognized that inflammation is one of the main pathogenic mechanisms in the PAH as well as in other cardiovascular diseases [24, 45]. The inflammatory process in pulmonary arterioles can injure pulmonary arterial endothelial cells, stimulating the proliferation of Pulmonary Arterial Smooth Muscle Cells (PASMC) [22]. However, it is observed that cytokines such as interleukin-1 and interleukin-6, are important mediators of inflammation that will contribute to the development of PAH [46, 47]. Still, we highlight that the dosage 3 mg/kg and 25 mg/kg were used in less than 21 days, both in experimental models induced by monocrotaline [38, 48].

The chemoprotective properties of RES can be observed on oxidative stress parameters that were significantly reduced also at doses of 3 mg/kg, 25 mg/kg, and 40 mg/kg, noting that these results were found in different models of PAH (monocrotaline and normoxia/hypoxia) and the dosage of 40 mg/kg not only reduced the plasma levels of the cytokines but also decreased the expression of their mRNA significantly. In a previous study, it was evidenced that the free radical-scavenging capacity of resveratrol depends on concentration. The author showed that the antioxidant properties of resveratrol are increased with higher levels of this nutraceutical [49].

Anti-proliferative effects of RES in PASMCs were observed at doses of 10 μmol/L, 30 μmol/L, 40 μmol/L, 80 μmol/L, 100 μmol/L, 2.5 mg/kg, 20 mg/kg, and 100 mg/kg. Studies have demonstrated that apoptosis of PASMCs is necessary for the regression of pulmonary vascular changes in the developed of PAH [50, 51]. However, the precise mechanisms related to the proliferation and apoptosis of these cells in the pathogenesis of PAH remain unclear. In this sense, Guan *et al.* (2017) [52] showed that resveratrol might inhibit the hypoxia-induced proliferation and migration of PASMCs, inhibiting the phosphatidylinositol 3-kinase (PI3K) and protein kinase B (AKT) signaling pathway, thus attenuating pulmonary arterial remodeling. It is important to emphasize that the anti-proliferative effects of RES were observed in a relatively low dose (10-100 μmol/L) in a dose-dependent manner.

Regarding right ventricular hypertrophy was observed significant differences at doses of 40 μmol/L, 80 μmol/L, 100 μmol/L, 3 mg/kg, 25 mg/kg, and 100 mg/kg. We were emphasizing that the low doses (40-100 μmol/L) and high dose (100 mg/kg) were effective against hypertrophy in myocardial cells of neonatal rats exposed to hypoxia. The relationship between right ventricular hypertrophy and monocrotaline-induced PAH is well established [53]. However, Zhou *et al*. [54] showed the role of hypoxia in the development of right ventricular hypertrophy and the preven-

tive effect of RES on this parameter. Since, that acute or chronic alveolar hypoxia is a potent stimulus for pulmonary vasoconstriction leading to the proliferation and migration of PASMCs and thickening of the vessel wall, as well as compensatory right ventricular hypertrophy.

The benefits of the cardiovascular function were evidenced in all the studies included in this review. Thus, we observed that in lower doses (10-100 μmol/L) and high doses (2.5-100 mg/kg), RES protects in a dose-dependent manner against the development of PAH-induced through monocrotaline, normoxia, and hypoxia models. Therefore, the authors show improvements in variables such as right ventricular hypertrophy (RVH), right ventricular hypertrophy index (RVHI), right ventricular systolic pressure (RVSP), mean pulmonary artery pressure (mPAP), and right ventricular contractility (+dP/dtmax). In this sense, we observe that such results also benefit the endothelial function, promoting reduction of pulmonary cells apoptosis, an increase of acetylcholine-induced relaxation, and improved endothelial nitric oxide synthase (eNOS) expression [30, 55]. These results demonstrate the efficacy of RES as a primary therapy in the treatment of PAH, promoting cardiovascular benefits through their chemoprotective, anti-inflammatory, antioxidant, and anti-proliferative properties, in different experimental models in a dose-dependent manner.

CONCLUSION

The results observed here showed that resveratrol, in low and high doses, protects PAH-induced through different models, as well as possesses chemoprotective, anti-inflammatory, antioxidant, and anti-proliferative properties. In the case of pulmonary injury related to PAH, resveratrol protects cells from apoptosis, thereby working as an anti-apoptotic agent. Therefore, we can observed that resveratrol presents protective heart action still in a relatively low dose (10-100 μmol/L). Besides, the results indicate that this polyphenol provides cardiovascular benefits, attenuating the development of PAH in a dose-dependent manner.

LIST OF ABBREVIATIONS

PAH	=	Pulmonary Arterial Hypertension
RES	=	Resveratrol
PASMC	=	Pulmonary Arterial Smooth Muscle Cell
MCT	=	Monocrotaline
HPH	=	Hypoxia-induced Pulmonary Hypertension
hPASMC	=	Human Pulmonary Artery Smooth Muscle Cells
mPAP	=	Mean Pulmonary Artery Pressure
RVHI	=	Right Ventricular Hypertrophy Index
RVHI	=	Right Ventricular Hypertrophy Index
RVSP	=	Right Ventricular Systolic Pressure

AUTHORS' CONTRIBUTIONS

ACF, JSS, RD, JMPC, AWSM and ASMV: conducted literature research and wrote the draft of the manuscript. CAADF, CJD and MRQB provided editorial input and revision for the final version. CTM and JOBM: read and approved the final manuscript.

CONSENT FOR PUBLICATION

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CONFLICT OF INTEREST

The authors declare no conflict of interest, financial or otherwise.

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REFERENCES

[1] Boucly A, Weatherald J, Savale L, *et al.* Risk assessment, prognosis and guideline implementation in pulmonary arterial hypertension. Eur Respir J 2017; 50(2): 1700889.

[2] Farber HW, Loscalzo J. Pulmonary arterial hypertension. N Engl J Med 2004; 351(16): 1655-65.

[3] Jayasekera G, Crowe T, Mackenzie A, *et al.* Left atrial function by cardiac MRI in idiopathic pulmonary arterial hypertension (IPAH) and pulmonary hypertension due to heart failure with preserved ejection fraction (PH-HFpEF). In: C106 Money Don't Matter Tonight: Pulmonary hypertension assessment, prognostication, and treatment. American Thoracic Society 2017; pp. A6903-3.

[4] Hoeper MM, Galiè N. Letter by Hoeper and Galie regarding article,"Hemodynamic, functional, and clinical responses to pulmonary artery denervation in patients with pulmonary arterial hypertension of different causes: phase II results from the pulmonary artery denervation-1 study. Circ Cardiovasc Interv 2016; 9(1): e003422.

[5] Paffett ML, Hesterman J, Candelaria G, *et al.* Longitudinal in vivo SPECT/CT imaging reveals morphological changes and cardiopulmonary apoptosis in a rodent model of pulmonary arterial hypertension. PLoS One 2012; 7(7): e40910.

[6] Meloche J, Courchesne A, Barrier M, *et al.* Critical role for the advanced glycation end-products receptor in pulmonary arterial hypertension etiology. J Am Heart Assoc 2013; 2(1): e005157.

[7] Bazavan M, Macaluso GP, Pauwaa S. Understanding Cardiovascular Hemodynamics. In: Ventricular-Assist Devices and Kidney Disease. Springer 2018; pp. 15-32.

[8] Klings ES, Machado RF, Barst RJ, *et al.* An official American Thoracic Society clinical practice guideline: Diagnosis, risk stratification, and management of pulmonary hypertension of sickle cell disease. Am J Respir Crit Care Med 2014; 189(6): 727-40.

[9] Hoette S, Jardim C. Souza Rd: Diagnosis and treatment of pulmonary hypertension: An update. J Bras Pneumol 2010; 36(6): 795-811.

[10] Valerio CJ, Schreiber BE, Handler CE, Denton CP, Coghlan JG. Borderline mean pulmonary artery pressure in patients with systemic sclerosis: Transpulmonary gradient predicts risk of developing pulmonary hypertension. Arthritis Rheum 2013; 65(4): 1074-84.

[11] Siafakas N, Vermeire P. Pride N, *et al.* Optimal assessment and management of chronic obstructive pulmonary disease (COPD). The European Respiratory Society Task Force. Eur Respir J 1995; 8(8): 1398-420.

[12] Barreto AC, Franchi SM, Pereira AC, Lopes AA. Pulmonary arterial hypertension: Physiopathology, genetic aspects and response to the chronic use of sildenafil. Arq Bras Cardiol 2005; 85(2): 147-54.

[13] Ling Y, Johnson MK, Kiely DG, *et al.* Changing demographics, epidemiology, and survival of incident pulmonary arterial hypertension: results from the pulmonary hypertension registry of the United Kingdom and Ireland. Am J Respir Crit Care Med 2012; 186(8): 790-6.

[14] Steendijk P. Right ventricular function and failure: Methods, models, and mechanisms. Crit Care Med 2004; 32(4): 1087-9.

[15] Kovacs G, Maier R, Aberer E, *et al.* Borderline pulmonary arterial pressure is associated with decreased exercise capacity in scleroderma. Am J Respir Crit Care Med 2009; 180(9): 881-6.

[16] Lapa M, Dias B, Jardim C, *et al.* Cardiopulmonary manifestations of hepatosplenic schistosomiasis. Circulation 2009; 119(11): 1518.

[17] Graham BB, Bandeira AP, Morrell NW, Butrous G, Tuder RM. Schistosomiasis-associated pulmonary hypertension: Pulmonary vascular disease: the global perspective. Chest 2010; 137(6): 20S-9S.

[18] Opitz CF, Hoeper MM, Gibbs JSR, *et al.* Pre-capillary, combined, and post-capillary pulmonary hypertension: A pathophysiological continuum. J Am Coll Cardiol 2016; 68(4): 368-78.

[19] Simonneau G, Gatzoulis MA, Adatia I, *et al.* Updated clinical classification of pulmonary hypertension. J Am Coll Cardiol 2013; 62(25)(Suppl.): D34-41.

[20] Scalia GM, Scalia IG, Kierle R, *et al.* ePLAR—the echocardiographic pulmonary to left atrial ratio—a novel non-invasive parameter to differentiate pre-capillary and post-capillary pulmonary hypertension. Int J Cardiol 2016; 212: 379-86.

[21] McLaughlin VV, Archer SL, Badesch DB, *et al.* ACCF/AHA 2009 expert consensus document on pulmonary hypertension: a report of the American College of Cardiology Foundation Task Force on expert consensus documents and the American Heart Association developed in collaboration with the American College of Chest Physicians; American Thoracic Society, Inc.; and the Pulmonary Hypertension Association. J Am Coll Cardiol 2009; 53(17): 1573-619.

[22] Chen Y, Jowett S, Barton P, *et al.* Clinical and cost-effectiveness of epoprostenol, iloprost, bosentan, sitaxentan and sildenafil for pulmonary arterial hypertension within their licensed indications: a systematic review and economic evaluation. In: NIHR Health Technology Assessment programme: Executive Summaries. NIHR Journals Library 2009.

[23] Zha LH, Zhou J, Li TZ, *et al.* NLRC3 inhibits MCT‐induced pulmonary hypertension in rats *via* attenuating PI3K activation. J Cell Physiol 2019.

[24] Vendrov AE, Lozhkin A, Madamanchi NR, Runge MS. Nox4 deletion attenuates age-associated vascular inflammation and atherosclerosis burden in hyperlipidemic mice by modulating macrophage phenotype. Circulation 2018; 138(1): A17163-3.

[25] Dorfmüller P, Perros F, Balabanian K, Humbert M. Inflammation in pulmonary arterial hypertension. Eur Respir J 2003; 22(2): 358-63.

[26] Budhiraja R, Tuder RM, Hassoun PM. Endothelial dysfunction in pulmonary hypertension. Circulation 2004; 109(2): 159-65.

[27] Ramos RP, Ferreira EVM, Arakaki JSO. Estratégias do tratamento da hipertensão arterial pulmonar. Pulmão RJ 2015; 24(2): 71-7.

[28] Rich S, Brundage BH. High-dose calcium channel-blocking therapy for primary pulmonary hypertension: Evidence for long-term reduction in pulmonary arterial pressure and regression of right ventricular hypertrophy. Circulation 1987; 76(1): 135-41.

[29] Singh TP, Rohit M, Grover A, Malhotra S, Vijayvergiya R. A randomized, placebo-controlled, double-blind, crossover study to evaluate the efficacy of oral sildenafil therapy in severe pulmonary artery hypertension. Am Heart J 2006; 151(4): e851-5.

[30] Xu D, Li Y, Zhang B, *et al.* Resveratrol alleviate hypoxic pulmonary hypertension *via* anti-inflammation and anti-oxidant pathways in rats. Int J Med Sci 2016; 13(12): 942.

[31] Mishra V, Shuai B, Kodali M, *et al.* Resveratrol treatment after status epilepticus restrains neurodegeneration and abnormal neurogenesis with suppression of oxidative stress and inflammation. Sci Rep 2015; 5: 17807.

[32] Szkudelska K, Szkudelski T. Resveratrol, obesity and diabetes. Eur J Pharmacol 2010; 635(1-3): 1-8.

[33] Rich S, Brundage BH, Levy PS. The effect of vasodilator therapy on the clinical outcome of patients with primary pulmonary hypertension. Circulation 1985; 71(6): 1191-6.

[34] Chan CC, Cheng LY, Lin CL, Huang YH, Lin HC, Lee FY. The protective role of natural phytoalexin resveratrol on inflammation, fibrosis and regeneration in cholestatic liver injury. Mol Nutr Food Res 2011; 55(12): 1841-9.

[35] Tian W, Yang L, Liu Y, *et al.* Resveratrol attenuates doxorubicin-induced cardiotoxicity in rats by up-regulation of vascular endothelial growth factor B. J Nutr Biochem 2020; 79: 108132.

[36] Berman AY, Motechin RA, Wiesenfeld MY, Holz MK. The therapeutic potential of resveratrol: A review of clinical trials. NPJ Prec Oncol 2017; 1(1): 35.

[37] de Sá Coutinho D, Pacheco MT, Frozza RL, Bernardi A. Anti-inflammatory effects of resveratrol: Mechanistic insights. Int J Mol Sci 2018; 19(6): 1812.

[38] Paffett ML, Lucas SN, Campen MJ. Resveratrol reverses monocrotaline-induced pulmonary vascular and cardiac dysfunction: a potential role for atrogin-1 in smooth muscle. Vascul Pharmacol 2012; 56(1-2): 64-73.

[39] Wilson DN, Schacht SE, Al-Nakkash L, Babu JR, Broderick TL. Resveratrol prevents pulmonary trunk remodeling but not right ventricular hypertrophy in monocrotaline-induced pulmonary hypertension. Pathophysiology 2016; 23(4): 243-50.

[40] Kwon Y-J, Chung T-H, Shim J-Y, Lee Y-J. The association of pulse pressure with metabolic syndrome in Korean elderly: A nationwide population-based study. Diabetes Res Clin Pract 2017; 123: 75-81.

[41] Shi W, Zhai C, Feng W, *et al.* Resveratrol inhibits monocrotaline-induced pulmonary arterial remodeling by suppression of SphK1-mediated NF-κB activation. Life Sci 2018; 210: 140-9.

[42] Yang DL, Zhang HG, Xu YL, *et al.* Resveratrol inhibits right ventricular hypertrophy induced by monocrotaline in rats. Clin Exp Pharmacol Physiol 2010; 37(2): 150-5.

[43] Chen B, Xue J, Meng X, Slutzky JL, Calvert AE, Chicoine LG. Resveratrol prevents hypoxia-induced arginase II expression and proliferation of human pulmonary artery smooth muscle cells *via* Akt-dependent signaling. Am J Physiol Lung Cell Mol Physiol 2014; 307(4): L317-25.

[44] Yu L, Tu Y, Jia X, *et al.* Resveratrol protects against pulmonary arterial hypertension in rats *via* activation of silent information regulator 1. Cell Physiol Biochem 2017; 42(1): 55-67.

[45] Florentin J, Coppin E, Vasamsetti SB, *et al.* Inflammatory macrophage expansion in pulmonary hypertension depends upon mobilization of blood-borne monocytes. J Immunol 2018; 200(10): 3612-25.

[46] Humbert M, Monti G, Brenot F, *et al.* Increased interleukin-1 and interleukin-6 serum concentrations in severe primary pulmonary hypertension. Am J Respir Crit Care Med 1995; 151(5): 1628-31.

[47] Steiner MK, Syrkina OL, Kolliputi N, Mark EJ, Hales CA, Waxman AB. Interleukin-6 overexpression induces pulmonary hypertension. Circ Res 2009; 104(2): 236-44.

[48] Csiszar A, Labinskyy N, Olson S, *et al.* Resveratrol prevents monocrotaline-induced pulmonary hypertension in rats. Hypertension 2009; 54(3): 668-75.

[49] Gülçin İ. Antioxidant properties of resveratrol: A structure–activity insight. Innov Food Sci Emerg Technol 2010; 11(1): 210-8.

[50] Mandegar M, Fung Y-CB, Huang W, Remillard CV, Rubin LJ, Yuan JX-J. Cellular and molecular mechanisms of pulmonary vascular remodeling: Role in the development of pulmonary hypertension. Microvasc Res 2004; 68(2): 75-103.

[51] Akagi S, Matsubara H, Nakamura K, Ito H. Modern treatment to reduce pulmonary arterial pressure in pulmonary arterial hypertension. J Cardiol 2018; 72(6): 466-72.

[52] Guan Z, Shen L, Liang H, *et al.* Resveratrol inhibits hypoxia-induced proliferation and migration of pulmonary artery vascular smooth muscle cells by inhibiting the phosphoinositide 3-kinase/protein kinase B signaling pathway. Mol Med Rep 2017; 16(2): 1653-60.

[53] Steven S, Oelze M, Brandt M, *et al.* Pentaerythritol tetranitrate in vivo treatment improves oxidative stress and vascular dysfunction by suppression of endothelin-1 signaling in monocrotaline-induced pulmonary hypertension. Oxid Med Cell Longev 2017; 2017: 4353462.

[54] Zhou S, Li M-T, Jia Y-Y, *et al.* Regulation of cell cycle regulators by SIRT1 contributes to resveratrol-mediated prevention of pulmonary arterial hypertension. BioMed Res Int 2015; 2015: 762349.

[55] Hsieh T-C, Wu JM. Unraveling and trailblazing cardioprotection by resveratrol: Actionable targets and mechanisms of resveratrol. in: resveratrol: State-of-the-Art science and health applications. 2018; pp 348.