



Changes of respiratory system in mice exposed to PM_{4.0} or TSP from exhaust gases of combustion of cashew nut shell



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ABSTRACT

Air pollution is a topic discussed all over the world and the search for alternatives to reduce it is of great interest to many researchers. The use of alternative energy sources and biofuels seems to be the environmentally safer solution. In this work, the deleterious effects on the respiratory system of mice exposed to PM_{4.0} or TSP, present in exhaust gases from the combustion of CNS were investigated, through data from respiratory system mechanics, oxidative stress, histopathology and morphometry of the parenchyma pulmonary. The results show changes in all variables of respiratory system mechanics, in oxidative stress, the histopathological analysis and lung morphometry. The results provide experimental support for epidemiological observations of association between effects on the respiratory system and exposure to PM_{4.0} or TSP from CNS combustion exhaust gases, even at acute exposure. It can serve as a basis for regulation or adjustment of environmental laws that control the emissions of these gases.

1. Introduction

Environmental problems resulting from atmospheric pollution are mainly caused by the use of fossil fuels as an energy source. By providing less aggression to the environment, the use of alternative sources or biofuels in the energy matrix has been listed as a worldwide solution to reduce air pollution. The use of a cleaner energy matrix is an alternative to guarantee economic development with less effects on the environment and human health. Following this trend, the renewable energy sector increased between 15% and 55% per year between 2005 and 2012 (REN 21, 2013), highlighting the use of biomass as a source for energy production, with the highest potential of Growth in the following years (WEC, 2010).

Brazil is one of the largest agricultural producers in the world and has great potential for the production of residual biomass. The utilization in the energy matrix of the residual biomass originated from processes such as sugarcane, rice, or cashew nut processing stand out as a promising alternative to the environmentally correct reuse of these biomasses before disposal. The energy utilization of residual products from the processing of cashew nut, such as cashew nuts shell (CNS), is already a reality in some industries. However, the adverse health effects of exposure to exhaust gases from the CNS combustion process are not

completely explained, and are the subject of this study.

The cashew tree (*Anacardium occidentale* L.) is a tropical plant, native to Brazil and dispersed in almost all of its territory (Severino, 2008). Cashew tree is among the most cultivated fruit trees, standing out in the socioeconomic context of some underdeveloped tropical regions, around the globe, for the high nutritional and commercial value of its products, and also for the intensive work, whose production and Industrialization guarantee income for the population, making this culture an important source of jobs (Ramos et al., 2011).

The processing of the cashew nut starts by separating the commercially valuable almond from the CNS, which holds great potential for fuel (Lima, 2008). The separation occurs by heating at temperatures near 800 °C in tanks containing 10% of the liquid extracted from the CNS itself.

Since the disposal of CNS in landfills is not recommended due to its high volume of production, several processes to exploit CNS's energy potential have been investigated in order to find increasingly economical and environmentally sustainable routes (Alcocer et al., 2015). A promising alternative is the use of CNS as a biofuel in the feed of boilers during the processing of cashew nuts. The high calorific value of CNS (22.48 MJ/kg) is higher than other residual biomasses such as: rice straw (16.35 MJ/kg) and sugarcane bagasse (18.61 MJ/kg). However,

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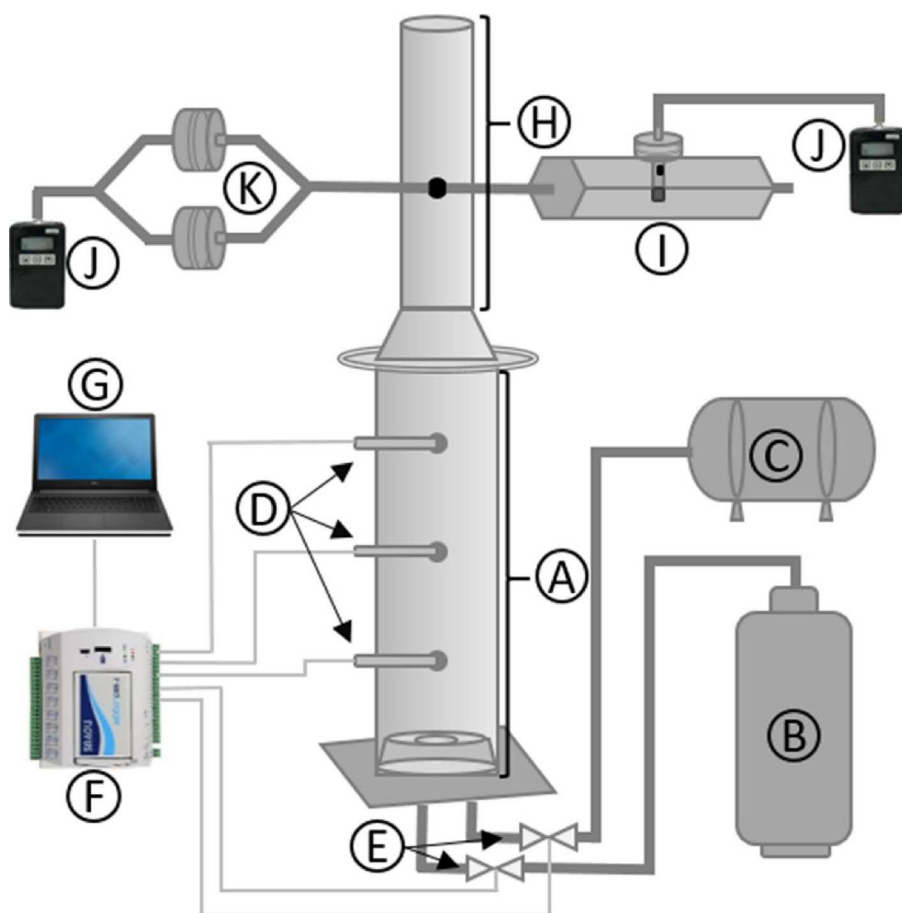


Fig. 1. Combustion system and collection of filters with $PM_{4.0}$ or TSP from combustion exhaust gases from CNS. A – Biomass combustion reactor; B-LPG; C – Air compressor; D – Thermocouples; E – Flow transducers; F – Data acquisition system (Fieldlogger); G – Notebook; H – Chimney; I – Cyclone chamber for collecting filters with $PM_{4.0}$; J – Suction pump; K – System for collection of TSP.

this requires the use of filters to contain toxic emissions and pollutants exhausted from exhaust gases from their combustion (EMBRAPA, 2003).

In view of the above, there is an urgent need to evaluate the effects of CNS's combustion emissions on health. This information may provide greater security in its use as biofuel. In this work, the deleterious effects on the respiratory system of mice exposed to particulate matter with aerodynamic diameter less than 4 micrometres ($PM_{4.0}$) and total suspended particles (TSP) present in CNS combustion exhaust gases, are investigated. To conduct this research, analyzes of the mechanics of the respiratory system, oxidative stress, histopathology and morphometry of the pulmonary parenchyma, and the chemical analysis of the composition of the particles coming from this combustion were performed.

2. Materials and methods

2.1. Collection of $PM_{4.0}$ and TSP

A CNS combustion system was developed to collect $PM_{4.0}$ and TSP from its exhaust gases (Fig. 1). For this collection, the CNS's (400 g) were first placed in a cylindrical stainless steel burner (Fig. 1A). Then, the initial combustion ignition of CNS was performed by supplying liquefied petroleum gas (LPG-Figure 1B) and ambient air from an air compressor (Fig. 1C). The combustion process of the CNS was accompanied by thermocouples (Fig. 1D) and flow transducers (Fig. 1E) connected to a data acquisition system (FieldLogger-Figure 1F) for the analysis and control of temperature and LPG and air flows (Unpublished data), transferring the information to a notebook (Fig. 1G).

The exhaust gases generated by CNS combustion were directed by a chimney (Fig. 1H) to a chamber containing a coupled cyclone (SKC®-Aluminum Cyclone – Fig. 1I), which selects for a glass fiber filter

(0.8 μm of porosity and 37 mm in diameter), only $PM_{4.0}$ particles. The cyclone system was fed with a suction pump (AirChek® XR5000 – Fig. 1J) using a flow of 2.5 L/min. To collect the TSP filters, the flue gases were directed to a system containing 2 glass fiber filters (Fig. 1K), where the flow of 5 L/m was maintained by another suction pump (AirChek® XR5000 – Fig. 1J).

2.2. Identification and quantification of polycyclic aromatic hydrocarbons (PAHs) and carbonyl compounds associated with $PM_{4.0}$ and TSP

The $PM_{4.0}$ and TSP filters from the CNS combustion exhaust gases collected were extracted with 200 mL of the acetone:hexane (1:1) mixture for 10 h. After extracting the PAHs, the extract was concentrated to a final volume of 1.0 mL using a spin evaporator (Buchi, Flawil, Switzerland). PAHs analysis was performed by high performance liquid chromatography according to thermo scientific application note 20572 adapted from EPA 610 (Thermo Scientific, 2012) using a Varian ProStar HPLC with diode arrangement detector (DAD), hypersil green PAH column (25 cm $\times$ 4.6 mm $\times$ 5 μm), wavelength 225 nm, mobile phase CH_3CN/H_2O at a flow rate of 1.25 mL/min under elution gradient. Individual concentrations of the 16 PAHs were estimated by the external standard method using a PAH calibration mix (10 $\mu g \cdot mL^{-1}$ in acetonitrile) purchased from Sulpeco (Bellefonte, PA, USA).

The carbonyl compounds (aldehydes and ketones) were collected and analyzed according to the EPA method TO-11A. LpDNPH S10da Cartridges of supelco analytical containing 350 mg of high purity silica gel coated with DNPH were used in *in situ* adsorption and derivatization of the carbonyl compounds. Elution of the carbonyl compounds was accomplished by the passage of acetonitrile through the cartridges, the percolation occurring by gravity. The procedure was performed with

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