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Indoor air pollution in rural north-east India: Elemental compositions, changes in haematological indices, oxidative stress and health risks

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ABSTRACT

Chronic smoke exposure, emitted by biomass fuel burning leads to many diseases, which are originated due to oxidative stress. The present study investigated the levels of $PM_{2.5}$, PM_{10} and $PM_{2.5}$ bound trace metals released during cooking with fuelwood and subsequent changes in haematological parameters along with oxidative stress in rural tribal women of northeast India exposed to wood smoke. The levels of $PM_{2.5}$, PM_{10} and trace metals associated with $PM_{2.5}$ (nickel, cobalt, manganese, zinc, cadmium, lead and copper) were measured. In addition, blood samples were analyzed for concentrations of different blood related parameters (haemoglobin, platelet count, red blood cells and white blood cells) and levels of antioxidants (reduced glutathione, superoxide dismutase, and catalase). Plasma malondialdehyde (MDA) was measured as a biomarker of lipid peroxidation. Health risk assessment was done to assess the potential risk posed by inhalation of fine particles emitted from cooking with fuel wood. Levels of both $PM_{2.5}$ and PM_{10} were higher in wood users compared to LPG users during cooking period (644.4 ± 368.3 μg/m^3 vs 50 ± 23.8 μg/m^3; 915 ± 441.3 μg/m^3 vs 83.3 ± 33 μg/m^3) and it exceeded the permissible limits of WHO. Levels of trace metals during the cooking period in fuel wood users were significantly higher than LPG users ($p = 0.01$). After controlling possible confounders, both platelet count and white blood cells (WBC) had a significant positive association with $PM_{2.5}$and PM_{10}. Similarly, haemoglobin had a negative association with both $PM_{2.5}$ and PM_{10}. Depleted levels of antioxidant enzymes and increase in lipid peroxidation (MDA) suggest a close association with pollutants released from wood smoke, indicating oxidative stress in women who used fuelwood for cooking. The total hazard quotient (HQ) of 0.11 was within the acceptable limit (i.e., 1.0). The total excess lifetime cancer risk (ELCR) was 5.4×10^{-6} which is five times higher than the acceptable limit of 1.0×10^{-6}. Individual carcinogenic risk of Ni (2.3×10^{-6}) and Cd (3.1×10^{-6}) were also higher compared to acceptable limit. These results indicate that tribal women cooking with wood are at greater risk of developing cancer and also give support to the positive association between wood smoke and oxidative stress.

1. Introduction

Considering worldwide distribution, 2.8 billion people rely on solid fuels, in the form of wood, dung, crop residues, charcoal, coal etc. for cooking (World Health Organisation, 2014). According to Census of India 2011, 86.7% households in rural areas and 26.3% households in urban India depend on solid biomass as the primary source of energy for cooking (TERI, 2014). Type of fuel, ventilation in the kitchen, proximity to the stove/cooker and the amount of time spent in cooking are key parameters for exposure to indoor air pollution from cooking (Pant et al., 2016). Furthermore, indoor air pollution from combustion of biomass fuel has emerged as a potential risk to health. Studies provides evidence to link indoor air pollution due to biomass fuel combustion through an increased occurrence of respiratory infections (such as pneumonia, tuberculosis and chronic obstructive pulmonary disease), low birth weight, cataracts, cardiovascular morbidity and mortality, and mortality both in adults and children (Bruce et al., 2002; Fullerton et al., 2008; Pérez-Padilla et al., 2010).

It is not easy to burn biomass fuels in simple household cookstoves due to the need of intricate premixing of fuel and air for proper burning. Using these cookstoves actually resulted in emission of harmful pollutants such as carbon monoxide (CO), sulphur dioxide (SO_2), nitrous oxides, respirable particulate matter ($PM_{2.5}$ and PM_{10}), polycyclic aromatic hydrocarbon (PAH), formaldehyde and metals (Bruce et al., 2002; Sehgal et al., 2014; Zhang and Smith, 2007) that are injurious to health. According to WHO Air Quality Guidelines for household fuel

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combustion interim target 1 (IT-1), permissible limit for $PM_{2.5}$ and PM_{10} are 35 μg/m^3 and 50 μg/m^3, while the concentrations of particulate matter in households, which used biomass fuel, far exceeded those of WHO permissible limits. A study in Kenya reported that PM_{10} concentration was as high as 2000 μg/m^3 in households using biomass fuels (Ezzati and Kammen, 2001). A similar study in South India reported high concentrations of respirable particulate matter in biomass fuel using households during cooking time that ranged from 500 to 2000 μg/m^3 (Balakrishnan et al., 2002). Particulate matter is responsible for oxidative stress and trigger activation of antioxidant defense, inflammation, and toxicity in cells (Li et al., 2008). Oxidative stress induced by particulate matter is responsible for causing many degenerative and non-degenerative diseases like pulmonary diseases, cardiovascular diseases and Alzheimer's disease (Lodovici and Bigagli, 2011). Trace metals present in wood smoke can induce oxidative stress through the formation of reactive oxygen species (ROS) as superoxide ion, hydrogen peroxide, and hydroxyl radical which subsequently leads to DNA damage, lipid peroxidation, and protein modification (Jomova and Valko, 2011; Stohs and Bagchi, 1995). These are precursors of diseases like cancer, cardiovascular disease, diabetes, atherosclerosis, neurological disorders (like Alzheimer's and Parkinson's disease) and chronic inflammation. Further, the International Agency for Research on Cancer (IARC) has classified many metals as carcinogenic in nature viz. Cd and Ni has been classified as Group 1 carcinogens. Studies have shown that household burning of solid biomass fuels emit many transition metals that exceed the acceptable limits posing a serious threat to human health (Matawle et al., 2016; Pervez et al., 2012).

Therefore, burning of solid biomass fuel in households is a major source of indoor particulate pollution that poses a serious health risk for the exposed population. Studies on the status of indoor air pollution from biomass fuel combustion and its subsequent health effects in north-eastern India are scarce. Most studies, conducted in India, were in Southern and Northern India and a few in Eastern India (Ansari et al., 2010; Balakrishnan et al., 2004; Nayek and Padhy, 2017; Pandey, 2012). In addition, research data on the effects of particulate matter and particulate matter bound trace metals from wood smoke on haematological parameters and antioxidants are very few. Therefore, the present study aims to investigate the effects of particulate matter and particulate matter bound trace metals on haematological parameters and antioxidants status of rural tribal women of northeast India exposed to wood smoke. Additionally, health risk from exposure to trace metals in particulate matter was also assessed.

2. Material and methods

2.1. Study area and subjects

This study was conducted in Assam, a north-eastern state in India, from October to December 2014. Three adjacent tribal villages namely, Bhaiskhuli (26°06′35.86″N, 90°36′35.45″E), Kuruwabhasha (26°06′10.11″N, 90°36′47.87″E) and Goraimari (26°05′48.45″N, 90°37′39.21″E) were selected which are located approximately 8 km from the district headquarter Goalpara in Assam. All the three villages are similar in socio-economic backgrounds and dietary habits. People from the Rabha tribe inhabit these three villages. Agriculture is the main occupation for them where wood and barks of the tree are the primary source of energy for cooking. These people collect fuelwood from the nearby reserve forest and use traditional cookstoves made of mud and bricks. Most of the households have a separate kitchen, without any proper ventilation. The only ventilation was a small space between the roof and the wall. A control group, for comparison, was considered from Goalpara town area as people using LPG in the villages were rare. The control group featured those households where only LPG was in use as a fuel for cooking purpose.

A total of 120 non-smoker healthy tribal women from these three villages participated in this study along with 80 non-smoker healthy women from the control group. A questionnaire based survey was conducted to collect data on demographic information, socioeconomic status, education, kitchen characteristics and cooking history. Cooking index (CI) was calculated by multiplying years of exposure to an average number of hours of exposure per day. Only 30 women each from the study and control group gave their permission for blood collection. Ethical clearance was obtained from the Institutional Ethical Committee of Visva-Bharati University. Informed consent was obtained from the participants before conducting data and sample collections.

2.2. Blood collection

Venous blood (5 ml) was collected by the help of a phlebotomist who was hired for the job, in tubes containing K_2EDTA as anticoagulant after obtaining informed consent from the participants. A portion of the sampled blood was used for determining routine haematological parameters. Another portion of the sampled blood was used for separating plasma and erythrocytes. The blood was centrifuged at 2500 g for 15 mins at 4 °C. Plasma and erythrocytes were separated and stored at −70 °C until analysis, while the buffy coat was discarded. Erythrocyte lysate or haemolysate was used to determine the levels of catalase (CAT), superoxide dismutase (SOD) and reduced glutathione (GSH), while plasma was used for analyzing malondialdehyde (MDA). Erythrocyte lysate was prepared by washing the erythrocytes three times with 0.9% saline water and then lysed by addition of cold isotonic saline (pH 7.4). Then it was centrifuged at 3000 g for 15 min at 4 °C and the haemolysate obtained was stored at −70 °C until analysis.

2.3. Measurement of haematological indices

Routine haematological parameters such as haemoglobin (Hb), differential count of white blood cell (WBC) and red blood cell (RBC), and platelet count were measured following the standard procedures (Bain et al., 2011).

2.4. Measurement of oxidative stress and lipid peroxidation

2.4.1. Malondialdehyde (MDA)

Levels of MDA, a product of lipid peroxidation, were measured spectrophotometrically in blood plasma using the double heating method by Draper and Hadley (1990). Plasma (0.5 ml) was mixed with 2.5 ml 10% (w/v) trichloroacetic acid and kept in boiling water bath for 15 min. After cooling at room temperature, the mixture was centrifuged at 1000 g for 10 min. Then 2 ml of the supernatant was transferred to a test tube containing 0.67% (w/v) of thiobarbituric acid (TBA). The tube was again placed in boiling water bath for 15 min. After cooling to room temperature the absorbance was measured at 532 nm using a spectrophotometer. Results were expressed in nmol/ml plasma.

2.4.2. Reduced glutathione (GSH)

Erythrocyte GSH was determined as per the method by Kuo et al. (1983). TCA (0.2 ml) was added to 0.4 ml of haemolysate and centrifuged at 3000 g for 15 mins at 4 °C. Then, 0.5 ml of supernatant from it was added to 2 ml Na_2HPO_4 (3 M, pH 8) and 0.5 ml solution of 0.04% 5,5`-dithiobis (2-nitrobenzoic acid) (DTNB) in 10% sodium citrate. The absorbance was then measured at 412 nm to calculate the GSH levels. GSH level is expressed in nmol/gHb.

2.4.3. Superoxide dismutase (SOD)

The activity of superoxide dismutase (SOD) in erythrocyte was checked using the method by Beauchamp and Fridovich (1971), which is based on the inhibition of nitrobluetetrazolium (NBT) reduction. Washed erythrocyte of 0.1 ml was treated with 0.25 ml chloroform and 0.5 ml ethanol and then centrifuged at 3000 g for 60 min at 4 °C. Then, 0.025 ml of the supernatant was collected and added to a mixture containing 0.2 ml 0.1 M EDTA, 0.1 ml 1.67×10^{-4} M NBT, 2.29 ml