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Tradescantia pallida as a biomonitoring tool to assess the influence of vehicle exhaustion and benzene derivatives

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Air pollutants affect their quality, making, inappropriate, harmful and damaging to health in general. The air quality can be measured by the use of higher plants as bioindicators in environmental biomonitoring. In this study, we evaluated the increased frequency of micronuclei (MN) in *Tradescantia pallida* exposed to potentially toxic environments. The vegetables were implanted in five points (4 test and 1 control) for 15, 30 and 60 days. After this period of exposure, the young inflorescences were analyzed and the observation frequency of MN using the technique of *Tradescantia-micronucleus* (TRAD-MCN). The results showed an increased frequency of MN in pollen grains of *T. pallida* in environment with severe vehicular exhaust and manipulation of benzene derivatives, indicating that the compounds present in the atmosphere of these environments have genotoxic potential.

Key words: Atmospheric pollutants, bioindicators; genotoxicity, pollen grains, *Tradescantia pallida*.

INTRODUCTION

According to Conama Resolution (n° 3) 28/06/1990, smog refers to the presence of compounds or substances that promote interference in the air quality. The air quality standards, primary and secondary, suffer direct

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interference from vehicular exhaust, considered the main cause of air pollution. Volatile organic compounds, particulate materials, nitrogen oxides, carbon and sulfur, and benzene derivatives, such as polycyclic aromatic hydrocarbons (PAHs) cause damage to the environment, affecting the flora, fauna, and people, for making improper air, inconvenient, harmful and detrimental to health in general (Pereira et al., 2013; Inomata et al., 2015).

The chemical modification air quality standard can be measured through the use of bioindicators, plant or animal beings, characterized by the use in biomonitoring of air quality (Billet et al., 2015). As a result of, the plants are inexpensive, easy to use and cultivation, have shown as the most effective means of an assessment and response quality of the environment, presenting practical use mainly related to air pollution control (Rodríguez et al., 2015).

The variation in biological monitoring techniques is sufficient to encompass the use of various species, believed to be active biological monitors (Güez et al., 2012; Ávila et al., 2013). The use of superior plants such as angiosperms, in biomonitoring studies has grown sharply (Santos, 2015). The *Tradescantia pallida*, is popularly known as tetrapoeraba-roxa. It is an ornamental plant widely distributed in Brazil, as it has a great adaptability to climate variations. It is a small angiosperm in the Commelinaceae family for presenting genetic characteristics favorable to studies, such as large chromosomes, it is often used as a test system in biomonitoring (Carvalho, 2005; Crispim et al., 2012).

One of the parameters for biomonitoring is the comparative analysis of the structural pattern of leaves with anatomical features arising from the exposure to a particular agent. These features can be macro or microscopic, as the decrease in leaf size and stomata, organelles responsible for gas exchange (Mott et al., 2014).

Other trials from *Tradescantia* are cytogenetic tests, related to the analysis of the cellular genome. Two of these tests are widely accepted on the observation of somatic mutations or changes in the structure of chromosomes: The test with analysis of mutations in stem hairs (Trad-SHM) and bioassay of MN in pollen grains of stem cells (Trad-MCN) (Carvalho, 2005; Savóia et al., 2009; Andrade-Vieira et al., 2011).

Thus, this study aimed to evaluate the toxic effects by increasing frequency of micronuclei (MN) in *T. pallida* exposed to few zones in the city of Teresina, Piauí, Brazil.

MATERIALS AND METHODS

T. pallida was identified by the Herbarium Graziela Barroso - TPB (Federal University of Piauí) with the voucher number 28339. Then 50 seedlings of *T. pallida* were cultivated in small pots with fertile soil and regular irrigation. We distributed the plants in 4 points in the city of Teresina - Piauí, during the period of August to October in 2012. It is because, August to October is the dry season with more dust, temperature, and vehicle exhausts in this state of Brazil. The municipality is located on the right bank of the Parnaíba River and occupies a total area of 1,809 km^2, with geographical coordinates: 05º05'12" south latitude and 42º48'42" west longitude (Branco, 2003).

The areas selected for the exhibition were: a petrol filling station (5°04'12.54"S, 42°81'26.22"W), an auto body repair of vehicles (5°11'21.47"S, 42°79'13.78"W), responsible for handling solvents and coating materials, to be submitted to exhaust environments of automotive vehicles and handling benzene derivatives; a via of access, avenue Frei Serafim (5°08'37.82"S, 42°79'75.33"W), and a vehicle parking in an institution of higher education (5°06'89.23"S, 42°74'70.64"W); by having heavy automobile traffic, thus being subjected to oxidation fuel and large amounts of particulate materials (Figure 1). In addition to the 4 test points, 1 was also exposed to an area with low frequency of urban pollution, operating as a negative control (NC) (5°11'73.89"S, 42°70'98.56"W).

After the cultivation, 10 seedlings were sent for display in each of their previously cited environments, and they were weekly watered with deionized water. Plant structures used in laboratory analysis were young inflorescences (Figure 2), where pollen grains appear more abundantly on tetrad stage. The inflorescences withdrawals were carried in aqueous solution (for maintenance adequate humidity and integrity) to the laboratory and analyzed in the range of periods: 15, 30 and 60 days after implantation. This procedure related to frequency of MN found in pollen grains of stem cells with the time of exposure to clastogenic effects of pollutants being studied.

Once young inflorescences were removed, they were fixed in methanol: acetic acid (3:1) for 24 h and stored in ethanol (70%). The flower bud clumps were dissected out and macerated in glass slide for staining acetic carmine (2%) and further heating to about 60°C for impregnating the dye. In an optical microscope at magnification of 400 X, 3000 tetrads were viewed for each exposure period. The occurrence frequency of MN was obtained from the ratio between the number of MN in 300 tetrads for each exposure place at all points, including the NC, in each time period described above, and follows the method described by Andrade-Vieira et al. (2011).

Statistical analysis was performed using GraphPad Prism (version: 5.0). values are mean ± standard deviation (SD). Analysis of variance (ANOVA) followed by Bonferroni and Dunnett's test, considering $p<0.05$, $p<0.01$ and $p<0.001$.

RESULTS

The results allowed the increase in the frequencies of MN in *T. pallida*, from exposure to gaseous pollutants originating from the vehicle combustion and benzene derivatives action in periods of 15, 30 and 60 days of exposure as shown in Figures 3 to 5.

Figure 6 shows all points with the NC, total period of 60 days, allowing an analytical correlation between the genotoxic influence of air pollutants, absorption time and accumulation of these air pollutants by plants and environments more significance for induction of MN.

The median frequency of MN in *T. pallida* obtained by tetrads counts 3000 per point in each period demonstrated great variance as compared to the NC as shown in Table 1. Exposure times were compared to determine the presence or absence of relevant statistics on the test points as shown in Table 2.

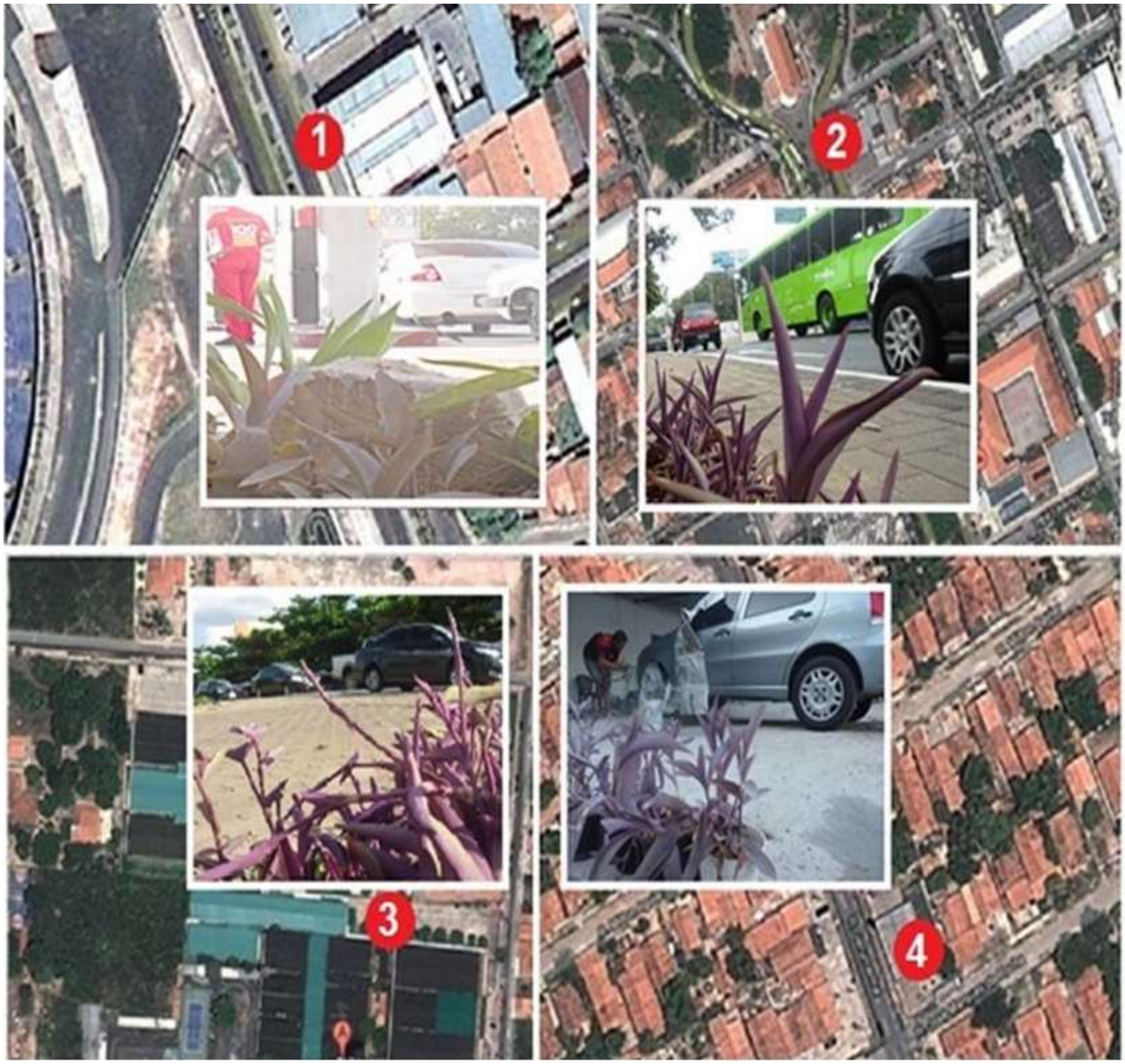

Figure 1. *Tradescantia pallida* exposure places in the city of Teresina: fuel refueling point (point 1), access way, avenue Frei Serafim (point 2), vehicle parking lot of a Higher Education Institution (point 3), and workshop manipulation of coating materials and solvents (point 4).

DISCUSSION

The plants are more sensitive to pollution than animals and man. Thus, they favor subsidies and an easier study of the effects of pollutants in the environment (Klumpp et al., 2006; Maioli et al., 2008). Chemical and physical agents exert influence vastly on the rate of change of biochemical activities in plants, thus increasing the frequency of changes in DNA molecules has been widely studied (Carvalho, 2005).

Frequency of MN showed elevated in all points when compared to the NC group. According to Figure 1, although the increase in the percentage of this type of chromosomal damage was not significant in points 1 and 3 within 15 days after implantation of *T. pallida*, points 2 and 4 had high statistical significance.

Low MN rates found in sections 1 and 3, respectively petrol station and parking, are justified by the reduced exposure to vehicle exhaust, the main cause of air pollution. Since the high frequency of MN in points 2 and 4, respectively an avenue of the great movement of vehicles and a garage, it is justified by the greater presence of toxic compounds due to the intense vehicular exhaust and direct manipulation of benzene derivatives in these environments.

The amount of carrier flow and handling content of compounds derived from benzene led to a wide variability in the results between the test points. The tests, as well

Figure 2. Young inflorescence of *Tradescantia pallida* (white arrow).

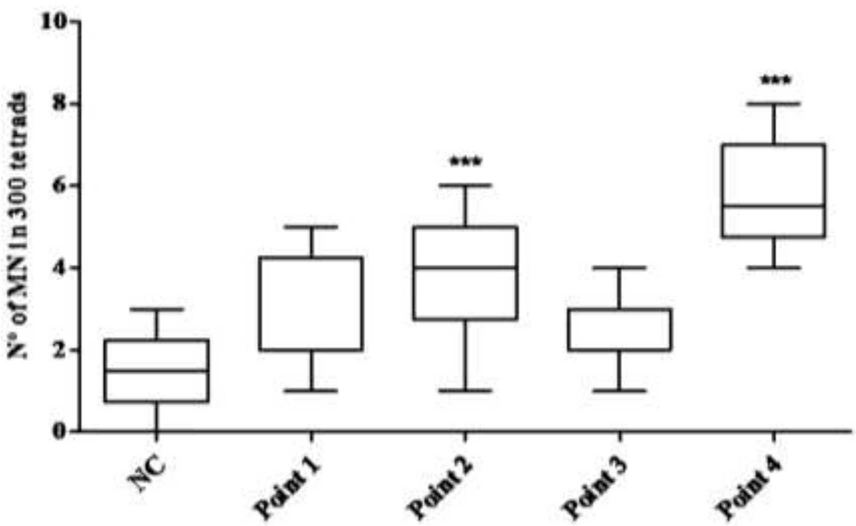

Figure 3. Frequency the micronuclei (MN) in (mean ± SD) of young *T. pallida* inflorescences after 15 days of seedlings exposure to local testing and control test. ANOVA followed by Dunnett's multiple comparison test; frequency of MN at significance of $p<0.001$*** compared to the NC.

literature showed a direct and proportional relation between the significant number of chromosomal damage observed in *T. pallida* and elevation in the presence of these toxic gases, according to Figures 2 to 4 (Gábelová et al., 2004).

It was evident that local with exceeding the flow of vehicles has higher clastogenic effects and aneugenic indices correlated to Table 1. The longer period of exposure to polluting compounds induced an increase in the percentage of genetic damage of *T. pallida* as is

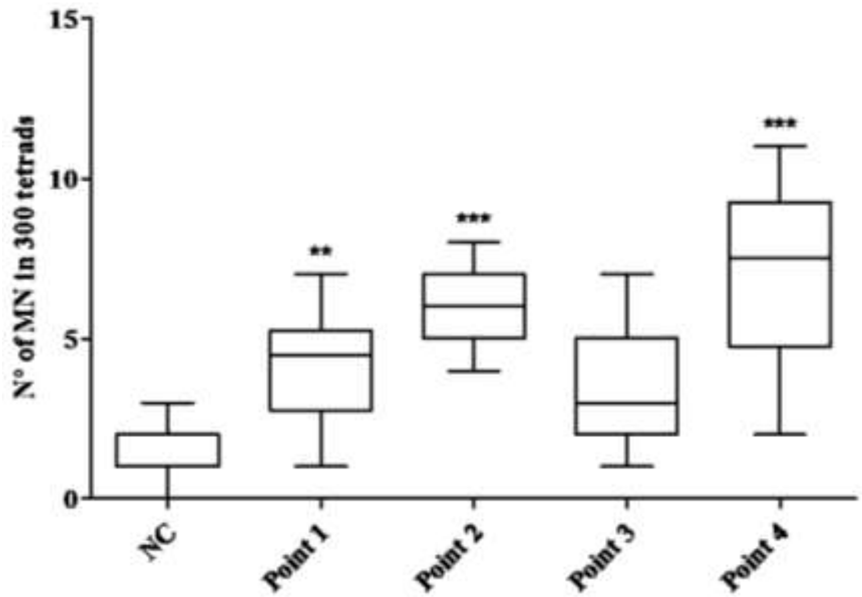

Figure 4. Frequency the (MN) in 300 tetrads (mean ± SD) of young *T. pallida* inflorescences after 30 days of seedlings exposure to local testing and control test. ANOVA followed by Dunnett's multiple comparison test; frequency of MN at significance of p<0.001** and p<0.001*** compared to the NC.

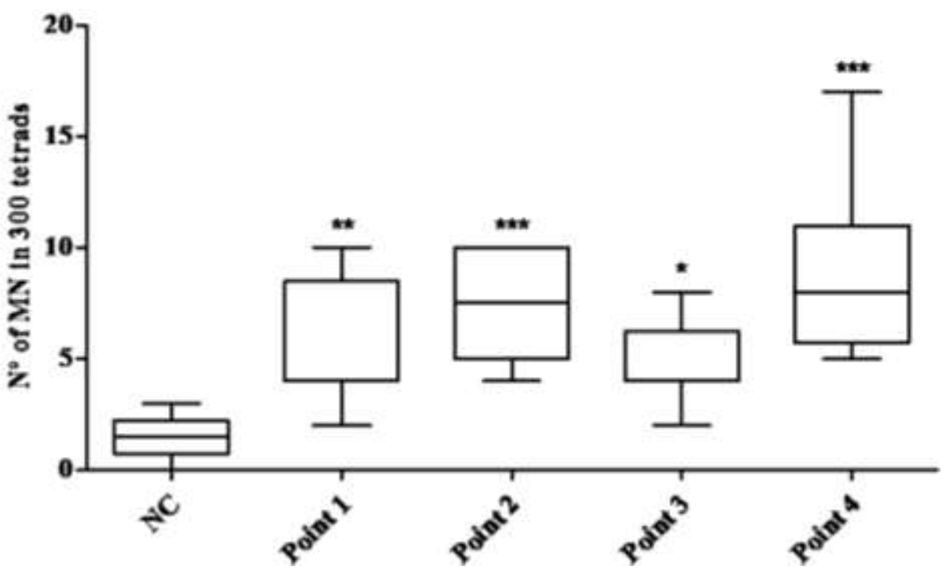

Figure 5. Frequency the (MN) in 300 tetrads (mean ± SD) of young *T. pallida* inflorescences after 60 days of seedlings exposure to local testing and control test. ANOVA followed by Dunnett's multiple comparison test; frequency of MN at significance of p<0.01** and p<0.001*** compared to the NC.

shown in Table 2. In the 15 days analysis only point 4 was significant in relation to the other, as in the analysis of 30 days was observed that point 2 became significant to the point 4. In consonance, the analysis to 60 days of exposition shown that all the points has become significant according to the increasing on MN.

Points 2 and 4 showed high relevance, the fact can be proven by the index and type of pollutants in these places is more intense. What determines the influence of the amount and the constant movement of vehicles in point 2

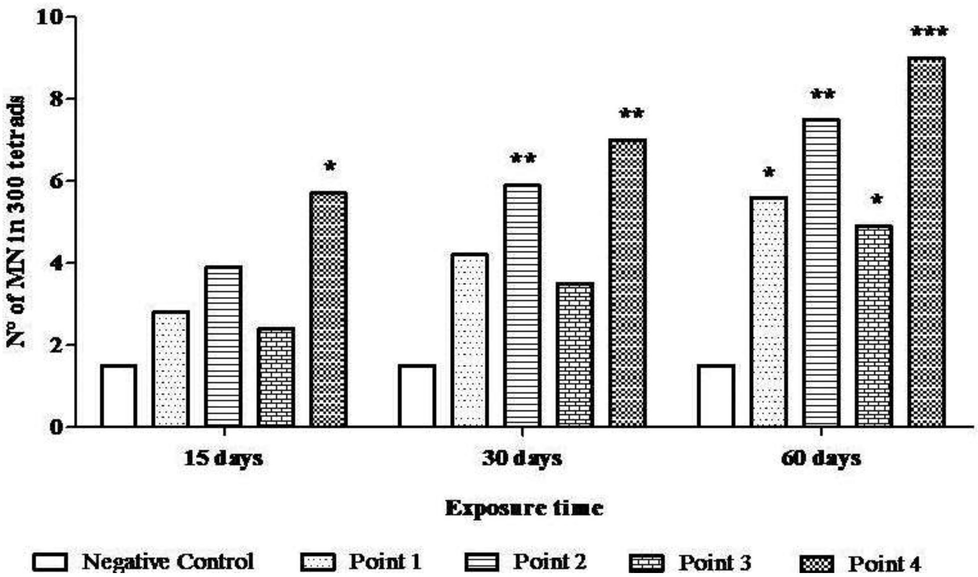

Figure 6. Frequency of MN in 300 tetrads of young (mean ± SD).*T. pallida* inflorescences in all periods of exposure, 15, 30 and 60 days. Two-way ANOVA followed by Bonferroni *post-hoc* test; comparison between the exposure days and points at p<0.05*, p<0.01** and p<0.001*** compared to the NC.

Table 1. Micronuclei (MN) formation in tetrads of young *T. pallida* inflorescences in all periods of exposure, 15, 30 and 60 days.

Point	15 days	30 days	60 days
	MN (3000 tetrads)	MN (3000 tetrads)	MN (3000 tetrads)
NC	1.54 ± 1.08 (0.0 - 3.0)	1.54 ± 1.08 (0.0 - 3.0)	1.54 ± 1.08 (0.0 - 3.0)
Point 1	2.8 ± 1.39 (1.0 - 5.0)	4.2 ± 0.81** (1.0 - 7.0)	5.6 ± 2.79** (2.0 - 10.0)
Point 2	3.9 ± 1.52 *** (1.0 - 6.0)	5.9 ± 1.19 *** (4.0 - 8.0)	7.5 ± 2.27 *** (4.0 - 10.0)
Point 3	2.4 ± 0.84 (1.0 - 4.0)	3.5 ± 1.78 (1.0 - 7.0)	4.9 ± 1.79 * (2.0 - 8.0)
Point 4	5.7 ± 1.33 *** (4.0 - 8.0)	7.0 ± 2.78 *** (2.0 - 11.0)	9.0 ± 3.9 *** (5.0 - 17.0)

Directed using ANOVA and Dunnett's multiple comparison test. Mean ± standard deviation (SD) with a significance of 15-days p<0.001*** in points 2 and 4; for 30 days of p<0.01** in point 1 and p<0.001*** in points 2 and 4; and 60-days p<0.05* in point 3, p<0.01** in point 1 and p<0.001*** in points 2 and 4 in the test group, compared to the NC.

is the constant manipulation of benzene derivatives at point 4, that influence the expressively increase in MN frequency.

The frequency of MN found at points with higher vehicular traffic is according to records of Carreras et al. (2006) and Klumpp et al. (2006) who observed a direct relationship between mutagenic index and expressively increase of population concentration in urban areas. Metropolitan areas tend to present more atmospheres contaminated by pollutants than areas with less flow of vehicles and people, leading to a higher incidence of genetic damage in plants used as bio-indicators.

The data cited by Carreras et al. (2006) from a study with the same focus, using 3 test points in the city of Cordoba, Argentina, showed values between 2.4 ± 2.08 in distant area from the center and 4.2 ± 2.6 in region central city, revealing lower rates in inducing genetic damage in *T. pallida* compared with our study. The results showed mean values of 5.6 ± 0.7 and 7.1 ± 1.0, equating to the average percentage of this study, since the highest percentage found in sections that had only the pollutant factor of vehicle exhaust it was 7.5 ± 2.27, as shown in Table 1.

In a study, Costa and Droste (2012) suggested that rainfalls may decrease the genotoxic impacts in *T. pallida*. For this reason, we targeted dry season in this study to avoid the rain effects on this plant. Our study confirms the genotoxic effects in *T. pallida* at the points exposed in the city of Teresina.

It is noteworthy that during the analysis of *T. pallida* other structural changes in the cells of pollen grains were identified. These findings understood increased formation

Table 2. Influence of exposure time to compare the percentage of micronuclei (MN) in *T. pallida* between test groups, assuming the significant value of each.

Days	Point 1	Point 2	Point 3	Point 4
15	NS	NS	NS	p<0.05 *
30	NS	p<0.01 **	NS	p<0.01 **
60	p<0.05 *	p<0.01 **	p<0.05 *	p<0.001 ***

Performed using two-way ANOVA and Bonferroni posttests. Comparison between days of exposure and exposure points, with significant results at p<0.05*, p<0.01** and <0.001***, compared to the negative control. NS, Non-significant.

of non-viable pollen (characterized by the absence or reduction in the cytoplasm and a weak staining), which involves the interference of the fertility of the plant organism. According to Costa and Droste (2012), reduced pollen viability and increased in the frequency of MN, it is resulting from the influence of the vehicle pollution and extent of drought, factors that correlate to the points tested in research and high temperature in the city, Teresina.

In urban areas, air pollution is demonstrated by the photochemical smog resulting from the interaction between nitrogen oxides, hydrocarbons such as benzene and sunlight to form oxidation products, which can cause harm to humans and plants. As cars emit carbon monoxide, sulfur dioxide, aromatic compounds and hydrocarbons, contribute directly to the formation of photochemical smog and other pollution (Proncove, 2006; Semace, 2011).

The growing demand for new technologies makes the benzene is one of the industrial substances produced in higher volume. It comes mainly in production and oil refining, one of the constituents of gasoline. The exhaust of automotive vehicles accounts for about 5% of total hydrocarbons emitted into the environment. It is understood that the primary means of benzene poisoning in the workplace, either by direct contact with the substance or its derivatives (Abiquim, 1999; Costa and Costa, 2002). Thus, the high frequency of MN in *T. pallida* is related in point 4 with the studies of Alves et al. (2011), which found a positive association between high frequency of MN in pollen mother cells *T. pallida* exposed at different times of the large amount of polycyclic aromatic hydrocarbons - HPA's. Among the molecules considered mutagenic and / or carcinogenic, the HPA's have a prominent place in environmental and biological studies (Conpet, 2006; Jung et al., 2009; Brito et al., 2013; Silva et al., 2015).

When exposed to environmental pollutants, *T. pallida* suffers in short, physiological and structural changes resulting from the genotoxic potential. Thus, many injuries can be caused by these compounds, serving as an excellent indication, for example, its clastogenic effects, chromosomal breakage, and aneugenic, partial or total loss of chromosomes, detected by exposing the plant to the environment (Ma and Grant, 1982; Savóia, 2008; Sisenando, 2011; Crispim et al., 2014).

According to Thewes et al. (2011), chromosomal variations found in this experiment are explained by environmental changes that affect the errors in the process of formation and development of cellular structures of these vegetables (Rodríguez, 2015)

Identification of the potential harm in the genome of *T. pallida*, through exposure of polluted air *in situ* may be an important indicator for genotoxic events in mammals, since these eukartotic cells are continuously entering into different divisional phases and constantly mutate (Carvalho, 2005).

Conclusion

Air pollution is an emerging problem in society; it's leading to major changes in ecosystems thus a significant number of diseases in humans, animals and plants in general. The *T. pallida* is an important form to detect air pollution. TRAD-MCN bioassay is a relatively simple technique, easy reproducibility and high efficiency in the evaluation of chromosomal damage arising from the interference of the pollutants. The results showed the genotoxic potential of the four tested environments when compared to the negative control. Thus, increased frequency of micronuclei was correlated with the local exposure time and, as the largest potential to cause chromosomal aberrations occurred in most vehicle flow and manipulation of benzene derivatives environments. These data, although relevant, are early. Necessitating the need for investigations in different seasons and chemical analyzes of atmospheric air of the places to be studied.

Conflicts of Interests

The authors have not declared any conflict of interests.

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REFERENCES

Abiquim (1999). Anuário da Indústria Química Brasileira. Associação

Brasileira da Indústria Química. São Paulo. P 250.

Alves ON, Loureiro ALM, Santos FC, Nascimento KH, Dallacort R, Vasconcellos PC, Hacon SS, Artaxo P, de Medeiros SRB (2011). Genotoxicity and composition of particulate matter from biomass burning in the eastern Brazilian Amazon region. Ecotoxicol. Env. Safety 74:1427-1433.

Andrade-Vieira LF, Davide LC, Gedraite LS, Campos JM, Azevedo H (2011). Genotoxicity of SPL (spent pot lining) as measured by *Tradescantia* bioassays. Ecotoxicol. Environ. Saf. 74(7):2065-9.

Ávila RID, Guerra MT, Borges KDAS, Vieira MDS, Oliveira Júnior LMD, Furtado H, da Mota MF, Arruda AF, Valadares MC (2013). *Punica granatum L.* Protects mice against hexavalent chromium-induced genotoxicity. Braz. J. Pharm. Sci. 49:689-697.

Billet S, Al Zallouha M, Landkocz Y, Fougere B, Lepers C, Delepee R, Cazier F, Martin PJ, Verdin A, Ledoux F, Kelly FJ (2015). Urban air pollution fine particulate matter exposure: Biomarkers of carcinogenesis modulated by people ageing. Toxicol. Lett. 238:S63.

Brito KCT, Lemos CT, Rocha JAV, Mielli AC, Matzenbacher C, Vargas, VMF (2013). Comparative genotoxicity of airborne particulate matter (PM2.5) using Salmonella, plants and mammalian cells. Ecotoxicol. Env. Safety 94:4-20.

Branco MA (2003). A árvore: paisagismo e clima em Teresina-PI. Cadernos de Teresina (PI). XV.

Carreras HA, Pignata ML, Saldiva PHN (2006). *In situ* monitoring of urban air in Córdoba, Argentina using the Tradescantia-micronucleus (Trad-MCN) bioassay. Atmospheric Env. 40:7824-7830.

Carvalho HA (2005). *Tradescantia* as bioindicadtor plant in monitoring the clastogenic effects of ionizing radiation. Radiol. Brasil 38:459-462.

Conama Resolução n° 3, de 28/06/1990. Ministério do Meio Ambiente. Dispõe sobre padrões de qualidade do ar, previstos no PRONAR. Publicação DOU, de 22/08/1990. 15937-15939. Disponível em <http://mma.gov.br>Acesso em 20/12/2013.

Conpet (2006). Consultoria e Serviços do Petróleo Ltda. A Saúde da População e a Poluição Atmosférica.. Disponível em http://www.conpet.gov.br.

Costa MAF, Costa MFB (2002). Benzeno: uma questão de saúde pública. Interciência 27:201-204.

Costa GM, Droste A (2012). Genotoxicity on *Tradescantia pallida* var. *purpurea* plants exposed to urban and rural environments in the metropolitan area of Porto Alegre, southern Brazil. Braz. J. Biol. 72:801-806.

Crispim BA, Spósito JCV, Mussury RM, Seno LO, Grisolia AB (2014). Effects of atmospheric pollutants on somatic and germ cells of *Tradescantia pallida* (Rose) D.R. HUNT cv. *purpurea.* Ann. Braz. Acad. Sci. 86:1899-1906.

Crispim BA, Vaini JO, Grisolia AB, Teixeira TZ, Mussury RM, Seno LO (2012). Biomonitoring the genotoxic effects of pollutants on *Tradescantia pallida* (Rose) D.R. Hunt in Dourados, Brazil. Env. Sci. Pollut. Res. 19:718-723.

Gábelová A, Valovičová Z, Horváthová E, Slameňová D, Binková, B, Šrám RJ, Farmer PB (2004). Genotoxicity of environmental air pollution in three European cities: Prague, Košice and Sofia. Mutat. Research/Genet. Toxicol. Environ. Mutagen. 563(1):49-59.

Güez MC, Waczuk EP, Pereira KB, Querol MVM, Rocha JBT, Oliveira LFS (2012). *In vivo* and *in vitro* genotoxicity studies of aqueous extract of *Xanthium spinosum.* Braz. J. Pharm. Sci. 48:461-467.

Inomata S, Fushimi A, Sato K, Fujitani, Y, Yamada H (2015). 4-Nitrophenol, 1-nitropyrene, and 9-nitroanthracene emissions in exhaust particles from diesel vehicles with different exhaust gas treatments. Atmospheric Environ. 110:93-102.

Jung D, Cho Y, Collins LB, Swenberg JA, Giulio RT (2009). Effects of benzo[*a*]pyrene on mitochondrial and nuclear DNA damage in Atlantic killifish (*Fundulus heteroclitus*) from a creosote-contaminated and reference site. Aquat. Toxicol. 95:44-51.

Klumpp A, Ansel W, Klumpp G, Belluzzo N, Calatayud V, Chaplin N *et al* (2006). EuroBionet: a Pan-European biomonitoring network for urban air quality assessment. Env. Sci. Pollution Res. 9:199-203.

Semace (2011). Plano de Controle da Poluição por Veículos em uso-PCPV. Superintendência Estadual do Meio Ambiente.

Ma TH, Grant WF (1982). The Tradescantias-adventurousplants. Herbarist 48:36-44.

Maioli OLG, Santos JM, Júnior NCR, Cassini STA (2008). Parâmetros bioquímicos foliares das espécies *Licania tomentosa* (benth.) E Bauhiniaforficata (link.) Para avaliação da qualidade do ar. Quim. Nova 31:1925-1932.

Mott KA, Berg DG, Hunt SM, Peak D (2014). Is the signal from the mesophyll to the guard cells a vapour-phase ion? Plant Cell Environ. 37:1184-1191.

Pereira TS, Beltrami LS, Rocha JA, Broto FP, Comellas LR, Salvadori DM, Vargas VM (2013). Toxicogenetic monitoring in urban cities exposed to different airborne contaminants. Ecotoxicol. Environ. Safety 90:174-182.

Proncove (2006). Avaliação do Programa de Controle da Poluição do Ar por Veículos Automotores. Ministério do Meio Ambiente.

Rodríguez YA, Christofoletti CA, Pedro J, Bueno OC, Malaspina O, Ferreira RA, Fontanetti CS (2015). *Allium cepa* and *Tradescantia pallida* bioassays to evaluate effects of the insecticide imidacloprid. Chemosphere 120:438-442.

Santos AP, Segura-Muñoz SI, Nadal M, Schuhmacher M, Domingo JL, Martinez CA, Magosso Takayanagui AM (2015). Traffic-related air pollution biomonitoring with *Tradescantia pallida* (Rose) Hunt. cv. purpurea Boom in Brazil. Environ. Monitoring Assess. 187:1-10.

Savóia EJL, Domingos M, Guimarães ET, Brumati F, Saldiva PH (2009). Biomonitoring genotoxic risks under the urban weather conditions and polluted atmosphere in Santo Andre, SP, Brazil, through Trad-MCN bioassay. Ecotoxicol. Environ. Safety 72:255-260.

Silva CS, Rossato JM, Rocha JAV, Vargas MVF (2015). Characterization of an area of reference for inhalable particulate matter (PM2.5) associated with genetic biomonitoring in children. Mutat. Res. 778:44-55.

Sisenando HA, Medeiros SRB, Saldiva, PHN, Artaxo P, Hacon SS (2011). Genotoxic potential generated by biomass burning in the Brazilian Legal Amazon by *Tradescantia* micronucleus bioassay: a toxicity assessment study. Environ. Health 10:1-9.

Thewes MR, Endres JD, Droste A (2011). Genotoxicity biomonitoring of sewage in two municipal wastewater treatment plants using the *Tradescantia pallida* var. *purpurea* bioassay. Genet. Mol. Biol. 34:689-693.