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OZONE BIOMONITORING: A NOVEL APPROACH OF ENVIRONMENTAL ASSESSMENT

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Biomonitoring has emerged as an important tool to assess the environmental health using living organisms. It is a reliable, economical and easy method and has far more advantages as compared to the conventional physico- chemical methods that require more capital, technically sound instruments and workers to work upon. With ozone concentration on a unprecedented rise, ozone biomonitoring has become a demand of the present time. Although the different protocols of ozone biomonitoring are now well established in the developed countries, we still need to work upon it in the developing and underdeveloped countries. The present article explains the different methods utilized in ozone biomonitoring and the different biomonitoring programmes that are being conducted by different agencies.

Keywords: biomonitoring, economical, protocols, programmes

The fluctuations in the climate and environmental contamination have imposed a tremendous amount of stress on ecosystems as a whole. Although, environmental stress can act as a major driving force in the evolutionary process, but in the last few decades, owing to the excessive anthropogenic interferences, the imposed stress has crossed its threshold, resulting in harmful metabolic changes in plant functioning. The last few years have seen a considerable concern about the increasing environmental stress imposed upon the plants. In lieu of this, concept of biomonitoring has emerged as very significant strategy which has made it possible to meet the requirement for protection of the environment against the increasing number of environmental stress factors and is an efficient technique for the assessment of environmental quality during the last few decades (De Temmermann *et al.* 2001). Biomonitoring can be simply defined as the systematic use of organisms or their response to determine the conditions or changes in environment Until now, the evaluation of the environmental quality was primarily based upon chemical and physical measurements. These methods, although gave us an idea about the presence and concentration of the environmental contaminants, it did not describe their actual impact on the biota or ecosystem. Information related to the emission of pollutants from the source, the transmission within the atmosphere and the ambient air concentration close to the soil surface where transfer to the receptors takes place can be obtained via physical and chemical analysis. These data can be used for risk assessments using simulation models when dose-response relationships are known for a given pollutant. However, these measurements can only predict a possible conclusion, but do not give any idea about the real incidence of the environmental contamination. This gap is filled in by biomonitoring as an additional system to be used besides concentration measurements with respect to emissions and ambient air concentrations (De Temmerman *et al.* 2001). Unlike physical monitoring, biomonitoring is not susceptible to temporal variations in the pollutants over daily, monthly or seasonal scales and captures the integrated response to these changes. Therefore, through biomonitoring, the long term effect of pollutants can be captured by measurements taken in the field on single occasion. Further, biomonitoring technique is considered to be particularly suitable for developing countries, where economical and infrastructural limitations allow only a very limited air monitoring network and biomonitoring offers the opportunity to determine the large scale pattern of pollutant distribution, as well as

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temporal changes.

OZONE BIOMONITORING

Tropospheric ozone has now been established as the main culprit causing significant negative effects on crop performance around the globe (Danh *et al.* 2015, Feng *et al.* 2015). As several studies have proved that the concentration of ozone will further increase, ozone biomonitoring becomes an essential feature. Ozone biomonitoring can be done via two ways, by observing changes in naturally occurring vegetation (passive biomonitoring), or by transplanting specific plants/cultivars with known ozone sensitivities (active biomonitoring) (Mukhopadhyay *et al.* 2020, Boquete *et al.* 2017). Although, a variety of vegetative entities can be used for biomonitoring, vascular plants are the commonly used bioindicators (Harmens *et al.* 2015, Pellegrini *et al.* 2014, Klumpp *et al.* 2006). The important characteristics that make vascular plants the most suitable bioindicators are that they can develop ozone induced visible foliar injuries at relatively low ozone concentrations. Tobacco, white clover, snap beans, poplar etc are few of the bioindicator plants that develop clear visible injury symptoms and have been used in several biomonitoring programmes (Fig 1). In passive biomonitoring, the naturally growing vascular plants are used. However, in active biomonitoring, plants with known ozone sensitive genotypes and the ozone insensitive genotypes are used.

Important plants/cultivars that are commonly used for ozone biomonitoring are:-

(i)Snap bean (*Phaseolus vulgaris* L.): The snap bean genotypes S156 (ozone sensitive) and R123 (ozone resistant) has been commonly used for large scale biomonitoring programmes across Europe by International Co-operative Programme on Effects of Air Pollution on Natural Vegetation and Crops (ICP Vegetation) (Agathokleous *et al.* 2017, Drapikowska *et al.* 2016).

(ii)White clover (*Trifolium repens* L.): The two biotypes of white clover NC-S (ozone sensitive) and NC-R (ozone resistant) have been extensively used in different biomonitoring experiments conducted by ICP Vegetation (1996-2006) and by individual research group in Italy and Greece (Saitanis *et al.* 2015, Harmens *et al.* 2015). The responses of NC-S/NC-R biotypes have established very strong correlation with ozone concentration across Europe, not only in terms of visible foliar injury, but also through the biomass ratio (Saitanis *et al.* 2015, Harmens *et al.* 2015). On the basis of different experiments, it has now been proved that white clover is more efficient than snap bean for ozone biomonitoring studies.

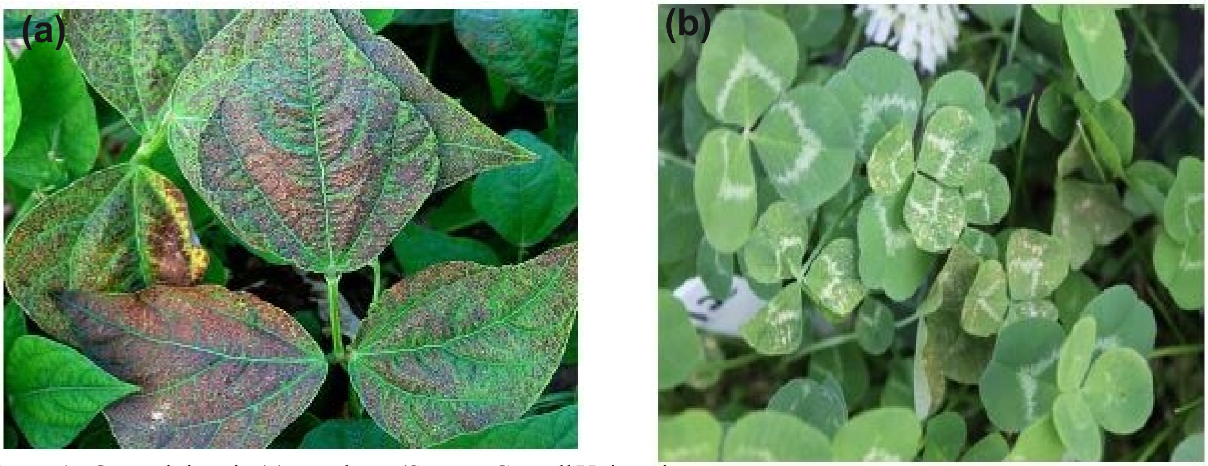

Figure 1: Ozone injury in (a) snap bean (Source: Cornell University, http://blogs.cornell.edu/livegpath/gallery/beans/ozoneinjury/ (b) white clover (Source: ICP Vegetation)

(iii)Tobacco (*Nicotiana tabaccum* L.): The tobacco cultivars that are commonly used in ozone biomonitoring programmes are Bel W3 (ozone sensitive) and Bel B (ozone resistant). These cultivars were developed at tobacco breeding programme at the Agricultural Research Services (ARS), USDA, Agricultural Research Centre, Beltsvile, MD, USA, and is the oldest and most successful ozone biomonitoring systems (Saitanis *et al.* 2020, Pellagrini *et al.* 2014). However, studies done till now have relied only upon the appearance of foliar injuries in response to ozone (Sandrin *et al.* 2018, Saitanis *et al.* 2015) and it has been suggested that the tobacco biomonitoring protocol can be improved by incorporating other leaf traits and morphological characters like Bel W3/Bel B biomass ratio etc. (Kaffer e*t al.* 2019). Further experiments are needed to strengthen the ozone dose/exposure relationships needed for more successful biomonitoring programmes (Agathokleous *et al.* 2020).

The response of these above mentioned plants/cultivars have provided significant information related to the concentration of ozone across the biomonitoring sites. However, it has been proved that their performance can be highly influenced by the variable climatic conditions like high temperature (Agatholeous *et al.* 2017, Harmens *et al.* 2015) and interference with other pollutants specifically nitrogen di oxide and suspended particulate matter (Sandrin *et al.* 2018). Therefore, there is an urgent need to develop new biomonitoring protocols wherein the different aspects of air quality and climate conditions should be included to provide more reliable and stable results so that the ultimate objective of biomonitoring is obtained (Agathokleous *et al.* 2020, Silva *et al.* 2012).

OZONE BIOMONITORING PROGRAMMES: Different environmental agencies like United Nations Economic Commission for Europe (UNECE), United States Environmental Protection Agency (USEPA), European Environmental Agency (EEA) etc. have focused on ambitious biomonitoring programmes over the last decade. Several of these biomonitoring programmes have focused on evaluating the response of plants towards tropospheric ozone (surface ozone), which is now established to be the major culprit causing harmful physiological effect to biota, posing serious threat to plant productivity and ecosystem functioning (Unger *et al.* 2020, Feng *et al.* 2019, Mill *et al*. 2018). Since concentration of ozone is continuously increasing in remote and sub urban areas (Paoletti *et al*. 2014), ambient ozone should be monitored not only in urban and sub urban areas, but also in forested, agricultural and other remote areas. Certain plant species, also called as bioindicator plants develop ozone specific injury symptoms which can be utilized in detecting and monitoring ozone stress via biomonitoring programmes. The first reported biomonitoring studies were initiated by US Forest Service (USFS) along with US Environmental Protection Agency (US EPA) in north east and north central USA during 1994-2009 (Smith 2012). This programme included 24 states and 450 biosites, where foliar injury response of ozone sensitive plants (approximately, 46,000 in number) was evaluated for ozone injury (Smith *et al.* 2008). This biomonitoring programme provided important information on the regional quality of ozone across north east and north central region of USA and provided important regional information on ozone air quality (Coulston 2011). It was observed that the selected bioindicator species showed higher injury percent at sites with high ozone concentration (Smith 2012). This biomonitoring programme not only gave important information about ozone concentration along the north eastern and north central transact of USA, it also established that foliar injury can be used as an important tool for assessment of ozone concentration over a large area. This biomonitoring programme depicted a declining trend of peak ozone concentration during the growing season; however, seasonal ozone concentration was on rise (Smith 2012).

Another biomonitoring programme, the EuroBioNet (European Network of Air Quality by use of Bioindiator Plants), initiated in Europe in 1999 as a network of research institutes and municipal environmental authorities from 12 urban sites in eight EU member states.

This programme used extremely ozone sensitive tobacco (*Nicotiana tabaccum* L.) cultivar Bel W3 to evaluate the phytotoxic effect of ozone at different sites. Other bioindicator plants used in this biomonitoring programme were poplar (*Populus nigra* 'Brandaris'), spiderwort (*Tradescantia* sp. clone 4430), Italian rye grass (*Lolium multiflorum italicum*) and curly kale (*Brassica oleracea acephala*) were exposed to ambient air at 90 monitoring sites according to standardized methods. In one of the studies related to this biomonitoring programme, the ozone sensitive cultivar of tobacco (Bel W3) was studied at 100 different sites and a relationship between ozone concentration and ozone induced visible foliar injury was recognized (Klummp *et al.* 2006). This study clearly established a significant correlation between foliar injury levels and ozone concentrations (Klumpp *et al.* 2006). Similar correlation was also observed between ozone concentration and foliar injury on tobacco was also observed in a biomonitoring study done at the rural sites in Spain (Ribas and Penuelas, 2003). These biomonitoring studies clearly depicted an ozone gradient with low levels of ozone-induced foliar injury in north and North West Europe, and medium to high values in the southern and central regions.

Apart from visible injury, other characters that are utilized in the biomonitoring studies of ozone are biomass and yield. National wise assessment programmes like NCLAN (National Crop loss assessment Programme, USA) and EOTP (European Open Top chamber Programme, Europe) initiated in early1980s were important landmark biomonitoring programmes in which ozone concentrations were assessed on the basis of biomass and yield responses of the experimental plants. Results of 41 NCLAN studies involving 14 plant species grown across USA during a 7 year period showed that 7 species suffered a 10 % yield loss when exposed to average ozone concentration ($7hd^{-1}$) below 50 ppb (Heagle 1989, Murphy *et al.* 1999). These biomonitoring programmes indicated that 20 % of Europe crop production could be at a risk of yield losses of 5 % or more due to ozone pollution (Mills *et al.* 2007). Similarly several other Biomonitoring programmes like EU/EC International Cooperative Programme on Assessment and Monitoring of Air Pollution Effect (ICP Forests) and on Natural Vegetation (ICP Vegetation) in Europe, and Forest Health Biomonitoring Programmes in North America provided reliable and substantial information related to ozone concentration and its effect on natural vegetation in respective areas (Harmens *et al.* 2015).

Apart from ozone, several other biomonitoring programmes for other environmental contaminants have also been conducted. European Moss Survey initiated in late 1980s by ICP Vegetation gave valuable information regarding the deposition of heavy metals across Europe. In 2005, this programme included the monitoring of atmospheric nitrogen deposition as well and on the basis of the results areas at the risk of high atmospheric nitrogen deposition were identified. Biomonitoring programmes have played a structural role in framing the pollution abatement policies such as Gothenberg Protocol (UNECE 1999), NCE Directives (EU 2001), Protocol on heavy metals, Aarhus Protocol on persistent organic pollutants etc.

CONCLUSION

Ozone biomonitoring programmes have now been adopted in all the developed countries and a lot of efforts are ongoing to establish these networks in the developing countries.

However, ozone biomonitoring still remains a challenging aspect in underdeveloped countries. The most important aspect of biomonitoring is to develop its societal significance by involving the general public in these programmes. The biomonitoring programme of EuroBioNet has taken the initiative of accommodating school students in their programme through different school projects. Further, the biomonitoring programmes should also be extended to forest and agricultural areas rather than confining them to rural and urban areas. In the developed countries, where extensive biomonitoring networks exist, there is a need to develop some new approaches for biomonitoring that could improve on the currently limited capabilities of existing schemes. The use of biosensors to study the negative effects of xenobiotics in the environment is yet another aspect which can be utilized in the biomonitoring studies. DNA sequencing is another important aspect of environmental genomics which can be exploited for improved results of biomonitoring studies. The objective of machine learning algorithms (ecoinformatics) is yet another promising approach to establish a new routine biomonitoring framework. DNA metabarcoding combined with the concepts of ecoinformatics will allow scaling up both spatial and temporal resolution for large and more ambitious biomonitoring programmes in future.

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REFERENCES

Agathokleous E Saitanis CJ Burkey KO Ntasti G Vougeleka V Mashaheet AM and Pallides A 2017 Application and further characterization of snap bean S156/R13 ozone biomonitoring systems in relation to ambient temperature. *Sci Total Environ* **580** 1046- 1055.

Boquete MT Aboal JR Carballeira A and Fernandez JA 2017 Do mosses exist outside of Europe? A biomonitoring reflection. *Sci Total Environ* **593**-594 567-570.

Coulston JW 2011 Modeling ozone bioindicator injury with microscale and landscape-scale explanatory variables: a logistic regression approach. In: Conkling BL (ed) *Forest health monitoring: 2007 national technical report, Gen. Tech. Rep.* SRS-147. U.S. Department of Agriculture Forest Service, Southern Research Station, Asheville, NC

Danh NT, Huy LH and Oanh NTK 2016 Assessment of rice yield lossdue to exposure to ozone pollution in Southern Vietnam. *Sci Total Environ* **566-567** 1069- 1079.

De Temmermann L, Bell JNB, Garrec JP, Klumpp A, Krause GHM and Tonneijck AEG 2001 Biomonitoring of air pollutants with plants-considerations for the future. In: *EuroBionet -Urban Air Pollution*, Bioindication and Environmental Awareness. 337-373.

Drapikowska M, Drapikowski P, Borowiak K, Hayes F, Harmens H, Dziewiatka T and Byczokowska K 2016 Application of novel image based estimation of invisible leaf injuries in relation to morphological and photosynthetic changes of *Phaseolus vulgaris* L. exposed to tropospheric ozone. *Atmos Pollut Res* **7** 1065- 1071.

EU European Union 2001 Directive 2001/81/EC of the European parliament and the council on national emission ceilings for certain atmospheric pollutants. Off *J Euro Commun L.* 30922-30.

Feng H, Hu E, Wang X, Jiang L and Liu X. 2015. Ground level ozone pollution and its impacts on food crops in China: A review. Environ Pollut **199** 42-48.

Feng Z, De Marco A, Anav A, Gualtieri M, Sicard P, Tian H, Fornasier F, Tao F, Guo A and Paoletti E 2019 Economic losses due to ozone impacts on human health, forest productivity and crop yield across China. *Environ Int* **131** 104966.

Harmens H, Mills G, Hayes F, Norris DA and Sharps K 2015 Twenty eight years of ICP Vegetation: an overview of its activities. *Ann di Bot* **5** 31-43.

Heagle AS 1989 Ozone and crop yield. *Ann Rev Phytopath* **27** 397-423.

Klumpp A, Ansel W, Klumpp G, Vergne P, Sifakis N, Sanz MJ, Rasmussen S Ro-Poulsen H Ribas A Penuelas J, Kam bezidis HD, Shang H, Garrec JP and Calatayud 2006 Ozone pollution and ozone biomonitoring in European cities Part II. Ozone induced plant injury and its relationship with descriptors of air pollution. *Atmos Environ* **40** 7437-7448.

Kuffer MI, Domingos M, Lieske I and Vaargas VMF 2019 Predicting ozone levels from climate parameters and leaf traits of Bel W3 tobacco variety. *Environ Pollut* **248** 471-477.

Mills G, Buse A, Gimeno B, Bermejo V, Holland M, Emberson L and Pleijel H. 2007. A synthesis of AOT40-based response functions and critical levels of ozone for agricultural and horticultural crops. *Atmos Environ* **41** 2630 - 2643.

Mills G,Sharps K, Simpson D, Pleijel H, Broberg M, Uddling J, Jaramillo F, Davies WJ, Dentener F, Van den Berg M, Frei M, Burkey K, Emberson L, Feng Z, Kobayashi K and agarwal M 2018 Ozone pollution will compromise efforts to increase Global wheat production. *Glob Chang Biol* **24** 3560-3574.

Mukhopadhyay S Dutta R and Das P 2020 A critical reviewon plant biomonitors for determination of polycyclic aromatic hydrocarbons (PAHs) in air through solvent extraction techniques. *Chemos* (in press).

Murphy JJ, Delucchi MA, McCubins DR and Kim HJ 1999 The cost of crop damage caused by ozone air pollution by motor vehicles. *J Environ Manag* **55** 273-289.

Paoletti E, De Marco A, Beddows DCS, Harrison RM and Manning W. 2014. Ozone levels in European and USA cities are increasing more than at rural sites, while peak values are decreasing. *Environ Pollut* **192** 295-299.

Pellegrini E, Campanella A, Lorenzini G and Nali C 2014 Biomonitoring of ozone: A tool to initiate the young people into scientific method and environmental issues. A case study in Central Italy. *Urban For Urban Green* **13** 800-805.

Ribas A and Penuelas J 2003 Biomonitoring of tropospheric ozone phytotoxicity in rural Catalonia. *Atmos Environ* **37** 63-71.

Saitanis CJ, Burkey KO, Agathokleous E Hung YT 2020 Ambient ozone alternative monitoring and biomonitoring with higher plants. In: *Handbook of Environment and Waste Management.* Vol **3**. Edited by Hung Y, Wang LK, Shammas N. World Scientific Publishing Co. (in press).

Saitanis CJ, Panagopoulos G, Dasopoulou V, Agathokleous E and Papatheohari Y 2015 Integrated assessment of ambient ozone phytotoxicity in Greece's Tripolis plateau. *J Agric Meteorol* **71** 55-64.

Sandrin CZ, Goncalves da Silva, Engela MR, Sacramento GN and Domingos M 2018 Ozone biomonitoring with white clover. *Clean-Soil, Air, Water* **46** 1800377.

Silva DT, Meirelles ST and Moraes RM 2012

Relationship between ozone, meteorological conditions, gas exchange and leaf injury in *Nicotiana tabacum* Bel W3 in a sub tropical region. *Atmos Environ* **60** 211-216.

Smith GC, Coulson JW and O'Connell BM 2008 Ozone bioindicators and forest health: A guide to evaluation, analysis and interpretation of ozone injury data *In the forest inventory and analysis programme.* Gen Tech resp. NRS-34. Newtown Square, PA: US department of Agriculture. Forst service, Northern Research station. pp. 34.

Smith GC 2012 Ambient ozone injury to forest plants in North east and North central USA: 16 years of biomonitoring. *Environ Monit Assess* **184** 4049- 4065.

UNECE 1999 Air pollution and vegetation. Annual Report 998/1999. *ICP-vegetation Coordination Centre*, CEH Bangor, University of Wales, Bangor.

Unger N, Zheng Y, Yue X and Harper KL 2020 Mitigation of ozone damage to the world's land ecosystems by source sector. *Nat Clim Chang* **10** 134- 137.