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Review

Impacts of particulate matter pollution on plants: Implications for environmental biomonitoring

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ABSTRACT

Air pollution is one of the serious problems world is facing in recent Anthropocene era of rapid industrialization and urbanization. Specifically particulate matter (PM) pollution represents a threat to both the environment and human health. The changed ambient environment due to the PM pollutant in urban areas has exerted a profound influence on the morphological, biochemical and physiological status of plants and its responses. Taking into account the characteristics of the vegetation (wide distribution, greater contact area etc.) it turns out to be an effective indicator of the overall impact of PM pollution and harmful effects of PM pollution on vegetation have been reviewed in the present paper, covering an extensive span of 1960 to March 2016. The present review critically describes the impact of PM pollution and its constituents (e.g. heavy metals and poly-aromatic hydrocarbons) on the morphological attributes such as leaf area, leaf number, stomata structure, flowering, growth and reproduction as well as biochemical parameters such as pigment content, enzymes, ascorbic acid, protein, sugar and physiological aspect such as pH and Relative water content. Further, the paper provides a brief overview on the impact of PM on biodiversity and climate change. Moreover, the review emphasizes the genotoxic impacts of PM on plants. Finally, on the basis of such studies tolerant plants as potent biomonitors with high Air Pollution Tolerance Index (APTI) and Air Pollution Index (API) can be screened and may be recommended for green belt development.

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1. Introduction

In recent Anthropocene era, rapid pace of industrialization and urbanization has given birth to dust or particulate matter (PM) pollution (Rai, 2013,2016a). Dust or PM is actually the solid matter, not only of anthropogenic origin, but also natural origin (Ferreira-Baptista and DeMiguel, 2005; Rai, 2013,2016a). Environmental contamination and human exposure with respect to dust or PM pollution have dramatically increased during the last decade, particularly in developing countries like India (Rai, 2013; Rai and Panda, 2014; Rai, 2016a). The size fractionation of PM and its adverse human health impacts have been well documented in literatures (Brook et al., 2003; Mcdonald et al., 2007; Thomas and Richard, 2010; Delfino et al., 2011; Rai, 2011a,b; Ulrich et al., 2012; Rai, 2013; Rai and Panda, 2014; Rai and Singh, 2015; Rai, 2016a,b).

The ambient air pollutants have a potential adverse impact on biochemical parameters, which further leads to a reduction in the overall growth and development of plants. The impact of various atmospheric pollutants on plants both in terms of physiology and biochemistry has been under investigation for many years (Agrawal and Agrawal, 1989; Rai, 2011a,b; Rai et al., 2013; Rai and Panda, 2014; Rai and Singh, 2015; Rai, 2016a,b). Plant adaptation to changing environmental factors involves both short-term physiological responses and long-term physiological, structural and morphological modifications. These changes help plants minimize stress and maximize use of internal and external resources (Dineva, 2004). The changed ambient environment due to the PM pollutants in urban area has exerted a profound influence on the morphological, biochemical physiological and genetic status of plants (Farooq et al., 2000a,b; Seyyednejad et al., 2011; Prajapati, 2012a,b; Younis et al., 2013; Rai et al., 2013; Rai and Panda, 2014; Rai and Singh, 2015; Rai, 2016a,b). Present review provides a critical literature review on these multifaceted aspects covering an extensive span of 1960 to March 2016. We will try to confine our review on PM; however, at some places some important references pertaining to gaseous pollutants may also appear.

Since plants are constantly exposed to air, they are the primary receptors for both gaseous and particulate pollutants of the atmosphere. In terrestrial plant species, the enormous foliar surface area acts as a natural sink for pollutants especially the particulate ones. Vegetation is an effective indicator of the overall impact of air pollution particularly in context of particulate matter (PM). The harmful effects of PM on vegetation have already been noted by several researches (Dasgupta, 1957; Keller, 1983; Agrawal et al., 1991; Rayyappa and Singara Charya, 1993; Garg et al., 2000a,b; Shrivastava and Joshi, 2002; Chandawat et al., 2011; Rai, 2011a,b; Rai, 2013; Rai and Panda, 2014; Rai and Singh, 2015; Rai, 2016a,b). Table 1 lists the important attributes of vegetation particularly road-side vegetation, which may change due to exposure of PM.

Trees have a very large surface area and their leaves function as an efficient pollutant-trapping device. Leaves, susceptible and highly exposed parts of a plant, may act as persistent absorbers for PM in a polluted environment (Maiti, 1993; Samal and Santra, 2002). The use of higher plants for air monitoring purposes is becoming more and more widespread. The main advantages are greater availability of the biological material, simplicity of species identification, sampling and treatment and ubiquity of some genera, which makes it possible to cover large areas. Lichens and mosses are characterised by irregular and patchy distribution, and their sampling should be done by specialists capable of differentiating between similar-looking species (Maiti, 1993). These limitations become more pronounced in industrial and densely populated areas, where several anthropogenic pressures may cause scarcity or even lack of indicator species at some sampling points. Therefore, the search of alternative biological indicators becomes especially important. Trees, in view of their tough physiognomy may survive in urban areas where lichens are too sensitive to survive. Therefore, trees are particularly relevant in urban areas where lichens are often missing (Rucandio et al., 2010; Rai, 2013; Rai and Singh, 2015; Rai, 2016a,b). As trees have a larger collecting surface area than other land cover types and also promote vertical transport by enhancing turbulence, there is a greater opportunity for particles to be collected on the trees surface. Trees are therefore more efficient at capturing particles from the atmosphere by dry deposition relative to short vegetation (Gallagher et al., 1997; Mcdonald et al., 2007). The sticky PM emitted from the automobile exhausts is the major constituent of PM pollution, which is deposited on the leaf surface of common roadside plants. PM reduces growth, yield, flowering, and reproduction of plants (Saunders and Godzik, 1986). PM from different sources impact on the chemical composition of plants is often used as an indicator of and a tool for monitoring environmental pollution (Rao, 1977; Posthumus, 1984, 1985; Agrawal and Agrawal, 1989; Kulump et al., 1994; Dmuchowski and Bytnerowricz, 1995; Rai, 2011a,b; Rai, 2013; Rai et al., 2013; Rai and Panda, 2014; Rai and Singh, 2015; Rai, 2016a,b).

2. Use of plants as biomonitors

Ecological investigation of impact of PM on morphological, physiological and biochemical parameters of plants assist in identifying the suitable biomonitors through calculation of air pollution tolerance index (APTI) and Anticipated Pollution Index (API). The effect of air pollution on the plants can be quantified using a parameter *i.e.* APTI. APTI is a species dependent plant attribute which expresses the inherent ability of plant to encounter stress emanating from pollution (Tiwari et al., 1993). APTI was proposed by Singh and Rao (1983) to assess the tolerant/resistance power of plants against air pollution. The APTI was calculated using the formula:

$$\mathrm{APTI} = (A(T+P)+R)/10$$

where: A=Ascorbic Acid (mg/g)
T=Total Chlorophyll (mg/g-f w)
P=pH of the leaf extract
R=Relative water content of leaf (%).

Table 1
List of structural and functional properties of roadside vegetation which may change due to PM (Modified after Sigal and Suter, 1987; Grantz et al., 2003).

Particulate Matter Affects roadside vegetation (through alteration in structural and functional properties)	Structural property of roadside vegetation	Functional property of roadside vegetation
	At organism/individual level: Leaf area, Shoot morphology, Root morphology, Individual biomass, Allometry, Age distribution	Photosynthesis, Respiration, Nutrient acquisition, Nutrient leaching from foliage, Carbon allocation, Individual mortality
	At population level: Population distribution, Population dispersion, Genetic diversity, Species diversity	Competitive vigor, Reproductive success, Biomass productivity, Redundancy and resilience
	At community level: Canopy leaf area index, Root distribution, Biomass	Succession, Soil stabilization, Productivity
	At ecosystem level: Element pool sizes, Soil type	Nutrient cycling, Hydrologic cycling

Table 2
Grouping of plants using APTI (Source: Lakshmi et al., 2009).

SI no.	APTI	Response
1	30–100	Tolerant
2	29–17	Intermediate
3	16–1	Sensitive
4	<1	Very sensitive

The plant species can be conveniently grouped based on the APTI values (Table 2). The APTI determination provides a reliable method for screening large number of plants with respect to their susceptibility to air pollutants. This is a simple method and very easy to apply in all type of field conditions without adopting any costly environmental monitoring gadgets. The sensitive species can be used as bio indicators and tolerant species can be used as a sink for air pollutants (Kuddus et al., 2011; Krishnaveni et al., 2013). The APTI index shows the effect of the pollutants only on the biochemical parameters. In order to combat air pollution by planning the green belt development in a particular area, many socio-economic factors are also considered. By combining the resultant APTI values with some relevant biological and socio-economic characters (plant habit, canopy structure, type of plant, laminar structure and economic value), the API is calculated for different species. Based on these characters, different grades (+ or −) are allotted to plants. Based on the current grading system, a tree can secure a maximum of 16 positive points. These points are scaled to a percentage system and based on the score obtained, the category has been assessed. Tables 3 and 4 shows the parameters used to grade the performance and assessment categories along with the scores of a particular tree species. Thawale et al. (2011) observed biochemical changes in four selected plant species, namely *Azadirachta indica* (neem), *Mangifera indica* (mango), *Delonix regia* (gulmohar), and *Cassia fistula* (amaltas) of residential, commercial, and industrial areas of Nagpur city in India and found the suitability of *A. indica* as the most tolerant species which may be recommended for green belt plantation.

Table 3
Gradation of plant species on the basis of APTI and other biological and socio-economic characters* (After Shannigrahi et al., 2003; Mondal et al., 2011).

Attributes				
(a) Tolerance air pollution tolerance index (APTI)		7.0–8.0		+
		8.1–10.0		++
		10.1–11.0		+++
		11.1–12.0		++++
		12.1–13.0		+++++
(b) Biological and socio-economic	(i) Plant habit	Small		−
		Medium		+
		Large		+++
	(ii) Canopy structure	Sparse/Irregular/Globular		−
		Spreading crown/Open/Semidence		+
		Spreading dense		++
	(iii) Type of plant	Deciduous		−
		Evergreen		+
	(iv) Laminar structure	Size	Small	–
			Medium	+
			Large	++
		Texture	Smooth	−
			Coriaceous	+
		Hardiness	Delineate	−
			Hardy	+
	(v) Economic value	Less than three uses		−
		Three or four uses		+
		Five or more uses		++

*Maximum API score that can be attained: 16.

Table 4
API of plant species (After Shannigrahi et al., 2003; Mondal et al., 2011).

Grade	Score (%)	Assessment of plant species
0	Up to 30	Not recommended for plantation
1	31–40	Very poor
2	41–50	Poor
3	51–60	Moderate
4	61–70	Good
5	71–80	Very good
6	81–90	Excellent
7	91–100	Best

Controlling pollutants is a very complex problem. Sources and emissions have to be identified and it is necessary to find sufficiently sensitive and low-cost techniques which permit the simultaneous measurement of many contaminants. Risks have to be assessed, critical emissions have to be controlled, and economical aspects have to be integrated (Wolterbeek, 2002).

Monitoring with the help of biological indicators is simple, cheap, and convenient method to ensure the state of local environment. Biomonitoring allows continuous observation of an area with the help of bioindicators, an organism (or part of it) that reveals the presence of a substance in its surroundings with observable and measurable changes (e.g. accumulation of pollutants), which can be distinguished from the effects of natural stress (source: Bradley, David-Biomonitors-sciencebase.com; (accessed 10.11.13.)). Physicochemical studies can offer a quantitative image of the various substances that are dispersed in the environment and sometimes an indication of their toxicity on the human body, but only biological monitoring may give an insight into their effects on organisms. From this point of view, living organisms are very important to highlight the effects of pollution, because it presents some additional advantages compared to physical and chemical analysis (source – Greenlifeuniverse.com; (accessed 10.11.13.)). Regarding indicators of pollution, they are of two types: ***sensitive species***, indicating the presence of a pollutant by the appearance of lesions or malformations and ***accumulator species***, which concentrates the pollutant in their bodies. There is also another category, i.e. species that proliferate and become abundant in polluted areas. Several plant species are necessary for monitoring the large number of persistent contaminants in urban environments. Rucandio et al. (2010) observed that all of these plant species such as *Cedrus deodara*, (Deodar Cedar), *Cupressus sempervirens* (Italian cypress), *Pinus pinea* (Stone Pine), *Nerium oleander* (oleander), *Ligustrum ovalifolium* (Californian privet) and *Pittosporum tobira* (Japanese pittosporum) has a good capability to act as biomonitor of the pollution of certain elements, and they are able to distinguish among different places with varied contamination degree. Plant response to air pollution can be used to assess the quality of air that may provide early warning signals of air pollution trends. Biomonitoring tools may range from lichens to higher plants (Table 5). Table 5 lists certain important biomonitors and bioindicators as visualized through extensive literature review.

3. Effects of PM on plants

There are two main types of direct injury that PM pollution can cause on plants: acute and chronic injury. Acute injury results from exposure to a high concentration of gas for a relatively short period and is manifested by clear visible symptoms on the foliage, often in the form of necrotic lesions. While this type of injury is very easy to detect (although not necessarily to diagnose), chronic injury is much more subtle: it results from prolonged exposure to lower gas concentrations and takes the form of growth and/ or

Table 5
List of potent biomonitors and bioindicators under different sources of PM pollution.

Source of pollution	Plant/s as biomonitor/s	Impact	Reference
Coal burning station in Massachusetts	Lichens on *Acer saccharinum* and *Populus deltoides*	Lichen cover decreased near coal burning station	Murphy et al. (1999)
Automobile pollution in Lucknow, India	*Ficus religiosa* and *Thevetia neriifolia*	Changes in photosynthetic pigments, protein and cysteine contents; also changes in leaf area and foliar surface architecture of plants	Verma and Singh (2006)
Vehicular traffic and domestic heating in Siena (central Italy)	57 species of lichens	Acted as bioindicator and revealed amelioration of air quality over 1995.	Loppi et al. (2002)
Particulate pollution due to urban traffic and other human activities in European and North-American regions	*Ficus nitida* and *Eucalyptus globulus, Quercus petraea, Alnus glutinosa, Fraxinus excelsior, Acer pseudo-platanu, Pseudotsuga menziesii*	Efficient in dust capturing potential in developing and developed countries with arid and semi arid conditions	Freer-Smith et al. (2004)
Vehicular traffic and major industrial plants within the urban area of Naples	*Quercus ilex*	High levels of Cu, Cd, Ni and other elements in plant leaves	Alfani et al. (2000)
Air pollution impact in Man and Biosphere Reserve Wienerwald, Austria	Bryophytes-*Scleropodium purum, Hypnum cupressiforme* and *Abietinella abietina*	Distribution of bryophytes was greatly influenced by air quality in reserve as revealed by Index of Atmospheric Purity-method IAP	Krommer et al. (2007)
Air pollution biomonitoring (spatial and temporal variation in elemental concentration) at six sites in northern Minnesota, USA	Lichens – *Cladina rangiferina*, *Evernia mesomorpha*, *Hypogymnia physodes*, and *Parmelia sulcata*	Biomonitoring with lichens is strongly species dependent as corticolous species were more enriched in heavy metals than the terricolous species when monitored over a span of 11 years	Bennett and Wetmore (1999)
Vehicular traffic in the town of Montecatini Terme (central Italy)	*Flavoparmelia caperata*	As biomonitor of heavy metals	Loppi et al. (2004)
Air pollution in an urban site of Northern Italy	Lichens-*Hypogymnia physodes, Parmelia sulcata, Pseudevernia furfuracea* and *Usnea gr. hirta*	In relation to 29 elements studied *H. physodes, P. furfuracea* and *U. gr. hirta* have a similar accumulation capacity, while that of *P. sulcata* is lower	Bergamaschi et al. (2007)
Industrial complex of Cubatão, Brazil	*Tibouchina pulchra*	Adverse impacts were visible injuries, reductions in net photosynthesis, growth parameters, and ascorbate concentrations, and increased F, N, and S foliar concentrations	Moraes et al. (2003)
Heavy metal pollution in in Palermo city (Sicily, Italy)	*Nerium oleander L.*	*N. oleander* can be considered as a means of assessing dust contamination in the urban environment	Mingorance and Oliva (2006)
Global problem of carbon dioxide enrichment (climate change) on crop physiology and food quality	Crop plants	Adverse impacts of carbon dioxide enrichment may be decreased protein and mineral nutrient concentrations, as well as altered lipid composition in food of crop plants	DaMatta et al. (2010)
Heavy metal pollution in Hong Kong	*Bauhinia blakeana*	Acted as biomonitors of metals, sulfur dioxide and total suspended particulates	Lau and Luk (2001)
Polycyclic aromatic hydrocarbons problem in Naples, Italy	*Quercus ilex*	The total PAH contents in *Q. ilex* leaves were quite high	Alfani et al. (2001)
Trace metal pollution in Galicia, NW Spain	*Quercus robur* leaves and *Pinus pinaster* needle	Limited efficiency as bioaccumulators of the metals studied	Aboal et al. (2004)
Problem of air pollution in industrial complex of Cubatão, SE Brazil	*Psidium guajava*, *Psidium cattleianum* and *Mangifera indica*	*P. cattleyanum and P. guajava* may be used as an accumulative indicator in tropical climates	Moraes et al. (2002)

yield reductions, often with no clear visible symptoms. Plants that are constantly exposed to environmental pollutants absorb, accumulate and integrate these pollutants into their systems. It reported that depending on their sensitivity level, plants show visible changes which would include alteration in the biochemical processes or accumulation of certain metabolites (Agbaire and Esiefarienrhe, 2009). Pollutants can cause leaf injury, stomatal damage, premature senescence, decrease photosynthetic activity, disturb membrane permeability and reduce growth and yield in sensitive plant species (Tiwari et al., 2006). The long term, low-concentration exposures of air pollution produces harmful impacts on plant leaves without visible injury (Joshi et al., 2009). Several studies have been conducted to assess the effects of pollution on different aspects of plant life such as overall growth and development (Gupta and Ghouse, 1987; Misra and Behera, 1994), foliar morphology (Farooq et al., 2000; Pal et al., 2000; Shrivastava and Joshi, 2002; Gostin, 2009; Sukumaran, 2012), anatomy (Garg et al., 2000), and biochemical changes (Garty et al., 2001; Mashitha and Pise, 2001; Gavali et al., 2002; Rai et al., 2013; Rai et al., 2013; Rai and Singh, 2015; Rai, 2016a,'b). Effects of cement, petro-coke dust, fly ash, coal dust, automobile exhaust and other air-borne particulates on various morphological and physiological parameters in different plants were well-studied by many workers (Naidoo and Chirkoot, 2004a,b; Verma and Singh, 2006; Prajapati and Tripathi, 2008a,b,c,d,e,f; Saha and Padhy, 2011; Rai et al., 2013; Rai and Panda, 2014; Rai and Singh, 2015; Rai, 2016a,'b).

4. Morphological impact of PM on plants

Plant leaves are the primary receptors for both gaseous and PM pollutants of the atmosphere. Before these pollutants enter the leaf tissue, they interact with foliar surface and modify its configuration. Dust deposition on leaf surface, consisting of ultrafine and coarse particles, showed reduction in plant growth (Bender et al., 2002) through its effect on leaf gas exchange (Ernst, 1982), flowering and reproduction of plants (Saunders and Godzik, 1986), number of leaves and leaf area, one of the most common driving variables in growth analyses (Lambers et al., 1998). Reduction in leaf area and leaf number may be due to decreased leaf production rate and enhanced senescence (Seyyednejad et al., 2011). Due to alkaline cement dust pollution on Pine and Spruce, needle density and number of needle scars were higher for shoots formed in the period of higher pollution than for the shoots grown under a considerably lower pollution load (Ots et al., 2011).

Dust deposition also affect the stomata causing occlude stomata (Hirano et al., 1995) as the particles enter the leaf through stomatal opening and their toxicity disturb the physiological activity of plants (Farmer, 1993) such as the inhibition of plant growth, rate of photosynthesis (Armbrust, 1986), late flower and the hormonal imbalance (Farooqui et al., 1995). The inhibition of net photosynthesis would inhibit the assimilate translocation and eventually leaf area would decrease. It has been demonstrated that cement dust treated plants of *Brassica campestris* (Mustard) showed a consistent reduction in growth, photosynthetic pigments, yield and oil content over control plants (Shukla et al., 1990) and these factors together decreased the biomass of this plant.

Literature revealed that that stomatal closure can help to protect plants against pollution damage (Mansfield and Majernik, 1970). Apart from PM, Mansfield and Majernik (1970) in their concise review described the impact of CO_2 and SO_2 on stomatal behavior in several plants. Black and Unsworth (1980) observed and described the *Phaseolus vulgaris* (Kidney Bean), show stomatal opening in response to SO_2 fumigation. However, In general, there appear to be threshold concentrations above which all pollutants usually cause stomatal closure (Freer-Smith and Taylor, 1992) which may be attributed to direct damage to the stomatal apparatus along with that of other leaf tissues. Krajickova and Mejstrik (1984) observed that in *P. vulgaris* (Kidney Bean), and *Zea mays* (corn), gaseous diffusion in stomata was increased on application of fly ash from power plant; however, they were not plugged.

The dust particles or PM reduce photosynthetically active radiation (PAR) by altering its optical properties particularly the surface reflectance in the visible and short wave infrared radiation range (Eller, 1977; Hope et al., 1991; Keller and Lamprecht, 1995) and increase the leaf temperature (Naidoo and Chirkoot, 2004). The cement dust as the source of PM, deposits on the plants and hence producing significant effect. Cement kiln dust decreased height, biomass and net productivity in plants (Prasad et al., 1991). Jahan and Iqbal (1992) observed reduction in leaf blade area of five tree species as a result of extensive dust and SO_2 pollution.

The structure and morphology of epi-cuticular wax is a reliable indicator of plant health (Neinhuis and Barthlott, 1998) by creating a barrier between the plant and the environment and regulating the resistance to pollution stress. Deposition of road dust as well as cement dust containing high levels of MgO and PM causes degradation of epicuticular wax (Sauter and Pambor, 1989; Bermadinger et al., 1988) by increasing the erosion rate of wax structure (Huttunen, 1994) and inducing changes in leaf wettability (Saneoka and Ogata, 1987), inhibiting transpiration which could have far-reaching physiological consequences, such as prevention of gas exchange and photosynthesis (Sauter and Voß, 1986; Sauter et al., 1987) and loss of solutes from leaf cells (Bystrom et al., 1968). PM resulting from stone dust due to quarrying activities caused foliar anomalies and injury symptoms such as tissue necrosis, brown and yellow patches, black spots and in extreme cases,death of leaves (Saha and Padhy, 2011). Presence of excess amount of heavy metals like copper and sulfur in the plants cause various physiological changes such as symptoms of chlorosis (Bergman, 1983), premature aging and dying off of leaves. Details regarding heavy metals have been discussed as separate head. Alteration of leaf morphology and anatomy as a result of PM pollution were also studied by Prasad and Rao (1981), Agrawal and Agrawal (1989), Verma and Singh (2006) and Rai et al. (2010). Acclimatization of plants to air pollutants might change their morphological structure such as thicker epidermal cells and longer trichomes (Rangkuti, 2003). Once deposited on the leaf surface some elements may be taken up into the leaf via the stomata (Reimann et al., 2001) affecting the overall plant development and reducing the resistance of plants to drought, frost, insect and fungi (Shanker et al., 2005).

5. Physiological and biochemical effect on plants or plant leaves

5.1. pH

The growth and development aspects of plants are adversely affected by airborne PM depending on their physical and chemical nature. Dust pH may alter the leaf extract pH of plants. The changes in leaf-extract pH might influence the stomata sensitivity due to air pollutants. Cement dust on hydration liberates calcium hydroxide which can raise leaf surface alkalinity in some cases to pH 12. This level of alkalinity can hydrolyze lipid and wax components, penetrate the cuticle, and denature proteins finally plasmolyzing the leaf (Guderian, 1986; Czaja, 1960, 1961, 1962). The low pH of the leaf extract showed a relationship with the type of air pollution. The more acidic nature demonstrates that the air pollutants, mostly gaseous types, namely SO_2, NO_X diffuse and form acid radicals in the leaf matrix by reacting with cellular water, affecting the chlorophyll molecules (Turk and Wirth, 1975). In the presence of an acidic pollutant the leaf pH is lowered and this decline is greater in sensitive plant species. Alkaline nature of cement dust reduces the absorption of mineral substances from the soil, inducing changes in the plant physiology and morphology (Raajasubramanian et al., 2011). Alkaline nature of cement dust is responsible for alteration of leaf pH, as it cause chloroplast damage (Singh and Shrivastava, 2002). Dusts with pH values of ≥ 9, may cause direct injury to leaf tissues on which they are deposited (Vardak et al., 1995) or indirectly through alteration of soil pH (Hope et al., 1991; Auerbach et al., 1997) and dusts that carry toxic soluble salts will also have adverse effects on plants (Prajapati and Tripathi, 2008a,b,c,d). Leaf-extract pH may also be a useful indicator as SO_2 entering the leaf is dissolved in inter-cellular water of mesophyll cells to form sulfurous acid (H_2SO_3), which depending on the pH of the medium dissociates into H^+ and HSO_3^- and SO_3^- , causing acidification of the cell (Puckett et al., 1973).

The exposure of SO_2 and NO_2 could decrease leaf extract pH as the plants with high sensitivity to SO_2 and NO_2 closed the stomata faster when exposed to the pollutants (Larcher, 1995). Consequently, sensitive plants had higher leaf-extract pH than tolerant plants. Table 6 demonstrates the impact of pH variations in different plants.

5.2. Relative water content

Water plays an important role in plant life. Relative water content expresses the balance of plant water uptake and release (Jones, 1994). High water content within a plant body may help to maintain its physiological balance under air pollution stress condition (Singh and Verma, 2007). High relative water content favors resistance in plants (Dedio, 1975). Plant with sufficient water content, still can expand the total leaf area as stated by Schuppler et al. (1998). If the leaf transpiration reduces due to the air pollution, plants cannot live well due to losing its engine that pulls water up from the root for photosynthesis. It has been reported that air pollutants increases cell permeability (Keller, 1986), which causes loss of water and dissolved nutrients, resulting in early senescence of leaves (Masuch et al., 1988). Therefore, leaf relative water content (RWC) is the appropriate measure of plant water status in terms of physiological consequence of cellular water deficit (Joshi and Bora, 2011).

5.3. Pigment content, photosynthesis and stomata

The photosynthetic pigment chlorophyll, found in the chloroplasts of green plants, is an index of productivity and is called a photoreceptor. The photosynthetic pigments are the most likely to

Table 6
Impact of pH variations on stomatal behavior and photosynthesis (Modified after Freer-Smith and Taylor, 1992).

pH range	Plants	Impact	Reference
4.0–5.6	Poplar (*Liriodendron tulipifera*)	Decreased photosynthesis and stomatal conductance.	Martens et al. (1989) and Freer-Smith and Taylor (1992)
4.0–5.6	Spruce (*Picea abies,* two clones)	Decreased photosynthesis and stomatal conductance in one clone.	Van Elsacker and Impens (1988) and Freer-Smith and Taylor (1992)
2.5–5	Spruce (*Picea rubens*)	Long-term depression of transpiration	Eamus et al. (1989) and Freer-Smith and Taylor (1992)
3.0–5.5	Beech (*Fagus sylvatica*)	Increased stomatal conductance and inhibition of night-time closure.	Fliickinger et al. (1988) and Freer-Smith and Taylor (1992)
3.5–5.5	Spruce (Picea abies)	Decreased maximum and increased minimum stomatal conductance.	Barnes et al. (1990) and Freer-Smith and Taylor (1992)
5.0–6.5	*Mangifera indica*	Affect chlorophyll and decrease photosynthesis	Thawale et al. (2011) and Chauhan (2010)
5.5–6.5	*Psidium guajava*	Decreased photosynthesis and stomatal conductance	Dwivedi and Tripathi (2007), Choudhury and Banerjee (2009) and Joshi and Bora (2011)
7.0–9.0	*Ficus religiosa*	Increases the conversion of hexosugar to ascorbic acid	Dwivedi and Tripathi (2007), Choudhury and Banerjee (2009) and Thambavani and Sabitha (2011).

be damaged by air pollution (Prusty et al., 2005). Air pollution-induced degradation in photosynthetic pigments was also observed by a number of workers (Bansal, 1988; Singh et al., 1990; Sandelius et al., 1995).

Chlorophyll measurement is an important tool to evaluate the effect of air pollutants on plants as it plays an important role in plant metabolism. The reductions in chlorophyll concentration correspond directly to the reduction in plant growth. Variable that has a larger or smaller effect on growth than on photosynthesis can lead to variations in starch accumulation and thereby “dilute” nutrient concentrations in organs such as leaves (Marinari et al., 2007).

Chlorophyll pigments exist in highly organized state, and under stress they may undergo several photochemical reactions such as oxidation, reduction, and reversible bleaching (Puckett et al., 1973) hence any alteration in chlorophyll concentration may change the morphological, physiological and biochemical behavior of the plant. The different pollutants play a significant role in inhibition of photosynthetic activity that may results in depletion of chlorophyll and carotenoid content of the leaves of various plants (Chauhan and Joshi, 2008). Critical analysis of the fluorescence properties of chlorophyll a (Chl a) in photosystem II (PSII) is an effective tool in the study of the physiological aspects of photosynthesis (Govindjee et al., 2004; Bussotti et al., 2010), and has been extensively applied in plant stress investigations (Maxwell and Johnson, 2000; Adams and Demmig-Adams, 2004; Bussotti et al., 2010). Bussotti et al. (2010) assessed the impact of ozone stress in woody plants with chlorophyll a fluorescence. Leaves of *Viburnum tinus* recorded decreased photosynthesis and gaseous exchange on the augmentation of black dust (Thompson et al., 1984). Further, limestone dust deposition decreased overall plant performance through loss of chlorophyll content, inhibition of CO_2 assimilation, uncoupling of the oxygen-evolving complex and decreased electron transport in Namib Desert shrub, *Zygophyllum prismatocarpum* (Tall zygophyllum) (van Heerden et al., 2007).

Impact of simulated acid rain and iron ore dust deposition in *Eugenia uniflora* (Surinam cherry) resulted in lowest values for net photosynthesis, stomatal conductance, transpiration, chlorophyll a content and electron transport rate through photosystem II (PSII) (Neves et al., 2009). Further, Neves et al. (2009) observed that catalase and superoxide dismutase activities were decreased by simulated acid rain. In *Schinus terebinthifolius* (Brazilian Pepper) deposition of iron ore PM increased chlorophyll content, the maximum quantum efficiency of photosystem II and the electron transport rate (Kuki et al., 2008). Iron solid PM on being deposited at leaf surface of *Clusia hilariana* (Clusia) caused significant reductions in photosynthetic rate, stomatal conductance, transpiration, organic acid accumulation, potential quantum yield of PSII, and changes in daily CAM photosynthesis pattern while increase in relative membrane permeability and reduction in catalase and superoxide dismutase activities; however, lipid peroxidation did not change (Pereira et al., 2009).

Zhang et al. (2010) observed physiological responses of a diploid honeysuckle (*Lonicera japonica* Thunb.) and its autotetraploid cultivar to elevated ozone (O_3) exposure and found decrease in net photosynthetic rate and stomatal conductance. Due to O_3 exposure, chlorophyll loss, increased membrane permeability, decrease in seed yield, loss of total sulfhydryl groups, reduction of soluble protein content, and increase in guaiacol peroxidase activity were observed in leaves of soybean cultivars (Chernikova et al., 2000).

Coal smoke pollution imposed inhibitory effects of pollution stress on leaf pigments concentrations, nitrate reductase activity and the contents of reducing sugars and total N content, whereas stimulatory effects were given on stomatal index and nitrate and sulfur contents in *A. indica* (neem) (Iqbal et al., 2010a,b). Moreover, Coal-smoke emissions adversely affected photosynthesis, N-metabolism and growth characteristics of *Triumfetta rhomboidea* (Chinese Burr), as observed by Iqbal et al. (2010a,b) at pre-flowering, flowering and post-flowering stages of plant growth.

Pollutant gases such as SO_2, NO_X and O_3 produce oxy-radicals in reactions with plant material (Shimazaki et al., 1980; Sakaki et al., 1983). These radicals cause widespread damage to membranes and associated molecules including the chlorophyll pigments (Sakaki et al., 1983; Malhotra and Khan, 1984). The reduction in chlorophyll concentration in the polluted leaves could be due to chloroplast damage (Pandey et al., 1991), inhibition of chlorophyll biosynthesis (Esmat, 1993) or enhanced chlorophyll degradation. Together, SO_2 plus NO_2 can decrease photosynthesis in *Betula pendula* (Silver Birch) and also alter the gas exchange of woody plants *Fagus sylvatica* (common beech), *Populus deltoids* (eastern cottonwood) *X Populus nigra* (Black Poplar), *Picea sitchensis* (Sitka Spruce) and *Picea abies* (Norway spruce) (Freer-Smith and Taylor, 1992). Mansfield and Freer-Smith (1984) reported that as low as 70 ppb of SO_2 inhibit photosynthesis in Silver birch (*B. pendula*). For O_3 similar depressions of photosynthesis are seen in both long-term and short-term exposures (Reich and Amundson, 1985; Freer-Smith and Taylor, 1992). Effects of NO and NO_2 on net photosynthesis have generally only been seen for exposure concentrations of more than 500 ppb (Freer-Smith and Taylor, 1992).

Air pollutants, specifically PM, make their entrance into the tissues through the stomata and cause partial denaturation of the chloroplast and decrease pigment contents in the cells of polluted leaves (Rao and LeBlanc, 1966) and hence the considerable loss in total chlorophyll, in the polluted leaves exposed to air pollution stress supports the argument that the chloroplast is the primary site of attack by air pollutants such as SPM, SO_2 and NO_X (Tripathi and Gautam, 2007). A conceptual diagram showing the summary of the relationships between stomatal uptake, metabolic changes and detoxification system under PM attack in plant cells is presented in Fig. 1. Rao and LeBlanc (1966) have also reported reduction in chlorophyll content brought by acidic pollutants like SO_2 which might be due to the replacement of Mg^{2+} by two hydrogen atoms and degradation of chlorophyll molecules to phaeophytin, a chlorophyll degradation product which, no longer, serves to trap the solar energy for photosynthesis. SO_2 readily diffuse and dissolves in mesophyll tissues and produces sulfite and bisulfite ions which in turn are photo-oxidized to less toxic SO_2^-–S involving free radical reactions (Asada and Kiso, 1973). The excess free oxygen radicals may affect cellular components including chlorophyll pigments (Shimazaki et al., 1980) and may inactivate enzymes involved in chlorophyll synthesis. Several studies with higher plants exposed to different SO_2 concentrations show decreases in chlorophyll content (Inglis and Hill, 1974; Hallgren and Huss, 1975; De Santo et al., 1979; Agrawal and Rao, 1982) whereas certain pollutants increase the total chlorophyll content; others decrease it (Agbaire and Esiefarienrhe, 2009). Several researches has exhibited increase in chlorophyll content under air pollution, such as Tripathi and Gautam (2007) reported that *M. indica* (Mango) leaves subjected to air pollution showed an increase in chlorophyll content (Tripathi and Gautam, 2007). Seyyednejad and Koochak (2011) also reported the increase of chlorophyll content of *E. Camaldulensis* (Red Gum) leaves in polluted site as compared with control (Seyyednejad and Koochak, 2011). Adverse effects of dust and other air pollutants on plants with reduction in photosynthetic pigments and yield have been shown by different workers in different crops (Lerman, 1972; Bytnerowitz et al., 1987; Farmer, 1993; Saquib and Khan, 1999; Rajput and Agrawal, 2005a, b; Lone and Khan, 2007). Dusted or encrusted leaf surface is responsible for reduced photosynthesis and thereby causing reduction in chlorophyll content. Decrease in chlorophyll content as a result of increased dust deposition was noticed by Prajapati and Tripathi (2008). Plants growing in polluted areas have been observed with a decrease in the chlorophyll 'a', chlorophyll 'b' and total chlorophyll contents (Raina and Sharma, 2003). The studies of Rajput and Agrawal (2005) and Joshi et al. (2009) further revealed that air pollution in urban areas adversely affect total chlorophyll, carotenoid, and ascorbic acid in wheat plants. Swami et al. (2004) have reported a significant reduction in chlorophyll content, carotenoid, ascorbic acid, pH and moisture content in the leaves of two species of trees viz. Sal (*Shorea robusta*) and Rohini (*Mallotus philippinensis*) exposed to road side automobile pollution. According to Yu (1988), at pH above 3.5, HSO^{-3}, which is formed during SO_2 metabolization, generates superoxide radical (O^{2-}) which causes the oxidation of carotenoid and in the absence of carotenoid protection, oxidation of chlorophyll molecule occurs, resulting in the reduction of photosynthetic ability of the plant.

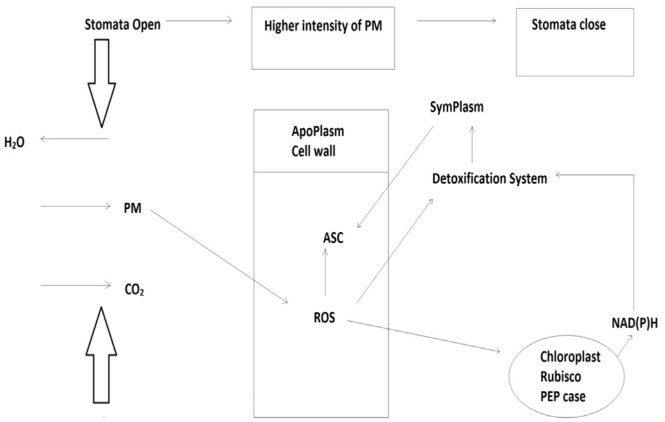

Fig. 1. Summary of the relationships between stomatal uptake, metabolic changes and detoxification system under PM attack in plant cells. Asc=ascorbate, PEPcase=phosphoenolpyruvate carboxylase, ROS=reactive oxygen species, Rubisco=ribulose-1,5-bisphosphate carboxylase (Modified after Dizengremel et al., 2008).

Sato et al. (1993) also reported that cement dust decreased the productivity and concentration of chlorophyll in a number of crops. Cement kiln dust over the leaf surface induce chloroplast damage resulting in decreased chlorophyll content. Reduction of total chlorophyll has been found in leaves of various annuals plants and conifers covered by cement dust (Pandey and Kumar, 1996; Nunes et al., 2004).

The shading effects due to deposition of suspended PM on the leaf surface might be responsible for the decrease in the concentration of chlorophyll in polluted area. It might clog the stomata thus interfering with the gaseous exchange, which leads to increase in leaf temperature which may consequently retard chlorophyll synthesis. Reduction in chlorophyll content may be due to the interference of all the metals with chlorophyll synthesis and fat metabolism, inhibiting root shoot growth, photosynthesis, nutrient uptake, leaf area, biomass etc. (Pollacco, 1987) as shown by decreased chlorophyll content in *A. Procera* (Rubber bush), leaves with increasing concentrations of metals (Pandey and Tripathi, 2011). The progressive decline of metabolites such as protein, amino acid, chlorophyll, carotenoid, and total sugar is probably due to decrease in chlorophyll content (Prasad and Inamdar, 1990). Darrall and Jager (1984) described chlorophyll as a bio-indicator for air pollution levels and effects.

Carotenoids are a class of natural fat-soluble pigments found principally in plants, algae and photosynthetic bacteria, where it play a critical role in the photosynthetic process. They act as accessory pigments in higher plants. Several researchers have reported reduced carotenoid content under air pollution (Joshi et al., 2009; Tiwari et al., 2006). Carotenoid protects chlorophyll from photoxidative destruction (Sifermann-Harms, 1987). The carotenoid contents of some crop plants were found to decrease in response to SO_2 (Pandey, 1978; Singh, 1981; Nandi, 1984) as they are more sensitive to SO_2 than chlorophyll (Shimazaki et al., 1980). carotenoids play an important function, particularly as photoprotective agents within the chloroplasts. But under stress condition, normal protective process may become overloaded and hence cellular destruction including pigment degradation occurs (Senser et al., 1990). Oxidation of carotenoids takes place through light-catalyzed reactions resulting in the formation of epoxide, which is further reduced in dark by an enzyme-catalyzed reaction (Calvin, 1955). Krinsky (1966) has confirmed the existence of such epoxide cycles and its role in protection of chlorophylls against photo-oxidation.

5.4. Ascrobic acid

Ascorbic acid, a natural antioxidant in plants plays significant role in pollution tolerance (Chen et al., 1990). Ascorbic acid, which is also called as vitamin C, is also another parameter that may decide the tolerance of plant to air pollution by checking its content in the leaf. It plays a significant role in light reaction of photosynthesis (Singh and Verma, 2007), activates defense mechanism (Arora et al., 2002) and under stress condition, it can replace water from light reaction II (Singh and Verma, 2007). Ascorbic acid, a natural antioxidant in plant has been shown to play important role in pollution tolerance (Joshi and Swami, 2007). Ascorbate was known as an antioxidant molecule able to detoxify air pollutants (Smirnoff, 1996) and it is also able to control cell expansion and cell division (Loewus, 1999; Conklin et al., 2000).

Ascorbic plays a role in cell wall synthesis, defense and cell division. It is also a strong reducer and plays important role in photosynthetic carbon fixation, with the reducing power directly proportional to its concentration (Seyyednejad et al., 2011). So it has been given top priority and used as a multiplication factor in the formula. High pH may increase the efficiency of conversion from hexose sugar to ascorbic acid, while low leaf extract show good correlation with sensitivity to air pollution (Escobedo et al., 2008, Pasqualini et al., 2001; Conklin, 2001; Liu and Ding, 2008).

Earlier report have shown that a definite correlation between ascorbic acid content and resistance to pollution exist in plants (Varshney and Varshney, 1984). Resistant plants contain high amount of ascorbic acid, while sensitive plants possess a low level of ascorbic acid. The level of this acid declines on pollutant exposure (Keller and Schwager, 1977). Thus, plants maintaining high ascorbic acid level even under polluted conditions are considered to be tolerant to air pollutants. Ascorbic acid being a strong reductant activates many physiological and defence mechanisms and its reducing power is directly proportional to its concentration (Lewin, 1976).

Pollution load dependent increase in ascorbic content of all the species may be due to the more rate of production of reactive oxygen species (ROS) such as SO^{3-}, HSO^{3-}, OH^{-}, and O^{2-} during photo-oxidation of SO^{3-} to SO^{4-} where sulfites are generated from SO_2 absorbed. The free radical production under SO_2 exposure would increase the free radical scavengers, such as ascorbic acid, super oxide dismutase, and peroxidase (Pierre and Queiroz, 1981). It also reacts with hydrogen peroxide and protects carotenes and tocopherols in plants which respond to various stress in plants exposed to pollutants (Perl-Treves and Perl, 2002). ROS such as singlet oxygen, hydrogen peroxide and hydroxyl radical are generally very reactive molecules possessing an unpaired electron, and in normal conditions the balance between the generation and diminution of ROS is controlled by the antioxidant defence system. However, when ROS are not adequately removed, an effect termed “oxidative stress” may result. Excess ROS formed within cells, can provoke oxidation and modification of cellular amino acids, proteins, membrane lipids and even DNA, creating oxidative injury that results in a reduction in plant growth and development (Hernandez-Jimenez et al., 2002; Ogawa and Iwabuchi, 2001).

In the literature there are contradictory results about the effects of individual pollutants or ambient air pollution on ascorbic acid contents of various plant species. In fumigation experiments with O_3 or SO_2 and in field experiments comparing trees growing in more or less polluted areas several authors detected increases (Bermadinger et al., 1990; Härtling and Schulz, 1995) others decreases (Keller and Schwager, 1977; Pandey and Agrawal, 1994) and others no alteration of the ascorbic acid contents (Hausladen et al., 1990; Madamanchi et al., 1991) as a result of the impact of air pollutants. Some authors (Tanaka et al., 1985; Nouchi, 1993) proved that the reaction to O_3 may vary with pollutant dose, with low concentrations causing an increase and high doses causing a loss of ascorbic acid.

5.5. Enzymes

It is well known that ascorbate-glutathione cycle occurs in chloroplasts (Foyer and Halliwell, 1976; Asada, 1984; Dalton, 1991; Foyer et al., 1991; Chernikova et al., 2000) which is responsible for scavenging toxic peroxide through SOD catalysis (Mehler, 1951; Foyer et al., 1991). Through his extensive lab investigation Lagriffoul et al. (1998) demonstrated that in contrast with growth parameters, the measurement of enzyme activities may be included as early biomarkers in a plant bioassay to assess the phytotoxicity of Cd-contaminated soils on maize plants. Doğanlar and Atmaca (2011) observed decreases in pigment and total soluble protein contents and increases in peroxidase enzyme activity in certain plants e.g. *Acer negundo* (Boxelder), *Platanus orientalis* (Oriental Plane) and *N. oleander* (Oleander) when exposed to industrial and urban pollution in Turkey. It has been demonstrated in *Pinus sylvestris* (Scotch pine) that enzyme scavenging system as well as antioxidants system like superoxide dismutase, guaiacol peroxidase, ascorbate peroxidase (Pukacka and Pukacki, 2000) can respond with raised activity to signals from environmental pollution peroxidise (Pukacka and Pukacki, 2000). Reactive oxygen species (ROS) damage plant cells by oxidizing membrane lipids, including the photosynthetic apparatus (Foyer and Harbinson, 1994; Aranjuelo et al., 2008); inhibit protoplast regeneration (Marco and Roubelakis-Angelakis, 1996; Aranjuelo et al., 2008); and damage proteins, chlorophyll, and nucleic acids (Foyer and Harbinson, 1994; Aranjuelo et al., 2008).

Singh et al. (2011) observed that in mustard, the ozone exposure considerably increased oxidative stress reduced leading to significant losses of photosynthetic pigments and protein in leaves while accelerated the antioxidative defense system in order to combat its impact. Sulfur dioxide, one of the major airborne pollutants, can penetrate plant foliage through stomata and can cause metabolic or physical injury (Khan and Malhotra, 1982). Sulphur fumigation as SO_3^{2-} and SO_4^{2-} inhibited RuBP carboxylase, but earlier proved to be more inhibitory (Khan and Malhotra, 1982). Ozone effects on the *Lamottea dianae* were studied under controlled condition by Calatayud et al. (2011) and they marked decline in the in vivo CO_2 fixation capacity of Rubisco. Apart from this other noticeable effects were callose accumulation, formation of pectinaceous wart-like cell wall exudates and phloem alterations (Calatayud et al., 2011).

5.6. Sugar content

Soluble sugar is an important constituent and source of energy for all living organisms. Plants manufacture this organic substance during photosynthesis and breakdown during respiration (Tripathi and Gautam, 2007). Pollutants like SO_2, NO_2 and H_2S under hardening conditions can cause more depletion of soluble sugars in the leaves of plants grown in polluted area. The decrease in total sugar content of damaged leaves probably corresponded with the photosynthetic inhibition or stimulation of respiration rate (Tzvetkova and Kolarov, 1996). Likewise, Bucker and Ballach (1992) found the level of soluble carbohydrates decreased due to fumigation with mixture of O_3, SO_2 and NO_2 both in young and mature leaves, this decrease in soluble sugars may be a consequence of increased metabolic consumption of energy under stress conditions. Higher respiration rate was reported in tree species resistant to air pollution (Lorenc-Plucinska, 1982). A higher inhibition of photosynthesis and photorespiration in the less resistant individuals were established in *P. sylvestris* (Scotch pine) by Lorenc-Plucinska (1982). Reduction in sugar content of crop around the cement dust polluted area can be attributed to increased respiration and decreased CO_2 fixation because of chlorophyll deterioration (Tripathi and Gautam, 2007). The reduction in total sugar reflects the interference of light absorption caused by deposition of dust over the surfaces of leaf.The low sugar levels may be due to lowered synthesis or diversion of the metabolites to other synthesis processes. The reaction of sulfite with aldehydes and ketones of carbohydrates can also cause reduction in carbohydrate content. The increase of soluble sugars was also observed following chronic exposure (Miller et al., 1969). The increase in soluble sugar was reported in *Albizia lebbeck* (Siris) and *Callistemon citrinus* (Red Bottlebrush) grown in industrial land (Seyyednejad et al., 2009). Increase in amount of soluble sugar is a protecting mechanism of leaves it has been shown in Pinto bean in exposure with different concentration of ozone (Dugger and Ting, 1970). It has been demonstrated in milkweed that the changes observed in sugars, amino acids, and phenols due to ozone exposure may alter plant-insect relationships as a result of shifts in nutritional quality for insect-herbivores (Bolsinger et al., 1991).

5.7. Protein

Protein is one of most essential foliar biochemical constituents of plants and is required for enzymatic activity in plant species. Protein content in plants exhibits both increasing and decreasing trend in response to pollution stress depending on the plant species and its inherent resistance against pollution. Reduction in protein content might be due to the enhanced rate of protein denaturation which is also supported by the findings of Prasad and Inamdar (1990). Constantinidou and Kozlowski (1979) found enhanced protein denaturation and breakdown of existing protein to amino acid as the main causes of reduction in protein content. A decline in the foliar protein content was also observed at the polluted sites. Kumar and Dubey (1998) have also concluded that pollutants coming out of auto-exhaust may cause inhibitory effect on protein synthesis. The reduction in the foliar protein content was probably due to either breaking down of existing protein and/or reduced *de novo* synthesis of protein (Iqbal et al., 2000; Singh and Jothi, 1999). Reduction in protein content could also be attributed to decreased photosynthesis (Constantinidou and Kozlowski, 1979; Singh et al., 1988). Rana et al. (2000) observed effect of various concentrations of SO_2 on the quantity of free proline in the anthers of *Brassica juncea* L (mustard). The effects of pollutants on plants include pigment destruction, depletion of cellular lipids and peroxidation of polyunsaturated fatty acid (Tiwari et al., 2006). The deleterious effects of the pollutants are caused by the production of reactive oxygen species (ROS) in plants, which cause peroxidative destruction of cellular constituents (Tiwari et al., 2006). proline accumulation often occurs in a variety of plants in the presence of different stresses which act as a free radical scavenger to protect plants away from damage by oxidative stress (Wang et al., 2009). There appears to be a relationship between lipid peroxidation and proline accumulation in plants subjected to diverse kinds of stress (Wang et al., 2009). If such a relationship exists, proline accumulation might play an important role in inhibiting air pollution-induced lipid peroxidation. For example, proline accumulation in leaves of plants exposed to SO_2 fumigation (Tankha and Gupta, 1992), heavy metals (Wang et al., 2009) and salt (Woodward and Bennett, 2005) stress has been reported (Tankha and Gupta, 1992; Wang et al., 2009; Woodward and Bennett, 2005). Seyyednejad and Koochak (2011) also demonstrated that under air pollution conditions, proline level of polluted leaves significantly increased. Copper is a micronutrient for plants and a component of various proteins, particularly those involved in both the photosynthetic (plastocyanin) and the respiratory (cyto -chrome oxidase) electron transport chain. In excess, the absorbed copper plays a cytotoxic role by generating ROS in Fenton-type reactions, leading to disturbance of metabolic pathways and macromolecular damage (Hegedus et al., 2001). Protein synthesis decreased due to the low chlorophyll and reduced leaf area surface similar findings was reported by Baszynski et al. (1980). Trivedi and Singh (1995) noticed significant reduction in protein content in a few plants as a result of fly ash PM. Marked alterations in photosynthetic pigments and protein content in foliar tissues as a result of auto-exhaust pollution were noticed by Verma and Singh (2006). Proline, a total free amino acid accumulated in plants when they experience moisture stress conditions and decline on release of stress (Pokhriyal and Raturi, 1984). Proline accumulation provides high stability in drought induced stress. Heavy metals reduced crude proteins in agricultural crops and in this study the effect was much pronounced on forestry tree species as compared to agricultural crops (Hemalatha et al., 1997). The decrease in protein could be understood that metal in all likelihood would interfere with sulfur containing amino acid and crude protein resulted in decreased protein content (Somasundaram et al., 1994). Amino acids play a central role in plant primary metabolism. Being early products of photosynthesis and nitrogen assimilation, they represent an important link between nitrogen and carbon metabolism (Durzan and Steward, 1989).

Metals and PAH may be an inextricable component of PM and may impose adverse effect on vegetation.

6. Impact of metals

Heavy metals are classified among the most deleterious groups of anthropogenic environmental pollutants due to their toxicity and persistence in the environment (Sayyed and Wagh, 2011; Tiwari, 2011; Anim et al., 2012; Gyamfi et al., 2012; Prajapati et al., 2012). Presence of heavy metals in the road-side and particulate matter in the ambient air is serious and has adverse human health effect (Rai, 2015). Presence of heavy metals above their threshold level also could be potentially harmful on local vegetation and environment. Elevated levels of heavy metals may cause oxidative stress either by inducing the generation of reactive oxygen species (ROS) within sub cellular compartments or by decreasing enzymatic and non enzymatic antioxidants due to an affinity with sulfur-containing group (–SH) (Gupta et al., 2012, Benavides et al., 2005). Heavy metals like Zinc, Copper, Iron and Sulfur are required for biosynthesis of enzymes, auxin and some protein, essential for normal growth and development of plants (Onder and Dursun, 2006). Disturbance in their concentration can cause significant modification of biochemical processes in plants leading to loss of production (Bucher and Schenk, 2000), lower yield and quality of agricultural crops. Loss of hazel nut production due to high zinc concentration was shown in the leaves of *Corylus avellana* (Hazel). Excessive copper may destroy subcellular structure of plants (Sresty and Madhava, 1999). High concentration of sulfur become toxic to plant as it produces various biochemical changes in plants such as destruction of the Chlorophyll and cells causing reduction in the thickness of the annual rings of the trees (Kantarci, 2003).

Photosynthetic efficiency of most of the plants is affected by heavy metals (Krupa and Baszyński 1995; Burzynski and Klobus, 2004). Metals have multifaceted impact on photosynthesis (Krupa and Baszyński, 1995; Prasad and Strzałka, 1999; Burzynski and Klobus, 2004). The excess of Cu, Cd, or Pb inhibits directly the photosynthetic electron transport (Krupa and Baszyński, 1995; Myśliwa-Kurdziel et al., 2002) as well as the activities of Calvin-Benson cycle enzymes or net assimilation of CO_2 (Prasad and Strzałka, 1999; Burzynski and Klobus, 2004). Ce in conjunction with UV-B UV-B radiation caused the decrease in chlorophyll content, net photosynthetic rate, Hill reaction activity, photophosphorylation rate and Mg^{2+}-ATPase activity in in soybean (*Glycine max* L.) (Liang et al., 2010). Cu was more toxic than Cd and Pb for photosynthesis in cucumber leaves in context of net photosynthetic rate and stomatal conductance in leaves (Burzynski and Klobus, 2004). Higher plants function as biomonitors of aerial metal contamination due to their accumulation properties. Keane et al. (2001) and Prajapati and Tripathi (2007) have biomonitored the trace metals present in PM_{10} using leaves of *Saraca indica* (Ashoka), *Lantana camara* (Lantana) and dandelion leaves. *N. oleander* (oleander) (Sawidis et al., 1995; Aksoy and Ozturk, 1997; Rossini Oliva and Mingorance, 2006; Doğanlar and Atmaca, 2011), *Quercus ilex* (Oak) (Monaci et al., 2000), *Robinia pseudoacacia* (Acacia) (Celik et al., 2005), *P. pinea* (Pine) (Mignorance and Olivia, 2006), *Pyracantha coccinea* (Firethorn) (Akguc et al., 2008), and *Murraya paniculata* (Orange Jasmine) (Titseesang et al., 2008) are reported as biomonitors for various heavy metal pollutants (Doğanlar and Atmaca, 2011).

7. Impact of poly-aromatic hydrocarbons (PAH)

PAHs influence physicochemical properties of membrane, which may cause inhibition of photosynthetic and respiration processes too (Huang et al., 1996; Duxbury et al., 1997; Tukaj and Aksmann, 2007; Va´ňova et al., 2009). PAHs and/or products of their biotransformation and photomodification influence biochemical and physiological processes taking part in growth and development of plants (Va´ňova et al., 2009). At higher concentration fluoranthene (used to correlate the impact of polyaromatic hydrocarbons) inhibited growth of shoot, callus and the content of photosynthetic pigments (chlorophyll a and b, carotenoids) in 21-day-old pea (*Pisum sativum* L.) (Va´ňova et al., 2009).

8. Impact of PM on biodiversity and climate change

PM may adversely affect the biodiversity particularly in relation to urban forests. It has been reported that deposition of fine PM to forests may act as a source of nutrients, but also changes leaf surface properties, increasing the duration of surface wetness and modifying the habitat for epiphytic organisms, leading to increased risks from pathogens (Cape, 2008). Further, PM can directly affect photosynthesis, through abrasion, stomata blockage and smothering of the leaves, once the PM settle down on the organ surface (Hirano et al., 1995; Das et al., 2012). Indirect effects may involve chemical and physical modification of the soil properties. Germination and the early growth stages are the most vulnerable periods of a plant life cycle; thus, any environmental stress, combined with the sensitivity of the species, can interfere with a species establishment success (Fan and Wang, 2000; Grantz et al., 2003). This condition is likely to affect the vegetation dynamics, causing further ecological problems (Narayan et al., 1994). Also, it has been reported that PM deposited directly to the soil can influence nutrient cycling, especially that of nitrogen, through its effects on the rhizosphere bacteria and fungi (Grantz et al., 2003). It is reported that deposition of PM affects the microbial community living in the phyllosphere that plays an important role in decomposition of litter fall (Prajapati et al., 2012; Miller et al., 1982). Since fungi are important decomposers, changing the fungal community on the needles finally weakens the decomposer community, hence, decreasing the rate of litter decomposition. All these processes alter nutrient cycling (Bruhn, 1980). Slowly decomposing litter influences nutrient availability within the ecosystem because of accumulation of carbohydrates and mineral nutrients (Cotrufo et al., 1995). Apart from PM, secondary air pollutants also suspected to have profound adverse impact on forest ecosystem (Cape, 2008).

It is worth to mention that increased PM may reduce radiation interception by plant canopies and may reduce precipitation through a variety of physical effects and hence may contribute to the climate change (Grantz et al., 2003). PM affects climate change by scattering incoming and, to a lesser degree, outgoing radiation. Black carbon and other dark particles absorb radiative energy. Coarse particles and cloud droplets formed by the condensation of water vapor on particles also have radiative effects, which can have local and global impacts on climate change (source: The North American Mosaic: An overview of key environmental issues, cec.org; (accessed 10.11.13.)). Paleomagnetic studies particularly in the context of paleosols may provide an insight to past climate changes (Rai, 2013). Elevated CO_2 and N fertilization treatments may result in an unbalanced nutritional status of leaves in three poplar species (Marinari et al., 2007). It has been demonstrated that elevated CO_2 reduced carboxylation capacity, induced photosynthetic acclimation and reduced enzymatic and/or nonenzymatic antioxidant activities, suggesting that changes in electron flow did not cause any photooxidative damage (Aranjuelo et al., 2008). The influences of increases in CO_2 levels on plant growth and physiology have been studied by a several researchers (Wolfe et al., 1998; Urban, 2003; Long et al., 2004; Aranjuelo et al., 2005a, 2006; Erice et al., 2006; Aranjuelo et al., 2008). Under the condition of CO_2 enrichment, it has been observed that after an initial stimulation of photosynthetic rates, the carboxylation capacity of plants decreases after long-term exposure (Ainsworth et al., 2004; Long et al., 2004; Aranjuelo et al., 2005b, 2008). PM plays a role in various environmental issues, especially ground-level ozone and climate change. $PM_{2.5}$ and ground-level ozone are closely related through common precursors, sources and meteorological processes. Because of this close relationship, changes in the emissions of one pollutant can lead to changes in the concentrations of PM or ground-level ozone PM and their precursors can be carried long distances by the wind and eventually be deposited on the ground or in water. Their deposition makes lakes and streams acidic, changes the nutrient balance in coastal waters and large river basins and encourages eutrophication, depletes the nutrients in soil, damages sensitive forests and farm crops, and affects the diversity of ecosystems. PM also carry toxic components such as mercury, which can degrade water quality and hence aquatic biodiversity.

9. Genotoxic effects of PM on plants

The genotoxic effect of PM pollutants on the ecosystem, including the build-up of resistant species, is also of considerable concern. The effects of toxic compounds, and the subsequent genotoxic effects on plants, are of particular importance as plants comprise a large portion of our biosphere and constitute a vital link in the food chain (Grant, 1998; Rajput and Agrawal, 2005). Estimating PM genotoxicity is therefore crucial to evaluating risk to the environment and plant health.

Also, most of these PM pollutants e.g., polycyclic aromatic compounds (PACs), heavy metals and halogenated aliphatic hydrocarbons, have been shown to be genotoxic (Grant, 1998; Prajapati and Tripathi, 2008a,b,c,d,e,f; Rai, 2015; Rai, 2016a,b). Higher plants can be considered sensitive and efficient indicators of genotoxicity when compared to other physical and chemical treatment methods. The benefit of using plants as a bioindicator of genotoxicity is that they are sensitive to exposure periods ranging from a few minutes to weeks, are easy to handle and deploy, and are inexpensive compared to conventional indicator methods. Furthermore, although plant response to air toxics cannot be extrapolated directly to predict the effect on human health, the findings of plant bioassays can provide an indication of air pollution and environmental stress (Guimarães et al., 2000; Prajapati and Tripathi, 2008a,b,c,d,e,f; Rai, 2015; Rai, 2016a,b).

Various PM pollutants have been demonstrated to cause genotoxicological impact on plants as well as humans (Rai, 2015). For instance, although polycyclic aromatic hydrocarbons (PAHs) are relatively chemically inert compounds, however, through metabolic activation to electrophilic derivatives (e.g. diolepoxides, quinones, conjugated hydroxyalkyl derivatives) these are capable of covalent interaction with nucleophilic centres of DNA. These adducts of PAH to DNA cause base pair substitutions, frameshift mutations, deletions, S-phase arrest, strand breakage and a variety of chromosomal alterations (Singer and Grunberger, 1983; Dipple, 1985; EC, 2002; Piraino et al., 2006; Rai, 2015; Rai, 2016a,b).

Tradescantia pallida, *Allium cepa*, *Arabidopsis thaliana*, *Vicia faba* (Grant, 1998) and *Trifolium repens* (Piraino et al., 2006) are only some of the plant species that have been used successfully in air genotoxicity biomonitoring through the application of cytogenetic tests.

Tradescantia can assess mutagenic effects by the formation of micronuclei in meiotic or mitotic cells and pink mutations in stamen hairs. The Trad-MCN bioassay has been used extensively for monitoring environmental genotoxicity, and it is particularly sensitive to chemical mutagens (Ma et al., 1994; Batalha et al., 1999; Monarca et al., 1999; Guimarães et al., 2000; Carreras et al., 2006; Prajapati and Tripathi, 2008a,b,c,d,e,f).

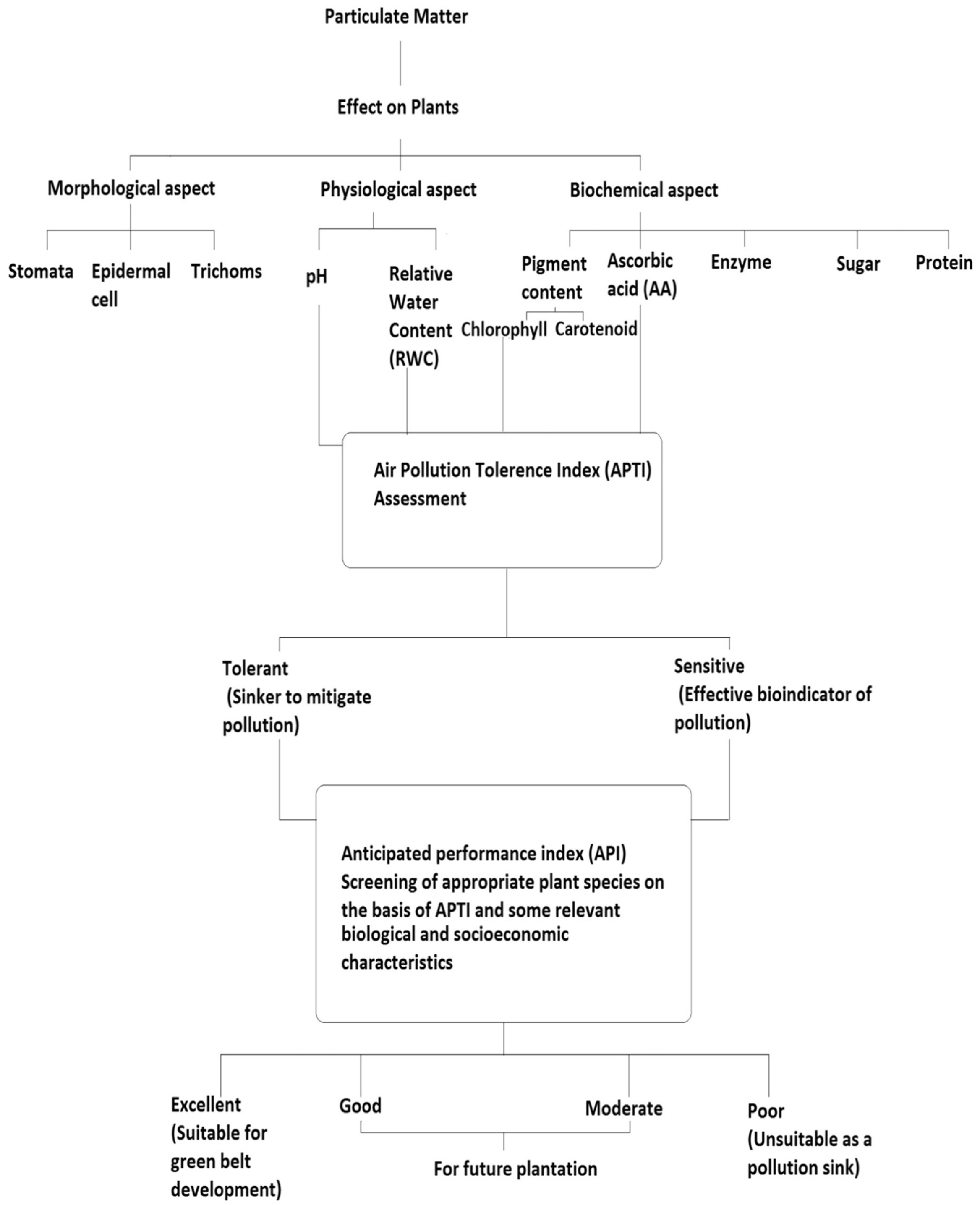

Fig. 2. A conceptual diagram of APTI and API showing the screening of tolerant and sensitive plant species.

10. Conclusions

Although plants possess some stress-tolerant mechanisms within them, considerable amount of damage is caused to them as a result of PM or dust deposition leading inhibition of photosynthetic activities and protein synthesis as well as susceptibility to injuries caused by microorganisms and insects. Metabolically, physiological, biochemical and genotoxic responses of plants to PM pollution can be viewed as potentially adaptive changes. Evaluation of morphological, physiological and biochemical changes in plants on exposure to PM pollution is an important step to isolate and screen tolerant plants from sensitive ones. A conceptual diagram of APTI and API showing the screening of tolerant and sensitive plant species is presented in Fig. 2. Tolerant biomonitors may be used and recommended for green belt development to cope up with the problem of PM pollution.

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