A review on current status of essentiality and toxicity of nickel in food crops

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***Abstract*: Nickel (Ni) is an essential micro-nutrient for plants. However, very low amount of nickel is required for normal plant growth. Therefore, it is important to understand the function and toxic effects of nickel in plants as the level of nickel pollution in the environment increases. Based on available data, Ni has been identified as a component of various enzymes in plants and has a decisive metabolic role in certain enzymatic activities, such as maintaining proper cellular redox state. This review paper is based on an overview of the data available over the past 20 years, with the core content including the adverse morphological, physiological and biochemical effects of Ni stress in plants. The purpose of this review is to cover the occurrence and sources of nickel, its dimensions of essentiality and toxic effects due to oxidative stress in food crops.**

***Keywords*: Nickel, Antioxidant system, Chlorophyll contents, Heavy metals, Photosynthesis, Plant stress**

Introduction:

Nickel is a silvery white, hard, malleable, ductile and ferromagnetic metal which is the twenty eighth element in periodic table [13]. Nickel has several oxidative states which is widely found in the environment [35,58]. The name "Nickel" comes from the word "*Kupfenickel*" meaning "Old Nick's Copper", named by German miners as niccolite emits toxic fumes when heated [20]. Nickel in the divalent state has been observed to be the main broad analogue of nickel in biological systems. Most of nickel exists as solid hydroxide at pH 6.7 whereas all nickel complexes soluble at pH 6.5 [22]. It is usually combined with sulphur and iron in pentlandite and millerite accordingly [2]. Nickel is a trace metal like other heavy metals which is emitted into the environment from both natural and anthropogenic sources [7,44]. Smelting, industrial wastes, volcanoes, fertilizer applications, electroplating industries, organic manures, sewage sludge, urbanization etc. are the sources of nickel which is emitted to the atmosphere [4,26]. On the other hand, some anthropogenic sources which are responsible for nickel outflow in the environment are residual fuel oils, mines of nickel and municipal waste calcination [6]. Nickel pollution is reported across the world including Asia, Europe and North America [47]. Nearly thousand tons of nickel are produced and released into the environment every year [36]. Nickel has become a serious problem over the past few decades as its concentration in contaminated soil has reached 26,000 ppm, i.e., 20% higher than uncontaminated areas. Nickel is an essential micronutrient for the growth and development of plants and some microorganisms, though there is no existing evidence of essentiality of nickel in human body [29]. It plays a vital role in wide range of morphological and physiological functions such as germination of seeds and productivity. Nickel is an essential component which helps in the production of the enzyme urease, which in turn helps in metabolism in plants. Nickel also helps in the degradation of methylglyoxal which is a cytotoxic compound naturally produced by cellular metabolism [37,56]. Deficiency of nickel may produce an effect on growth and metabolism of plants and result in chlorosis, necrosis of leaf tip, barrier N metabolism and Fe uptake [18]. However, the higher amount of nickel may cause of inhibiting enzymatic activity, photosynthetic electron transport, mitotic activity, chlorophyll biosynthesis and reduction in plant growth [55]. Nickel toxicity is reported to be more prevalent than its deficiency [12]. In the present review, we get an overview of role of nickel as a micronutrient and its toxicity at higher level in different food crop plants.

Nickel as a micronutrient:

Nickel being one of the essential micro elements in low concentration for biochemical activities, which helps in nitrogen metabolism that stimulates plant growth and seed germination [17,46]. In most cases nickel is retained in root, but it is also found in vacuoles and cell walls of stems and leaves [2]. Plant growth and nitrogen metabolism can be observed by applying nickel at low concentration which prove that nickel is a beneficial micronutrient for crops [7]. Certain enzyme activities such as urease, glyoxalase, methyl–CoM reductase, superoxide dismutase and hydrogenases are metabolized by low concentration of nickel absorption [3]. Multiple experiments performed on different plant species showed that Ni and urease are vital for plant activity. Low concentration or deficiency of nickel hampered the nitrogen metabolism and exaggerate the concentration of urea in shoots [13,51]. Low concentration of nickel helps in hydrolysis, germination, nitrogen metabolism, water relations, photosynthesis, plant growth, disease resistance and senescence [4,58]. Hyper accumulation of Ni in plant roots makes it toxic and in turn defends it from pathogens and herbivores [31].The uptake of other heavy metals like Zn, Fe, Cu and Mn are decreased because of accumulation of Ni in plants roots [6].

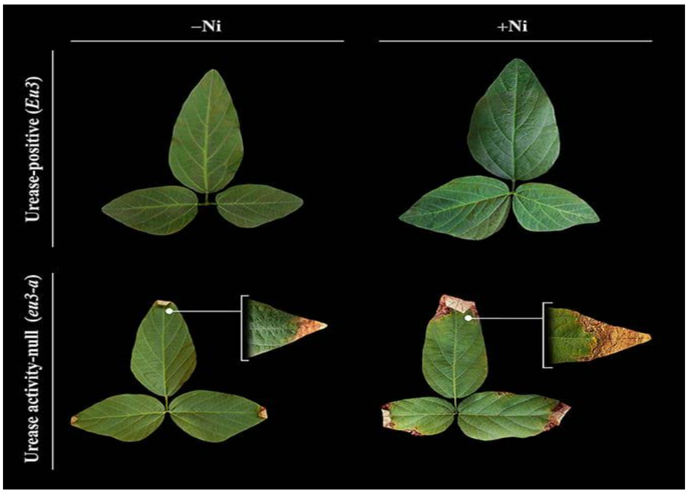

Figure 1: Deficiency of nickel [8]

Transport and distribution of Nickel in plants:

Nickel as a micronutrient cannot absorb directly by plants. There are three major absorption process such as– root interruption, mass flow and diffusion [3].Nickel is deported from roots to shoots and leaves through xylem by passive diffusion and active transport [13].Primary transport in xylem, re-transport in phloem, and transfer from xylem to phloem are important processes of element redistribution within plants. Transport of xylem is directed from roots to aboveground plant parts in transpiration flow, whereas transport of phloem proceeds from source to sink and is more selective [50]. Nickel absorption depends on nickel binding proteins and metal-ligand complexes such as nicotianamine (NA), histidine (his) and organic acids [2]. Concentration of Ni^{2+} in soil, plant metabolism, pH, redox condition, hydrous oxide and the acidity of the soil are the factors responsible for Ni uptake by plants [12]. It is reported that Cu^{2+} and Zn^{2+} inhibit Ni^{2+} uptake competitively, as these three metal ions are absorbed by same transport process [13]. Nickel is absorbed more in acidic soil rather than alkaline soil as in the case of *Lathyrus sativus* L.(Fabaceae) where it is reported that the Ni absorption is increased at pH 5.0 but is decreased at pH 8.0. Secondary active transport of chelated Ni^{2+} has been reported with corresponding proteins that specifically bind Ni^{2+}, such as HoxN (high-affinity nickel transport protein, a permease), metallothionein and metallochaperones [2]. The specific mechanism of Ni^{2+} uptake is still not understood. It is not clear whether the hyper accumulator species absorb Ni due to its specific tolerance level or on the characteristic pattern of particular absorption mechanisms on the lowered Ni^{2+} uptake which depends mostly on the characteristic patterns of translocation and distribution and binding Ni into insoluble complexes.

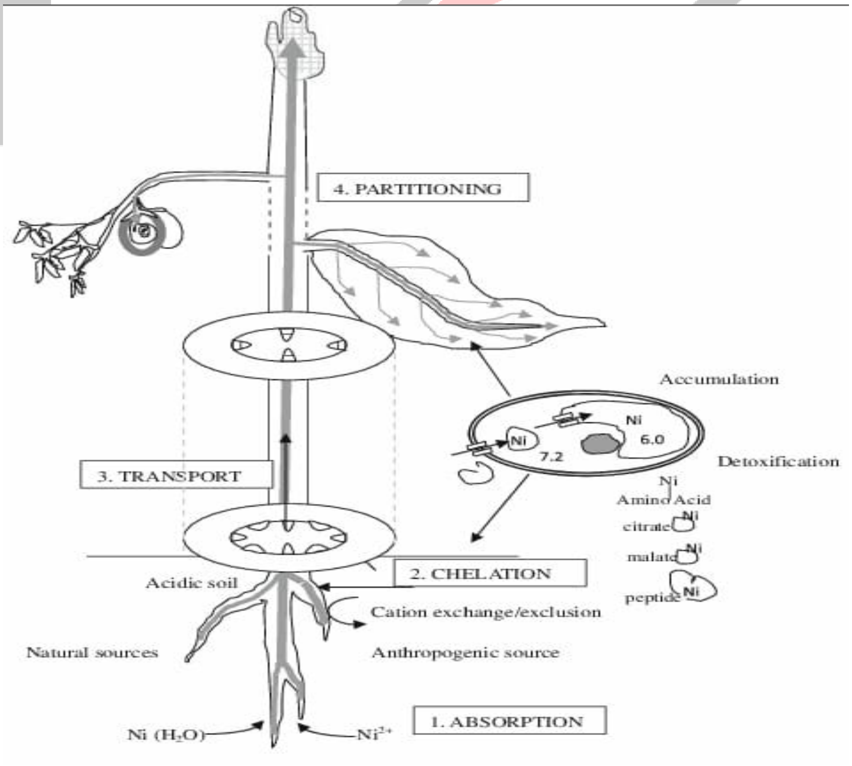

Figure 2: Nickel uptake, transport & distribution in plant [13]

Antagonistic effect of Nickel on plants:

Heavy metals such as Cr, Cd, Ni, Zn, Cu & Pb are the most significant environmental pollutant. These anthropogenic components exaggerated in the environment by industrialization and urbanization [39]. Nickel, one such toxic metal propels adverse physiological and metabolic processes in plants [32,53]. High concentration of nickel in soil interrupt normal growth process in wheat (*Triticum aestivum* L.), cabbage (*Brassica oleraceae* L.), pigeon pea (*Cajanus cajan* L.) and many other plants. Due to the interference of all physiological processes: water absorption, transpiration, respiration, photosynthesis, absorption and distribution of essential elements, nitrogen metabolism, etc., have documented a reduction in plant growth at toxic concentrations of Ni [30].

Effect on growth, development and physiological characteristics:

Plant growth is a very necessary for sustaining life on earth. Minerals and microelements which are present in environment are responsible for the internal and external development of plants. In above studies it is mostly confirm that excessive absorption of nickel is toxic for the growth and metabolism of plants. It does not only affect the metabolism but also influence the plant morphology, chlorophyll content, antioxidant enzyme activity, malondialdehyde (MDA) content and electrolyte leakage [11].The cell membrane, the first living structure of plant cells, is the target of heavy metal toxicity. Membrane permeability measured in terms of electrolyte leakage has been reported to increase for plants exposed to various heavy metals, including Ni [49]. Mainly the roots absorb and accumulate the nickel and as a result root length is inhibited more than shoot length. Therefore, to evaluate of heavy metal toxicity, roots are mainly tested.

The wheat seedlings showed significantly decline in growth after applying 250 µM and 500 µM concentration of nickel. The root length, shoot length and total biomass clearly inhibited on high concentration of nickel treatment in hydroponics procedure [12]. In other experiment, after 7days continuous exposer in 0.43mM Ni treatment, the root of *Nicotina tabacum* L. became dark brown and growth of roots and shoots are severely inhibited [12]. Application of 200 mM Ni on barley seedlings severely affected the biomass of roots and in the case of 400 mM Ni concentration the seedlings didn't survive. Germination rate of seeds and growth of seedlings of *Brassica juncea* L. were decreased under 50 and 100 mg dm^{-3}[13]. The above-mentioned toxic effects of nickel on seed germination and seedling growth may be due to the adverse effects produced by Ni and disrupted cell proliferation, elasticity of cell wall and poor metabolic procedure [40].

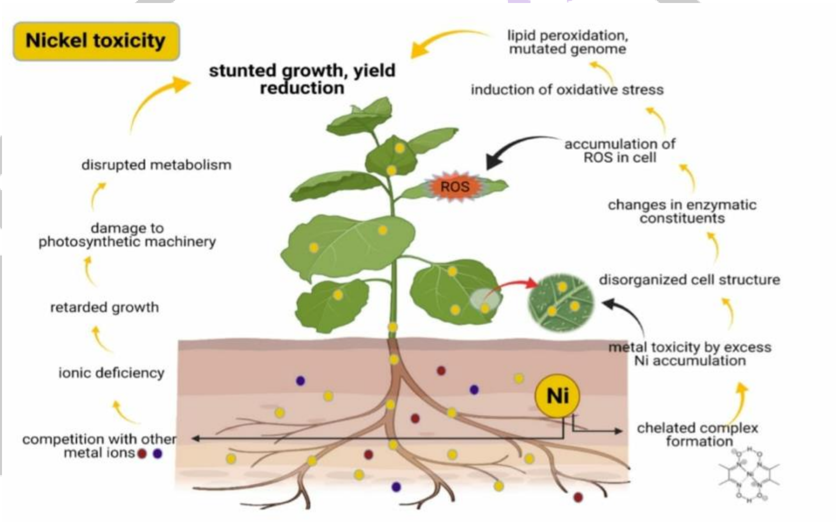

Figure 3: Toxic effect of nickel in plants [18]

Ni concentration	Plant Species	Visual symptoms
100 mM	Onion (*Allium cepa* L.)	Decrease root growth [13]
>10 mM	Onion roots (*Allium cepa* L.)	Observed irregular shape of nuclei [13]
Ni alone or combined with other Heavy metals	Maize roots (*Zea mays* L.)	Reduce of dry weight [13]
100 µM	Black nightshade (*Solanum nigrum* L.)	Membrane damage & Ni accumulation in root [6]
200 µM	Cabbage *(Brassica oleracea* L.*)* and wheat *(Triticum aestivum* L).	Visible chlorosis, necrosis & wilting [6]
0.5 mM	Arugula (*Eruca sativa* L.)	Decrease biomass production and chlorophyll content [41]
1.0 mM	Rye grass (*Lolium perenne* L.)	Decrease plant nutrition and biomass of shoot, chlorosis [24]
150 µM	*Kalmegh (Andrographis. paniculatus* (Burm. f.) Wall.	Reduction of biomass [38]

Table 1: Symptoms shown in variable Ni concentration

Suppression of photosynthesis and effect on chlorophyll content:

The fundamental basis of competitive success in green plants is photosynthesis. In higher plants the principal organ of photosynthesis is leaf. Many studies showed that concentration of Ni determines and influence the photosynthesis process in plants [15]. The adverse impact of toxic level of Ni inhibits photosynthetic protein complexes directly or indirectly [13]. Ni interferes in the activity of some antioxidant enzymes which is involved in various physiological processes such as Calvin cycle and chlorophyll synthesis, and may even hamper photosynthetic activity [21]. Phototoxicity depends on the different concentration of Ni present in soil and its toxicity dominates the measurement of root elongation and biomass production in hydroponics culture [23]. The inhibition rate of photosynthesis increases chlorosis, necrosis and wilting in leaves [58]. Ni stress not only damages photosynthetic pigments and apparatus it also affects organization of chlorophyll contents including chloroplast structure, mesophyll cells, epidermal tissues [56]. We conclude that, Ni toxicity interrupts electron transport, obstructs chlorophyll synthesis, declines Calvin cycle enzymes and makes deficiency of CO_2 by stomatal closure [3]

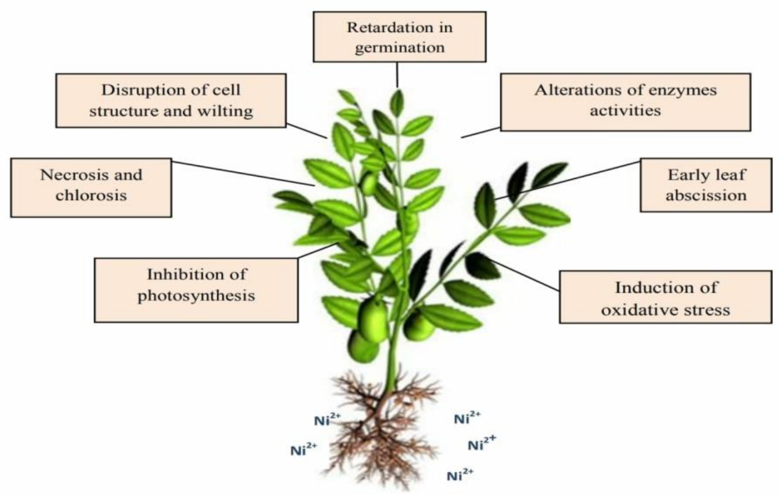

Figure 4: Induction of growth and other physiological characteristics cause Ni stress [6]

Ni stress destroys the size and structure of grana and reduces the number of thylakoids in chloroplasts. Reducing the size of grana, the non-appressed lamellae is increased and disrupted the functional mechanism of photosynthesis [6]. *In vitro* study showed that the oxidative Ni stress significantly hampers the activity of Photosystem II (PSII) and it targets the photosynthetic Electron Transport Chain (ETC) whereas, *in vivo* study showed Ni stress mainly inactivates Photosystem I (PSI) [10]. Light Harvesting Complex II (LHCII) is the main active pigment- protein complex of PSII where the oxidative stress of Ni damages LHCII and obstructs with energy transfer between proteins and pigments [9].

Ni stress affects the mechanism of chlorophyll which is essential part of photosynthesis and damages the structure of xanthophyll and carotenoids. The accumulation of Ni in cells changes chloroplast structure, decreases chloroplast size and chlorophyll contents (chlorophyll a and chlorophyll b). Excess nickel destroys the content of cytochromes as well as ferredoxin and plastocyanin in the thylakoids which mainly involve in electron transport chain [6]. Nickel induces some changes such as decrease in cellular moisture content and also results in oxidative stress which leads to membrane lipid peroxidation [3]. Number of studies showed that increasing nickel concentration adversely influences photosynthetic protein complexes in maize (*Zea mays* L.) and in *Brassica oleracea* L. seedlings which were grown in agar with applying $NiSO_4.7H_2O$ (10-20 g/m^{-3}) and showing as a result of deformation of the plastoglobuli and disorganization in the membrane lipid composition [3,6**]**. Increasing Ni concentration 20 µM to 100µM on maize seedlings, the chlorophyll a decreased 50 % and chlorophyll b decreased 70 % onwards. In black gram (*Vigna mungo* L.), the photosynthetic pigments were clearly affected because of oxidative stress of nickel [1].

Plant Species	Visual Symptoms
Bean (*Phaseolus vulgaris* L)	Grey spots on leaves cause of necrosis and coalesce [4]
Cabbage (*Brassica oleracea* L)	Marginal chlorosis [4]
Barley (*Hordeum vulgare* L.)	Leaf chlorosis [4]
Oats (*Avena sativa* L.)	Pale yellow strips visible through the length of the leaves [4]
Soybean (*Glycine max* L.)	Leaf necrosis 150 µM Ni stress [8]

Table 2: Toxic symptoms on chlorophyll content in different plants

Impact on activity of antioxidant enzymes:

From two decades a number of studies proved that an inversely proportional relation between Ni stress and antioxidant enzymes in plants. Nickel is only toxic in higher concentration and being a heavy metal, it is considered as abiotic stress of plants [34]. Antioxidant enzyme activity may depend on the duration and type of stress treatment in the plants. The efficient antioxidant defence system of plants is compromised against the oxidative nickel stress which may convert into superoxide free radicals [6]. The enzymatic antioxidants comprise of catalase (CAT), catechol peroxidase (POD), superoxide dismutase (SOD), glutathione-s-transferase (GST), glutathione peroxidase (GSH-Px), glutathione reductase (GR), guaiacol peroxidase (GOPX) and ascorbate peroxidase (APX) which may help to remove reactive oxygen species [5,52].Increasing enzymatic activity is thought to be a metabolic response under various stress conditions, including water stress, air pollutants, and heavy metals, and is usually measured in crude extracts regardless of the intracellular distribution of these enzymes [48].

Both *in vitro* and *in vivo* studies reported that the antioxidant enzyme activity increases in the presence of low concentration of nickel but if the Ni concentration become increased, the activity of enzymes have repressed [2]. Excess of nickel in soil may reduce many other antioxidant activities and helps to generate accumulation of ROS and finally it is being an oxidative stress for plants. Since Ni is a redox active metal so it can generate ROS directly [13].

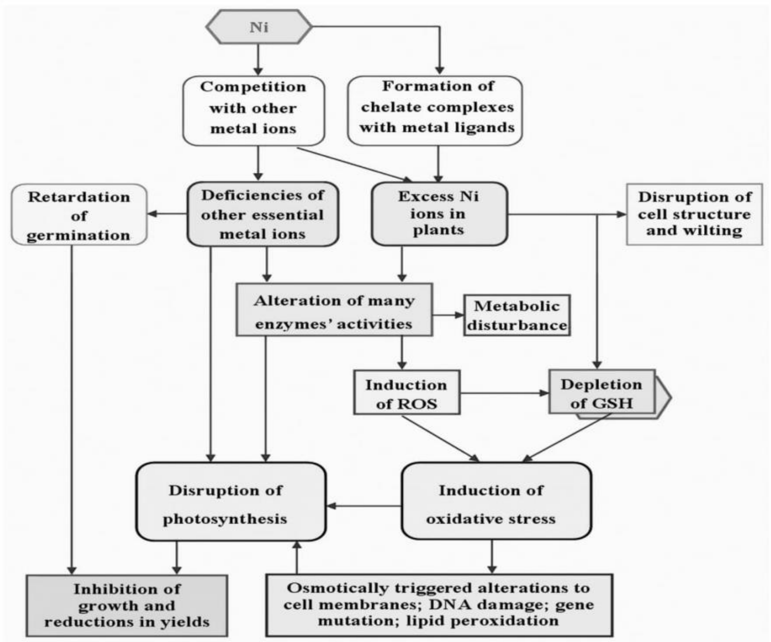

Figure 5: Enzymatic activity of excessive Ni in plants [42]

Plant Species	Nickel Concentration	Applied days	Mechanism of tolerance
Garden pea plant (*Pisum sativum* L.)	10, 100 and 200µM	14 days	The activity of SOD reduced but activity of CAT remains unchanged [1].
Pigeon pea plant (*Cajanus cajan* L.)	0.5 mM	6 days	Reduce the activity of CAT [1].
Jatropha curcas L. embryos	100, 200, 400 & 800 µM	-	At first SOD activity increase at 400µM Ni treatment but at 800µM the activity significantly decreased. POD activity reduced with excess Ni concentration [2].
Black gram seedlings (*Vigna mungo* L.)	20, 40, 60, 80 & 100µM	7 days	Nickel induced oxidative damage in black gram and reduces antioxidant enzyme activities [2].
Cabbage plant (*Brassica oleracea* L.)	0.5mM	8 days	POD activities significantly declined [12].
Hydrocharis dubia L. seedlings	0.5, 1, 2, 3 & 4 mM	3days	SOD, CAT & POD activities reduced [12]

Table 3: enzyme activities under different concentration Ni stress in various plant species

Reactive Oxygen Species (ROS) and antioxidant system :

Previous research has found that heavy metal toxicity is always responsible for oxidative stress for plants.ROS is generally produced by different metabolic reactions that occur in different cellular parts of plants, such as mitochondria and chloroplasts [1]. An overabundance of Ni produces categorical by products such as reactive oxygen species (ROS) in various cellular organelles [45].

Under stressful conditions, including exposure to excessive concentrations of heavy metals, an imbalance between ROS production and removal occurs in plant tissues.This may subsequently lead to oxidative damage to many important macromolecules such as lipids, proteins and nucleic acids. The most common indicator of oxidative stress is lipid peroxidation, which results in disturbance of membrane integrity and thus enhanced permeability[54].

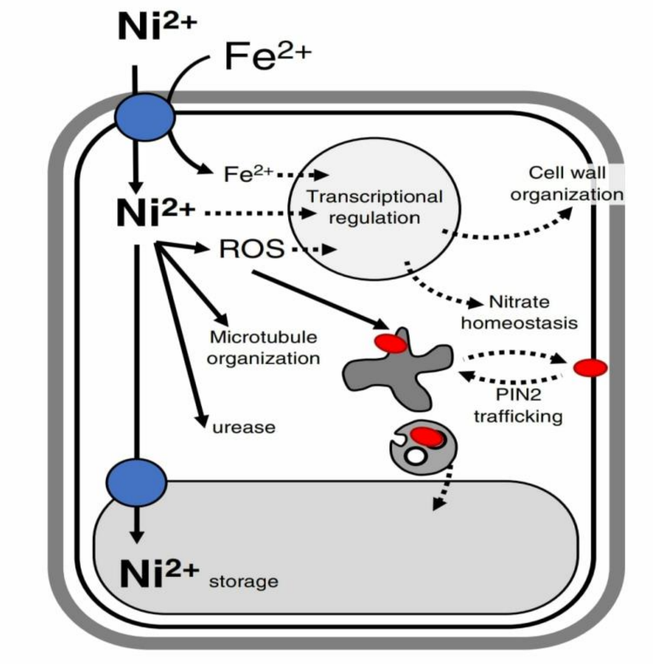

Figure 6: ROS Stress under Ni accumulation[14]

ROS can cause severe damage in the deficiency of scavenging capacity of cellular defence system [27]. Destruction of organic layers through the peroxidation of lipids is the most destroying impact of production of ROS. Nickel toxicity which is considered as an abiotic stress for plants leads to over production of ROS which helps to regenerate of peroxidation of lipids, disruption of cell homeostasis, oxidation of proteins, inactivation of antioxidant enzymes, destroying the ultra structure of DNA and other cellular constituents of plant cells [25]. This may induce to alter the properties of cells and compels to dysfunction of organelles and entire cells [16]. Singlet oxygens, superoxide radicals, hydrogen peroxide and hydroxyl radicals are considered as ROS of plants [2]. ROS formed because of inevitable leakage of electrons from the electron transport activities of chloroplasts, mitochondria, and plasma membranes. Nickel enhances the generation of ROS which may lead cellular homeostasis [43]. During respiration and photosynthesis of plant cells, the ROS is produced and mainly generated when electrons get transferred from reduced electron's sources to oxygen [2]. In wheat plant, ascorbate peroxidase (APX) is playing a vital role to decline H_2O_2 from the leaves under Nickel stress [3]. Both *in vitro* and *in vivo* studies showed that ROS is the main lead to generate oxidative stress under higher concentration of Nickel.

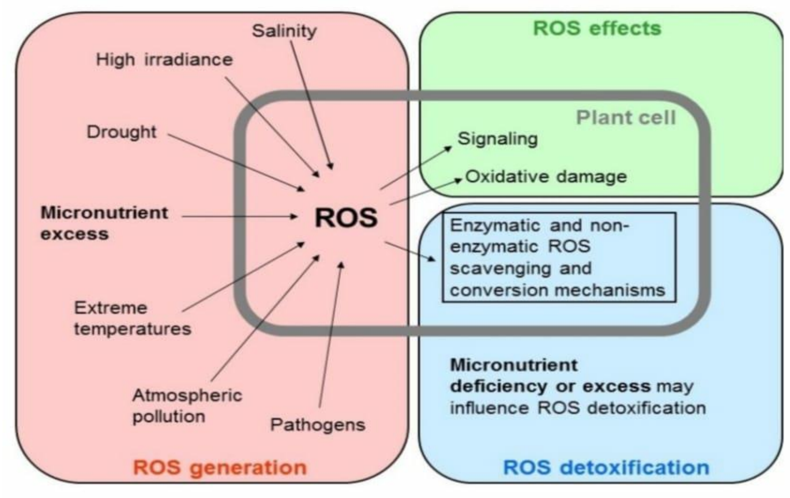

Figure 7: ROS generation and detoxification [28]

Conclusion:

This review article has provided instant information related to the importance of Ni in plant growth and development. Sufficient nickel plays a crucial role in a wide range of physiological processes from seed germination to productivity. Furthermore, without an adequate supply of this metal, plants cannot complete their life cycle. In addition to this, at elevated levels, it alters all metabolic activities of the plant, such as water relation and mineral nutrition, leading to enzyme inhibition, disruption of stomatal function, photosynthetic electron transfer and degradation of chlorophyll molecules, consequently minimize the photosynthetic rate and biomass yield. Excess nickel concentrations induce oxidative damage in plants, which is associated with the observed multiple toxic effects of the metal. Therefore, large amounts of ROS and lipid peroxides can damage many organelles and DNA, oxidize proteins and lipids, and also degrade photosynthetic pigments. However, plants are equipped with organized defence mechanism to combat toxic effects, including exclusion/restriction of metals from entering cells through the plasma membrane.

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