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REVIEW ON HEALTH EFFECTS OF CHROMIUM EXPOSURE: REFLECTION FROM OXIDATIVE STRESS TOWARDS CARCINOGENICITY

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ABSTRACT

Recently, the main theme of world is urbanization as well as industrialization as a result, there has been a growing concern about increase in toxic metals contamination *i.e*. chromium that affect the environment, living organisms and human health. Recently, laboratory and clinical reports about the pathogenesis of the toxicity, carcinogenicity and allergenicity of chromium compounds have been reviewed as a basis for consideration of the mechanisms involved for pathogenecity. Chromium compounds have been proofed as a redox cycling environmental carcinogenic agent that provoke apoptosis as the most important mode of cell death. Defects in apoptosis regulatory mechanisms contribute to carcinogenesis induced by Cr (VI). Activation of apoptosis signaling pathways is tightly linked with the generation of reactive oxygen species (ROS). Exposure to Cr [VI] can result in various point mutations in DNA and to chromosomal fragment, and also the oxidative alteration in proteins. In recent report, we have collected the previous reports and overviewed the cytotoxic as well as genotoxic mechanisms of Cr [III] and Cr [VI]. In conclusion, chromium induced cytotoxicity can be attributed to oxidative stress started from glutathione mediated metal reductive activation and continued by mitochondrial/lysosomal toxic interaction.

KEY WORDS: **Chromium, oxidative stress, Glutathione, mitochondrial damage, Apoptosis, Genotoxicity**

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INTRODUCTION

On the increase of urbanization as well as industrialization, living bodies as well as human are exposed in polluted environment. Among several pollutants, heavy metals' including chromium, shows adverse effects on human health.[1] In foremost, chromium, seventh most abundant metal in the earth's crust and a potent environmental contaminant, primarily in the form of a range of alloys, has been in wide industrial use for long times. In environment, chromium (Cr) stables in two oxidation states; trivalent Cr (Cr III) and hexavalent Cr (Cr VI). Cr (III) compounds are also in extensive use in certain industrial processes, have been found as irritants, but not as carcinogens or allergens whereas Cr VI can take action as a sensitive irritant, as a carcinogen and also as an allergen to human. [3] Excessive exposure has in association with chromate production. [1] In nature, chromate elements are toxic to cells, toxic to genes and also carcinogenic. In response to cytotoxic, genotoxic, and carcinogenic effect, chromates are proposed to affect through generation of reactive oxygen species (ROS), DNA damage and several changes like DNA-protein cross-link, DNA strand breaking, and chromosomal instability. [2,3] The Cr [VI] ion has been accepted as the principal cause of these toxic responses, and there have been many studies of the occurrence and prevention of dermatitis, asthma and cancer in these and allied industries. Many experimental and biochemical studies of the mechanisms of these effects have been reported, both to explore their pathogenesis and to try to understand the basic biological processes involved. The world literature published from 1985 to mid-2000 has been reviewed for information about the mechanisms of the allergenic, carcinogenic and irritant effects of chromium. [3] Our review is focused on theories about the mechanisms of the three major toxic properties of chromium ions, irritancy, carcinogenicity and the related genetic toxicity, and allergenicity, concentrating on biochemical and other mechanisms likely to be involved according to the laboratory and clinical literature.

Physicochemical properties of chromium

Chromium and its principal ions [atomic number 24, relative atomic mass 51.996] arise in each of the oxidation states from +2 to +6, but only the 0 [elemental metal form], +2, +3 and +6 states are common. Divalent chromium [+2] is unstable in most compounds as it is easily oxidized to the trivalent form by air. Accordingly, only the trivalent Cr [III] and hexavalent Cr [VI] forms are important for human health [Fig. 1]. Valid generalizations of the biological effects of chromium in its elemental form cannot be made. This review is important to realize the +3 and +6 oxidation states which have very different chemical and hence biological properties. The difference in electric potential between Cr [VI] and Cr [III] reflects the strong oxidizing potential of hexavalent chromium. The substantial energy [1.33 eV] required to oxidize the trivalent Cr to the hexavalent form. This oxidation of Cr[III] never occurs in biological systems. In contrast, reduction of Cr [VI] occurs spontaneously in the organism unless present in insoluble form. For example, in blood, Cr [VI] is rapidly reduced to Cr [III]. Once Cr[VI] has penetrated the membrane of the red blood cell, it is reduced and Cr[III] becomes bound to cellular constituents for building it incapable to depart the erythrocyte. [3]

Absorption, distribution, excretion and metabolism

After human exposure to Cr [III] by breathing, urinary absorption of Cr[III] were found to be increased indicating respiratory absorption. However, the degree of the pulmonary uptake of Cr [III] is influenced by the nature of the compound. [4] Studies in experimental animals are compatible with the poor absorption of Cr[III] salts following exposure by oral, dermal and inhalation routes.[5,6] Following oral administration of sodium chromate in tracer doses to humans indicated that about 10% of the administered dose had been absorbed from the gastrointestinal tract. After duodenal administration approximately half of the administered radioactivity appeared to have been absorbed on the basis of faecal excretion where as 10% found in the urine during the first 24 h. Reduction of Cr [VI] to the trivalent form was demonstrated. [7] The correlation between respiratory exposure to Cr [VI] and urinary excretion of chromium has been demonstrated in welders and in workers in the chrome plating industry. [8,9] The respiratory uptake rate depends on the solubility of the chromium compound. [8] Cr [VI] is reduced in the lower respiratory tract by the epithelial lining fluid and by pulmonary alveolar macrophages. In contrast to Cr [III], which is bound to plasma proteins, such as transferrin, Cr [VI] entering the blood stream is taken up selectively by erythrocytes, reduced, and bound to haemoglobin. Reduction of Cr [VI] during transport in the blood is consistent with the finding that only Cr [III] is present in the urine. Aitio et al. [8] reviewed the results of biological monitoring of exposure to chromium [predominately soluble Cr [VI]] and identified three half-lives for excretion of 7 h, days and 3 to 5 years. The best estimates for the sizes of these different compartments are 40%, 50% and 10%, respectively. Retention of chromium on the skin was found following topical application of sodium chromate. [3]

Human exposure

In case of adult human, the lethal oral dose is considered to be 50–70 mg soluble chromates per kilogram body weight. Studies in man and experimental animals have established the essential role of ''trace amounts'' of Cr[III] [50–200 µg/day] for the maintenance of normal glucose metabolism. The intake of the chromium by the oral route does not represent toxicity problem. The clinical features of acute poisoning are vomiting, diarrhoea, haemorrhage and blood loss into the gastrointestinal tract, causing cardiovascular shock. [10] If the patient survives for more than about 8 days; the major effects resulting from oral ingestion of toxic doses of chromium are liver and kidney necrosis. Although parenteral administration of chromium to experimental

animals can lead to teratogenic effects but birth defects have not been related with human exposure to Chromium. [11] *Dermal exposure:* Chronic ulcers of the skin and acute irritative dermatitis have been consistently reported in workers exposed to chromium containing materials. [11] Chromates and Cr [VI] released from alloys and chromium plated objects have been allied with the induction of allergic contact dermatitis. Generally, it is assumed that Cr [VI] is necessary for the sensitization, while both chromium [VI] and Cr [III] may cause dermatitis in sensitized individuals. Intracellular reduction of Cr [VI] to Cr [III] seems to be a prerequisite for this effect. The strong acid and powerful oxidizing properties of soluble chromium ions are regarded as the primary grounds of its irritant action on epithelia. [3] *Respiratory exposure:* Irritation of the respiratory tract causes due to inhalation of Cr [VI] compounds. Not only irritation but ulceration and also perforation of the nasal septum occur frequently in workers employed in the chromate producing and hexavalent chromium-using industries. International Agency for Research on cancer reported that sinonasal cancer possibly indicating a pattern of excess risk for these rare tumours. Rhinitis, bronchospasm and pneumonia have also been reported in workers exposed to Cr [VI] together with impairment of respiratory dynamics during respiration. Though the precise role of chromium is uncertain as such workers are often exposed to other chemicals. [10] Nevertheless, bronchial asthma has been reported to be due to exposure to chromates. Epidemiological studies of workers in the chromate production industry have consistently shown excess risks for lung cancer. Studies in experimental animals have confirmed Cr [VI] to be carcinogenic by inhalation but not by ingestion or skin contact. There is inadequate evidence in experimental animals and man for the carcinogenicity of Cr [III] compounds or metallic chromium. [1]

General mechanisms of chromium toxicity

It is known to us that, Cr [VI] is much more toxic than Cr [III]. Cr [VI] enters cells more readily than Cr [III] compounds and is eventually reduced to Cr [III]. Cr [VI], tested as sodium dichromate, was transported through mammalian cell membranes by the carboxylate, sulphate and phosphate carrier systems. The kinetics of uptake also implicated the rate of reduction to Cr [III]. The intracellular reduction of Cr[VI] implies the generation of short-lived species of pentavalent and tetravalent chromium with affinities for cellular constituents. It may differ from that of Cr [III]. The pentavalent form is stabilized by glutathione. [3] Thus, the reduction of Cr [VI] is considered to serve as a detoxification process. Once absorbed and retained in biological tissue chromium compounds occur as Cr [III]. Glutathione and cysteine seem to be the most important cofactors for the intracellular reduction of Cr [VI], but ascorbic acid, microsomes in the presence of NAD/NADH, microsomal cytochrome P450, mitochondria and proteins such as haemoglobin and glutathione reductase may also be active in the reduction process. [1]

Genotoxicity by chromium

Cr [VI] compounds of various solubility in water. It is consistently active in numerous studies covering a wide range of tests for genetic and related effects [WHO, 1990. [10] With purified DNA and isolated nuclei Cr [III] compounds were generally more reactive than Cr [VI] compounds. However, with cellular test systems Cr [III] compounds of various solubility gave positive results in only a minority of studies, often under particular treatment conditions or at very high concentrations, which were usually commands of enormity higher than those needed to obtain the same effects with Cr [VI] compounds. In particular, DNA damage was not observed in the cells of animals after administration of chromic chloride and micronuclei were not found in the cells of animals given chromic nitrate. The DNA damage, gene mutation, sister chromatid exchange or cell transformation in cultured animal and human cells is not generally produced by Cr [III] compounds but chromosomal aberrations were often observed with high concentrations of Cr[III] compounds. Assessments of viscosity, ultraviolet absorption spectra and thermal denaturation of purified DNA and RNA showed that, at variance with Cr[VI], which [as an oxidizing agent] breaks the polynucleotide chain, Cr[III] is responsible for physicochemical alterations of nucleic acids by interacting with the phosphate groups and nitrogen bases. DNA strand breaking, DNA-DNA and DNA-protein cross-linking and modification of nucleotides, such as 8-hydroxyguanine, indicative of oxygen radical formation is caused by Cr [VI]. However, these reactions do not occur in cell-free systems in the absence of reducing agents and current consensus is that the highly reactive intermediates such as Cr [V] and Cr [IV] formed during cellular Cr [VI] reduction are primarily responsible for the observed genotoxicity.41 Cellular reducing agents that may be of importance for Cr [VI] reduction includes ascorbate and sulphydryl compounds such as cysteine and glutathione. [3] Although hydroxyl, cysteinyl and thionyl radicals may be formed during the reduction of Cr[VI] it is not known if these intermediates are of relevance in chromium induced carcinogenesis. [12,13] The oxygen free radicals generated by chromium-mediated reactions activate a transcription factor known as NFkB, which is a critical activator of genes involved in inflammation, immunity and apoptosis. Thus, chromium carcinogenicity is showing to be complex matter. [3] Accordingly, although it is now accepted that inhalation exposure to Cr [VI] can cause cancer, the mechanism[s] involved, the involvement of other valence states and persuade of solubility are still very much in dispute.

Reactive oxygen species

Reactive oxygen species [ROS] are the most important group of radical species. [14,15] ROS and reactive nitrogen species [RNS] are known to play a dual role in bio-logical organisms, since they can be either harmful or beneficial to living systems. [16] One further beneficial example of ROS at low concentrations is the induction of mitogenic reaction. In disparity, at elevated concentrations, reactive oxygen species can be

imperative intermediaries of break to cell structures, including lipids in cell membranes, proteins and nucleic acids. [17] Antioxidant actions of non-enzymatic antioxidants and antioxidant enzymes correct the adverse effects of ROS. [18] Oxidative stress can be defined most simply as the imbalance between the production of ROS, RNS capable of causing peroxidation of lipid layer of cells and the body's antioxidant defense. [19] There is some evidence showing metals, like iron, cadmium, chromium, copper, mercury, lead, nickel, aluminum and vanadium is able to generate ROS and RNS through lipid peroxidation, DNA damage, depletion of sulfhydryls, and altered calcium homeostasis. [20,21] The mitochondrial transcription could be sensitive to free radical harass, to lipid per oxidation yields, or mutually. It has been anticipated that mitochondrial impairment being the result of oxidative-induced damage plays a critical role in the metals toxicity. [22-27] The overall objective of this paper is to provide a concise and current review of the effects of metals toxicity on mitochondrial function and oxidative stress.

Metals-induced oxidative stress

Free radicals are defined as atoms or molecules that contain one or more unpaired electrons; the toxicity of many xenobiotic, generally metals, especially chromium are associated with the production of free radicals, which are in turn toxicant and implicated in the pathophysiology of many diseases. [19] The possible role of oxidative damage in pathology of metals may throw in to their toxicity. [28] Increased rates at ROS generation have often been suggested to contribute to the toxicity of high levels of heavy metals, including lead, cobalt, mercury, nickel, cadmium, vanadium, molybdenum, chromium and aluminum, as well as other elements such as selenium and arsenic. [29] However, the evidence for a primary role of oxidative stress in toxicity for the elements in question is not particularly convincing. For example, although increased lipid per oxidation has often been demonstrated in isolated cells uncovered to metals, or in tissues from animals disillusioned by metals, this per oxidation may be a consequence of tissue injury and GSH depletion cause by the metals rather than on early contributor to the metal toxicity. [30,31] Several studies have focused on metal-induced toxicity and carcinogenicity, accentuating their role on the production of ROS / RNS in biological systems. [31]

Mitochondria: The key source of cellular oxidative stress

As estimated, [32] some 0.2–2% of the oxygen taken up by cells is converted by mitochondria to ROS, mainly through the production of superoxide anion. Mitochondria consume 85–90% of a cell's oxygen to support oxidative phosphorylation, the major-energy creation system in cells that works during oxidation of fuels through the synthesis adenosine triphosphate [ATP]. [33] Hence, the mitochondrial respiratory chain serves as a major store of ROS, ensuing as of the disproportionate superoxide anions. [34] Within mitochondria, it is the electron transport chain that is the main source of ROS. [35] The sites of ROS production along the chain have been subjected to many studies. [36,37] Topical conclusions show that two major sites of superoxide production are at complex I and complex III. [38-40] As described in previous reports, [19,41,42] the term oxidative stress refers to both oxidative damage and oxidative stress impact on signaling, transcriptional control and other normal processes within cells; the term has also encompassed the effects of oxidants such as RNS. In mammalian tissues, there are at least three distinct superoxide dismutase [SOD] isoenzymes, including one manganese form [Mn-SOD] present in the mitochondrial matrix and two copper and zinc forms [Cu,Zn-SOD], one of which is located only in the cytosol and the other one is in various extracellular fluids, respectively. [43] SOD plays a key role in catalyzing the dismutation of $O_2^{\bullet-}$ to O_2 and H_2O_2. Glutathione peroxidase [GPx] and catalase [CAT], remove hydrogen peroxide. In the presence of change over metals, H_2O_2 can be a bridged to the extremely reactive OH. [44,45] Metabolizing water and corresponding alcohols [ROH] need to reduce H_2O_2 and a wide range of organic hydroperoxides [ROOH] by some catalyzing reactions through GPx. Another plentiful reactive radical is Nitric oxide [NO°]. NO° acts as an significant oxidative biological signaling molecule, having an important responsibility on a large variety of diverse physiological processes. These include neurotransmission, blood pressure regulation, protection mechanisms, smooth muscle recreation and immune regulation. [46,47] NO is enzymatically generated by the actions of nitric oxide synthases [NOS] and has a half-life of only a few seconds in an aqueous background. Beneath these circumstances, Peroxynitrite anion $ONOO^-$ is an oxidizing free radical produced by NO^- and the $O_2^{\bullet-}$, being capable to reason DNA fragmentation and lipid per oxidation. [47,48]

Chromium induced mitochondrial dysfunction and oxidative stress

Chromium [Cr] is one of the imperative reasons of allergic dermatitis and has toxic and carcinogenic effects on humans and animals. [49,51] Chromate plating and other hexavalent Cr [VI] disclosure can occur in several industrial uses such as chromate pigments, chromate-based corrosion inhibitors, stainless steel machining and welding, etc. [50,52] Different scientific report currently suggest the in vitro and in vivo effects of oxygen scavengers, glutathione vitamin B2, vitamin E and vitamin C on chromate-induced injuries together with DNA damage, lipid oxidation, enzyme silence, cytotoxicity and mutagenesis. Also, Chromium overdoes occurs in the workplace primarily in the valence forms Cr [VI] and Cr [III]. [53] Inhalation of hexavalent chromium can result in several disorders such as pulmonary fibrosis, chronic bronchitis, lung cancer, occupational asthma and others. [54-58] Cr [VI] can also generate highly reactive oxidant such as peroxynitrite. In actuality, Cr [VI] diminution consequences in numerous oxidants: [a] Cr[V] can directly oxidize cell components, [b] Cr[IV] catalyzes robust hydroxyl radical [HO°] generation in Fenton-like reactions with H_2O_2; and some enzymes simultaneously

reduce Cr[VI]to Cr[V] and generate superoxide [$O_2^{\bullet-}$].[59-61] The reduction of mitochondrial core protein results in inhibits of electron transfer chain and thereby impaired oxygen decline. These phenomena escort to radicals' formation and oxidative stress.[62] The results of a study showed that total blood Cr intensity, SOD, lipid peroxidation activity and DNA damage were significantly higher and GSH level was significantly lower in exposed group as compared to the unexposed group.[63] Also, the studies showed that the toxicity of Cr [III] is largely allied with cross linking mechanism which leads to multiform DNA damages, like, strand breakage, DNA-protein interaction by cross linking, DNA-DNA interface, Cr-DNA adducts and base adjustments in cells.[64-67] Intercellular matrix the glutathione rapidly forms a complex with Cr [VI], followed by a slow reduction of Cr [VI] to yield Cr [V]. In addition, superoxide can further reduce Cr [VI] to Cr [V], which can further catalyze the demonstration of H_2O_2. Thus, it leads to the creation of DNA damaging hydroxyl radical. Also, Cr is a ROS promoting agent, resulting in mitochondrial damage that leads to apoptosis and carcinogenicity. For example, in vivo, Cr [VI] exposure results in apoptosis, mitochondrial instability, release of cytochrome c and at least initiation cell disruption [Fig 2]. All in all, effective Cr chelating or elevated cellular antioxidation is the most useful way of treating Cr-induced oxidative injury, leading to the prevention of neurodegenerative disorders and chronic diseases.[68]

Epigenetic and oncogene activation by Cr^{6+} exposure

Previously non-microarray based studies used to report the function of mutations in oncogene. The reports suggested that several oncogenes like *ras, p53, Bcl-2, cyclin-D1* altered their expression in Cr^{6+} carcinogenesis. This possibility has been supported after several research studies, was conducted in experimental test systems or cancer tissues of Cr^{6+} exposed workers. In lung cancer due to exposure of Cr^{6+}, activated *ras* oncogene was found. Though it is considered a rare event and not involved in Cr^{6+} carcinogenesis. Changes in *Bcl-2* and *p53* expression level were noted although these were found to be unspecific to Cr^{6+} carcinogenesis.[69] Further investigations revealed mutant *p53* gene in lung cancer of chromate exposed workers.[70] It was illustrated that the elevated serum levels of pantropic p53 [pan-p53] proteins in Cr^{6+} workers;[71] and induction of p53 level up to 6-fold in Cr^{6+} exposed human lung fibroblasts.[72] The key role of *p53* gene in chromate toxicity or carcinogenesis was demonstrated using *p53* deficient transgenic mice;[73,74] intervention studies showed that the loss of crucial gene *p53* increased the genomic DNA fragmentation.[73] The effect of short term high dose [0.05 and 0.25 µM] Cr^{6+} exposure on benzo alpha pyrene [B[a]P] [DNA damage] directed gene alteration in mouse hepatoma cells was investigated [51]. RT-PCR based analysis showed up regulation in genes related to apoptosis [*Aifm, Bid, Bak, Bcl2, Fas, Apaf1, Tnf, Bax*], cell cycle control [*Rad17, Mdc1*], tumor suppression [*p15, p16, p18, p19, p21, p27*], DNA damage [*Brca1, Brca2, ATM, Gadd45, Mgmt*] and down-regulation in genes related to drug metabolism [*Cyp1b1, Cyp1z2, Gsta1, Nqo1, Aldh3, Cyp1a1*]. In an earlier study, the exposure of Cr^{6+} was found to increase the carcinogen-DNA adduct formation in mouse hepatoma cells.[76] These observations indicated that Cr^{6+} exposure facilitated the carcinogen - DNA adducts formation causing DNA damage. With respect to epigenetic changes, Cr^{6+} induced methylation of p16 promoter and repression of DNA-mismatch-repair or tumour suppressor genes mut L homologue 1[*MLH1*] and *MLH2* has been reported besides the genetic instability in chromate lung cancer.[77,78] Sun *et al* [79] reported an increase in protein as well as mRNA level of G9a. G9a is a histone methyl-transferase enzyme that able to methylate *H3K9* [histone H3 lysine 9] and accounted for global elevation of its dimethylated type and silencing of tumour suppressor gene *MLH1* transcription. Others also showed that Cr^{6+} repressed the transcription co-activators.[80,81] Klein *et al* [82] showed methylation of genes and modulation of gene cyclin-D1 by Cr^{6+} in transgenic cells; study revealed the responsiveness of cell cycle regulation to the toxic metal. A crucial role of *cyclin D1* in Cr^{6+} toxicity was noticed in a study on ex-chromate workers affected with lung cancer wherein cyclin-D1 expression was found to be more as compared to non-exposed subjects harboring other disease like pneumoconiosis. The altered expression of ATM [ataxia telangiectasia mutated] gene,[83] aneuploidy and dysregulation in spindle assembly checkpoint bypass were reported in Cr^{6+} exposed cells;[84] these changes normally carry apoptosis, cell cycle ruling, because these are rudiments of cells responding to DNA damage and to genomic instability. In cell signaling [MAPK] pathway, activation of [Extra cellular signal regulated kinase] *ERK*, [C-Jun-N-terminal kinase] *JNK*, [mitogen activated protein kinase] *p38* [regulators of cell growth, proliferation, apoptosis, and differentiation.] was observed. The activation of changes depended on toxicant's profiles, resulting ROS generation or oxidative stress.[85-90] Their activation was also reported in Cr^{6+} exposed mouse embryonic stem cells;[91] lower level of toxicant activated *JNK* [c-Jun-N-terminal kinase] via leukocyte C-terminal Src kinase (*LCK*), a associate of the Src family unit of protein tyrosine kinases or the *Fyn-Cas-Crk* (*FAK/Src-Yes-Fyn/p130 CAS/CRK*) signaling cascade. *LCK* could activate *STAT3* [signal transducer and activator of transcription] and [interleukin-6) *IL-6* which contributed to inflammation and cancer.[92] Another studies investigating ROS dependent changes found that Cr^{6+} exposure activated nuclear factor kappa *β* [*Nfkβ*] and *p38* pathway. *Nfkβ* is essential for apoptosis. It was also measured an indicator of Cr^{6+} induced cytotoxicity.[93,94] Using cultured cells, investigators also showed activation of activator protein-1 (AP-1) but *HOGG1* (8-oxoguanine DNA glycosylase) gene was found to be unbiased. It is conditional that *Nfkβ* does not participate in tumourigenesis; it is somewhat linked with a decrease in cell proliferation and induction of apoptosis.[95] Over expression of inflammation specific *COX-2* via *N*fk*β* / *c-Jun / AP-1* dependent pathway was observed in normal human bronchial epithelial cells and mouse embryonic fibroblasts after Cr^{6+} exposure.[96] The signaling molecule [*VEGF*] vascular endothelial growth factor was found to be over expressed by Cr^{6+}. The *VEGF* is

involved in angiogenesis. It is usually over articulated in lung cancer, and used as prognostic marker. [97-101] One study on the divergent showed the repression of *VEGF* expression by Cr^{6+}. In signaling pathway, other types of genes that are activated in response to Cr^{6+} are *Fyn* and *LCK* and the initiation of an interferon signaling mechanism. [93,101,102] Activation of *AKT* (*α* serine-threonine protein kinase) was also noticed by Cr^{6+} in human lung fibroblast transformation. *AKT* is known to override G1/S checkpoint bypass, prevent Cr^{6+} induced decrease in localization of retinoblastoma protein and p27 (cycline dependent kinase inhibitor 1B) the key factors of G1/S checkpoint, and contribute to toxicant induced genomic instability. [104] Levels of *ApoJ / CLU* (a senescence biomarker apolipoprotien J and an oxidative stress responsive gene protein clusterin) in serum were noted to be high in shipyard welders during the oxidative stress and were found to be lower after worksite intervention. [105]

Genomic alteration by Cr exposure

In the foremost work on Cr^{6+} induced cellular gene expression modulation, Ye and Shi [106] examined genomics in human lung type II epithelial A549 cell. By following microarray of 2400 genes after using potassium dichromate as a source of Cr^{6+}, it has been investigated that the molecular basis of Cr^{6+} provoked ROS generation and the resultant oxidative anxiety, and established a significant dysregulation of 220 genes that were part of the pathways of oxidative stress, apoptosis, and also carcinogenesis. In stress response pathway [Table I], an unregulated transcription was seen in Cu/Zn superoxide dismutase (SOD), GPx, metal-regulatory transcription factor-1, *p53*, heat shock proteins [*HSP60, HSP70, HSP75*], and activating transcription factor-3. The oxidative stress reactive proteins secluded the right conformation of lately produced proteins; their main function was to protect cells from ROS / oxidative stress and preserve the vitality of cells. In apoptosis pathway Ye and Shi [106] reported up regulation of only the *hSIAH1* gene [Table I]. Functionally, this apoptotic gene facilitated to label the protein for proteasomal degradation and the programmed cell death through induction of p53 signaling during Cr^{6+} induced stress. In cell cycle regulatory pathway [Table I], the unregulated genes transcribed protein products significant for cellular endurance, cell division, cell proliferation and differentiation, G1 phase arrest in cell cycle, and tumour suppressor function. By underneath expression of the individual proteins, the down regulated genes deregulated cell cycle control via hold back of cyclin dependent kinase activation causing cell cycle arrest, and demanding epithelial cell veracity for apoptosis. In DNA repair and metabolism pathway [Table I], only three genes were found to be dysregulated. The up regulated gene encoded photolyase that was involved in DNA repair. The down regulated *CK2* [casein kinase 2] and cell division cycle [*CDC*] decreased the function of serine / threonine kinase. It is also required for DNA replication. To standardize the signaling mechanisms in cell propagation and growth, *CDK4* [cell cycle dependent kinase], *CDK5* binds with *CK2/CDC45* encoded a protein. [107]

Figure Legends

Figure 1

Diagrammatic presentation of chromium ion reduction and free radical generation.

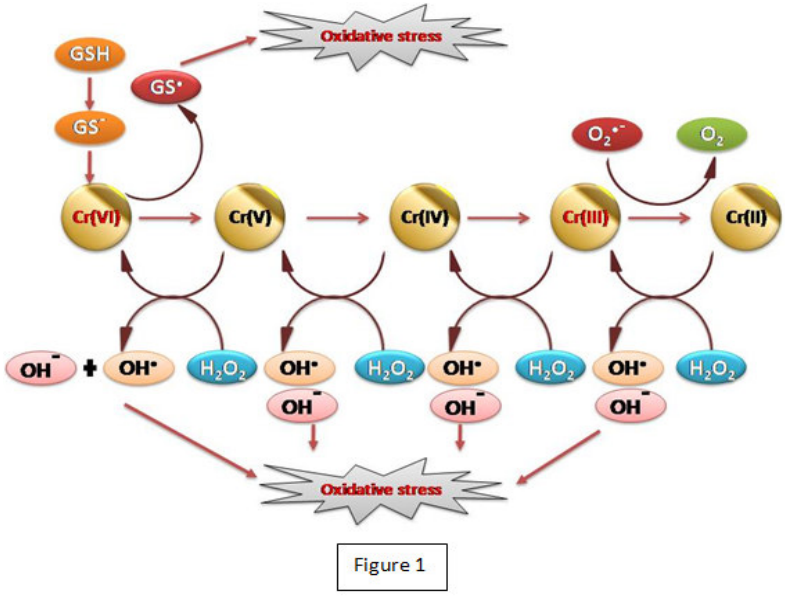

Figure 1

Figure 2

Schematic presentation of pathway of cell cycle arrest and apoptosis due to chromium ion exposure.

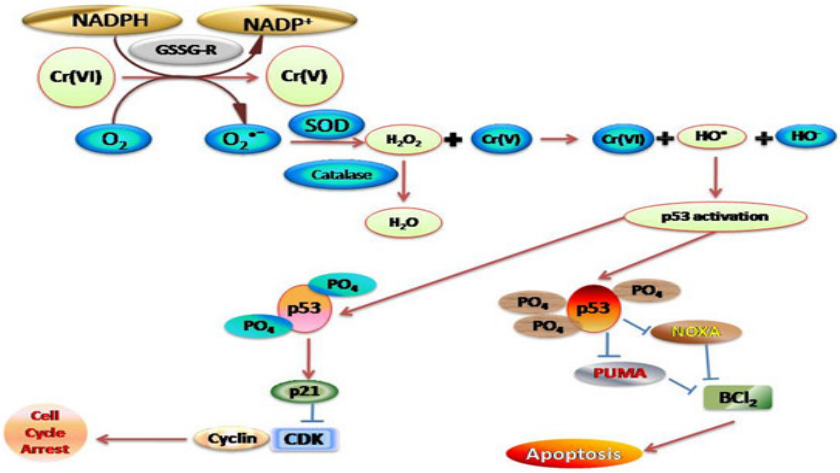

Figure 2

Table 1
Up regulation of several genes in Cr(VI) exposed cells

Name	Functions
Cu/Zn superoxide dismutase (*SOD*)	Catalyzing conversion of superoxide into hydrogen peroxide
Gluthathione-S-transferase subunit 5 theta	Cellular stress oxide
Glutathione peroxidase (*GPX1*)	Removal of hydrogen peroxide
Plasma glutathione peroxidase precursor	Oxidative Stress
Glutathione peroxidise 1,2 (*Gpx1, 2*)	Removal of hydrogen peroxide
Glutathione reductase (*Gsr*)	Maintains high levels of reduced glutathione in the cytosol
Nuclear factor (*Nrf2*)	Transcription activator that binds to antioxidant response (ARE) elements in the promoter regions of target genes.
Peroxiredoxin 1(*Prdx1*)	Involved in redox regulation of the cell
Activating transcription factor 3,4 (*ATF3,4*)	Stress response
Cellular glutathione peroxidase I	Stress
Caspase4	Activation cascade of caspases responsible for apoptosis
Cytoplasmic FMR1 interacting protein 2 (*CYF1P2*)	p53/TP53- dependent induction of apoptosis
Prolyl endopeptidase (*Prep*)	Expressed in apoptotic cell in p53 manner
Serine/threonine protein kinase *MST4*	Mediator of cell growth. Modulates apoptosis
Ring finger protein 144B (*RNF144B*)	Induces apoptosis via a p53/TP53-dependent but caspase-independent mechanism
Bcl-XL	Potent inhibitor of cell death. Inhibits activation of caspases
Death receptor 6 (DR6)	Induce apoptosis
Collagenase type 4	Breakdown of extracellular matrix
Cell death protein (RIPK1)	Activation of NF-kappa-B.
Apoptosis inducing serine/threonine protein kinase (*STK17B*)	Pro-apoptotic
Death inducer obliterator gene (*DIDO1*)	Pro-apoptotic
Phorbol-12-myrestate-13-acetate induced protein 1 (*PMAIPI*)	Pro-apoptotic
Oxidative stress induced growth inhibito (*OKL38*)	Proliferation of normal cells through the regulation of cell death
Transcription factor 1 (*E2F1*)	Cell cycle regulation and p53-dependent apoptosis
B Cell lymphoma6 (*BCL6*)	Anti-apoptotic
Tumour protein p53 inducible nuclear protein 1 (*TP53INP1*)	Induces apoptosis
Tumor necrosis factor receptor (*TNFRS10*)	Activation of NF-kappa-B
BCL2/adenovirus E1B 19kDa interacting protein 3 (*BNIP3*)	Apoptosis-inducing protein
Death effectors domain containing 2 (*DEDD2*)	May play a critical role in death receptor-induced apoptosis
Tumour necrosis factor receptor (TNFRS10)	Activation of NF-kappa-B
Siah E3 ubiquitin protein ligase 1 (hSIAH1)	Apoptosis
Proliferation associated protein (*Pag*)	Cell proliferation
Pigment epithelium differentiation factor (*PEDF*)	Cell differentiation
Cdk5 activator isoform *p39*	Cell growth
INK4p19	Induces G1 cell cycle arrest
CROC-1A	Cell growth
Cyclin E	Control of the cell cycle at the late G1 and early S phase
Cyclin G	G2/M phase arrest in response to DNA damage
Cyclin T2	Cell growth
Cyclin L1	Transcriptional regulator participates in pre-mRNA splicing
Cyclin-dependent kinase inhibitor 1B (*CDKN1B*)	Involved in G1 arrest
Cyclin B1interacting protein (*CCNB1IP1*)	Control of the cell cycle at the G2/M (mitosis) transition
CDC-like kinase 3(*CLK4*)	Role in the formation of spliceosomes

CDC-kinase 1,3,4	Involved in pre-mRNA processing
CDC2-related protein kinase (*CRKRS*)	Involved in RNA splicing
Cyclin D binding myb-like transcription factor 1 (*DMTF1*)	Growth arrest
Cyclin-dependent kinase inhibitor 1B (*CDKN1B*)	Involved in G1 arrest
Cyclin B1interacting protein (*CCNB1IP1*)	Control of the cell cycle at the G2/M (mitosis) transition
CDC-like kinase 3(*CLK4*)	Role in the formation of spliceosomes
Tubulin alpha-1 (*TUBA3*)	Major constituent of microtubules
Cell division-control protein 25B (*CDC25B*)	Tyrosine protein phosphatase which functions as a dosage-dependent inducer of mitotic progression.
Cyclin dependent kinase 4 (*CDK4*)	Regulate the cell-cycle during G(1)/S transition
Proliferating cell nuclear antigen (*PCNA*)	Involved DNA replication
GrpE-like1 (*Grpel1*)	Essential component of the PAM complex
Tetratricopeptide repeat domain (*Ttc27*)	Important to the functioning of chaperone, cell-cycle, transcription and protein transport complexes
Heat repeat containing 1(*Heatr1*)	Involved in nucleolar processing of pre-18S ribosomal RNA
Trefoil factor 1(*Tff1*)	Stabilizer of the mucous gel overlying the gastrointestinal mucosa that provides a physical barrier against various noxious agents
E2F transcription factor (E2F2)	Control of cell cycle and action of tumor suppressor proteins
Transcription factor Dp1 (*Tfdp1*)	Can stimulate E2F-dependent transcription
Photolyase	DNA repair
Apurinic/apyrimidinic endonuclease (*APEX*)	DNA base excision repair
Deoxyribonuclease I precursor (*DNASE I*)	DNA cleaving
DNA topoisomerase II alpha (*TOP2A*)	DNA Repair
DNA polymerase beta subunit (*POLB*)	DNA base excision repair
DNA polymerase alpha (*POLA*)	DNA replication initiation
DNA polymerase delta catalytic subunit	DNA replication and repair
Histone acetyltransferase B subunit 2 (*HATB2*)	Histone acetylation
DNA excision repair protein (*XPF*)	DNA repair endonuclease
Growth arrest and DNA-damage-inducible (*Gadd45a*)	DNA damage repair
Breast cancer1 (*Brca1*)	Maintain genomic stability and acts as a tumor suppressor
MutL homologue 1 (*Mlh1*)	Involved in DNA mismatch repair system
MutS homologue 2,6 (*Msh 2,6*)	Involved in DNA mismatch repair system
RAD51, RAD54 homologue (*Rad51, 54*)	Involved in DNA repair and mitotic recombination
Exonuclease 1 (*Exo1*)	Functions in DNA mismatch repair
XRCC6 binding protein (*Xrcc6bp 1*)	Has a role in chromosome translocation
Ercc8	Involved in nucleotide excision repair
BRCA1/BRCA2- containing complex, subunit 3(*Brcc3*)	Metalloprotease that specifically cleaves 'Lys-63'-linked polyubiquitin chains

CONCLUSION

From considering the different reports on Cr ion's toxicity on cellular system, in summary, it may be concluded that 300μM Cr^{6+} influenced the global cytogenomics and different signaling system, like; Src kinase, MAPK and CDK, and also oncogenes, bioenergetics, and hampered cell cycle from normal rhythm and regulated through the stress.

CONFLICT OF INTEREST

The authors declare that there are no conflicts of interest.

REFERENCES

1. Hopkins J. IARC Monographs on the evaluation of carcinogenic risks to humans: Volume 49. Chromium, nickel and welding: Lyon, 1990. pp. 677. Sw. fr. 95.00. ISBN 92 832 1249 5. Food and Chemical Toxicology. 1991 Jan 1;29(9):647-8.
2. World Health Organisation. Chromium (Environmental Health Criteria 61) International Programme on Chemical Safety, Geneva, Switzerland, (1990).
3. Dayan AD, Paine AJ. Mechanisms of chromium toxicity, carcinogenicity and allergenicity: review of the literature from 1985 to 2000. Human & Experimental Toxicology. 2001 Sep 1;20(9):439-51.
4. Kiilunen M, Kivistö H, Ala-Laurila P, Tossavainen A, Aitio A. Exceptional pharmacokinetics of trivalent chromium during occupational exposure to chromium lignosulfonate dust. Scandinavian journal of work, environment & health. 1983 Jun 1:265-71.
5. Mertz W. Chromium occurrence and function in biological systems. Physiol. Rev.;(United States). 1969 Apr 1;49(2).
6. Myers CR. Chromium toxicity: reductive activation by human enzymes. Final Technical Report F49620-98-1- 0079. Government Reports

Announcements Index. Part 22, AFOSR Report, 1999, 1–34.
7. Donaldson RM, Barreras RF. Intestinal absorption of trace quantities of chromium. J Laborat Clinic Med. 1966;68:484-93.
8. Aitio A, Jarvisalo J, Kiilunen M, Kalliomaki PL, Kalliomaki K. Chromium. Biological Monitoring of Toxic Metals. In: Clarkson TW, Friberg L, Nordberg GF, Sager PR (Eds), Plenum, New York; 1988. p. 369–82.
9. Lindberg E, Vesterberg O. Monitoring exposure to chromic acid in chromeplating by measuring chromium in urine. Scand j work, environhealth. 1983; Aug 1:333-40.
10. World Health Organisation. Chromium (Environmental Health Criteria 61) International Programme on Chemical Safety, Geneva, Switzerland, 1990. Available from: http://www.who.int/water_sanitation_health/dwq/chemicals/chromium.pdf
11. Clarkson TW, Nordberg GF, Sager PR. Reproductive and developmental toxicity of metals. Scand j work, environ health. 1985 Jun 1:145-54.
12. Standeven AM, Wetterhahn KE. Is there a role for reactive oxygen species in the mechanism of chromium (VI) carcinogenesis? Chem Res Toxicol 1991; 4: 616–625.
13. Wetterhahn K. Mechanism of chromium (VI) carcinogenesis: Reactive intermediates and interaction with DNA. Journal of Inorganic Biochemistry. 1989 Aug 31;36(3):344.
14. Halliwell B. Biochemistry of oxidative stress. Biochemical Society Transactions. 2007 Nov 1;35(5):1147-50.
15. Ranjbar A, Khorami S, Safarabadi M, Shahmoradi A, Malekirad AA, Vakilian K, Mandegary A, Abdollahi M. Antioxidant activity of Iranian Echium amoenum fisch & CA Mey flower decoction in humans: A cross-sectional before/after clinical trial. Evidence-based Complementary and Alternative Medicine. 2006;3(4):469-73.
16. Guo T, Cui L, Shen J, Wang R, Zhu W, Xu Y, Qian X. A dual-emission and large Stokes shift fluorescence probe for real-time discrimination of ROS/RNS and imaging in live cells. Chemical Communications. 2013;49(18):1862-4.
17. Dröge W. Free radicals in the physiological control of cell function. Physiological reviews. 2002 Jan 1;82(1):47-95.
18. Halliwell B, Gutteridge J, Cross CE. Free radicals, antioxidants, and human disease: where are we now?. The Journal of laboratory and clinical medicine. 1992 Jan 6;119(6):598-620.
19. Mittler R. Oxidative stress, antioxidants and stress tolerance. Trends in plant science. 2002 Sep 1;7(9):405-10.
20. Valko M, Rhodes CJ, Moncol J, Izakovic MM, Mazur M. Free radicals, metals and antioxidants in oxidative stress-induced cancer. Chemico-biological interactions. 2006 Mar 10;160(1):1-40.
21. Ranjbar A, Khani-Jazani R, Sedighi A, Jalali-Mashayekhi F, Ghazi-Khansari M, Abdollahi M. Alteration of body total antioxidant capacity and thiol molecules in human chronic exposure to aluminum. Toxicological & Environmental Chemistry. 2008 Jul 1;90(4):707-13.
22. Cadenas E, Davies KJ. Mitochondrial free radical generation, oxidative stress, and aging. Free Radical Biology and Medicine. 2000 Aug 31;29(3):222-30.
23. Ott M, Gogvadze V, Orrenius S, Zhivotovsky B. Mitochondria, oxidative stress and cell death. Apoptosis. 2007 May 1;12(5):913-22.
24. Orrenius S, Gogvadze V, Zhivotovsky B. Mitochondrial oxidative stress: implications for cell death. Annu. Rev. Pharmacol. Toxicol.. 2007 Feb 10;47:143-83.
25. Golden TR, Melov S. Mitochondrial DNA mutations, oxidative stress, and aging. Mechanisms of ageing and development. 2001 Sep 30;122(14):1577-89.
26. Lin MT, Beal MF. Mitochondrial dysfunction and oxidative stress in neurodegenerative diseases. Nature. 2006 Oct 19;443(7113):787-95.
27. Krumschnabel G, Manzl C, Berger C, Hofer B. Oxidative stress, mitochondrial permeability transition, and cell death in Cu-exposed trout hepatocytes. Toxicology and applied pharmacology. 2005 Nov 15;209(1):62-73.
28. Huang X, Moir RD, Tanzi RE, Bush AI, Rogers JT. Redox-Active Metals, Oxidative Stress, and Alzheimer's Disease Pathology. Annals of the New York Academy of Sciences. 2004 Mar 1;1012(1):153-63.
29. Valko MM, Morris H, Cronin MT. Metals, toxicity and oxidative stress. Current medicinal chemistry. 2005 May 1;12(10):1161-208.
30. Ercal N, Gurer-Orhan H, Aykin-Burns N. Toxic metals and oxidative stress part I: mechanisms involved in metal-induced oxidative damage. Current topics in medicinal chemistry. 2001 Dec 1;1(6):529-39.
31. Jomova K, Valko M. Advances in metal-induced oxidative stress and human disease. Toxicology. 2011 May 10;283(2):65-87.
32. Hansford RG, Hogue BA, Mildaziene V. Dependence of H2O2 formation by rat heart mitochondria on substrate availability and donor age. Journal of bioenergetics and biomembranes. 1997 Feb 1;29(1):89-95.
33. Requejo R, Chouchani ET, Hurd TR, Menger KE, Hampton MB, Murphy MP. Measuring mitochondrial protein thiol redox state. Methods enzymol. 2010 Dec 31;474:123-47.
34. Brand MD. The sites and topology of mitochondrial superoxide production. Exp geront. 2010 Aug 31;45(7):466-72.
35. Ranjbar A, Ghahremani MH, Sharifzadeh M, Golestani A, Ghazi-Khansari M, Baeeri M, Abdollahi M. Protection by pentoxifylline of malathion-induced toxic stress and mitochondrial damage in rat brain. Hum exp toxicol. 2010 Oct 1;29(10):851-64.
36. Brand MD. Uncoupling to survive? The role of mitochondrial inefficiency in ageing. Experimental gerontology. 2000 Sep 30;35(6):811-20.
37. Starkov AA, Fiskum G, Chinopoulos C, Lorenzo BJ, Browne SE, Patel MS, Beal MF.

Mitochondrial α-ketoglutarate dehydrogenase complex generates reactive oxygen species. The Journal of neuroscience. 2004 Sep 8;24(36):7779-88.
38. Jang YC, Lustgarten MS, Liu Y, Muller FL, Bhattacharya A, Liang H, Salmon AB, Brooks SV, Larkin L, Hayworth CR, Richardson A. Increased superoxide in vivo accelerates age-associated muscle atrophy through mitochondrial dysfunction and neuromuscular junction degeneration. The FASEB Journal. 2010 May 1;24(5):1376-90.
39. Weisiger RA, Fridovich I. Mitochondrial superoxide dismutase site of synthesis and intramitochondrial localization. Journal of Biological Chemistry. 1973 Jul 10;248(13):4793-6.
40. Liang LP, Ho YS, Patel M. Mitochondrial superoxide production in kainate-induced hippocampal damage. Neuroscience. 2000 Nov 15;101(3):563-70.
41. Sies H, Cadenas E, Symons MC, Scott G. Oxidative stress: Damage to intact cells and organs [and discussion]. Philosophical Transactions of the Royal Society of London B: Biological sciences. 1985 Dec 17;311(1152):617-31.
42. Abdollahi M, Ranjbar A, Shadnia S, Nikfar S, Rezaiee A. Pesticides and oxidative stress: a review. Medical Science Monitor. 2004 Jun 3;10(6):RA141-7.
43. Young IS, Woodside JV. Antioxidants in health and disease. Journal of clinical pathology. 2001 Mar 1;54(3):176-86.
44. Yon JM, Baek IJ, Lee SR, Jin Y, Kim MR, Nahm SS, Kim JS, Ahn B, Lee BJ, Yun YW, Nam SY. The spatio-temporal expression pattern of cytoplasmic Cu/Zn superoxide dismutase (SOD1) mRNA during mouse embryogenesis. Journal of molecular histology. 2008 Feb 1;39(1):95-103.
45. Fantel AG, Mackler B, Stamps LD, Tran TT, Person RE. Reactive oxygen species and DNA oxidation in fetal rat tissues. Free Radical Biology and Medicine. 1998 Jul 1;25(1):95-103.
46. Sies H, Arteel GE. Interaction of peroxynitrite with selenoproteins and glutathione peroxidase mimics. Free Radical Biology and Medicine. 2000 May 15;28(10):1451-5.
47. Szabó C, Ischiropoulos H, Radi R. Peroxynitrite: biochemistry, pathophysiology and development of therapeutics. Nature Reviews Drug Discovery. 2007 Aug 1;6(8):662-80.
48. Webb AJ, Patel N, Loukogeorgakis S, Okorie M, Aboud Z, Misra S, Rashid R, Miall P, Deanfield J, Benjamin N, MacAllister R. Acute blood pressure lowering, vasoprotective, and antiplatelet properties of dietary nitrate via bioconversion to nitrite. Hypertension. 2008 Mar 1;51(3):784-90.
49. Stohs SJ, Bagchi D, Hassoun E, Bagchi M. Oxidative mechanisms in the toxicity of chromium and cadmium ions. Journal of environmental pathology, toxicology and oncology: official organ of the International Society for Environmental Toxicology and Cancer. 1999 Dec;19(3):201-13.
50. Barnhart J. Occurrences, uses, and properties of chromium. Regulatory toxicology and pharmacology. 1997 Aug 31;26(1):S3-7.
51. Vincent J, editor. The nutritional biochemistry of chromium (III). Elsevier; 2011 Oct 13.
52. Bagchi D, Bagchi M, Stohs SJ. Chromium (VI)-induced oxidative stress, apoptotic cell death and modulation of p53 tumor suppressor gene. In Molecular Mechanisms of Metal Toxicity and Carcinogenesis. Springer US. 2001.p. 149-58.
53. Mertz W. Chromium occurrence and function in biological systems. Physiol. Rev.;(United States). 1969 Apr 1;49(2).
54. Anderson RA. Chromium metabolism and its role in disease processes in man. Clinical physiology and biochemistry. 1985 Dec;4(1):31-41.
55. Wedeen RP, Qian LF. Chromium-induced kidney disease. Environmental health perspectives. 1991 May;92:71.
56. Hummel M, Standl E, Schnell O. Chromium in metabolic and cardiovascular disease. Hormone and metabolic research. 2007 Oct 1;39(10):743-51.
57. Wang ZQ, Cefalu WT. Current concepts about chromium supplementation in type 2 diabetes and insulin resistance. Current diabetes reports. 2010 Apr 1;10(2):145-51.
58. Gambelunghe A, Piccinini R, Ambrogi M, Villarini M, Moretti M, Marchetti C, Abbritti G, Muzi G. Primary DNA damage in chrome-plating workers. Toxicology. 2003 Jun 30;188(2):187-95.
59. Newbold RF, Amos J, Connell JR. The cytotoxic, mutagenic and clastogenic effects of chromium-containing compounds on mammalian cells in culture. Mutation Research/Genetic Toxicology. 1979 May 1;67(1):55-63.
60. Levis AG, Bianchi V, Tamino G, Pegoraro B. Cytotoxic effects of hexavalent and trivalent chromium on mammalian cells in vitro. British journal of cancer. 1978 Mar;37(3):386.
61. Vasylkiv OY, Kubrak OI, Storey KB, Lushchak VI. Cytotoxicity of chromium ions may be connected with induction of oxidative stress. Chemosphere. 2010 Aug 31;80(9):1044-9.
62. Rossi SC, Gorman N, Wetterhahn KE. Mitochondrial reduction of the carcinogen chromate: formation of chromium (V). Chemical research in toxicology. 1988 Mar;1(2):101-7.
63. Khan FH, Ambreen K, Fatima G, Kumar S. Assessment of health risks with reference to oxidative stress and DNA damage in chromium exposed population. Science of the total environment. 2012 Jul 15;430:68-74.
64. Bagchi D, Stohs SJ, Downs BW, Bagchi M, Preuss HG. Cytotoxicity and oxidative mechanisms of different forms of chromium. Toxicology. 2002 Oct 30;180(1):5-22.
65. Peterson-Roth E, Reynolds M, Quievryn G, Zhitkovich A. Mismatch repair proteins are activators of toxic responses to chromium-DNA damage. Molecular and cellular biology. 2005 May 1;25(9):3596-607.
66. Sugden KD, Stearns DM. The role of chromium (V) in the mechanism of chromate-induced oxidative DNA damage and cancer. Journal of

environmental pathology, toxicology and oncology: official organ of the International Society for Environmental Toxicology and Cancer. 1999 Dec;19(3):215-30.
67. Arakawa H, Weng MW, Chen WC, Tang MS. Chromium (VI) induces both bulky DNA adducts and oxidative DNA damage at adenines and guanines in the p53 gene of human lung cells. Carcinogenesis. 2012 Oct 1;33(10):1993-2000.
68. Chiu A, Shi XL, Lee WK, Hill R, Wakeman TP, Katz A, Xu B, Dalal NS, Robertson JD, Chen C, Chiu N. Review of chromium (VI) apoptosis, cell-cycle-arrest, and carcinogenesis. Journal of Environmental Science and Health, Part C. 2010 Aug 31;28(3):188-230.
69. Katabami M, Dosaka-Akita H, Mishina T, Honma K, Kimura K, Uchida Y, Morikawa K, Mikami H, Fukuda S, Inuyama Y, Ohsaki Y. Frequent cyclin D1 expression in chromate-induced lung cancers. Human pathology. 2000 Aug 31;31(8):973-9.
70. Kondo K, Hino N, Sasa M, Kamamura Y, Sakiyama S, Tsuyuguchi M, Hashimoto M, Uyama T, Monden Y. Mutations of the p53 gene in human lung cancer from chromate-exposed workers. Biochemical and biophysical research communications. 1997 Oct 9;239(1):95-100.
71. Hanaoka T, Yamano Y, Katsuno N, Kagawa J, Ishizu S. Elevated serum levels of pantropic p53 proteins in chromium workers. Scandinavian journal of work, environment & health. 1997 Feb 1:37-40.
72. Carlisle DL, Pritchard DE, Singh J, Owens BM, Blankenship LJ, Orenstein JM, Patierno SR. Apoptosis and P53 induction in human lung fibroblasts exposed to chromium (VI): effect of ascorbate and tocopherol. Toxicological sciences. 2000 May 1;55(1):60-8.
73. Bagchi D, Bagchi M, Stohs SJ. Chromium (VI)-induced oxidative stress, apoptotic cell death and modulation of p53 tumor suppressor gene. InMolecular Mechanisms of Metal Toxicity and Carcinogenesis 2001 (pp. 149-158). Springer US.
74. Bagchi D, Stohs SJ, Downs BW, Bagchi M, Preuss HG. Cytotoxicity and oxidative mechanisms of different forms of chromium. Toxicology. 2002 Oct 30;180(1):5-22.
75. Fan Y, Ovesen JL, Puga A. Long-term exposure to hexavalent chromium inhibits expression of tumor suppressor genes in cultured cells and in mice. Journal of Trace Elements in Medicine and Biology. 2012 Jun 30;26(2):188-91.
76. Schnekenburger M, Talaska G, Puga A. Chromium cross-links histone deacetylase 1-DNA methyltransferase 1 complexes to chromatin, inhibiting histone-remodeling marks critical for transcriptional activation. Molecular and cellular biology. 2007 Oct 15;27(20):7089-101.
77. Takahashi Y, Kondo K, Hirose T, Nakagawa H, Tsuyuguchi M, Hashimoto M, Sano T, Ochiai A, Monden Y. Microsatellite instability and protein expression of the DNA mismatch repair gene, hMLH1, of lung cancer in chromate-exposed workers. Molecular carcinogenesis. 2005 Mar 1;42(3):150-8.
78. Kondo K, Takahashi Y, Hirose Y, Nagao T, Tsuyuguchi M, Hashimoto M, Ochiai A, Monden Y, Tangoku A. The reduced expression and aberrant methylation of p16 INK4a in chromate workers with lung cancer. Lung Cancer. 2006 Sep 30;53(3):295-302.
79. Sun H, Zhou X, Chen H, Li Q, Costa M. Modulation of histone methylation and MLH1 gene silencing by hexavalent chromium. Toxicology and applied pharmacology. 2009 Jun 15;237(3):258-66.
80. Shumilla JA, Broderick RJ, Wang Y, Barchowsky A. Chromium (VI) inhibits the transcriptional activity of nuclear factor-κB by decreasing the interaction of p65 with cAMP-responsive element-binding protein-binding protein. Journal of Biological Chemistry. 1999 Dec 17;274(51):36207-12.
81. Kimura T. Molecular mechanisms of zinc-mediated induction and chromium (VI)-mediated inhibition of mouse metallothionein-I gene transcription. Journal of Health Science. 2010;56(2):161-6.
82. Klein CB, Su L, Bowser D, Leszczynska J. Chromate-induced epimutations in mammalian cells. Environmental health perspectives. 2002 Oct;110(Suppl 5):739.
83. Ha L, Ceryak S, Patierno SR. Chromium (VI) Activates Ataxia Telangiectasia Mutated (ATM) Protein requirement of atm for both apoptosis and recovery from terminal growth arrest. Journal of Biological Chemistry. 2003 May 16;278(20):17885-94.
84. Wise SS, Holmes AL, Xie H, Thompson WD, Wise JP. Chronic exposure to particulate chromate induces spindle assembly checkpoint bypass in human lung cells. Chemical research in toxicology. 2006 Nov 20;19(11):1492-8.
85. Qian Y, Jiang BH, Flynn DC, Leonard SS, Wang S, Zhang Z, Ye J, Chen F, Wang L, Shi X. Cr (VI) increases tyrosine phosphorylation through reactive oxygen species-mediated reactions. InMolecular Mechanisms of Metal Toxicity and Carcinogenesis 2001 (pp. 199-204). Springer US.
86. Hodges NJ, Smart D, Lee AJ, Lewis NA, Chipman JK. Activation of c-Jun N-terminal kinase in A549 lung carcinoma cells by sodium dichromate: role of dissociation of apoptosis signal regulating kinase-1 from its physiological inhibitor thioredoxin. Toxicology. 2004 Apr 15;197(2):100-11.
87. Tessier DM, Pascal LE. Activation of MAP kinases by hexavalent chromium, manganese and nickel in human lung epithelial cells. Toxicology letters. 2006 Dec 1;167(2):114-21.
88. Chuang SM, Liou GY, Yang JL. Activation of JNK, p38 and ERK mitogen-activated protein kinases by chromium (VI) is mediated through oxidative stress but does not affect cytotoxicity. Carcinogenesis. 2000 Aug 1;21(8):1491-500.
89. Chuang SM, Yang JL. Comparison of roles of three mitogen-activated protein kinases induced

by chromium (VI) and cadmium in non-small-cell lung carcinoma cells. Molecular and cellular biochemistry. 2001 Jun 1;222(1-2):85-95.
90. O'Hara KA, Klei LR, Barchowsky A. Selective activation of Src family kinases and JNK by low levels of chromium (VI). Toxicology and applied pharmacology. 2003 Aug 1;190(3):214-23.
91. Chen L, Ovesen JL, Puga A, Xia Y. Distinct contributions of JNK and p38 to chromium cytotoxicity and inhibition of murine embryonic stem cell differentiation. Environmental health perspectives. 2009 Jul 1;117(7):1124.
92. O'hara KA, Vaghjiani RJ, Nemec AA, Klei LR, Barchowsky A. Cr (VI)-stimulated STAT3 tyrosine phosphorylation and nuclear translocation in human airway epithelial cells requires Lck. Biochemical Journal. 2007 Mar 1;402(2):261-9.
93. Chen F, Ding M, Lu Y, Leonard SS, Vallyathan V, Castranova V, Shi X. Participation of MAP kinase p38 and IkappaB kinase in chromium (VI)-induced NF-kappaB and AP-1 activation. Journal of environmental pathology, toxicology and oncology: official organ of the International Society for Environmental Toxicology and Cancer. 1999 Dec;19(3):231-8.
94. Liu K, Husler J, Ye J, Leonard SS, Cutler D, Chen F, Wang S, Zhang Z, Ding M, Wang L, Shi X. On the mechanism of Cr (VI)-induced carcinogenesis: dose dependence of uptake and cellular responses. InMolecular Mechanisms of Metal Toxicity and Carcinogenesis 2001 (pp. 221-229). Springer US.
95. Kim YD, An SC, Oyama T, Kawamoto T, Kim H. Oxidative Stress, hogg1 Expression and NF-. KAPPA. B Activity in Cells Exposed to Low Level Chromium. Journal of occupational health. 2003;45(5):271-7.
96. Zuo Z, 左正宏, Cai T, Li J, Zhang D, Yu Y, Huang C. Hexavalent chromium Cr (VI) up-regulates COX-2 expression through an NF kappa B/c-Jun/AP-1-dependent pathway.
97. Papaioannou AI, Kostikas K, Kollia P, Gourgoulianis KI. Clinical implications for vascular endothelial growth factor in the lung: friend or foe. Respir Res. 2006 Oct 17;7(1):128.
98. Voelkel NF, Vandivier RW, Tuder RM. Vascular endothelial growth factor in the lung. American Journal of Physiology-Lung Cellular and Molecular Physiology. 2006 Feb 1;290(2):L209-21.
99. Kishiro I, Kato S, Fuse D, Yoshida T, Machida S, Kaneko N. Clinical significance of vascular endothelial growth factor in patients with primary lung cancer. Respirology. 2002 Jun 1;7(2):93-8.
100. Kaya A, Çiledag A, Gulbay BE, Poyraz BM, Çelik G, Sen E, Savas H, Savas I. The prognostic significance of vascular endothelial growth factor levels in sera of non-small cell lung cancer patients. Respiratory medicine. 2004 Jul 31;98(7):632-6.
101. Cressey R, Wattananupong O, Lertprasertsuke N, Vinitketkumnuen U. Alteration of protein expression pattern of vascular endothelial growth factor (VEGF) from soluble to cell-associated isoform during tumourigenesis. BMC cancer. 2005 Oct 4;5(1):1.
102. Nemec AA, Barchowsky A. Signal transducer and activator of transcription 1 (STAT1) is essential for chromium silencing of gene induction in human airway epithelial cells. Toxicological sciences. 2009 Jul 1;110(1):212-23.
103. Nemec AA, Zubritsky LM, Barchowsky A. Chromium (VI) stimulates Fyn to initiate innate immune gene induction in human airway epithelial cells. Chemical research in toxicology. 2009 Dec 8;23(2):396-404.
104. Lal MA, Bae D, Camilli TC, Patierno SR, Ceryak S. AKT1 mediates bypass of the G1/S checkpoint after genotoxic stress in normal human cells. Cell cycle. 2009 May 15;8(10):1589-602.
105. Alexopoulos EC, Cominos X, Trougakos IP, Lourda M, Gonos ES, Makropoulos V. Biological monitoring of hexavalent chromium and serum levels of the senescence biomarker apolipoprotein j/clusterin in welders. Bioinorganic chemistry and applications. 2008 Apr 22;2008.
106. Ye J, Shi X. Gene expression profile in response to chromium-induced cell stress in A549 cells. InMolecular Mechanisms of Metal Toxicity and Carcinogenesis 2001 (pp. 189-197). Springer US.
107. Nigam A, Priya S, Bajpai P, Kumar S. Cytogenomics of hexavalent chromium (Cr6+) exposed cells: A comprehensive review. The Indian journal of medical research. 2014 Mar;139(3):349.