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Review Article

Complementary/Alternative Medicine Section

Pesticides and Human Health: Antioxidants and Heat Shock Proteins as Modulators of Cell Survival Signal

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ABSTRACT

In the modern times, insecticides have grown to be an essential part of the atmosphere. Their full-size use in public health packages and agriculture has ended in capability environmental pollution and health risks, which relies upon their residual quantity and toxicity. The widespread uses cause general population to low dose of continual exposure of insecticides through meals and environment. The residue evaluation of human specimen suggests an increasing trend within the ranges of insecticides in serum, adipose tissue, breast milk, urine and others. Implications of pesticides residues on human fitness following subchronic publicity are but to be comprehensively answered. Subjection to insecticides can be closely associated with neurotoxicity, hepatotoxicity, immunotoxicity, genotoxicity and injurious reproductive effects. Pesticides leads to Reactive Oxygen Species (ROS) generation in significant quantities, resulting in oxidative stress and cellular damages. Findings of research have discovered a concomitant genotoxic and apoptotic effect of the pesticides in Peripheral Blood Mononuclear Cells (PBMCs). Since genotoxic consequences of pesticides on humans cannot be overlooked, therefore identification and implication of protective measures are urgently needed. This have been tested that PBMCs go through dose-structured apoptotic cell demise following pesticide exposure and additionally highlights the effectiveness of various antioxidants in counteracting pesticide-precipitated cytotoxicity. Heat Shock Proteins (HSPs) have emerged as an antiapoptotic molecules which counteract cytotoxicity. The inducible expression analysis of HSPs ought to make contributions to the human PBMCs to get over the toxic results of subchronic pesticide exposure. Though the linkage between cellular events of apoptosis is thought, the molecular mechanism highlighting the precise function of HSP in pesticide-mediated cytotoxicity yet stays to be comprehensively replied. To better understand this mechanism, different antioxidant and HSP inducers have been employed, and also highlighted their attenuating effects towards the apoptotic capacity of such pesticides. This review article therefore, focuses on the fact that antioxidants and HSP inducers efficiently protect cells, emphasising their role in pesticide-induced toxicity at molecular and cellular level as well as their possible use as therapeutic intervention.

Keywords: Apoptosis, Cytotoxicity, Environmental chemicals, Oxidative stress

INTRODUCTION

Pesticides are the chemicals useful in various agricultural practices. It has been estimated that in growing nations-pests, weeds and different plant diseases ruin about 50% of crops [1]. As a result, the use of pesticides has become the integral part of agriculture to increase crop production. Every year more than two million tons of pesticides are used across the world and large percentages of these affect the non target organism [1,2]. Excessive use of these pesticides posses potential risks to the ecosystem and human health [1,3]. Residues analysis of pesticides have detected significant levels of these chemicals in human body fluids and tissues like blood, placenta, amniotic fluid, and in secretions like semen, breast milk, and so for [3,4].

Authors have reported noticeably better levels of different pesticides residues and their associated with various diseases in human subjects [3,4-6]. In humans, these pesticides disturb both biochemical and physiological functions. Accumulation of these pesticide residues may cause subacute and chronic toxicity affecting the critical organs including liver, kidney, testes, spleen, thymus, and lymph nodes resulting in endocrine disruption, genotoxicity, neurotoxicity, nephrotoxicity, hepatotoxicity [7]. Though the ability of pesticides to cause genotoxicity is among one of the preeminent worries of damaging impact to human health, the underlying mechanism remains no longer yet surely delineated. Several pesticides were shown to purpose cytotoxicity or immunotoxicity in non target species making them more vulnerable to infections [8,9]. Earlier studies from the laboratory have also demonstrated that subchronic exposure to these pesticides modulates oxidative stress and immune response in experimental animals [10-12].

Cytotoxicity of these pesticides has been shown to involve several biochemical pathways like inhibition of protein synthesis, leakage of lactate dehydrogenase, inhibition of glucose metabolism and ATP depletion [13]. In-vitro studies have shown that pesticide could also induce apoptosis in diverge cells like murine thymocytes, human mononuclear cells, human T-cells leukaemic cells and neuroblastoma cells and subsequent increased risk for autoimmune disease and allergies [14-17].

Blood cells are the first to come across pesticides on the event of their entry into the body. Peripheral Blood Mononuclear Cells (PBMC) mainly comprises of B-lymphocytes and monocytes. These blood cells are a essential component in the immune system to fight contamination and adapt to intruders/xenobiotics. Several studies from the laboratories have demonstrated, the cytotoxic effects (apoptosis/necrosis) of pesticides in human peripheral blood mononuclear cells and the molecular mechanism associated with pesticide induced immunotoxicity [18-22].

Apoptosis and necrosis are two biochemically and morphologically diverse process by which cells die [23]. Cells under normal situation and in culture go through apoptosis whilst exposed to a ramification of cytotoxic agents [23]. Apoptosis is a highly regulated process characterised by cytoplasmic shrinkage, asymmetry of plasma membrane phospholipid to exposed phosphatidylserine, chromatin condensation, the release of cytochrome c, caspase cascade activation and DNA fragmentation [18-22,24].

Oxidative stress has been mentioned as a possible mechanism of xenobiotic mediated toxicity in people, considering that this

phenomenon has been involved in the aetiology of various diseases such as cancer and neurodegeneration [25]. It may additionally cause cell harm and malfunction through the free radical-mediated damage of crucial molecules [19,26,27]. Moreover, oxidative stress performs a key role within the induction of apoptosis [18,19]. Overproduction of Reactive Oxygen Species (ROS) and depletion of Glutathione (GSH) were found to both precede the onset of apoptosis or render cells more sensitive to cell death [21,22]. Several in-vitro studies have reported that ROS triggers cytochrome c release from mitochondria which may lead to induction of apoptosis [14,15,20,28].

Cells might also use several mechanisms to defend themselves from the ill-effects of toxicants. In reaction to a huge style of lessons of environmental and pathophysiological toxicants (like heat shock, heavy metals, pesticide, oxidants, pills, bacterial pollution), all cells preferentially synthesise numerous families of proteins known stress or Heat Shock Proteins (HSPs) [29]. Under regular physiological situations, participants of the HSP protein family are involved as molecular chaperones in the stabilisation of unfolded protein precursors, translocation of proteins across cell membranes, dissolution of protein aggregates and restore or degradation of damaged proteins [30]. Mammalian HSP's were classified into four major families according to their molecular size HSP90, HSP70, HSP60 and small HSP's such as HSP27 [30,31]. The HSP60 and HSP90 are constitutively expressed in mammalian cells at the same time as HSP27 and HSP70 are strongly precipitated by exceptional stresses which includes heat, anticancer drugs, radiation, amino acid analogues, mitogens, arsenite, transition metals and many polychlorinated bi-phenyls, polyaromatic hydrocarbons, pesticides and herbicides [32,33].

Studies have reported that some members of HSP family could regulate apoptosis means of interacting with key components of the apoptotic signaling pathway, like cytochrome c [34,35]. Furthermore, HSP27 has been proven to increase the antioxidant defence capacity of cells by increasing glutathione content of cells [36]. Members of the HSP70 family play a similar antioxidant role like the small HSP27 and capabilities to modulate the engagement and/or progression of apoptosis prompted with the aid of a extensive style by a wide variety of stimuli together with hyperthermia, hypoxia and exposure to poisonous chemicals [33,37,38].

Recent studies have reported the linkage between cellular events incited by way of damage and cell demise and the molecular mechanism highlighting the specific role of HSP in pesticide-mediated cytotoxicity [18,22]. In view of this, this it is an area of interest to establish the role of oxidative stress and HSP in pesticide-mediated cytotoxicity. Further, to better elucidate the mechanism, we have reported a combinatorial approach of antioxidants and HSP inducers to strengthen the proposed mechanism and to reveal their attenuating consequences against the apoptotic ability of these pesticides. We therefore, emphasise that pesticide-induced toxicity can be evaluated by both in-vivo and in-vitro approaches where studies may provide an insight into the molecular mechanisms underlying pesticide poisoning.

PESTICIDE

Pesticides are widely used chemicals with unique properties designed to have an intense biological effect on the target pests [39]. Pesticides improve quality of life by increasing crop productivity [40]. Also, pesticides play a significant role by checking many insect-borne diseases [41]. Thus, the application of a wide variety of pesticides is necessary in the tropics to combat pests and associated diseases.

Pesticide Classification

Pesticides can be charaterised by target organism, their chemical structure and their physical state [42]. Following is the classification of pesticides according to the types of pests:

1. Algicides or algaecides for the contend of algae.
2. Avicides for the contend of birds.
3. Bactericides for the contend of bacteria.
4. Fungicides for the contend of fungi and oomycetes.
5. Herbicides (like glyphosate) for the contend of weeds.
6. Insecticides (like organochlorines, organophosphates, carbamates, and pyrethroids) for the control of insects- these may be ovicides (substances that kill eggs), larvicides (substances that kill larvae) or adulticides (substances that kill adults).
7. Miticides or acaricides for the contend of mites.
8. Molluscicides for the contend of slugs and snails.
9. Nematicides are chemical substances used to kill nematodes.
10. Rodenticides to contend with rodents.
11. Virucides to contend with viruses.

Pesticide: Emergences and Challenges

According to a Green Peace Report, India is now generating 90,000 metric tons of pesticides as the largest industry of Asia and twelfth largest in the entire world [40]. India lies at the fourth position among the worldwide suppliers of agrochemicals, after USA, Japan and China. The Indian pesticide industry is the biggest in Asia and 12th in the world. According to the Indian Insecticides Act 1968, all pesticide products are to be registered before they are manufactured, sold, exported or imported which is govern through the Ministry of Agriculture, Government of India [40]. The worldwide usage of pesticides is near about two million tons per year. China is the main contributory country, followed by the United States of America and Argentina, which is enlarging rapidly. Global pesticide use has increased progressively from last three decades and currently around four million tonnes are being used. India ranks 12th in worldwide pesticide usage, with maximum uses of insecticides. India share approximately 1% of the global pesticide used. Out of total insecticides hired for pest control in India, 50% are employed to cotton pest control [43]. Pesticides industry in India was worth Rs. 214 billion in 2019 which is expected to reach about Rs. 316 billion by 2024 [43].

PESTICIDE RESIDUES AND HUMAN HEALTH

Humans are exposed to various environmental chemicals including pesticides, heavy metals, Polychlorinated Biphenyls (PCBs). Pesticides like endosulfan, Hexachlorocyclohexane (HCH), Dichlorodiphenyltrichloroethane (DDT), Dieldrin, heptachlor, dicofol, methoxychlor malathion, parathion, dimethoate, aldicarb, carbofuran, and ziram are ubiquitous in environment and organisms. Organochlorine Pesticides (OCPs) like endosulfan, HCH, DDT are persistent environmental organic pollutants in nature. Total 40% of all pesticides used in India belong to the Organochlorine (OCP) class magnificence of chemicals [40].

Pesticides have been proposed as Endocrine Disruptor Chemicals (EDCs) that alter the function(s) of endocrine system and thus cause adverse health effects [44,45]. Some OCPs, Organophosphates (OP) and carbamate pesticides such as DDT, HCH, malathion, adicarb and carbofuran are known EDCs. Especially, accumulation of these chemicals in fatty tissues is a problem of greater concern especially in women. Hormonal changes that occur during pregnancy, lactation and menopause, have been shown destructive effects of these OCPs, many years after their initial exposure. Also, newborns are exposed through placental transmission as well as breast-feeding [46]. In the laboratory, a significant level of OCPs in maternal and cord blood samples, and other diseased subjects have been detected [46-48]. The OCPs adversely affect reproductive health and fetal development as it functions xenoestrogen. Hormonal homeostasis (particularly oestrogen progesterone balance) is extremely important in maintenance of pregnancy and OCPs can

alter this hormonal homeostasis, which may result in termination of pregnancy [46]. Moreover, several studies have reported the association of pesticide residues with other neurodegenerative diseases like Parkinson's disease and other chronic diseases such as chronic kidney diseases and cardiovascular diseases [48,49]. Hence, knowledge of biological ranges of these contaminants in human tissues is vital for the hazard evaluation of unfavourable fitness outcomes and for the identification of vulnerable groups.

PESTICIDES AND CYTOTOXICITY

Cytotoxicity can be defined as the ability of a chemical compound to generate the cell death response. Previously, an awful lot of the acute toxicity data for environmental pollutants has been derived from the bioassays in experimental animals. However, such in-vivo bioassays are steeply-priced and time-ingesting. Thus, there is a need to develop rapid and powerful bioassays to screen the enormous variety of chemicals for their toxicity. Rachlin and Perlmutter (1968) first demonstrated the usage of cultured fish cells for the acute in-vitro cytotoxicity assays of aquatic pollutants. Later on, practice made by several investigators to further develop in-vitro cytotoxicity assays for assessment of the acute toxicities of environmental pollutants [50].

Several techniques are employ to assess cellular loss of life, the majority dealt with membrane permeability for example uptake of trypan blue by dead cells. The exclusion of the Deoxyribonucleic Acid (DNA)-binding fluorescent dye propidium and cellular retention of flourescein are also interpreted as indicators of cell viability. The release of ^{15}Cr (Chromium) is regularly used to assess cell lysis inflicted by way of immune effector cells and complement-mediated damage. Other cytotoxicity assays measure functional factors of living but not dead cells. The tetrazolium dye-based assay defined a living cell as one able to converting 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) to formazan through mitochondrial dehydrogenase activity and uptake of neutral red into lysosomes is taken as proof of cell viability. The MTT assay can be use to determine the general cytotoxicity of the selected pesticides in human PBMC [22].

Toxic cellular damage results in necrosis whilst endogenous systems fail to compensate. Alternatively, cellular loss of life can be initiated by programmed process called apoptosis.

PESTICIDE AND APOPTOSIS

Apoptosis has been proven to be suffering from numerous elements, which including subjection to pesticides [51]. Several studies using unique cell lines and blood cells have validated that pesticide or pesticide mixture were capable of set off apoptosis in-vitro [51,52]. Jia Z and Misra HP, tested that zineb and endosulfan were determined to result in cytotoxicity in SH-SY5Y cells via both apoptotic and necrotic pathways [15]. Several organochlorine and organophosphate pesticides such as endosulfan, melathion, and phosphamidon had been able to set off apoptosis in human peripheral blood mononuclear cells in-vitro [18-22]. Earlier Kaioumova D et al., confirmed that the broadly used herbicide DMA-2,4-D kills human lymphocytes by starting up apoptosis [53]. It has been reported that malathion, lindane, and piperonyl butoxide, individually or in combined mixtures, induce apoptosis in murine splenocytes in-vitro [52]. Therefore, it is well understood that many compounds are not directly cytotoxic; rather they cause sublethal damages, which might also cause innate suicidal sequence of activities within the cell.

PESTICIDES AND GENOTOXICITY

The capability of pesticides to induce DNA damage or genotoxicity are among the greatest concerns of ill effects to human health, however the exact mechanism is not yet clearly delineated. The DNA is also a major target of constant oxidative damage by endogenous oxidants [54]. The DNA contains a backbone of the sugar deoxyribose linked by phosphate groups. Attached to deoxyribose are the purine bases adenine and guanine, and the pyrimidine bases cytosine and thymine. So, DNA is stable, well-protected molecule. However, ROS can interact with it and cause several types of damage: modification of DNA bases, single-DNA and double-DNA breaks, loss of purines (apurinic sites), damage to the deoxyribose sugar, DNA-protein cross-linkage, and harming to the DNA repair system.

The substance, 8-hydroxy-2' -deoxyguanosine (8-OHdG) is an adduct formed as a result of reaction between free radicals and DNA. It establishes the link between intracellular free radicals accumulation and genotoxicity. One of the mutagenic DNA lesions, 8-OHdG if allowed to accumulate can perpetuate through the DNA replication process. Further, it can also retard the DNA repair mechanism [55]. This damage to DNA may lead to various observations like NADPH and ATP depletion resulting in cell death or can also lead to malignant transformation in surviving cells [56,57].

On large scale, the coupling of $HO^{\bullet}$ with the nucleobases of the DNA strand, such as guanine, results in the development of C8-hydroxyguanine (8-OHGua) or its nucleoside forming deoxyguanosine (8-hydroxy-2-deoxyguanosine OHdG undergoes keto-enol tautomerism, which favors the oxidised product 8-oxo-7,8-dihydro-2-deoxyguanosine (8-oxodG). Damaged DNA restores their function enzymatically in all living cells so that they can regain their function normally, whereas misrepaired DNA leads to mutations (base substitution, deletions, and strand fragmentation) resulting into genotoxicity [55]. Inspite of a huge range of DNA products produced during oxidative damage to DNA (bases and sugar modifications, covalent crosslink, single-stranded and double-stranded breaks), most interest focused on alteration in nucleobase and especially on the abundant lesion of 8-oxo-2-deoxyguanosine as it is formed in-vivo and can be estimated quantitatively in cells following hydrolysis of the DNA to component bases [56].

The damage to the DNA may be involved in the etiology of diverse pathological conditions such as immunosuppression, neurodegeneration and autoimmune diseases [57,58]. Several studies have shown that pesticides can cause genotoxicity including increase in the frequency of chromosomal aberrations and DNA adducts formation, and increased levels of reactive oxygen species that can disrupt the genetic integrity and alter the biochemistry of metabolic pathways, resulting in increased susceptibility to infections in non target animals [59].

Cytogenetic studies on Phosphamidon (PHO), an organophosphate pesticide, indicate that it can induce chromosome aberrations and Micronuclei (MN) formation in man and mice [60]. Analysis for chromosomal aberrations such as breaks, satellite associations and gaps is commonly used to monitor pesticide-induced genotoxic effects on chromosomes under in-vitro and in-vivo conditions [60].

PESTICIDE INDUCED CELLULAR OXIDATIVE STRESS

Cellular oxidative stress has been considered one of the mechanism by which pesticide causes neurotoxicity, hepatotoxicity, and immunotoxicity [14,15]. Reactive oxygen species are produced naturally and continuously within the cell. An imbalance between pro-oxidant (ROS) and antioxidant mechanisms in cells causes oxidative stress [27]. Oxidative stress can thus occur where the production of free radicals increases, scavenging of free radicals or repair of oxidative modified macromolecules decreases or both, which are nothing but the alteration of the redox state. Within the cellular context, the redox status depends on the relative amounts of the oxidised and reduced partners of major redox molecules, such as glutathione.

Glutathione, the most abundant intracellular free radical scavenger, protects cells against reactive oxygen species as well as many toxins, mutagens and chemicals [21]. Glutathione is a thiol protein of mammalian cell. It is a peptide composed of glutamate, cysteine and glycine that exist in thiol-reduced Glutathione (GSH) and

oxidised glutathione (GSSG); those are recycled by the Glutathione Reductase (GR) and Glutathione Peroxidase (GPx) and Glutathione S Transferase (GST) [61]. The ratio GSSG/GSH reflects the redox status within the cell. It usually averages 1%, which means that GSH prevails over GSSG [61]. The glutathione system acts as a homeostatic redox buffer. The ratio between reduced GSH and total GSH is a good indicator of oxidative stress (reduction with increased oxidative stress), as well as down regulation of GPx/GR ratio with increased oxidative stress [61]. This is an important cellular parameter, since the intracellular redox status monitors the reactive amounts of the oxidised and reduced species of each redox system within the cell, depending on its oxidation potential (E°). The GSH is also an important factor in the cellular defence against oxidative stress due to its own antioxidant capacity and because of its involvement in the recycling of other antioxidants.

Several studies have demonstrated oxidative stress induced by pesticides in experimental animals and humans [62,63]. For instance, the organochlorine insecticide dieldrin has been shown to induce hepatotoxicity via oxidative stress in mice [63]. Hsu CH et al., reported that the fumigant phosphine induced cytotoxic and mutagenic effects by increasing ROS levels [64]. Bagchi D et al., examined that 2,3,7,8-Tetrachlorodibenzo-p-Dioxin (TCDD), eldrin, naphthalene and sodium dichromate (IV) increased hepatic lipid peroxidation and DNA fragmentation [65]. Deltamethrin, pyrethroids insecticide, administration in rats resulted in DNA fragmentation in the testicular cells with increased plasma levels of nitric oxide [66]. Malathion has been shown to modulate oxidative stress and immune response in experimental animals [11,28,67]. Generation of oxidative intermediate has been implicated in the process of programmed cell death induced by various agents.

HEAT SHOCK PROTEINS- DIAGNOSTICS BIOMARKERS AND THERAPEUTICS TARGETS

Cells are often exposed to environmental or endogenous stresses that can questioned to cell vulnerability. In order to ensure tissue integrity and function, cells survive with cellular injuries by protecting intracellular constituents, restrain cell death signaling pathways and activating damage repair mechanism. Heat shock and other cellular stress factors, such as oxidising agents, pollutants, heavy metals, and infective microbes, can induce a conserved cell defence mechanism, the Heat Shock Response (HSR), to hold proteostasis in eukaryotic cells or can severely perturb protein homeostasis (proteostasis), thereby compromising cell processes and to improved ageing and the incidence of various proteotoxic prompted issues in body, along with neurodegenerative sicknesses, heart failure, cancer, diabetes, tissue atrophy and fibrosis, and immune deficiency [68]. The HSR basically entails the expression of HSP, also termed molecular chaperones, which facilitate the synthesis and ensure the structural stability of other intracellular proteins. The HSPs can also mediate the degradation of damaged intracellular proteins. This cell protective mechanism enables the celluler to survive under harsh environmental situations, predominantly at increased temperatures [69]. The cellular reaction to stress is represented at the molecular level, way of the accelerated synthesis of HSPs, of which molecular chaperones and proteases represent two well charaterised families of proteins. The stimulation of HSPs is particular accomplished by the heat shock transcription factor HSF1. Hence, HSF1 capabilities as a primary regulator of the HSR [70,71].

The heat shock proteins are a massive superfamily of proteins with molecular weights ranging from 8 to 170 kDa, with the principle participants called HSP27, HSP60, HSC 70 and HSP90 being the proteins classically indentified to be induced as a result of heat treatment of mammalian cells [72].

The synthesis of HSP is one mechanism that the cell could utilise to protect itself from cellular damage following exposure to xenobiotics. Several studies have demonstrated the increased levels of HSPs expression by evaluating the toxicity of different kinds of pesticide agents on diverse species [73,74]. It has been reported that the synthesis of HSPs were induced by exposure to insecticide AVM in Frankliniella occidentalis at different stages [75]. A similar study has reported that the expression of HSP60, HSP70, and HSP90 were induced by neurotoxic insecticides of chlorpyrifos and esfenvalerate in Chinook salmon (Oncorhynchus tshawytscha) [76]. The altered HSPs expression in response to environmental pollutant have also been described in various cell lines [77,78]. So that HSPs will be use as biomarkers in different animal species. In our laboratory, we have reported the increased synthesis of HSP27 in human peripheral blood mononuclear cells following exposure to malathion and endosulfan pesticides [18,22]. Xing H et al., found that the levels of HSP70 mRNA was significantly increased in brain, liver and kidney of common carp *(Cyprinus carpio L.)* [79]. These results successfully showed that HSPs could be considered as biomarkers of various pesticide exposure.

HSP as Antiapoptotic Proteins

HSP27 is an Adenosine triphosphate (ATP)-independent powerful chaperone, whose main role is to offer protection against protein aggregation [80]. In spite of a poor understanding of the mechanisms by which many stress proteins may contribute to protecting cells against stress, a role in tolerance to stress has been specifically demonstrated for HSP27. It has been suggested HSP27 protects cells from apoptotic cell death, triggered by hyperthermia, ionising radiation, oxidative stress, Fas ligand, and cytotoxic drugs. At the same time, such stimuli often induce HSP27 overexpression, providing an example of how proapoptotic stimuli, delivered below a threshold level, can elicit protective responses [81]. Several different mechanisms may account for HSP27's antiapoptotic activity. HSP27 can increase the antioxidant defense of cells by increasing cellular glutathione content and also neutralise the toxic effect of oxidised proteins by its chaperon activity [82,83]. HSP27-mediated regulation of GSH, or γ-glutamylcysteinylglycine, was recognised in a study to determine mechanisms of resistance to TNF α [84]. HSP27 overexpression will increase expression and activity of glucose-6-phosphate dehydrogenase, an enzyme that features to lessen $NADP^+$ all through cellular recycling and reduction of GSH [85]. Overexpression of HSP27 has also been shown to increase activity of other detoxifying enzymes like GST and GR. One study has reported that HSP27 actually protects these enzymes when exposed to H_2O_2 in-vitro [86]. It has been reported that over expression of HSP27 significantly inhibits programmed cell death triggered by short term exposure to low concentrations of H_2O_2 [87].

HSP27 can also interfere with procaspases 3 activation and can modulate DAXX and Akt signalling mechanisms [87]. HSP27 binds cytochrome c once it is released from the mitochondria and inhibits its ability to promote apoptosome activation [88]. Hassoun R et al., demonstrated that HSP27could prevent activation of procaspases-9 and procaspases-3 in hypertrophic cardiomyopathy patients [89].

HSP70, also can inhibit apoptosis and thereby increase the survival of cells from wide range of lethal stimuli [90]. Indeed, over expression of HSP70 protects cells from stress-induced apoptosis, by modulating both upstream and downstream events of the caspase cascade activation [91]. This antiapoptotic effect was explained by the HSP70-mediated modulation of the apoptosome including activation of caspase 8 and caspase 3, upregulating the expression of B-cell lymphoma 2 (Bcl-2) and decreases the release of cytochrome c from the mitochondria into the cytoplasm [91]. These studies have provided new insight into the molecular mechanisms of apoptosis inhibition as HSPs regulates apoptosis via multiple pathways.

CONCLUSION(S)

A spectrum of pathogenic outcomes, especially immunosuppression is a serious concern in pesticides-induced chronic toxicity assessment. Conceivably, an alteration of immune system by such environmental chemicals could affect the individual's ability to mount well-regulated immune responses to microbial and vaccine antigen, allergens, self and tumor antigens. There are some major issues especially concerned about the immunosuppressive consequences of pesticides on exposed populations in growing countries. An elaborate programme of research is required to investigate this potential risk and to design precautionary measures. Considering the widespread distribution and stability of certain groups of pesticides in the biosystem, it appears that present data on adverse health effects in humans may represent only tip of the iceberg. Risk to human health from pesticides should not be underestimated in any way and urgent steps should be taken to define the factors, which affect the evaluation of immunotoxicity. The precise mechanism by which immunosuppression occurs in many cases are not described, but the close interactions between neurological and immunological feature may leads to toxicity. A better understanding of these interactions and more clearly defined end points in different species remains a priority for future.

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