Chapter 6

Genotoxic Risk in Human Populations Exposed to Pesticides

Delia Aiassa

Additional information is available at the end of the chapter

http://dx.doi.org/10.5772/intechopen.77968

Abstract

The importance of early detection of genetic damage is that it allows taking the necessary measures to reduce or suppress the exposure to the deleterious agent when it is still reversible, thus decreasing the risk of developing diseases. For this reason, genotoxicity tests should be considered as indispensable tools in the implementation of a complete medical surveillance in people potentially exposed to various environmental pollutants and especially those who live in the same place with people who have already developed some type of neoplasia at early ages in order to prevent the occurrence of tumors of environmental origin and work-related. On the other hand, the application of these tests is useful to detect possible long-term effects of substances that are introduced to the market without knowing exactly their capacity to affect human and environmental health.

Keywords: genotoxicity, pesticides, Argentina

1. Introduction

1.1. Pesticides

Pesticides are a heterogeneous group of chemical compounds used in the production of food and considered one of the major sources of contamination by synthetic substances generated as a result of agricultural activity. For more than a decade, many of them have been classified as potential carcinogens [1, 2].

The Food and Agriculture Organization of the United Nations (FAO [3]) defines a pesticide as any substance or mixture of substances intended for preventing, destroying or controlling any plague, including vectors of human or animal diseases, unwanted species of plants and

animals that cause harm or interfere in any other way in the production, processing, storage, transportation or commercialization (marketing) of food, agricultural products, wood and its derivates [4].

The benefits obtained by the use of pesticides are certainly numerous, however, the dissemination of large amounts of these compounds to the environment, has led to problems affecting both the environment and human health [5]. Particularly, in agricultural activities, agrochemicals are widely used products, and its use without the necessary protection can lead to genetic alterations and the possible development of some types of neoplasia [6, 7].

Exposure to these substances results in acute poisoning. Poisoning is the body's reaction to a toxic agent, and it is described as acute poisoning when the symptoms occur after a recent exposure to the chemical. In this kind of intoxication, the diagnosis is relatively easy, fast and with an established treatment.

Chronic health effects have been associated to pesticide exposure, including neurological disorders, reproductive or developmental problems and cancer. Epidemiological studies on farmers, pesticide manufacturers, pesticide sprayers and on accidentally exposed industrial workers or residents have shown that exposure to pesticides may increase the risk of site-specific cancers. Also, increased risks have been detected for leukemia, Ewing's bone sarcomas, kidney cancer, soft tissue sarcoma, non-Hodgkin's lymphoma, and testicular, colorectal, endocrine glands and brain cancers in children exposed to pesticides in their home or whose parents were occupationally exposed to pesticides [8].

1.2. DNA damage

Experimental data reveal that the chemical substances used in food production contain many components that affect the genetic material of organisms—they are genotoxic agents—([9–16]) and they may be responsible for the high incidence of different types of cancer (both in children and adults), reproductive problems or malformations in the offspring of populations occupationally and/or environmentally exposed to these compounds.

It has been observed that the offspring of agricultural workers have a higher risk of congenital anomalies. However, congenital anomalies in the mid-1990s represented around 20% of deaths during the first year of life in some countries, and in other countries, they represented almost 40% of deaths [17].

A genotoxic agent is described as a physical, chemical or biological agent that can interact with the genetic material (DNA) of organisms causing alterations, damage or ruptures.

This term includes agents that interact both directly and indirectly with the DNA causing ruptures and, also, those that interfere with enzymatic processes of repair, genesis or polymerization of proteins involved in chromosome segregation. Consequently, they may change the structure of a specific genome. Genotoxic agents can bind directly to DNA or act indirectly by affecting the enzymes involved in the physiological modifications of DNA during replication or transcription. These alterations could lead to impaired embryonic development or be the initial steps in the development of cancer. Genotoxic agents are not necessarily carcinogenic, but most carcinogens are genotoxic.

Genomic damage is probably the most important and fundamental cause of neurodegenerative disorders, reproductive effects and developmental problems [8]. It is also well established that genomic damage is produced by exposure to environmental contaminants (e.g., metals, pesticides), medical procedures (e.g., radiation and chemicals), micronutrient deficiency (e.g., folate), lifestyle factors (e.g., alcohol, smoking, drugs and stress), and genetic factors such as inherited defects in DNA metabolism and/or repair (Holland et al [18–20]).

Therefore, in recent years, there has been an increase in the number of studies that seek to understand and evaluate, using biomarkers, the possible consequences that exposure to pesticides has on the environment and mainly on human beings [21–23].

1.3. Genotoxicity biomarkers

Biomarkers are biological parameters that provide information about normal or pathological states of an individual or a population, and they are used for monitoring different aspects of a disease such as: treatment, prevention, diagnosis and progression of the disease, responses to the therapy, experimental toxicological evaluation of drugs or pesticides, environmental and epidemiological risk measurement, as well as evaluation of therapeutic intervention, among others [24].

In this sense, the use of genotoxicity biomarkers—chromosomal aberrations (CA), micronuclei (MN), sister chromatid exchanges (SCEs) and comets (CO)—has been relevant to analyze the potential risk of a substance, as they reveal the damage to the DNA, the molecule that transmits genetic information through generations. Therefore, they are considered suitable biomarkers to evaluate the risk of a potentially harmful substance and, in addition, their carcinogenic risk [66].

The chromosomal aberration test detects numerical (aneugenic effect) or structural (clastogenic effect) alterations at the chromosomal level. The importance of this test lies in experimental and epidemiological evidence suggesting that structural aberrations are involved in the carcinogenesis process, and, therefore, a high frequency of chromosomal aberrations is associated with an increased risk of developing cancer in the future ([26–30]).

Micronucleus test detects breaks at the chromosomal level and alterations of the mitotic apparatus, allowing the identification of compounds with aneugenic and clastogenic effects. The simplicity of the test is an advantage and the number of cells scored (1000 cells) gives statistical significance to the study.

The prospective analysis of a database of 6700 subjects from 20 laboratories representing 10 different countries have confirmed that a high frequency of micronuclei is predictive of an increased risk of cancer (Bonassi et al. [31–33]).

Sister chromatid exchanges (SCEs) are another cytogenetic assay to evaluate alterations at the chromosomal level. The exchange between sister chromatids occurs precisely by the reciprocal exchange of DNA between two sister chromatids in a duplicated chromosome. The frequency of exchange in eukaryotic cells is increased by the exposure to genotoxic agents that induce DNA damage by interfering with its replication, but it is not increased by those agents that only induce breaks in the DNA strands [28]. However, the formation mechanisms of these alterations are not completely elucidated, and, therefore, their biological significance is still uncertain [28].

The main molecular studies are (1) molecular cytogenetics to detect inversions, translocations, or to identify the chromosomal origin of micronuclei and (2) comet assay in lymphocytes.

The comet assay is an electrophoresis technique in agarose microgels considered to be highly sensitive to detect DNA damage in single cells. It detects DNA single- and double-strand breaks, labile alkali sites, and DNA-DNA or DNA-protein cross linking associated with repair sites by incomplete excision. When the nucleus is subjected to electrophoresis, the DNA fragments migrate in a pattern that resembles a comet, hence the name of this assay [34].

The studies that define the mechanisms of action and/or the cytotoxic and genotoxic effects can be performed at different levels of complexity. In vitro assays are very useful to detect the genotoxic effects of various agents in human cellular systems. Although these models do not include the toxicokinetics of substances (absorption, distribution, metabolization and excretion), it is possible to evaluate their potential effects using a wide range of biomarkers [25].

In vivo genotoxicity, studies provide a physiological framework to the activity of different agents with genotoxic potential. This allows to evaluate, under controlled conditions, a systemic response to the agent in question and to discern the effects according to the route of entry of the agent to the organism. These studies bring the results one step closer to real human exposure.

Finally, epidemiological studies use different genotoxicity biomarkers for the study of populations exposed to toxic agents. At the international level, there are numerous studies evaluating the effect of pesticides on the genetic material of exposed populations; however, in Argentina, these are still scarce [21, 35, 36].

1.4. Populations human exposed to pesticides

Studies conducted in populations exposed to pesticides, mostly in European applicators, show positive association between exposure to a complex mixture of agrochemicals and the presence of CA, SCEs, MN and/or CO [21, 37–39].

Argentine populations are exposed to complex mixtures of pesticides. In the province of Córdoba, the most commonly used mixtures contain glyphosate, cypermethrin, chlorpyrifos, and others as active ingredients ([40, 41]). Evaluating the genotoxic potential of components of the mixtures is the initial step to study its behavior to check possible antagonistic or synergistic effects that could modify the effect.

There are few reports regarding the genotoxic potential of glyphosate, cypermethrin and chlorpyrifos. Glyphosate herbicide has been studied in our research group by [12], Bosch et al. [16] and Barbosa et al. [42]. On the other hand, Kocaman and Topaktaş [43] reported on the effects of a commercial formulation of cypermethrin on peripheral blood lymphocytes, this is the only genotoxic and cytotoxic study in the available literature. Rahman et al. [44] and Vindas et al. [45] analyzed the genotoxic effects of chlorpyrifos on human cells performing the comet assay.

Therefore, there is a clear need to assemble a set of tests that cover different complexity levels so that we can have a more accurate approximation of the genotoxic potential of an agent on human population.

In this sense, it is important to highlight that a large part of the toxicity of many chemical substances is explained by their capacity to generate oxidative processes that can damage various cellular structures, including DNA; oxidative damage is, therefore, an important cause of genotoxicity [46]. One of the most commonly used techniques to evaluate the capacity of a substance to generate oxidative damage is through the quantification of thiobarbituric acid reactive substances (TBARS). A large number of reports in the literature show the oxidative effects of pesticides used in food production ([14, 47, 48]).

Given the impact of the problem raised, it is necessary to approach it not only from the biological sciences aspect (toxicological genetics), as discussed here, but to support it from the social sciences' perspective (legislation and environmental education).

The evidence of genetic risk as a result of exposure due to the intensive use of pesticides indicates the need to review the law enforcement, in order to develop educational programs aimed to control the use of these substances and/or implement prevention and protection measures.

Argentine legislation on pesticides is, in some cases, incomplete, permissive and/or obsolete [49, 50]. On the other hand, there is no participation of the Ministry of Health in the approval of pesticides registration for agricultural use. To the gaps or defects in legislation and the lack of control, is added the deficiency of measures that contemplate the effects of pesticides and their mixtures.

In the light of the foregoing, it is necessary to increase the scientific evidence regarding the toxicity and genotoxicity effects of chemical substances applied in our country. This will allow extending the legislation, adapting it to the real problems and, if necessary, modifying the permitted levels of pesticides and its mixtures in the environment.

In many countries, measuring the frequency of genetic damage in human groups exposed to environmental agents has been, for decades, a priority in public health studies, and the increased rates of chromosomal aberrations (CA) is commonly interpreted as evidence of genotoxic exposure and early biological effect on DNA.

It has long been known that there is a strong link between DNA alterations and cancer or chronic degenerative diseases. The carcinogenic process is initiated and promoted by alterations/mutations in areas where oncogenes, tumor suppressor genes and DNA repair systems are located [51, 52].

1.5. Genotoxic effects in children

Regarding the age groups and the DNA effects caused by these chemical substances, we must differentiate between adults and children. Children may be more sensitive to toxic agents compared to adults and the genetic damage occurring at an early age may represent adverse effects on adolescent or adult health (Landrigan et al. [53]; Roberts and Karr [54]). However, the information about the genotoxic effects in children is scarce, although in recent years, the number of studies has increased [35, 55, 56].

Children are a high-risk group concerning the effects of air pollution on health [53, 54, 57–59]. Some studies suggest that early childhood exposure to pollutants can lead to the development of chronic diseases in adulthood. The earlier the exposure, the higher the risk of developing a chronic disease, cancer included [60].

Among the adverse effects in children exposed to various environmental hazards, genetic damage receives special attention after it has been shown that an increased frequency of DNA damage in childhood is predictive of the development of cancer in healthy adults [61]. Children are still in an active development phase, and in this condition, their response to environmental risks may differ from that of the adults. The effects of this environment could manifest themselves many years, even decades, after exposure.

The clinical symptoms of acute pesticide poisoning are rarely pathognomonic; they can simulate an acute respiratory disease, conjunctivitis, gastrointestinal disease, cutaneous manifestations, among others.

In this sense, it agrees with Salameh et al. [62], Salam et al. [63], Alarcón et al. [64] and other authors who indicate that pesticide poisoning are commonly under-diagnosed.

Several studies show that an increase in the risk of developing cancer has been observed at high rates of both chromosomal aberrations and micronuclei [31, 32, 65, 66].

The presence of MN represents alterations that are the result of cell exposure to genotoxic contaminants.

The MNi originate from chromosome fragments or whole chromosomes that are lag behind during cell division and left outside the daughter nuclei. MNi can be assessed in different tissues such as blood and epithelial tissue, and they can be easily visualized through the optical microscope. In particular, nasal and buccal exfoliated epithelial cells have been used as biological control in people exposed to airborne contaminants since they are similar to epithelial cells of the respiratory tract and are easier to collect [18–20, 67–69].

The oral mucosal epithelial cells are the first barrier for substances introduced into the body by inhalation or ingestion; therefore, it is a suitable tissue to detect the genotoxic effects induced by airborne contaminants. Studies have shown a strong correlation between the MN frequency in buccal epithelial cells and blood cells, also related to the subsequent risk of developing cancer. Collecting the samples from this tissue is especially recommended for pediatric population due to the ease of the procedure ([70–72]).

The results from international and national publications are mostly consistent with the conclusion that environmental contaminants lead to increasing the MN frequency in children [35, 49, 55, 56, 73, 74]).

Gómez Arroyo et al. [36] evaluated the potential genotoxic risk in two groups: one group of 125 children (52 female and 73 male) from the state of Sinaloa (Mexico) whose houses are close to areas of intense agricultural activity which are sprayed with mixtures of pesticides; and a control group of 125 children (57 female and 68 male) living in the city of Los Mochis, Sinaloa; in both groups, micronuclei (MN) test in oral mucosal cells was used as a biomarker. The results showed a significant increase in the frequency of MN. Other nuclear abnormalities

associated with cytotoxicity or genotoxicity were detected; in all cases, the differences were significant when compared to the control group.

Benitez-Leite et al. [75] analyzed oral mucosa samples of 48 children from Paraguay potentially exposed to pesticides and 46 children not exposed, in order to determine genetic damage through the frequency of micronuclei (MNi). These authors found that the mean frequency of micronuclei was higher in the group potentially exposed to pesticides. This research provides evidence of genetic damage in a population of children potentially exposed to pesticides sprayed in the environment.

Our research group GeMA, performed monitoring studies in children from different locations in the Province of Córdoba through the micronuclei test (cytoma approach) in oral mucosa samples. All the locations are surrounded by fields which are sprayed with chemical agents and where there is a perception of damage caused by the use of agrochemicals. We studied 19 children between 5 and 12 years old, from the towns of Oncativo and Marcos Juárez (Province of Córdoba) which are surrounded by fields cultivated with soy and corn and with regular applications of pesticides [35]. Significant differences were found in the frequency of cells with nuclear abnormalities -buds- and the frequency of MN among the exposed groups and between them and the control groups. This work concludes that genotoxic monitoring constitutes the basis to a proper medical surveillance in populations at risk due to occupational or environmental exposure to chemical substances, such as pesticides.

Another study from Argentina [55] compares the MNi frequencies in oral exfoliated epithelial cells of environmentally exposed (by inhalation) children from urban areas with children that live in urban regions far from the area sprayed. The studied population consisted in 50 children from the town of Marcos Juárez (Córdoba), living at different exposure distances to the spraying site, and 25 children from Río Cuarto city (Córdoba), considered not exposed to such products. The MNi frequency was significantly different between the exposed children (500 m or less) when compared to the group of children not exposed. Forty percent of exposed individuals suffer some kind of persistent condition, which could be associated with chronic exposure to pesticides. The results indicated genetic damage in the group of children exposed to genotoxic substances when compared to the other group, and highlight the relevance of the MN assay in buccal mucosa for genetic biomonitoring and public health surveillance. The performed test detects a level of damage that is still reversible.

The limited number of published articles may indicate that carrying out adequately designed studies in children populations is difficult, more research in this segment of the human population is necessary.

Overall, the evidence of genetic damage in children due to early environmental exposure is strong, and every effort should be made to prevent them and pregnant women from such exposures and protect their health.

The most studied adult populations are those occupationally exposed to pesticides.

Several studies have been carried out in the agricultural sector, since it is considered the group with the highest risk of exposure to these compounds, with the purpose of evaluating the genotoxic risk they imply, especially for agricultural workers [76].

Aiassa et al. [21] and Gómez-Arroyo et al. [77] conducted a review work from studies performed on groups of people exposed to pesticides in Latin America.

In this paper from Aiassa et al. [21], we reviewed the main concepts in the field, the usefulness of genotoxicity assays and we compiled studies of genetic monitoring performed in the last 20 years in people occupationally exposed to pesticides. We think that genotoxicity tests, that include chromosomal aberrations, micronuclei test, sister chromatid exchanges and comet assays, should be considered essential tools for a complete medical monitoring in people exposed to potential environmental pollutants, particularly for those living in the same place as others who have already developed some type of malignancy. This screening is particularly important at early stages to prevent the occurrence of tumors, especially from environmental origins.

This work reviews 100 reports from different parts of the world, including 21 investigations from South America in the following countries: Argentina (6), Bolivia (2), Chile (3), Brazil (7), Colombia (2), Ecuador (1); 14 from North America: Mexico (7), United States (6), British Columbia (1); 4 from Central America: Costa Rica (3), Cuba (1); 37 from Europe: Hungary (4), Czech Republic (1), Russia (2), Spain (8), Greece (5), Italy (8), Denmark (1), Portugal (1), former Yugoslavia (1), Finland (1), Croatia (3), Poland (1), European Countries (1); 2 from Oceania: Australia (2); 2 from Africa: Egypt (2); and 10 from Asia: Syria (1), Turkey (3), Pakistan (1), Taiwan (2), Israel (1), India (2).

According to the analysis of these publications, 90% of the workers exposed to spraying are in contact with mixtures of pesticides, therefore, it is difficult to establish a correlation between a single pesticide and the damage observed. This leads to the issue of evaluating the risk of pesticides mixtures. The combined action of mixtures may result in noninteraction or interaction. If the toxicological capacity of each component of the mixture is different, the interaction between them may result either in an enhancement, when the combined effect is greater than the additive effect; or in an antagonistic effect, when the combined effect is less than the additive effect. The studies regarding the problem of pesticide mixtures are rare.

So far in Argentina, eight studies were carried out using CA, MN, SCEs and CO as biomarkers [35].

Dulout et al.'s study [78] is the first reported study from Argentina and one of the first carried out in Latin America, which was performed in floriculturists using CA and SCEs tests, obtaining negative results for CA and positive for SCEs. In 1987, the same author [79], in another population of floriculturists, analyzed CA, obtaining negative results and subsequently [80], in a new study reported positive results for SCEs. These results are followed by studies on rural workers (pesticide applicators) performed by Mañas et al. [60] that evaluate CA with positive results, Simoniello et al. [81] analyzed CO with positive results, Peralta et al. [82], who study CA, MN (in blood samples) and CO in both occupationally and environmentally exposed people, reported positive results for all biomarkers. Gentile et al. [41] also analyzed MN in blood samples with positive results. In all cases, the workers were exposed to several pesticides mixtures, making difficult to attribute the damage to a single compound and also impeding the comparison between different investigations due to the large number and variety of products applied. In none of these works was reported any exposure to other sources (confounding factors) that could interfere with the expressed results.

Gentile et al. [41] concluded that the exposure to pesticides in the study group of rural workers could induce levels of genetic damage detectable in peripheral blood lymphocytes by micronucleus assay. Age is a factor that increases both the frequency of binucleated cells with micronuclei and the total amount of micronuclei. Another factor, such as the years of exposure, does not affect these variables. Notwithstanding the above, all the potentially confounding factors must be considered when performing a cytogenetic evaluation.

As shown, in Argentina, problems derived from the use of pesticides have little attention in the health system. This situation is related to an underreporting of intoxications [83]. A high percentage of the Argentine population is engaged in agriculture and lives in rural areas where large quantities of substances are used to control plagues. It is also known that a high proportion of the population is actually and potentially exposed to these pesticides not only because they participate directly in work activities, but also because of the sprays that involuntarily reach urban areas, increasing the possibilities of harmful effects on their health.

According to Gómez Arroyo et al. [36], several studies about the genotoxic effects of pesticides have been carried out in diverse countries of Latin America from 1985 to 2013, using the four biomarkers; 41 of these studies were analyzed: 6 corresponding to Argentina, 2 to Bolivia, 10 to Brazil, 4 to Colombia, 5 to Costa Rica, 1 to Cuba, 2 to Chile, 3 to Ecuador, and 8 to Mexico. In most of the cases, workers from different countries of Latin America were in contact with products that are included in the list of highly dangerous pesticides, and it is remarkable that such individuals were mostly agricultural workers who were exposed to mixtures of pesticides. Results obtained in the studies performed in human populations demonstrate that CA, MN, SCE and CO are suitable tests to evaluate the risk associated with exposure to pesticides, showing a high percentage of positive results. Moreover, the studies carried out using CA and MN biomarkers have been correlated as predictors of cancer risk.

The genetic, molecular and biochemical methodologies that currently exist facilitate us to detect changes or alterations that act as a warning signal, allowing us to implement appropriate measures to minimize the risk on health [52].

One of the problems regarding adults population monitoring is confounding factors such as the habit of smoking, the consumption of alcohol and the occupational risk, that interfere in the analysis of the obtained results. These confounding effects are minimized and even absent during childhood. However, monitoring of children populations may consider potentially genotoxic factors including indoor tobacco smoke, regional ozone level, airborne nanoparticles, food contaminants such as pesticide residues and compounds generated by cooking (Holland et al. [84]), natural sources of ionizing and non-ionizing radiation, environmental pollutants, fuel and hydrocarbon emissions, which can vary significantly between rural or urban environmental settings [85].

2. Conclusions

Early detection of genetic damage is crucial to implement the necessary measures to reduce or suppress the exposure to deleterious agent when the damage is still reversible, thus reduce the

risk to suffer diseases. Therefore, genotoxicity tests should be considered as essential tools for a complete medical surveillance of people potentially exposed to environmental pollutants and, especially for those who inhabit places where other people have already developed any type of neoplasia at young ages, in order to prevent the occurrence of tumors of environmental or occupational origin. On the other hand, the application of these tests is useful to detect possible long-term effects of substances that are introduced to the market without knowing their capacity to affect human and environmental health.

The large number of studies performed in both occupationally exposed populations and environmentally exposed children provides important information to create a list of recommendations in order to avoid the genetic damage due to the exposure to pesticides and other contaminants.

It is recommended against the situations mentioned below:

- Disrupt the exposure to the potential risk in the workplace or in the proximity of residential areas until further studies are performed, and protection measures can be implemented to preserve the health of people (especially children) and the environment where they live.
- Control the sources of pollution, with the main objective of decreasing, removing or, ideally, eliminating the exposure.

The removal of sources of pollution away from residential areas is a matter of wide discussion, since it is difficult to control the drift of pesticides due to the environmental conditions in some provinces of Argentina such as Córdoba. Despite the toxicological classification of pesticides, the damage they cause to the genetic material of populations exposed to these chemicals should be taken into account.

The available literature shows an increased damage to the genetic material of children from Paraguay exposed to pesticides and living 50 m from the source of contamination [75].

The study carried out in Argentina with children from Marcos Juárez (Córdoba) did not found statistically significant differences in the micronuclei frequencies between the group that lives less than 500 m from the contamination source and the group that lives 500–1500 m from it. The results of Marcos Juárez study showed that the pesticides could spread by air and reach the entire town. Therefore, the vulnerable population of children is subject to an extremely high concentration and continuous exposure of pesticides, given that they live surrounded by crops fields. Up to 1095 m from the sprayed site, no significant differences were found in the MNi frequencies between the children of both groups. This information should be considered when establishing environmental safeguards in locations that are surrounded by crops fields and regularly sprayed [55].

Agricultural industry is one of the main economic activities in many regions of our country; however, despite the benefits it provides, it is responsible of environmental issues, risk to human health and damaging other organisms, so the result is a negative overall balance.

Reducing the environmental pollutants that affect human health, such as metals, pesticides, organic solvents, food additives, some natural products, and especially their derivative effects, would produce a remarkable improvement in health conditions of exposed populations.

- Establishing a monitoring protocol that tracks genotoxicity biomarkers to determine whether the biological indicators of cell damage are persistent through time, at least for 1 year and with two sampling in the absence of contaminants.

Genotoxicological monitoring in humans is a useful tool for estimating the genetic risk of exposure to a compound or complex mixtures of chemicals and constituting an early warning system for genetic diseases, reproductive problems and cancer. It also allows developing adequate control measures that can be implemented to protect human populations and the environment.

- Conduct studies of contaminants in urine, blood and environmental matrices such as air, water, sediments and soil.
- Educate the community with information campaigns about human and environmental health to promote a culture of care, foresight and prevention in the area.

It is essential to focus our attention on the children population. The WHO Task Force for the Protection of Children's Environmental Health has stated: "Children are not small adults," the premise behind this principle is that children have an exceptional vulnerability to the acute and chronic effects of environmental hazards and that they are disproportionately susceptible compared to adults [60, 86]. It has been recognized that children are a group, within the population, that has particular characteristics of exposure and special vulnerability to environmental toxins, and it is required a strategy for risk assessment that considers their particular features [53, 54].

The health of a society can be judged by the health of its children. This implies the early identification of preventable risks and the immediate translation of this knowledge into effective protection policies.

The evidence of the effects of environmental exposure at an early age is so substantial that every effort should be made to avoid such exposures in children and pregnant women as well as to protect their present and future health (Holland et al. 2011).

Author details

Delia Aiassa

Address all correspondence to: delia.aiassa@gmail.com

Departamento de Ciencias Naturales, Laboratorio GeMA – Genética y Mutagénesis Ambiental, Universidad Nacional de Río Cuarto, Argentina

References

[1] IARC International Agency for Research on Cancer. Monographs on the evaluation of carcinogenic risks to humans: 84. World Health Organization; 2002

[2] IARC International Agency for Research on Cancer 2015. Monographs on the evaluation of carcinogenic risks to humans: 112. World Health Organization

[3] FAO. 2004. El estado de la inseguridad alimentaria en el mundo 2004. Roma

[4] Martínez-Valenzuela C, Gómez-Arroyo S. Riesgo genotóxico por exposición a plaguicidas en trabajadores agrícolas. Revista internacional de contaminación ambiental. 2007;**23**(4):185-200

[5] Bhalli JA, Khan QM, Haq MA, Khalid AM, Nasim A. Cytogenetic analysis of Pakistani individuals occupationally exposed to pesticides in a pesticide production industry. Mutagenesis. 2006;**21**(2):143-148

[6] Alavanja M, Sandler D, McDonnell C, Lynch C, Pennybacker M, Zahm S, Mage D, Steen W, Wintersteen W, Blair A. Characteristics of pesticide use in a pesticide applicator cohort: The agricultural health study. Environmental Research. 1997;**80**:172-179

[7] Kohen R, Nyska A. Oxidation of biological systems: Oxidative stress phenomena, antioxidants, redox reactions, and methods for the quantification. Toxicology Pathology. 2002; **30**(6):620-650

[8] Bolognesi C, Creus A, Ostrosky-Wegman P, Marcos R. Micronuclei and pesticide exposure. Mutagenesis. 2011;**26**(1):19-26

[9] Dearfield KL, McCarroll NE, Protzel A, Stack HF, Jackson MA, Waters MD. A survey of EPA/OPP and open literature on selected pesticide chemicals. II: Mutagenicity and carcinogenicity of selected chloroacetanilides and related compounds. Mutation Research. 1999;**443**:183-221

[10] Ambulkar P, Ghosh S, Ingole I, Pal A. Genotoxic and cytotoxic effects of antibacterial drug, ciprofloxacin, on human lymphocytes in vitro. Nepal Medical College Journal. 2009; **11**(3):147-151

[11] Gorla N, García Ovando H, Larripa I. Chromosomal aberrations in human lymphocytes exposed to enrofloxacin and ciprofloxacin. Toxicology Letters. 1999;**104**:43-48

[12] Mañas F, Gonzalez Cid M, Weyers A, Ugnia L, García Ovando H, Larripa I, et al. Evaluación De Genotoxicidad "In Vivo" Mediante El Ensayo Cometa Y De Micronúcleos En Ratones Tratados Con Glifosato. Theoria. 2007. 2007;**15**:53-60

[13] Mañas F, Peralta L, Gorla N, Bosch B, Aiassa D. Aberraciones Cromosómicas En Trabajadores Rurales De La Provincia De Córdoba Expuestos A Plaguicidas. Journal of Basic Applied Genetics. 2009;**20**(1):9-13

[14] Mañas F, Peralta L, Raviolo J, García Ovando H, Weyers A, Ugnia L, et al. Genotoxicity of Ampa, environmental metabolite of glyphosate, assessed by the comet assay and cytogenetic tests. Ecotoxicology and Environmental Safety. 2009a;**72**:834-837

[15] Mañas F, Peralta L, Raviolo J, García Ovando H, Weyers A, Ugnia L, et al. Genotoxicity and oxidative stress of glyphosate: In vivo and in vitro testing. Environ Toxicol Pharmacol. 2009b:37-41

[16] Bosch B, Mañas F, Gorla N, Aiassa D. Micronucleus test in post metamorphic Odontophrynus cordobae and Rhinella arenarum (Amphibia: Anura) for environmental monitoring. Journal of Toxicology and Environmental Health Sciences. 2011;**3**(6):154-163

[17] Regidor E, Ronda E, García AM, Domínguez V. Paternal exposure to agricultural pesticides and cause specific fetal death. Occupational and Environmental Medicine. 2004;**61**: 334-339

[18] Holland N, Bolognesi C, Kirsch-Volders M, Bonassi S, Zeiger E, et al. The micronucleus assay in human buccal cells as a tool for biomonitoring DNA damage: The HUMN project perspective on current status and knowledge gaps. Mutation Research. 2008a;**659**:93-108

[19] Holland N, Bolognesi C, Kirsh-Volders M, Bonassi S, Zeiger E. y S. Knasmueller. 2008b. The micronucleus assay in human buccal cells as a tool for biomonitoring DNA damage: The HUMN project perspective on current status and knowledge gaps. Mutation Reserach. 656(1–2):93-108

[20] Holland et al. The micronucleus assay in human buccal cells as a tool for biomonitoring DNA damage: The HUMN project perspective on current status and knowledge gaps. Mutation Research. 2008c;**659**(1–2):93-108

[21] Aiassa D, Mañas F, Bosch B, Gentile N, Bernardi N, Gorla N. Biomarcadores de daño genético en poblaciones humanas expuestas a plaguicidas. Acta Biológica Colombiana. 2012;**17**(3):485-510

[22] Alavanja MC, Hoppin JA, Kamel F. Health effects of chronic pesticide exposure: Cancer and neurotoxicity. Annual Review of Public Health. 2004;**25**:155-197

[23] Reffstrup TK, Larsen JC, Meyer O. Risk assessment of mixtures of pesticides. Current approaches and future strategies. Regulatory Toxicology and Pharmacology. 2010;**56**:174-192

[24] Lock EA, Bonventre JV. Biomarkers in translation; past, present and future. Toxicology. 2008;**245**(3):163-166

[25] Mudry M, Carballo M. Genética Toxicológica. Buenos Aires, Argentina: Ed. De Los 4 Vientos; 2006

[26] Hagmar L, Brogger A, Hansteen IL, Heim S, Hogstedt B, Knudsen L, et al. Cancer risk in ¨humans predicted by increased levels of chromosomal aberrations in lymphocytes: Nordic study group on the health risk of chromosome damage. Cancer Research. 1994;**54**: 2919-2922

[27] Hagmar L, Bonassi S, Stromberg U, Brogger A, Knudsen LE, Norppa H, Reuterwall C. Chromosomal aberrations in lymphocytes predict human cancer: A report from the European Study Group on Cytogenetic Biomarkers and Health (ESCH). Cancer Research. 1998; **58**:4117-4121

[28] Albertini RJ, Anderson D, Douglas GR, Hagmar L, Hemminki K, Merlo F, et al. IPCS guidelines for the monitoring of genotoxic effects of carcinogens in humans. Mutation Research. 2000;**463**:111-172

[29] Bonassi C, Abbondandolo A, Camurri L, Dal Pra L, De Ferrari M, Degrassi F. Are chromosome aberrations in circulating lymphocytes predictive of a future Cancer onset in humans preliminary results of an Italian cohort. Study. Cancer Genetics and Cytogenetics. 1995;**79**(2):133-135

[30] Mitelman F, Mertens F, Johansson F. A breakpoint map of recurrent chromosomal rearrangements in human neoplasia. Nature Genetics. 1997;**15**:417-474

[31] Bonassi S, Znaor A, Ceppi M, Lando C, Chang WP, Holland N, et al. An increased micronucleus frequency in peripheral blood lymphocytes predicts the risk of cancer in humans. Carcinogenesis. 2007a;**28**(3):625-631

[32] Bonassi S, Znaor A, Ceppi M, Lando C, Chang WP, et al. An increased micronucleus frequency in peripheral blood lymphocytes predicts the risk of cancer in humans. Carcinogenesis. 2007b;**28**(3):625-631

[33] Bonassi et al. The HUman MicroNucleus project on eXfoLiated buccal cells (HUMN(XL)): The role of life-style, host factors, occupational exposures, health status, and assay protocol. Mutation Research. 2011c;**728**(3):88-97

[34] Rojas E, López MC, Valverde M. Single cell gel electrophoresis assay: Methodology and applications. Journal of Chromatography B. 1999;**722**:225-254

[35] Aiassa D, Mañas F, Bernardi N, Gentile N, Méndez Á, Roma D, Gorla N. Monitoreo de Genotoxicidad en personas expuestas a plaguicidas. Estudio preliminar en niños. Cuestiones de Población y Sociedad. 2014;**4**(4):73-84

[36] Gómez Arroyo S, Martinez Valenzuela C, Carbajal-López A, Martínez-Arroyo MC-S. R., Villalobos-Pietrini y S. Waliszewski, Riesgo genotóxico por la exposición ocupacional a plaguicidas en América Latina. Revista Internacional de Contaminación Ambiental. 2013; **29**:159-180

[37] Bolognesi C. Review Genotoxicity of pesticides: A review of human biomonitoring studies. Mutation Research. 2003;**543**(3):251-272

[38] Ergene S, Çelik A, Çavaş T, Kaya F. Genotoxic biomonitoring study of population residing in pesticide contaminated regions in Göksu Delta: Micronucleus, chromosomal aberrations and sister chromatid exchanges. Environment International. 2007;**33**(7):877-885

[39] Pastor Benito S. Biomonitorización citogenética de cuatro poblaciones agrícolas europeas expuestas a plaguicidas mediante el ensayo de micronúcleos. Tesis doctoral. Universitat Autónoma de Barcelona. Disponible en; 2002 http://www.tdx.cat/bitstream/handle/10803/3858/spb1de5.pdf?sequence=1

[40] López SL, Aiassa D, Benítez-Leite S, Lajmanovich R, Mañas F, Poletta G, Sánchez N, Simoniello MF, Carrasco AE. Pesticides Used in South American GMO-Based Agriculture: A Review of Their Effects on Humans and Animal Models. 2012 In James

[41] Gentile N, Mañas F, Bosch B, Peralta L, Gorla N, Aiassa D. Micronucleus assay as a biomarker of genotoxicity in the occupational exposure to agrochemiclas in rural workers. Bulletin of Environmental Contamination and Toxicology. 2012;**88**(6):816-822

[42] Barbosa MC, Aiassa y D, Mañas YF. Evaluación de daño al ADN en leucocitos de sangre periférica humana expuestos al herbicida Glifosato. Revista Internacional de Contaminación Ambiental. 2017;**4**(33):403-410

[43] Kocaman A, Topaktaş M. The in vitro genotoxic effects of a commercial formulation of α-cypermethrin in human peripheral blood lymphocytes. Environmental and Molecular Mutagenesis. 2009;**50**(1):27-36

[44] Rahman M, Mahboob M, Danadevi K, Banu B, Grover P. Assessment of genotoxic effects of chloropyrifos and acephate by the comet assay in mice leucocytes. Mutation Research. 2002;**516**:139-147

[45] Vindas R, Ortiz F, Ramírez V, Cuenca P. Genotoxicidad de tres plaguicidas utilizados en la actividad bananera de Costa Rica. Revista de biologia tropical. 2004;**52**:197-205

[46] Bandyopadhyay U, Das D, Banerjee R. Reactive oxygen species: Oxidative damage and pathogenesis. Current Science. 1999;**77**:103-121

[47] Martínez-Cayuela M. Toxicidad de xenobióticos mediada por radicales libres de oxígeno. Ars Pharmaceutica. 1998;**39**:5-18

[48] Beuret C, Zirulnik F, Giménez M. Effect of the herbicide glyphosate on liver lipoperoxidation in pregnant rats and their fetuses. Toxicology Reports. 2005;**19**:501-504

[49] Gómez Miralles J, Bevilacqua S, Mañas F, Bosch B, Gentile N, Peralta L, Aiassa D. Los plaguicidas en Argentina: la genotoxicidad de los agroquímicos y la falta de prevención penal. Abeledo Perrot Córdoba. 2012;**2**:128-139

[50] Gómez Miralles J, Bevilacqua V, Bevilacqua S. ¿Cuál es la normativa sobre agroquímicos en la República Argentina? Cap. 10. En Aiassa, D., B. Bosch y F. Mañas (comp). Plaguicidas a la carta: daño genético y otros riesgos. Miguel Tréspidi Editores. 2012a. 216pp

[51] Au WW, Badary OA, Heo MY. Cytogenetic assays for monitoring populations exposed to environmental mutagens. Occupational Medicine. 2001;**16**(2):345-357

[52] Bonassi S, Au WW. Biomarkers in molecular epidemiology studies for health risk prediction. Mutation Research. 2002;**511**(1):73-86

[53] Landrigan PJ, Kimmel CA, Correa A, Eskenazi B. Children's health and the environment: Public health issues and challenges for risk assessment. Environmental Health Perspectives. 2004a;**112**(2):257-265

[54] Roberts JR, Karr CJ. Pesticide exposure in children. Pediatrics. 2012;**130**:6

[55] Bernardi et al. Assessment of the level of damage to the genetic material of children exposed to pesticides in the province of Córdoba. Archivos Argentinos de Pediatría. 2015;**113**(1):126-132 ISSN: 0325–0075

[56] Neri M, Ugolini D, Bonassi S, Fucic A, Holland N, Knudsen LE, et al. Children's exposure to environmental pollutants and biomarkers of genetic damage II. Results of a comprehensive literature search and meta-analysis. Mutation Research. 2006;**612**(1):14-39

[57] Grigg J. Particulate matter exposure in children: Relevance to chronic obstructive pulmonary disease. Proceedings of the American Thoracic Society. 2009;**6**(7):564-569

[58] WHO. Children's Health and the Environment in Europe: A Baseline Assessment. Copenhagen, Denmark: World Health Organization Regional Office for Europe; 2007

[59] European Respiratory Society (ERS). 2010 Annual Congress 22; Barcelona, Spain; 2010

[60] Wild CP, Kleinjans J. Children and increased susceptibility to environmental carcinogens: Evidence or empathy? Cancer Epidemiology, Biomarkers & Prevention. 2003;**12**:1389-1394

[61] Gallo V, Khan A, Gonzales C, Phillips DH, Schoket B, Györffy E, et al. Validation of biomarkers for the study of environmental carcinogens: A review. Biomarkers. 2008;**13**:505-534

[62] Salameh PR, Baldi I, Brochard P, Raherison C, et al. Respiratory symptoms in children and exposure to pesticides. The European Respiratory Journal. 2003;**22**(3):507-512

[63] Salam MT, Li YF, Langholz B, Gilliland FD. Early-life environmental risk factors for asthma: Findings from the Children's health study. Environmental Health Perspectives. 2004;**112**(6):760-765

[64] Alarcon WA, Calvert GM, Blondell JM, Mehler LN, Sievert J, Propeck M, Tibbetts DS, Becker A, Lackovic M, Soileau SB, Das R, Beckman J, Male DP, Thomsen CL, Stanbury M. Acute illnesses associated with pesticide exposure at schools. Journal of the American Medical Association. 2005;**294**:455-465

[65] Bonassi S, Coskun E, Ceppi M, Lando C, Bolognesi C, et al. The HUman MicroNucleus project on eXfoLiated buccal cells (HUMN(XL)): The role of life-style, host factors, occupational exposures, health status, and assay protocol. Mutation Research. 2011a;**728**(3):88-97

[66] Bonassi S, El-Zein R, Bolognesi C, Fenech M. Micronuclei frequency in peripheral blood lymphocytes and cancer risk: Evidence from human studies. Mutagenesis. 2011b;**26**(1):93-100

[67] Coronas MV, Pereira TS, Rocha JA, Lemos AT, Fachel JM, et al. Genetic biomonitoring of an urban population exposed to mutagenic airborne pollutants. Environment International. 2009;**35**(7):1023-1029

[68] Kashyap B, Reddy PS. Micronuclei assay of exfoliated oral buccal cells: Means to assess the nuclear abnormalities in different diseases. Journal of Cancer Research and Therapeutics. 2012;**8**(2):184-191

[69] Samanta S, Dey P. Micronucleus and its applications. Diagnostic Cytopathology. 2012;**40**: 84-90

[70] Ceppi M, Biasotti B, Fenech M, Bonassi S. Human population studies with the exfoliated buccal micronucleus assay: Statistical and epidemiological issues. Mutation Research. 2010;**705**(1):11-19

[71] Bonassi S, Coskun E, Ceppi M, Lando C, Bolognesi C, Burgaz S, Holland N, Kirsh-Volders M, Knasmueller S, Zeiger E, Carnesoltas D, Cavallo D, da Silva J, de Andrade VM, Demircigil GC, Domínguez Odio A, Donmez-Altuntas H, Gattas G, Giri A, Giri S, Gómez-Meda B, Gómez-Arroyo S, Hadjidekova V, Haveric A, Kamboj M, Kurteshi K,

Martino-Roth MG, Montero Montoya R, Nersesyan A, Pastor-Benito S, Favero Salvadori DM, Shaposhnikova A, Stopper H, Thomas P, Torres-Bugarín O, Yadav AS, Zúñiga González G, Fenech M. The Human MicroNucleus project on eXfoLiated buccal cells (HUMN(XL)): The role of life-style, host factors, occupational exposures, health status, and assay protocol. Mutation Research. 2011;**728**(3):88-97

[72] Gentile N, Bernardi N, Bosch B, Mañas F, Aiassa D. Estudios de genotoxicidad en trabajadores rurales y familias. Revista Cubana de Investigaciones Biomédicas. 2016;**35**(3):228-239

[73] Ghosh P, Basu A, Singh KK, Giri AK. Evaluation of cell types for assessment of cytogenetic damage in arsenic exposed population. Molecular Cancer. 2008;**7**:45

[74] Suruda A, Schulte P, Boeniger M, Hayes RB, Livingston GT, Steenland K, et al. Cytogenetic effects of formaldehyde exposure in students of mortuary science. Cancer Epidemiology, Biomarkers & Prevention. 1993;**2**:453-460

[75] Benítez-Leite S, Macchi ML, Fernández V, Franco D, Ferro EA, Mojoli A, Cuevas F, Alfonso J, Sales L. Daño celular en una población infantil potencialmente expuesta a pesticidas. Pediatria. 2010;**37**(2)

[76] Castillo-Cadena J, Tenorio-Vieyra LE, Quintana-Carabia AI, García-Fabila MM, Ramírez-San Juan E. y Madrigal-Bujaidar E. Determination of DNA damage in floriculturists exposed to mixtures of pesticides. Journal of Biomedicine & Biotechnology. 2006. DOI 10.1155/ JBB/ 2006/97896

[77] Gómez-Arroyo S, Martínez-Valenzuela C, Calvo-González S, Villalobos-Pietrini R, et al. Assessing the genotoxic risk for mexican children who are in residential proximity to agricultural areas with intense aerial pesticide applications. Revista internacional de contaminación ambiental. 2013;**29**(3):217-225

[78] Dulout FN, Pastori MC, Olivero OA, Gonzales CID, Loria D, Matos E, et al. Sister-chromatid exchanges and chromosomal aberrations in a population exposed to pesticides. Mutation Research. 1985;**143**:237-244

[79] Dulout FN, Pastori MC, González Cid M, Matos E, Von Guradze HN, Maderna CR, et al. Cytogenetic analysis in plant breeders. Mutation Research. 1987;**189**(4):381-386

[80] Dulout FN, López Camelo JS, Guradze HN. Analysis of sister chromatid exchanges (SCE) in human populations studies. Rev Brazil Genetics. 1992;**15**(1):169-182

[81] Simoniello MF, Kleinsorge EC, Carballo MA. Evaluación bioquímica de trabajadores rurales expuestos a pesticidas. Medicina (Buenos Aires). 2010;**70**(6):489-498

[82] Peralta P, Mañas F, Gentile N, Bosch B, Méndez A, Aiassa D. Evaluación del daño genético en pobladores de Marcos Juárez Expuestos a plaguicidas: estudio de un caso en Córdoba, Argentina. Rev Cient Psicol Ciencias Soc Human Ciencias Salud. 2011;**2**(1):7-26

[83] Souza CJ. La problemática del uso de los agroquímicos y sus envases, su incidencia en la salud de los trabajadores, la población expuesta y el ambiente: Estudio colaborativo multicéntrico. Ministerio de Salud de la Nación. Argentina: Buenos Aires; 2007

[84] Holland N, Fucic A, Merlo DF, Sram R, Kirsch-Volders M. Micronuclei in neonates and children: effects of environmental, genetic, demographic and disease variables. Mutagenesis. 2011;**26**(1):51-56

[85] Gajskia G, Gerića M, Oreščaninb V, Garaj-Vrhovaca. Cytogenetic status of healthy children assessed with the alkaline comet assay and the cytokinesis-block micronucleus cytome assay. Mutation Research. 2013;**750**:55-62

[86] Garry VF. Pesticides and children. Toxicology and Applied Pharmacology. 2004;**198**:152-163