Life Sciences 203 (2018) 177–183

Contents lists available at ScienceDirect

Life Sciences

journal homepage: www.elsevier.com/locate/lifescie

Occupational exposure to pesticides in family agriculture and the oxidative, biochemical and hematological profile in this agricultural model

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ARTICLE INFO

Keywords:
Family farming
Rural worker health
Pesticides
Biomarkers
Occupational exposure
Biochemical profile
Hematologic profile
Oxidative stress

ABSTRACT

Aims: Family agriculture represents 80% of the Brazilian rural population, and rural workers linked to this agricultural model are also occupationally exposed to pesticides, which causes health risks for this part of the population. Thus, assessing health conditions becomes important and necessary as a form of biomonitoring of markers that can be used in effective health programs for agricultural workers.
Materials and methods: A total of 152 farmers, with an average age of 52 years, who answered the questionnaire and performed anthropometric, blood pressure, and blood dosages to determine the biochemical profile, hematological, markers of oxidative stress and mutagenicity. The sample was divided according to the interview report in two groups: volunteers who use pesticides in agricultural work ($n = 84$), and volunteers who do not use pesticides ($n = 68$).
Key findings: The group that uses pesticides presented higher values for measuring the circumference of the neck, and significant changes for the markers of oxidative stress, TBARS and Carbonyl, as well as significant reduction of antioxidant enzymes SOD, GPx e GSH, and in the dosages of total cholesterol, alkaline phosphatase, albumin, total leukocytes, monocytes and platelets. No mutagenic damage was verified according to the micronucleus test (MN).
Significance: The parameters that presented changes for the group that uses pesticides prove that even the low exposure can cause alteration of biomarkers, likely to compromise the health of the rural population. The results indicate the need for biomonitoring, and evaluation of its long-term impacts on farmers' health.

1. Introduction

Brazilian agriculture, in evident growth in recent decades, has consolidated Brazil as the world's second largest agricultural exporter [1]. With an agricultural model based on monocultures for export, the increase in productivity is linked to agricultural mechanization and increased use of pesticides [2] positioning the country as the world's largest consumer of pesticides [3].

Inserted in this context is family farming, where property management is shared by family members and rural activity is the main source of income [4]. Characterized by rural properties with a small territorial extension, family agriculture represents 84.4% of Brazilian rural establishments, where 74% of the workforce is linked to the agricultural sector [5]. Most of these farmers are linked to short production chains, responsible for about 70% of the food consumed in the country [6]. Distributed in agricultural agglomerates very diversified as to variables such as colonization, schooling and the types of production for which they are, these farmers are also involved in the process of "green revolution", in which the use of pesticides was driven by government incentives in the search for greater profitability [2].

The increase in pesticide use is strongly linked to increased productivity, both in production for export and in family agriculture. Nonetheless, the financial rise generated by agribusiness masks the negative impacts linked to the use of pesticides. In addition to the environmental impact, several studies aim to evaluate the damage to the health of rural workers [2]. The increase in the number of acute intoxications caused by the inappropriate use of these chemicals is in addition to numerous studies that seek to evaluate the chronic health effects caused by pesticides. Pesticides are being linked to changes in the endocrine and reproductive systems, lung disorders, degenerative diseases, and increased incidence of cancer in exposed populations [7–15]. The use of these products in public health for pest control by

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https://doi.org/10.1016/j.lfs.2018.04.038
Received 31 January 2018; Received in revised form 11 April 2018; Accepted 20 April 2018
Available online 22 April 2018
0024-3205/

disinfecting companies together with the excessive contamination in the food produced, expands the problem also for the urban population [12,16,17].

Considering that these exposed populations have very different characteristics regarding the type and mode of exposure, are the epidemiological studies that may point out possible biomarkers of occupational exposure. Biomarkers are important tools in epidemiological studies that seek to establish the relationship between exposure to chemical agents and their effects on exposed individuals [18]. By correlating the use of pesticide mixtures with health changes and their long-term chronic effects, these biomarkers can aid in the early diagnosis processes and prevention and control of chronic diseases, whether or not linked to pesticide exposure [12,19].

The main objective of the study was to determine the health conditions of a rural population linked to family farming in their real working conditions, and to identify possible biomarkers useful in the process of early diagnosis and prevention of chronic diseases.

2. Material and methods

2.1. Study population

The study was carried out in 9 localities of the rural zone of the municipality of Santiago (South region of Brazil). All participants are linked to family farming whose rural properties are of small territories, with low mechanization, and production directed to the own consumption or to commercialization in the nearest municipality. Seeking to assess the impact of pesticide use on the health of these farmers, and minimizing the interference of environmental exposure, the use of pesticides during labor practices was used as a criterion through self-report in a previously prepared questionnaire. The sample was divided into two groups: 84 farmers reported using pesticides at work (study group) and the control group consisted of 68 farmers, residents in the same localities who do not use pesticides in their agricultural work.

2.2. Identification and application of the questionnaire

This study was approved by the Ethics and Research Committee. After signing the Informed Consent Form an identification questionnaire was applied, and anthropometric data, health conditions and work practices of the participants were collected. The types of crops worked, the pesticides used and their frequency of use, as well as the individual protection conditions of each participant were also part of this questionnaire.

2.3. Collection of biological material

The biological samples were collected by venipuncture and adequately refrigerated for the transport of biological material. All volunteers were instructed to 12 h of fasting. Some aliquots were processed (hemogram, platelets and biochemical measurements), and others were frozen for further processing (markers of oxidative stress and antioxidant defenses). The preparation of the slides for analysis of Micronuclei was performed at the time of the hemogram. The study was conducted from August 2015 to April 2016.

2.4. Hematologic evaluation

Hemogram and platelet counts were performed on automated Sysmex KX 21 equipment using standard commercial kits and whole blood with K_3EDTA anticoagulant. At the time the blades were prepared for staining and analysis of the micronuclei test.

2.5. Biochemical evaluation

Using serum, glucose, total cholesterol, HDL, LDL, triglycerides, uric acid, urea, creatinine, aspartate aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatase, GT Range, total proteins and albumin measurements were performed in equipment ChemWell T (Labtest®), using commercial kits Labtest® and/or Bioclin®.

2.6. Evaluation of oxidative stress

Using plasma or whole blood, the following tests were carried out to evaluate the oxidative stress and antioxidant defenses of the groups studied: lipid peroxidation (TBARS), carbonylation of proteins (Carbonil), superoxide dismutase (SOD), catalase (CAT), glutathione reductase (GSH), and glutathione peroxidase (GPx).

2.7. Assessment of mutagenicity

The frequency of micronuclei (%) was evaluated in leukocytes. Global blood was collected and a sample was placed on the surface of the blade and made a smear, the blood was spread over the surface of the blade. After 24 h, the slides were fixed in 96% ethanol for 30 min. The slides were stained with anoptic dye and washed in water and put to dry. After drying the cells analyzed were considered as micronuclei the particles in relation to the main core: not exceed 1/3 of their size, are clearly separated with discernible edges and with the same color and refringence core [20,21].

2.8. Statistical analysis

The results were tabulated and analyzed in the statistical program SPSS version 20.0. The descriptive analyzes were done through frequency (%) or means (±) and standard deviation. To compare the means between the groups, the t-student-test was used and significant values of $p \leq 0.05$.

3. Results

A total of 152 farmers engaged in family agriculture, aged between 18 and 80 years (52.5 ± 13.7 years) participated in the study. The main characteristics of this population are presented in Table 1.

Smoking (10.5%) and regular consumption of alcoholic beverages (8.6%) were reported with low frequency by the study population and were not different among the groups.

Most of the participants (90.1%) own their work area. The group that uses pesticides presents, on average, a greater extension of the property in relation to the group that does not use pesticides. Farmers are engaged in diverse crops as soy, corn, beans, tobacco, fruits, vegetables and animal breeding.

Of the total sample of farmers, 87% reported knowing about personal protective equipment. The characteristics of work, occupational exposure and use of Personal Protective Equipment (PPE) of the group that declared to use pesticides, are presented in the Table 2.

Most participants have been exposed to pesticide mixtures for > 10 years, mainly during soybean cultivation (September to January). The pesticides used by the group, cited during the interview, are listed in the Table 3.

The herbicide Glyphosate was the pesticide most cited by farmers, and the main forms of application used are the costal applicator and the sprayer coupled to the tractor.

As for the markers, the results are grouped in biochemical markers, hematological and oxidative stress and micronucleus.

The biochemical determinations evaluated are shown in Fig. 1.

Table 4 presents the results of hematological parameters evaluated in the studied group.

The evaluation of the level of oxidative stress in the groups studied was performed based on the results obtained, presented in the Fig. 2.

The mutagenicity was evaluated using the NDI, calculated based on the Micronucleus test results, which can be visualized in Table 5.