



TECHNICAL REASSESSMENT OPINION NO. 08/GGTOX/Anvisa, of June 13, 2016

Analysis of the contributions to the proposed reassessment of the active ingredient Paraquat supported by the Technical Reassessment Opinion no. 01 of 2015/GGTOX/Anvisa, object of the Public Consultation no. 94 of October 8, 2015.

SUMMARY

The reassessment of the active ingredient Paraquat was determined by Resolution – RDC No. 10 dated February 22, 2008 due to the existence of studies showing the high acute and chronic toxicity of this active ingredient. The Oswaldo Cruz Foundation (Fiocruz) was hired to elaborate the technical note addressing all the relevant toxicological aspects of the active ingredient Paraquat. In 2015, the General Toxicology Office (GGTOX) analyzed the technical reassessment note of the active ingredient Paraquat elaborated by Fiocruz to evaluate the content and legal basis presented and conclude on which toxicological effects of Paraquat to humans were relevant from the regulatory point of view. This analysis supported the technical note of the National Health Surveillance Agency (Anvisa) and the proposition of technical regulation indicating the prohibition of this active ingredient in Brazil, which was included in the Public Consultation (CP) in the end of 2015. In 2016, GGTOX concluded the consolidation of the contributions of the CP and reviewed the contested toxicological aspects as well as analyzed new data and documents presented during this regulatory phase.

The technical note elaborated by Fiocruz concluded that Paraquat classified under all the prohibitive criteria for registration related to health established by Act no. 7.802 of June 1989 and by Decree no. 4.074 of January 4, 2002, because it considered the evidences in scientific literature related to acute intoxication, mutagenicity, endocrine deregulation, carcinogenesis, reproductive toxicity, teratogenesis and Parkinson's Disease to be sufficient. However, the conclusions made by Fiocruz were not necessarily based on an analysis with evaluation of the quality of the studies, according to regulatory criteria, or with correct interpretation regarding the suitability of the indication of the toxicological aspects as being prohibitive criteria for registration. Therefore, after analyzing the technical note of Fiocruz, Anvisa concluded that there was sufficient weight of evidence and legal basis for the prohibition of Paraquat in Brazil only for the following aspects: severity of the cases of occupational and accidental intoxications, due to the fact that the use of protection equipment (IPEs) does not guarantee full protection against intoxication by Paraquat, with the real level of exposure of workers exceeding the acceptable occupational exposure levels (AOELs); due to the existence of evidences of the triggering of Parkinson's Disease and evidences of its mutagenic potential. The technical note with this understanding was included in the CP, as well as the technical regulation proposal prohibiting Paraquat in Brazil.

Majority of the contributions received in the CP highlighted the impacts of the prohibition or severe restriction of Paraquat related to the agronomic aspects (lack of substitutes, problems with weed resistance, difficulty in pest control, increased use of other herbicides), economic aspects (loss of productivity, increased production cost, negative impacts on neighboring countries) and environmental aspects (impossibility of direct planting, increased soil degradation, increased emission of CO₂).

The analysis of the scientific literature studies showed that there are cases of severe acute intoxication of exposed workers but majority occurs from the incorrect use of Paraquat. Regarding the national scenario of Paraquat intoxication, it was observed that death is predominant in cases of suicidal and accidental intoxication. Therefore, considering the real possibility of intoxication by the product when used correctly in farming, severe acute intoxication is not common and the cases of acute occupational intoxication tends to be either moderate or mild, with effective antidotes and treatments



able to revert the health damages resulting from occupational exposure to Paraquat-based products.

Therefore, it is concluded that Paraquat, in relation to acute intoxication, is not classified under any prohibitive criterion for pesticide registration in Brazil. However, it is worth emphasizing that occupational exposure to Paraquat is relevant, especially due to evidences of higher human sensitivity to dermal exposure to this pesticide, with possible systemic absorption by this route of exposure. Furthermore, the cases of severe acute intoxication by accidental ingestion of Paraquat or suicide attempt are disturbing. Soon, considering all these aspects related to acute intoxication by Paraquat, even if it is concluded that this active ingredient classifies under other criteria considered prohibitive for registration, very restrictive risk mitigating measures would be imperative for it to remain in the country.

Although it was concluded to not include Paraquat in prohibitive criteria for registration due to acute intoxications by this pesticide, the conclusions on mutagenicity and Parkinson's Disease were kept, even after in-depth analyses, because the conclusions presented initially by Anvisa were confirmed in the consolidation phase of the CP.

The scientific evidences currently available on the mutagenic potential of Paraquat show that there is sufficient weight of evidence to prove its potential of inducing chromosomal aberrations in somatic cells *in vitro* and *in vivo* in different species and by different exposure routes, including dermal. Considering that the effect observed in somatic cells is enough to characterize its mutagenic potential in germinative cells, because it was possible to prove the inexistence of mutation in germinative cells and, in these cases, this is the scientific and regulatory understanding currently being used. Furthermore, the exposure to Paraquat reaches the testicles, affecting the spermatozooids, causing more concern with the mutagenicity of Paraquat in germinative cells, and the dermal exposure of humans is relevant and more important than the exposure of animals, thus rural workers may be susceptible to the mutagenic effects of Paraquat.

Based on the currently available evidences, there is a strong weight of evidence (animal studies and epidemiological studies) indicating that Paraquat is associated to the triggering of Parkinson's Disease in humans. Considering that the consequences of human exposure to Paraquat is more relevant than animal exposure, because Parkinson is a serious progressive disease whose treatment only delays the symptoms but is not able to revert the condition, that is, there is no cure and, due to the known severity of this disease to humans, it is possible to conclude that Paraquat may prove to be more dangerous to humans than what is shown by tests with laboratory animals and there is no antidote and efficient treatment. Although monitoring studies do not show exposures above the AOEL, it is known that Paraquat can be absorbed through the dermal route and even low doses are a concern due to the evidences associating this active ingredient to parkinsonism. It is not possible to state that the currently accepted AOEL is a safe limit in relation to the effects of Paraquat on dopaminergic neurons resulting from long-term exposure of rural workers.

Considering the current scientific knowledge, it is concluded that Paraquat is mutagenic and is a risk factor in Parkinson's Disease, with evidence of causality between exposure and the disease, being classified under the following prohibitive criteria for registration: (a) Scientific evidences of mutagenicity according to the updated results of experiences of the scientific community (Section c, Paragraph 6, Article 3 of Act no. 7.802, of 1989); (b) Lack of antidote or efficient treatment (Item II of Article 31 of Decree no. 4.074 of 2002 and Section b, Paragraph 6, Article 3 of Act no. 7.802, of 1989); (c) Evidences that the active ingredient is more dangerous to humans than what is shown in laboratory tests with animals (Item VII of Article 31 of Decree no. 4.074 of 2002 and Section c, Paragraph 6, Article 3 of Act no. 7.802, of 1989).

Based on the scientific data currently available and due to the classification of the toxicological aspects of Paraquat under the prohibitive criteria for registration established in the Brazilian legislation, the prohibition of the production, export, import, marketing and use of products made with this active pesticide ingredient in the country is suggested.

I – REPORT

The Technical Reassessment Opinion no. 01 on the toxicological reassessment of the active ingredient Paraquat considered that this pesticide presents unacceptable acute



toxicity, it has no antidote or efficient treatment, it is mutagenic and potentially capable of triggering parkinsonism in exposed workers, suggesting the cancellation of the registrations of the products made with this active ingredient.

During the public consultation (CP) no. 94 of 2015, referring to the Collegiate Board Resolution (RDC) proposal on the reassessment of the active ingredient Paraquat, a total of 2,879 contributions were received of which 39 were supported by files attached to the CP form (Table 1). Anvisa also received new documents filed by the Task Force referring to the CP that supported its argument and a document sent as an entity letter from abroad (Gremiagro), which was duly included electronically in the FormSUS. It was observed that 106 of these contributions agreed with the Resolution proposal, 57 partially agreed with the proposal and the other 2,716 contributions completely disagreed with the proposal or disagreed with the need for regulation.

Other contributions were received outside the deadline or mentioned in public consultations that occurred simultaneously with CP no. 94 of 2015. These contributions were not included in the CP results of Paraquat but their contents were addressed in this opinion since their arguments were very similar to that of the other contributions received.

Table 1. List of files attached to the contributions of the Public Consultation no. 94 dated October 8, 2015.

File	Topic
3655996_218117.pdf	Article on the appearance of glyphosate-resistant weeds.
3666610_218117.docx	Agronomic and economic importance - attached to the topics with the benefits of Paraquat.
3682503_218117.pdf	Agronomic and economic importance - attached to the topics with the benefits of Paraquat.
3688648_218117.pdf	Efficiency in the early harvest of corn.
3688648_218117.pdf	Letter to Anvisa from Fundação MS Para Pesquisa e Difusão de Tecnologias Agropecuárias [MS Foundation for Research and Diffusion of Agricultural Technology] – Technical arguments on the agricultural importance of Paraquat (weeds, handling of resistance, direct planting, harvest planning, environmental safety).
3688691_218117.pdf	Letter to Anvisa from Fundação MS Para Pesquisa e Difusão de Tecnologias Agropecuárias [MS Foundation for Research and Diffusion of Agricultural Technology] – Technical arguments on the agricultural importance of Paraquat (weeds, handling of resistance, direct planting, harvest planning, environmental safety).
3693722_218117.ods	Price of primary products – Out of scope.
3698496_218117.pdf	Agronomic and economic importance - attached to the topics with the benefits of Paraquat.
3700257_218117.pdf	Article on the impact of the prohibition of Paraquat in the suicide rates of South Korea.
3702195_218117.pdf	Agronomic and economic importance - attached to the topics with the benefits of Paraquat.
3703887_218117.docx	Toxicological aspects: Use of odorizing and emetic substances, herbicide applied in the direction of the soil, application only once or twice in the crop cycle, many times used in the mechanized form. The cases of death predominantly result from suicide and not related only to Paraquat. The distribution of Fuller's Earth and the training of medical doctors are important. Agronomic Aspects: Direct planting with reduced soil erosion, voluntary plants, lack of alternatives with similar characteristics.
3703923_218117.docx	Resistance and increased use of other herbicides.
3706548_218117.docx	Agronomic and economic importance - attached to the topics with the benefits of Paraquat.
3706762_218117.pdf	Agronomic and economic importance - attached to the topics with the benefits of Paraquat.
3709443_218117.pdf	Agronomic importance.



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3709559_218117.jpg	Photo of plant.
3711856_218117.docx	Importance of Paraquat in citrus crops.
3718279_218117.pdf	Direct planting, sustainable agriculture and use of Paraquat – masters thesis.
3721336_218117.pdf	Pack insert.
3724859_218117.pdf	Agronomic importance – Presentation, Fernando Adegas.
3726078_218117.docx	Data on medical care of cases of humans exposed to PARAQUAT PESTICIDE registered at the Toxicological Information Center of Santa Catarina (CIT/SC) in the period of 2003 to 2014.
3726398_218117.pdf	Document of the Brazilian Herbicide Resistance Action Committee (HRAC-BR) clarifying the importance of Paraquat in weed control, direct planting and resistance handling.
3726922_218117.docx	Agronomic and economic importance - attached to the topics with the benefits of Paraquat.
3727106_218117.docx	Agronomic and environmental importance.
3730205_218117.pdf	Minutes of the meeting of CONAPRE (Argentina) on the implications of the Rotterdam Convention on the use of Paraquat in Argentina.
3730600_218117.docx	Personal data – Out of scope.
3730672_218117.pdf	Document from Sungenta treatment of Paraquat intoxications. Main points addressed: lack of specific antidote, occupational exposure not resulting in systemic intoxication, medical treatment for acute intoxications is accepted by the medical community and is contained in the pack insert of the products, USEPA publications recognize this treatment.
3730840_218117.jpg	Personal photo – Out of scope.
3731564_218117.pdf	Agronomic importance, Ias [Healthy Eating Index] association.
3731608_218117.pdf	Out of scope – Article: Effect of the association of herbicide clomazone to chitosan-alginate nanospheres in the absorption in soils.
3731622_218117.xlsx	Intoxication data. Antonio Pedro University Hospital of the Fluminense Federal University.
3731739_218117.pdf	Document on behalf of the Brazilian Association of Toxicology Information Centers and Clinical Toxicologists (Abracit) presenting a summary of the major scientific evidences to support the removal of the Paraquat herbicide from the Brazilian market. Morbimortality data: Between the years 2007 and 2014, SUS [Unified Health System] registered 25,919 cases of intoxications by pesticides of agricultural use, of which 1283 of these cases resulted in the death of the intoxication victims. According to the data of the Brazilian Intoxication Registration System (Datatox), there were a total of 117 cases of Paraquat intoxication only in the year 2014 and 2015, of which 29 resulted in death. The document highlighted it as the potential chronic damage of the occupational exposure due to leading to the death of neurons through the induction of oxidative stress and reproduction of pathological characteristics of Parkinson's Disease in animal models. These damages would not be a real problem to farmers if the absorption of these agents in occupational circumstances were not plausible. However, there are references indicating high levels of Paraquat in exposed workers. It also highlights that a systematic review showed that the mortality of patients intoxicated by Paraquat was 85% and the use of cyclophosphamide reduced this mortality to 60%. In other words, even after receiving the best treatment, more than half of the people died from Paraquat intoxication. Reviews with positive results in the treatment of intoxications were cited but



	the lethality still remains very high (higher than 50%). It is worth highlighting that the effectiveness of this treatment in real scenarios would be very low since cyclophosphamide is a high cost chemotherapy drug that requires manipulation by a pharmacist in controlled chemical safety environments (laminar flow hood, sterile manipulation room with suitable exhaust and particle count). It also made reference to a study indicating that the number of suicides in South Korea reduced after the removal of Paraquat from the market; this information was considered important because it debunks that the removal of Paraquat from the market will have no impact on health. It was emphasized that it was necessary to evaluate the economic impact not only in relation to the increase in cost and loss of productivity in agriculture but the expenses with health and productivity loss of the intoxicated people must also be considered, citing Sri Lanka and China as countries where the banishment of Paraquat did not cause significant economic loss, despite being countries with intensive use of this pesticide and the damages from the permanence of Paraquat in the market exceeded the burden from its banishment.
3733582_218117.pdf	Review of the factors that place pollinators and the European agriculture at risk - Out of scope.
3733957_218117.pdf	Comments on epidemiological studies relating Paraquat to Parkinson's disease.
3735272_218117.docx	Analytic opinion on the Regulatory Rules (Economic Monitoring Department of the Ministry of Finance) indicating the lack of competitive impact.
3735905_218117.pdf	Embrapa Soja opinion on direct planting and agronomic importance.
3736418_218117.pdf	Addresses the impacts of the use of pesticides to health, citing the pesticide intoxication data registered at the Toxicological Information Center of Santa Catarina between the years 2003 and 2009 (discussed in the opinion).
3737088_218117.pdf	Bibliographic review on intoxication by Paraquat, Efficacy of various treatments with different approaches. It did not specifically talk about the penta conjugate treatment but addressed the reduction of mortality in treatments that are included in some of the steps of the penta conjugate treatment protocol. It highlighted the best survival rates in the use of immunosuppressants, antioxidants, hemoperfusion, methylprednisolone, vitamins B and C, propfol, vitamin E.
3737168_218117.pdf	List of substitutes.

Among the contributions received that disagreed with the proposal or the need for regulation, majority presented arguments related to the agronomic, environmental and economic importance of this active ingredient and the losses of its removal from the market: lack of substitute, resistance, impossibility of direct planting, difficulty in pest control, economic loss to the country, productivity loss, increased production costs, increased use of other types of herbicides, difficulty in soil preservation, and negative impacts on neighboring countries. Some of these contributions suggest reconciliation between maintenance of the Paraquat-based products in the market and mitigation of the health risks, proposing some actions that can be used to achieve this objective:

- Conduct risk assessment;
- Assessment of the possibility of partial cancellation of the registration only for crops with higher risk of contamination of the applicator (for example, perennial crops and crops that require manual application);
- Intensification of the inspection of the sale and use of the product;
- Maintenance of the registration until the existence of substitutes;
- Incentive to register substitutes;



- f) Assessment of the efficiency of possible substitutes;
- g) Improvement in the monitoring of its use and residues;
- h) Intensification of the training of manipulators of the products;
- i) Accreditation of applicators;
- j) Restriction of use (packaging, sites permitted for aerial application, back spraying, maintenance of only mechanized application);

The importance of conducting a more in-depth regulatory impact analysis was also mentioned in the CP. It is worth highlighting that few contributions presented arguments related to the analyses of the effects of Paraquat to health.

In addition to the CP, the good regulatory practices can contemplate additional steps like the complementary instruction in which, among other options, panels can be performed to allow the in-depth analysis with assessment of additional information that can support the final decision. In the reassessment of the active ingredient Paraquat, based on the importance of the RDC proposal resulting from the reassessment of this active ingredient, the need to expand the debate on the contributions received was observed and hence a Technical Panel with experts was organized after the CP.

The Technical Reassessment Panel of the active ingredient Paraquat was planned to deepen the discussions on occupational exposure, mutagenicity and parkinsonism, toxicological aspects prohibitive for registration considered relevant in the reassessment of Paraquat, with the main purpose of detailing the arguments presented in the CP to support the consolidation of the understanding of Anvisa and the elaboration of a final decision on the reassessment of Paraquat. Furthermore, the analysis of the contributions of the CP showed the need to expand the debate on the impacts of the results of the reassessment of Paraquat in agriculture, in the economy and in the environment.

The following experts in the areas of toxicology and agronomy affiliated with institutions of reference were invited to the panel to discuss all these aspects:

- a) Estefânia Moreira, researcher of the Londrina State University;
- b) Ligia Fruchtgarten, physician of the São Paulo Intoxication Control Center;
- c) Fernando Adegas, researcher of Embrapa;
- d) Hamilton Ramos, researcher of the Agronomic Institute of Campinas.

Representatives of the Paraquat Task Force and other bodies involved in the reassessment of pesticides (Ministry of Agriculture, Livestock and Supply (MAPA) and Brazilian Institute of Environment and Renewable Natural Resources (IBAMA)) were also invited.

After the analyses of the contributions and with the discussions in the panel of experts on the reassessment of Paraquat, some aspects of the Reassessment Opinion no. 1/GGTOX/Anvisa were reviewed and will be presented and discussed below.

1. International Status

After the CP, the table with the international status of the technical reassessment opinion was updated (table 1).

Table 1. International status of paraquat.

Country	Regulatory status
United States	Restricted use. The application should be monitored by a certified applicator. Application close to schools and homes, in parks, golf courses and playgrounds is prohibited. Re-entry interval of 12 to 24 hours. Aerial application allowed. In March 2, 2016, EPA initiated a public consultation on the proposal of even more restrictive measures after concluding that the intoxication data were disturbing. The measures proposed were: packaging in closed systems that allowed the direct transfer to the application equipment and that cannot be opened without destroying the packaging; special training for certified applicators, emphasizing that transfer of container and inappropriate storage cannot occur; changes in the label and distribution of supplementary material, with warnings to emphasize the



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	toxicity and associated risks of Paraquat-based products; prohibition of manual and back application; use restricted to certified applicators, that is, individual workers under the supervision of applicators will be prohibited.
Canada	Restricted use. Aerial application allowed. Reassessment concluded in December 2015. Restrictions adopted: classified in the restricted class, application only by certified or licensed applicators, addition of IPEs for back and tractor-driven application, inclusion of specifications for IPEs and tractor cabins, inclusion of warnings, medical cares and additional precautions in the pack insert, reduction of Paraquat concentration in the end-use product, use of the same mixing tank only for other pesticides specified in the pack insert, modification of the packaging to facilitate volume measurement without having to transfer the liquid to another container, implementation of handling program by the company.
Australia	Restricted use. Only terrestrial application. Under reassessment.
New Zealand	Restricted use. Only terrestrial application.
European Union	Prohibited since 2009, but the import of products with residues of this herbicide is allowed.
Switzerland	Prohibited since 1983.
Norway	Prohibited. Voluntary cancellation of the products.
Japan	Restriction in the concentration of the product.
Bosnia-Herzegovina	Prohibited.
Indonesia	Use for certain crops only by professionals with special permission.
Kuwait	Prohibited since 1985.
Cambodia	Prohibited since 2003.
Laos	Prohibited since 2010.
Arab Emirates	Prohibited since 2005.
Syria	Prohibited since 2005.
South Korea	Prohibited since 2012.
Philippines	Restricted use.
China	Prohibited. Cancellation of the registrations and production in 2014. Production allowed only for export. Use permitted until 2016.
Sri Lanka	Prohibited.
Columbia	Aerial application is prohibited.
Uruguay	Restricted use. Only products with packages of 1 to 30 liters and with dye are allowed.
Belize	Restricted use. Only terrestrial application.
Chile	Restricted use. Aerial application is prohibited.
Costa Rica	Restricted use. Aerial application is prohibited.
Jamaica	Restricted sale.
Ivory Coast	Prohibited since 2004.
Cape Verde	Prohibited since 2011.
Senegal	Prohibited since 2011.
Nigeria	Prohibited since 2011.
Mauritania	Prohibited since 2011.
Mali	Prohibited since 2011.
Republic of Chad	Prohibited since 2011.
Burkina Faso	Prohibited since 2007.
Guinea Bissau	Prohibited since 2011.
Gambia	Prohibited since 2011.
Malaysia	After the period of restricted use, there is currently registration only for formulations with concentrations of 100 g/L

Restricted use: use is allowed but with restrictions (limited concentration of the active ingredient, exclusively terrestrial application, application exclusively by accredited persons, among others).



Paraquat was prohibited in the entire European Union (EU) after judicial determination that contested the decision of the reassessment by the European Commission in maintaining the use of this active ingredient (UE, 2007). The reasons that led the Court to remove Paraquat from the list of authorized products were:

- 1) Existence of studies in scientific literature relating Paraquat to Parkinson's disease that were not considered in the reassessment;
- 2) The reassessment concluded that there was no neurotoxicity but did not satisfy all the necessary process requirements;
- 3) There was no assessment of a French study on the occupational exposure considered relevant;
- 4) In a Guatemalan study, the exposure of one of the operators was 118% above the permitted level, although he was using the product in the appropriate conditions;
- 5) The French study used as basis to allow the use of Paraquat, in its conclusions, warnings about the use of back sprayers in the application of products, reinforcing the doubts regarding the conclusion of harmlessness of the use of Paraquat-based products.

Although the use of Paraquat is prohibited in the EU, the export of products to the EU with the same maximum levels of residues considered acceptable before the decision to not include this active ingredient in the list of authorized products.

According to the contributions from Syngenta, representative of the Paraquat Task Force, products made with this active ingredient are registered and sold in various developed and developing countries around the world, including in major agricultural markets under demanding regulatory systems like the USA, Canada, Australia, Japan and New Zealand.

According to Syngenta, Paraquat is currently registered and sold in the following countries: Angola, Honduras, Peru, Antigua and Barbuda, India, Philippines, Argentina, Indonesia, Rwanda, Australia, Iran, Samoa, Bahamas, Iraq, São Tomé e Príncipe, Bangladesh, Israel, Singapore, Barbados, Jamaica, Solomon Islands, Belize, Japan, South Africa, Bolivia, Kenya, Saint Christopher and Nevis, Brazil, Kiribati, Saint Lucia, Cameroon, Madagascar, Saint Vincent and the Grenadines, Canada, Malawi, Sudan, Chile, Malaysia, Suriname, Marshall Islands, Tahiti, Colombia, Mauritius, Taiwan, Cook Islands, Mexico, Tanzania, Costa Rica, Morocco, Thailand, Cuba, Myanmar, Tonga, Dominica, Namibia, Trinidad and Tobago, Dominican Republic, Nauru, Turkey, Ecuador, New Zealand, Tuvalu, El Salvador, Nicaragua, Uganda, Ethiopia, Nigeria, Uruguay, Fiji-Niue, USA, Grenada, Pakistan, Vanuatu, Guatemala, Panama, Venezuela, Guyana, Papua New Guinea, Vietnam, Haiti, Paraguay, Zambia.

2. National Status

According to the contributions made by the Ministry of Agriculture, Livestock and Supply (MAPA), Paraquat is registered for weed control in pineapple, cotton, rice, banana, potato, coffee, sugarcane, citrus, kale, beans, apple, corn, rubber tree, soybean and wheat crops and it is also authorized as a drying agent for cotton, rice, potato, sugarcane, corn and soybean. In addition to these crops, there is a monograph for avocado, asparagus, beet, cocoa, coconut, pasture, pear, peach, sorghum (post-emergency and drying agent). Fifteen Paraquat-based technical products and twelve formulated products are registered as observed in tables 2 and 3.

Table 2. List of Paraquat-based technical products registered in Brazil.

Trademark	Registration Owner	Registration
Alamos Paraquat Technical	ALAMOS DO BRASIL	8314
CHN Paraquat Technical	ALLIERBRASIL AGRO LTDA.	11312
Helm Paraquat Technical	HELM DO BRASIL MERCANTIL LTDA	3808
Rainbow Paraquat Technical	RAINBOW DEFENSIVOS AGRÍCOLAS LTDA.	2513
Sinon Paraquat Technical	SINON DO BRASIL LTDA.	07805
Zeneca Paraquat Technical	SYNGENTA PROTEÇÃO DE CULTIVOS LTDA.	0678498
Paraquat Technical 500	STOCKTON - AGRIMOR DO BRASIL LTDA.	2108
Paraquat Technical ZY	ALLIERBRASIL AGRO LTDA.	11712



China Paraquat Technical	ALLIERBRASIL AGRO LTDA.	11212
Syngenta Paraquat Technical	SYNGENTA PROTEÇÃO DE CULTIVOS LTDA.	014507
Paraquat Technical Yn	ALLIERBRASIL AGRO LTDA.	11112
Ouro Fino Technical DCP	OUROFINO QUÍMICA LTDA	9814
Alta Paraquat Dichloride Technical	ALTA – AMÉRICA LATINA TECNOLOGIA AGRÍCOLA LTDA.	12414
Nufarm Paraquat Technical	NUFARM INDÚSTRIA QUÍMICA E FARMACÊUTICA S.A.	8914
Atanor Paraquat Technical	ATANOR DO BRASIL LTDA.	9214
CCAB II Paraquat Technical	CCAB AGRO S.A.	6916
CCAB Paraquat Technical	CCAB AGRO S.A.	13315

Table 3. List of Paraquat-based formulated products registered in Brazil.

Trademark	Registration Owner	Registration
Laredo	HELM DO BRASIL MERCANTIL LTDA	13309
Orbit	SINON DO BRASIL LTDA.	2010
Tocha	STOCKTON - AGRIMOR DO BRASIL LTDA.	13208
Gramocil	SYNGENTA PROTEÇÃO DE CULTIVOS LTDA.	1248498
Gramoxone 200	SYNGENTA PROTEÇÃO DE CULTIVOS LTDA.	1518498
Helmozone	HELM DO BRASIL MERCANTIL LTDA	14908
Paradox	SINON DO BRASIL LTDA.	5006
Pramato	AGROLI INDÚSTRIA QUÍMICA LTDA.	396
Gramoking	RAINBOW DEFENSIVOS AGRÍCOLAS LTDA	6115
Quatdown	RAINBOW DEFENSIVOS AGRÍCOLAS LTDA	5815
Sprayquat	RAINBOW DEFENSIVOS AGRÍCOLAS LTDA	5915
Nuquat	NUFARM INDÚSTRIA QUÍMICA E FARMACÊUTICA S.A.	1216
Flak 200	CROPChem LTDA	11415

* Access to Agrofit (http://agrofit.agricultura.gov.br/agrofit/principal_agrofit) on 06/08/2016.

The contributions made by MAPA in the CP no. 94 of 2015 are presented below and allows a deepening of the discussion on the existence of substitutes and on the importance of Paraquat-based products in Brazil. These contributions were made based on the technical positions of Embrapa and other researchers that considered the resistance aspect of weeds and maintenance of direct planting as relevant to justify the maintenance of the Paraquat registration.

According to the Researcher Fernando Adegas (Embrapa Soja), Paraquat is important for the stability of the agricultural reproduction system, which may be put at risk with the banishment. The use of Paraquat-based products occurs mainly on soy crops, performed in eight Brazilian states by medium-sized and big farmers that use advanced production systems and with high technology. There are still no plants resistant to Paraquat in Brazil and this has allowed it use as an alternative to fighting against Glyphosate-resistant weeds. The Glyphosate-resistant weeds are present in over 15 million hectares of agricultural area and distributed among the main Brazilian agricultural producing states: they have already reached the states in the South (RS, SC, PR), Southeast (SP, MG), Central West (MS, MT, GO, TO), Northeast (BA, PI) and North (MA), which is considered very disturbing to MAPA.

A registered company sent to MAPA a study conducted by Agricultural Engineer Prof. Dr. Alexandre Medonça de Barros (consultant of MB Agro and professor at the School of Economics of the Getúlio Vargas Foundation) in 2015 on the impact in the direct losses and in the production chain of soybean. This same study was sent to Anvisa during the Public Consultation made by the Paraquat Task Force. The major losses mentioned by this study were:

- Deceleration of the productivity gains of soybean crop in Brazil.
- Limitations to the use of transgenic crops tolerant to glyphosate (all the current varieties of transgenic soybean).
- Limitations to the adoption of direct planting.
- Increase in the use of herbicides and operating cost of its application.
- Reduction of the production value and profitability of the farmer.
- Negative effects on the socioeconomic conditions of the population.



- g) Reduction of tax collection.
- h) Reduction of exports and lowest increase of the commercial balance of the country.
- i) Increase in the environmental impact of soybean crop with the reduction of direct planting area and increased use of herbicides and machinery.
- j) Low soil protection (increased erosion) and increased emission of carbon due to the reduced direct planting area.
- k) Low planting speed, hindering the use of the best soybean sowing season and delaying the planting of second corn crop (associated to higher climatic risk).
- l) Reduction of the competitiveness of the Brazilian soybean farmer in relation to farmers of other countries, with possible reduction of the Brazilian share in the global production.
- m) Low food supply with increase in the production costs of meat and oils.

This same study showed that the potential impacts on the economy are significant, able to reach a reduction of about R\$27 billion in Gross Value of Production (GVP), with loss of two million jobs and reductions of R\$25 billion in labor income and R\$4.7 billion in tax collection (Barros and Hausknecht, 2015).

The direct planting technique is considered fundamental by MAPA and the Task Force for the country's agriculture and an excellent substitute of the traditional farming system because it is based on the principles of sustainability with social, economic and environmental dimensions, integrating productivity increase, reduced use of fossil fuels, increased atmospheric carbon sequestration and reduced soil erosion (Freitas and Landers, 2014). Other advantages to farmers and to the environment were mentioned in the CP and were included in table 5.

Table 5. Advantages in the use of Paraquat to the environment and farmers presented by MAPA in the CP, according to the study of Barros and Hausknecht (2015).

Advantages to farmers	Advantages to the environment
Efficient control of weeds.	Reduces soil erosion.
Rapid action.	Increases organic matter in the soil.
Safe for use on registered crops.	Reduces the emission of carbon gas.
Allows more than one crop per crop season.	Increases soil biodiversity.
Protects soil fertility.	Aids in preserving the quality of the water.
Increases productivity.	Avoids the use of harrowing and plowing.
Prevents workers from performing manual weeding.	
Higher quality of life to the worker.	

MAPA and Task Force also mentioned the Program for Reduction of Greenhouse Gases in Agriculture (ABC Program) as the main strategy for the mitigation and adaptation to climate changes for the sustainable development in agriculture, whose work was coordinated by the Presidential Staff, by MAPA and by the Ministry of Agrarian Development (MDA), with active participation and representation of the civil society. The objectives are to reduce the emission of greenhouse gases originating from agricultural activities with the following actions: direct planting system; reduction of deforestation; increase of agricultural production on sustainable bases, crop-livestock-forest integration, animal waste treatment; adaptation of rural properties to environmental legislation; expansion of cultivated forest area; and stimulation to the recovery of degraded areas.

The ABC Program is funding the adoption of direct planting in 8 million hectares until 2020. Seventy percent of the grain production area (83 million hectares in 2030) should adopt the direct planting system and prioritize the use of varieties and crops that perform biological nitrogen fixation.

The Task Force also reported in its contributions some advantages of using Paraquat in direct planting:

- a) Increase of the organic matter in soil, with consequent reduction of carbon in the atmosphere;
- b) Reduction of carbon gas in the atmosphere (sequestration of 0.15 to 1.7 tons of carbon/ha/year);
- c) Increase in productivity;



- d) Reduction of the use of diesel oil and consequent release of greenhouse gases;
- e) Reduction of erosion with consequent reduction of the contamination of water bodies and silting;
- f) Reduction of water loss in the soil;
- g) Greater use of nutrients by plants.

MAPA emphasized that it, Anvisa and Ibama, as competent federal authorities in the assessment of pesticides and similar products, have the important role in the society of reaching a balanced decision that provides the farmer with tools to produce while seeking food safety and protecting human health and the environment, based on the best science available internationally and using the risk assessment considering the Brazilian reality, always promoting good agricultural practices, as well as the responsible and safe use of these inputs. MAPA believes that Paraquat is a very important product to the Brazilian agriculture, not only in the control of infesting plants but for it being a tool with unique and fundamental characteristics for direct planting, aiming at environmental preservation with the control of soil erosion and avoiding loss of fertile soil and silting of lakes and rivers.

MAPA also clarified that the relevance of Paraquat to direct planting supported the commitments established by the Presidency at the United Nations Framework Convention on Climate Change (COP-21) with the commitment of greenhouse gas reduction. Among the proposed actions is the expansion of the use of Direct Planting System ratified in article 12 of Act 12.187 of October 2009, which establishes the National Policy on Climate Change.

In view of the importance of Paraquat to agriculture in Brazil, the suggestion made by MAPA in the CP was to proceed with the occupational risk assessment to identify the scenarios with unacceptable risks and apply the mitigation measures based on these results. This aspect will be discussed in the topics below because the relevance of this assessment should be made specifically for each toxicological aspect.

Like MAPA, the Brazilian Confederation of Agricultural and Livestock (CNA) recognizes the importance of Paraquat to the Brazilian agribusiness for being used on crops of high relevance to the Brazilian economy and for being responsible for the success of the Direct Planting System, a practice that reduces the emission of carbon in agriculture. As a total-action herbicide, it is one of the main options for handling the pre-seeding operation of plantations performed in direct planting, being an important product for the maintenance and expansion of this planting system in Brazil. Direct planting is a practice that revolutionized the Brazilian agriculture by proposing a more conservational farming that drastically reduces soil erosion compared to the conventional farming.

At the 15th Conference of the Parties, of the United Nations Framework Convention on Climate Change (UNFCCC), the Brazilian government disclosed the voluntary commitment to reduce the emissions of greenhouse gases by 2020. One of the proposed actions is the expansion of the use of direct planting in straw. Paraquat has a broad action spectrum and therefore acts rapidly and in different climate conditions as is the case of Brazil. In the contributions, the importance of this pesticide in locations with plenty of rain was highlighted. It also highlighted the need to not base the prohibition of this active ingredient on decisions made by other countries since Brazil has edaphoclimatic, soil and weed particularities, with the lack of alternatives as efficient as Paraquat to avoid losses. Brazil is a country with different climatic conditions and there is the need for a wider diversity of alternatives for pest control.

According to the report cited by MAPA and the Task Force in their contributions on the biological and economic aspects of the use of Paraquat in Brazil (Adegas and Osipe, 2015), the prohibition of this active ingredient in the utilization modalities will cause farmers to have an average annual increase of R\$ 407,672,429.53, representing an expenditure of 129.25% higher than the annual cost with the use of Paraquat due to the need to use alternative herbicides for drying, perennial crops and direct planting (Carfentrazone, Chlorimuron, diquat, 2,4-D, flumioxazin, glyphosate, ammonium glufosinate, saflufenacil, ametryn, bromacil+diuron, flazasulfuron, among others). Regarding the number presented in the report, it is worth highlighting that there are some errors in the calculations made, where the expenditure increase would be about 100% of the current cost.



Although the calculations overestimated the percentage increase of the average production cost with the use of other herbicides, the value of 100% continues to be high and represents almost double the exposure in the case of cancellation of the registration of Paraquat-based products. These values were obtained from the mean value of different substitute herbicides, thus the choice of cheaper products does not represent a very significant increase in costs. Furthermore, this study considered crops with permission in the monograph but for which the Task Force favorably manifested the cancellation in order to reduce the cases of acute intoxication. The Task Force represented by Syngenta emphasizes its contribution that, during the years, various crops were being removed to reduce the intoxications of small farmers. The Task Force also proposed the discontinuation of various crops to minimize the access to Paraquat and proposed maintaining the use of the herbicide only in cotton, rice, banana, potato, coffee, sugarcane, citrus, beans, corn, soybean and wheat crops and as a drying agent of sugarcane, soybean and potato. However, various crops removed by Syngenta still have established use in the monograph, requiring the review of their permanence since the Task Force warned about the need for their discontinuation.

Diverging from the proposal of the Task Force in the CP to withdraw the use for some crops, MAPA highlighted the importance of Paraquat to small farmers, stating that there was no alternative with the same efficiency and cost.

The Task Force also presented a study on the economic impact of the registration cancellation of Paraquat-based products in Australia, considering the different scenarios. This Australian study is more competent than the Brazilian report that was presented, but both studies have converging results and foresee an increase in production cost, increase in the resistance to Glyphosate, loss of productivity, reduction of production yield, unemployment, reduction in export, increase in expenditure with herbicides and manual labor and loss of soil structure.

The presentations of the Task Force, MAPA and the Embrapa researcher Dr. Fernando Adegas at the panel of experts observed all these arguments used in the contributions in CP no. 94 of 2015, reaffirming the agronomic and economic importance of Paraquat to Brazil and discussing some exposure mitigation measures. In the Panel, in addition to the previously presented measures, the Task Force only added the proposal of intensifying the training projects of the health professionals, the industry and the applicator.

MAPA also indicated that the removal of Paraquat from the market will compromise the competitiveness of the Brazilian agriculture. Among the main agricultural benefits of the use of paraquat, MAPA cited the following:

- a) Conservation of soil humidity, erosion reduction and consequent improvement in its nutrition and structure;
- b) Early harvest with better use of the land and greater yield to the farmer;
- c) Use of the ideal planting window for the subsequent crops, resulting in greater productivity;
- d) Sustainable handling of tolerant and resistant weeds;
- e) Preservation of the direct planting system and low carbon agriculture, reduction of risks and increase in income to the farmer;
- f) Competitiveness of the Brazilian agriculture and livestock production in the international market.

Dr. Fernando Adegas especially highlighted the importance of Paraquat due to the need for diversification of herbicides in chemical handling, concluding that:

- a) Paraquat is an important alternative to the handling of weeds resistant to other herbicides, especially to Glyphosate;
- b) As a total-action herbicide, Paraquat is one of the main options for handling the pre-seeding in direct planting, being important for the maintenance and expansion of this planting system in Brazil;
- c) There is no specific substitute for Paraquat, which is why the handling of weeds will be compromised by the non-use of the product, with likely increase in the quantity of herbicides in the systems where it is currently being used;
- d) The problems with voluntary plants should increase with the increase of crops resistant to herbicides. Even though Paraquat is registered for this purpose, it is one of the most promising products in the control of these infesting plants.



It can be observed that the discussion on the prohibition of Paraquat is very complex and deserves attention, because many contributions highlighted the problems that would be faced with the cancellation of the registration of Paraquat-based products related to the agronomic impacts (lack of substitutes, problems with weed resistance, difficulty in pest control, increased use of other herbicides), economic impacts (loss of productivity, increased production cost, negative impacts on neighboring countries) and environmental impacts (impossibility of direct planting, increased soil degradation, increased emission of CO₂). Furthermore, the need for reconciliation between the permanence of Paraquat-based products in the market and the mitigation of the health risks was constantly emphasized. The main measures proposed for this reconciliation to be possible were:

- a) Risk assessment;
- b) Intensification of the inspection of sales and use;
- c) Monitoring the use;
- d) Intensification of the monitoring of the wastes;
- e) Maintenance of the registration until the existence of substitutes;
- f) Incentive to register substitutes;
- g) Sale with electronic agronomic prescription control linked to pre-application and post-application assessment reports;
- h) Assessment of the efficiency of possible substitutes;
- i) Training, qualification and orientation programs on the use;
- j) Accreditation of applicators;
- k) Prevention of the development of resistance with the implementation of alternative handling;
- l) Restriction of the use (perennial crops, packages of 1L, aerial application, back spraying, maintenance of only mechanized application, maintenance only for pre-planting of annual crops, limitation of the quantity of product use to one or two crop seasons);
- m) Review of the pack insert, with inclusion of relevant information;
- n) Training of health professionals;
- o) Restriction of the sale to rural farmers;
- p) Monitoring of acute intoxications;
- q) After sales follow-up by the company to monitor the handling;
- r) In-depth analysis of the regulatory impact.

1. Acute Toxicity

In various contributions of CP no. 94 of 2015, a review of the cases of acute intoxication by Paraquat was requested, indicating the need for analysis of the relevance of uncommon accidental cases where Paraquat was used for a non-allowed purpose, such as to kill lice, or in cases of intoxication that occurred in unforeseen situations, such as a fallen airplane and the presence of fire during application of the product. Just like the cases of intoxication resulting from voluntary ingestion, these uncommon accidental cases occur from the incorrect use of the product or from unforeseen accidents and, therefore, cannot be determining factors for the banishment of the active ingredient, but should be the focus of preventive actions.

Besides pesticides, many other products can be used incorrectly and cause irreversible consequences to health, but this is not sufficient reason for them to be prohibited. Generally, many chemical products, when used incorrectly, enable the appearance of severe clinical manifestations different from the clinical effects expected from the suitable occupational use. Therefore, the concern should be on the need to prevent the accidental cases, voluntary ingestion and incorrect use.

When the occupational intoxications are analyzed, it is observed that there are cases of severe acute intoxication in exposed workers but they are mostly due to incorrect use: failure to use individual protection equipment, suction of the pesticide with the mouth, prolonged use of clothes with spilled Paraquat, failure to wash the body after dermal contact with the pesticide, use of the product even after the occurrence of dermal injury.

The issue to be raised is whether there are efficient ways of preventing acute intoxications resulting from misuse, since we know the Brazilian reality and know that the



adhesion to the use of IPE is extremely low and that applicators in general have low instruction and knowledge. In view of the serious complications that the use of Paraquat represents both from occupational exposure and from accidents and voluntary ingestions, it would be essential, in case it is maintained, to elaborate an acute intoxication prevention plan with monitoring mechanisms that is able to confirm that this plan is successful.

The contributions received in the Public Consult no. 94 of 2015 allow the assessment of the national scenario in relation to intoxications by Paraquat in some locations in Brazil. According to the data of the Toxicology Information Center of Santa Catarina (CIT/SC), sent with the contribution in the CP and referenced in the works of the University of Santa Catarina (Santello, 2009), it was observed that the intoxication by pesticides from 2003 to 2014 corresponded to about 6% of the cases of intoxication at CIT/SC, of which only 3.9% of these occurred due to exposure to Paraquat. The number of cases of intoxication by Paraquat is relatively small; however, from the total number of deaths by pesticides, 29% occurred from intoxication by Paraquat. The lethality of Paraquat is high since 23% of the cases of intoxication resulted in death.

This condition is more disturbing when only cases of suicide attempts are considered. It is observed that between 2003 and 2014, a total of 111 cases of suicide attempts by Paraquat were attended to at CIT/SC, of which 48% resulted in death, that is, the lethality of Paraquat significantly increases when only the suicide data is used. From the total of 58 cases of death by Paraquat intoxication, 53 cases occurred from suicide attempts, one due to an abortion attempt, two cases were accidental and one case had its cause ignored. In other words, during this period there was no record of any clearly occupational case that resulted in death and, as explained by Santello (2009), all the occupational cases were cured.

In the Technical Reassessment Panel for Paraquat, Dr. Lígia Fruchtengarten presented important data from the Intoxication Control Center of São Paulo (CCI/SP) of 2015, showing that from the 7,273 registered cases of intoxication only 1.04% was related to the use of agricultural pesticide. From 2006 to 2015, the intoxications by Paraquat corresponded to about 5% of the total cases of intoxication by agricultural pesticides (65 cases). Among the intoxications by Paraquat, 43 cases were suicide attempts (65%), 11 cases were occupational exposure (16%) and 8 cases were accidental (14%). The other three cases were of unknown cause. From these 65 cases of intoxication by Paraquat, twelve resulted in death and three patients recovered completely. The other 50 cases had unknown prognostics. The CCI/SP data showed that there was a very similar standard in the cases of intoxication by Paraquat in all the CITs, of which lack of data and low notification is a general problem that significantly compromises this assessment.

Dr. Lígia Fruchtengarten also presented some data from DATATOX, which shows the occurrence of 50 cases of intoxication by Paraquat in 2014 and 83 cases in 2015. From the cases of 2015, eleven were accidental (13%), 23 were occupational (28%) and 49 were suicide attempts (59%). From these 83 cases, 16 resulted in death, all from suicide attempts. There was only one case of severe intoxication after occupational exposure. The other accidental and occupational cases resulted in mild to moderate intoxication.

The contribution received by the coordinator of the Intoxication control center of the Antônio Pedro University Hospital of the Fluminense Federal University also showed that the severe intoxication by Paraquat is more common in suicide cases. Between January 2009 and November 2015, ten cases of intoxication by Paraquat were notified out of a total of 7,065 cases (6.8%). Six cases were suicide attempts, two cases were accidents involving children and two were occupational accidents. Three of these cases resulted in death, all from suicide, but one was likely associated to cocaine overdose (patient presented sudden cardiac arrest in less than 24 hours after exposure). This contribution highlighted that the kerosene sold without any restriction also caused death by accidental ingestion of two children, Bupropion was responsible for three suicidal deaths and the number of deaths by other pesticides was five, all suicidal, including some with low toxicity. Furthermore, lead was responsible for eight deaths (one accidental case, one homicidal and six suicidal), in other words, despite the cancellation of the registration of Aldicarb-based products, other substances were incorporated in the lead, which continues to be the means of committing suicide.

As can be observed in the presented data and as discussed by Dr. Lígia Fruchtengarten in her contribution to CP no. 94 of 2015, and in her presentation at the



Technical Reassessment Panel for Paraquat, the severe cases of intoxication by Paraquat notified and monitored at the intoxication centers generally result from the ingestion of Paraquat-based products in suicide attempts, for which there is really no specific antidote.

In view of the high number of suicidal deaths by Paraquat ingestion, various contributions received during CP no. 94 of 2015 considered it a priority to establish suitable strategies to avoid this type of death. The major strategy mentioned as efficient was the reduction of the access to this pesticide since changes in the concentration of addition of emetics has not proven to be efficient in reducing the deaths after ingestion. However, the intoxication data presented by the Intoxication Centers in CP no. 94 of 2015 showed how incoherent it would be to prohibit the sale of Paraquat exclusively due to the fact that it is associated to suicide attempts, because it is event that other pesticides or even other types of products are as important as it in the number of death cases and measures are not taken to prohibit their sale.

As explained by Dr. Lígia Fruchtingarten in her contribution to CP no. 94 of 2015 and in the Technical Reassessment Panel for Paraquat, the discussion on suicide should be addressed more comprehensively by the health system and should involve other aspects, including the social problems. It is also necessary to consider that the banishment may not improve the death rates by suicide, only force the use of another means. In other words, before making an extremely restrictive decision, it is necessary to create effective strategies in the prevention of cases of intoxication by Paraquat based on methodologies supported by internationally respected entities like the World Health Organization (WHO). The World Health Organization has a suicide prevention guide that indicates which alternatives should be adopted to reduce the cases of deaths. These alternatives will be discussed later in this opinion.

The participants of CP no. 94 of 2015 also showed concern with the lack of efficiency of the treatments for intoxications by Paraquat and with the lack of scientific background of the protocol proposed by the Task Force. Regarding the proposed protocol, it is worth highlighting that there is actually a scientific background but there is no consensus regarding its efficiency due to the shortage of data available. On the other hand, various contributions mentioned the need to standardize the treatment protocol in order to obtain greater efficiency with the use of measures that are newer and with greater treatment potential than some interventions currently performed, and also because a single guideline will allow the measurement of results in a more uniform way. The need for standardization and measurement of the efficacy of this protocol was addressed both in the CP and at the panel of experts.

With the contributions of the CP and with the discussion promoted at the Technical Reassessment Panel for Paraquat, it is observed that various isolated proposals have been made during the last decades for the treatment of intoxication by Paraquat, with the aim to reduce the absorption and impact of tissue lesion, especially the progressive pulmonary fibrosis. The elimination measure with extracorporeal removal by hemodialysis also proved to be efficient, but only if performed very early during the first hours after ingestion, which is not compatible with the reality of our health system. In general, there is a consensus on the great challenge of the treatment of severe intoxications by Paraquat and the need to try and improve the prognostics of the patients. Regarding this necessity, the reviews of the most recent literature indicate a possible treatment with positive results in the associated use of corticoids, immunosuppressants and N-acetylcysteine, but there is still the need for analysis of more results to conclude its efficiency. Protocol based on these evidences was proposed by the Task Force for Paraquat to be implemented as standard in the treatment of intoxications because it could be of greater contribution in the recovery attempt of patients than the practices adopted without any systematization.

In the Technical Reassessment Panel for Paraquat, Dr. Lígia and Dr. Sérgio Graff also highlighted the existence of few specific antidotes in the treatment of majority of the severe intoxications, such as the case of Paraquat. Even when available, the access in Brazil is difficult. The alternative in majority of the cases is a suitable support and decontamination treatment. Therefore, the penta-conjugate treatment protocol proposed by the Task Force may bring positive results, better than the current results, in the treatment of intoxicated patients. Furthermore, it was discussed that the local intervention for the treatment of workers and health professionals that treat these patients is of extreme importance to reduce the risk of exposure and improve the quality of the treatment.



Regarding the problems faced in the treatment of severe intoxications by Paraquat, Dr. Lúcia Fruchtgarten emphasized that there is considerable lack of knowledge by the health professionals regarding the severity of the intoxication and the need for early treatment in the intoxications by Paraquat, requiring the training of health professionals; there is also difficulty in the transfer of patients to hospitals of greater complexity and Specialized Dialysis Units. There is also low local availability of the qualitative test for confirmation of the diagnosis and the serum level for quantitative control of the intoxication cases, as well as lack of activated charcoal and medication suggested in the conjugate treatment at the health facilities (immunosuppressants, antioxidants and N-acetylcysteine). Other challenges for the assessment of the success of this protocol are the need to monitor the treatment until the final evolution of the cases of intoxication and the fact that there is still insufficient number of cases to assess the results of the proposed treatment.

The existence of an antidote or efficient treatment for acute intoxications by Paraquat is not considered. The clinical protocol presented by the Task Force cannot be considered efficient because it is not capable of reversing the intoxication condition of a patient to an acceptable healthy condition, and there is no reliable data on the reduction of the mortality of intoxicated patients. According to the report filed by the Task Force on June 29, 2015, the high mortality resulting from intoxications by Paraquat associated to the fact that there is no specific antidote or treatment is admitted, showing the need to seek for a more efficient treatment, which would be this proposed protocol. The treatment addressed in the report does not present any innovation in relation to what is already known in scientific literature, it only proposes a new approach, seeking to concomitantly treat or prevent the symptoms of various actions of Paraquat in the body. However, this protocol has not generated a treatment that can guarantee that the intoxicated patient will have real and great chances of recovery and, meanwhile, it is not considered an efficient treatment to revert the clinical condition of severe intoxication. If the broad action antidotes were sufficient to revert the severe intoxications by Paraquat it would have then been possible to consider that an efficient antidote or treatment existed.

In the contributions, many argued about the concept of antidote, but it would not be plausible for the entity responsible for the health of the population to accept such a broad concept of antidote for intoxications by pesticides because people's lives must be preserved. An antidote or treatment can only be considered efficient when it is able to revert the prognostics of death for at least majority of the cases of intoxication. Efficient treatment is that which favorably influences in the recovery of the patient, producing desired effects and, in the case of a severe intoxication such as one resulting from Paraquat ingestion and that leads to death, the desired effect is for at least majority of the cases to be reverted and for patients to not die or have severe and disabling aftereffects, reducing the quality of life.

Despite the shortage of specific antidotes, in general, the prognostics of intoxications by Paraquat improved considerably with the advances in the other treatment phases. However, severe intoxication by Paraquat is highly lethal even when these treatments are used. In the contributions of the Task Force, this concern is clear when it is said that "the systemic symptoms, involving liver and kidney failure, are self-limited and reversible, with the pulmonary fibrosis phase, when installed, is considered irreversible and responsible for the lethality of the cases of intoxication by oral route". Therefore, despite the efforts to standardize a treatment protocol for those intoxicated with Paraquat, there is still no efficient antidote or treatment for severe acute intoxication by Paraquat generally resulting from suicide.

The treatment protocol proposed for severe intoxications cannot be considered efficient; however, the cases where the patients present severe symptoms or those that result in death are mostly related to intentional ingestion of the product to attempt suicide. Normally, the dermal, eye and inhalatory routes are unable to produce severe systemic conditions such as those observed by intentional ingestion and exposure should be avoided with the use of PPE. Should intoxication occur through these routes, it tends to be recoverable and develops towards being cured. Therefore, considering the real possibility of intoxication by the product when used for agricultural purposes, according to the established correct use, the severe acute intoxication is not common and the cases of acute occupational intoxication tends to be either moderate or mild, with efficient antidotes and



treatment capable of reversing these types of damages to health resulting from occupational exposure to Paraquat-based products.

The contributions also highlighted the inclusion of warnings on labels and pack inserts warning about the danger, warnings against the transfer of the product to other packages and the inclusion of an indicative dye of substance improper for use as food or undesirable smell to warn the user, in addition to the inclusion of an emetic agent.

1.1. Prevention of cases of suicide by Paraquat

Some contributions received in the Public Consultation no. 94 of 2015 highlighted the importance of the prohibition of the product based on the cases of suicide. The suicide cases should be a constant concern to the health system. As can be observed in the Mortality Information System of DATASUS, Brazil has a suicide rate of about 5 cases/100 thousand inhabitants, lower than the global rate (11.4/100 thousand inhabitants) and lower than the rate of South Korea (24.8/100 thousand inhabitants) or Japan (19 cases/100 thousand inhabitants); nevertheless, it continues being a high rate. The data in Brazil are very disturbing especially when the rates of the States are analyzed individually because it is observed that they are very high in some states like Santa Catarina (13.2/100 thousand inhabitants) and Rio Grande do Sul (19.5/100 thousand).

According to WHO, the countries should establish strategic suicide attempt prevention plans involving a number of coordinated measures to enable the reduction of the cases. Limiting the access to the means used to commit suicide attempt is only one of the various actions that should be taken (WHO, 2012).

WHO discusses that the restriction to access is a key element in the prevention but it emphasizes that the restriction policy, such as limitation of access to pesticides, requires a clear understanding of the method preferences among the various susceptible groups and the cooperation and collaboration between various public and private sectors of the society in an integrated and synergistic action (WHO, 2012). It also reinforces that to take any measure in order to reduce the rate of suicides, it is necessary to have deep knowledge of the prevalence, characteristics and methods used because the suicidal behaviors are very different between the communities. Furthermore, the prevalent suicide method in a community may change rapidly and spread.

The access restriction is a key element in the prevention because it provides the individuals with the opportunity to reflect on their actions. However, restriction requires detailed understanding of the suicide methods used by the community. Among the actions cited by WHO as efficient is the limitation of the access to pesticides. It is worth highlighting that WHO does not recommend the banishment of the pesticides but the limitation of their use in order to hinder the obtainment of pesticides during the moment of despair (WHO, 2012).

Pesticides represent 30% of the deaths by suicide in the world and WHO defends that restriction measures would be very efficient for these cases, being easier to prevent this type of suicide with restrictive measures than the cases of hanging, for example.

According to WHO, the prevention of suicide requires many coordinated measures:

- a) Mental health policy;
- b) Policy for reduction of the excessive use of alcohol;
- c) Suitable access to health facilities for the treatment of mental disorders and disorders resulting from drug abuse;
- d) Restriction of the access to the methods used by the community for suicide attempt, editing of laws and national and local regulation.
- e) Responsive reports in the media;
- f) Increase in awareness campaigns on health disorders, drug abuse and suicide.

In the reassessment of a pesticide, Anvisa's role is to observe in which of these measures it should intervene in order to contribute in the reduction of the rates in Brazil. The restriction of the means used to commit suicide includes the reduction of availability, accessibility and attractiveness. Regarding the access to measures of restriction of the access to pesticides, WHO mentions the following measures as relevant:

- a) Ratification, implementation and application of international conventions on chemical products and hazardous wastes;



- b) Legislation to remove locally problematic pesticides;
- c) Compliance with the regulations on the sale of pesticides, reducing the access through safer storage and correct elimination, reduction of the toxicity and improvement of medical care.

Therefore, taking measures to limit the access to pesticides can help avoid more deaths by suicide. However, this action should be combined with other important measures.

A publication cited in the public consultation as relevant for showing reduction of the number of suicides in South Korea deserves a detailed discussion, considering the recommendations of WHO regarding how the prevention of suicide should be conducted (WHO, 2012; Myung et al., 2015).

According to Myung and collaborators, suicide is a very serious problem in South Korea, with the third highest rate in the world (24.8/100 thousand inhabitants). Paraquat was the most widely used pesticide for suicide in South Korea (35.5% of the deaths), causing strong concern in the country because it is a pesticide with high mortality rates. Myung and collaborators (2015) correlated the rate of suicide reduction to the prohibition of Paraquat in the country, because a significant reduction was observed in the suicides by intoxication with herbicides and fungicides soon after the prohibition. There was also significant reduction in the total suicide rate with the same standard. The reduction of these rates was explained by the fact that the use of Paraquat in suicide attempts resulted from impulsive acts; without the product available in the market, the person sought other ways that are generally less deadly.

The approach made by Myung and collaborators (2015) is relevant but some considerations should be classified because they highly weaken the association made in this publication. The reduction was observed immediately after the prohibition of Paraquat, during the period when its sale was still allowed. The author assumes that this occurred because the availability to consumers was reduced since farmers started storing Paraquat-based products. This hypothesis was raised by authors but there is not enough information to reach this conclusion. Furthermore, the study was conducted assessing the rates of suicide attempts by pesticides in a general manner and not the rates of specific suicides for Paraquat, which would have been essential to observe the sale soon after the prohibition. It is also necessary to consider that the prohibition of Paraquat is related to the change of the suicide prevention policy that occurred during that time, encouraging debates and prevention actions, which may have had a direct impact on this reduction in the rate of suicide attempt.

In addition to the considerations above, the analysis of these results should consider some other aspects because the authors themselves mentioned that other factors may have aided this reduction. Firstly, the authors discussed that the increased rate before the prohibition of Paraquat could have been associated to the concomitant financial crises in the country. Therefore, just like the increase in the rate of suicide attempt coincided with the emergence of the financial crisis, its reduction occurred concomitantly with the economic recovery of South Korea. In other words, it is plausible that these changes in the rate of suicide attempts had a higher correlation with the Korean financial crises than with the prohibition of Paraquat. The authors also mentioned various fragilities of this study: unknown factors were not accessed and considered in the statistical analysis, the time of analysis was too short, there was no investigation of the long-term effects of substitutes and it was not possible to access specific data on Paraquat.

In view of the above regarding suicide and the need for intervention through restrictive measures for access to Paraquat-based products, it is first necessary to assess the profile of suicides by Paraquat in Brazil and consider this data to propose regulatory measures. Damas and collaborators (2012) assessed the suicide attempt data in Santa Catarina, State with the second highest rate in Brazil. Between 1994 and 2006, it was observed that 55.7% of the suicide attempts occurred from the use of medication, 33.7% were pesticides, 5.2% were household cleaning and industrial chemical products and 3.5% were from drug abuse. It is worth highlighting that in 2006 it was already possible to observe a very different profile, with the use of medication in suicide attempts being more than twice the use of pesticides.

The epidemiological profile outlined by Schmitt and collaborators (2008) on the suicides in the extreme west of Santa Catarina showed a change of the means used, which



fits in the manifestation of WHO that changes in the preference method are constant. In the study of Schmitt and collaborators (2008), hanging occurred in over 70% of the cases for both men and women, the use of fire arm occurred in 9% for women and 13% for men and the ingestion of chemical substances was used by 11% of the women and 5% of the men. For this analysis, the ingestion of any substance was included in the cases of intoxication (pesticides, drugs, household cleaning products, among others). This standard was discussed as being very curious by the authors of the study because predominant economic activity in the extreme west of SC is based on agriculture and the use of pesticides in suicide attempts was expected to be the predominant method, but this was low. Meneghel and collaborators (2004) also observed that hanging was the major method used in the suicide attempts in Rio Grande do Sul (state with the highest number of suicides) in the period between 1980 and 1990. Fire arm appeared in second place. Drugs and unspecified means appeared in third place and sharp instrument appeared in fourth place.

The proposition of restrictive measures should also consider the actions with this same objective proposed by other countries. With the aim to reduce the cases of intoxication by Paraquat, USEPA published on March 2, 2016 the Public Consultation on its proposal for mitigation of intoxications by Paraquat (USEPA, 2016a). USEPA discussed that paraquat is extremely dangerous, the corresponding ingestion of 1.5 teaspoons being fatal and without any known antidote. It also considered that Paraquat-based products are associated to a disproportionately high number of incidents of intoxication, including accidental ingestions, typically fatal, as well as severe or moderate occupational exposure (spilling, splashes and leaks) after skin or eye contact. All the information on incidents available since 2000 were reviewed, observing during this period 27 deaths (11 intentional, 8 accidental, 2 intentional/accidental, 2 resulting from drifts, 3 without information); 22 severe intoxications (4 accidental ingestions, 0 occupational, 1 suicide attempt, 1 drift, 7 without information); 181 moderate (mainly occupational) and 352 mild (USEPA, 2014). Based on the number of incidents and on the severity of the cases, USEPA believes that the mitigation measures proposed are necessary for the continued registration of Paraquat-based products (USEPA, 2016b). It is worth emphasizing that Paraquat is still being reassessed by USEPA and the draft should be published in 2017 for comments. According to USEPA, all the measures proposed were discussed with the registrants of the technical products. Among the measures proposed by USEPA are:

- a) Changes in the labels emphasizing the toxicity of Paraquat and availability of additional warning leaflets.
- b) Development of education and handling projects and training materials for users of Paraquat-based products.
- c) Adoption of a closed system in the packaging.
- d) Restriction of the use of Paraquat to certified applicators, prohibiting application by workers monitored by the certified applicator.

In order to propose these measures, USEPA conducted an economic impact analysis and observed that the changes on the labels should not cause any negative impact, but the change to the closed packaging system will cost about 500 to 700 dollars per user, which means a total of 12.5 to 17.5 million dollars. The prohibition of manual application was not estimated because it will not be accurate due to the non-availability of data. The restriction of use to certified people will increase the costs by 1.3 million dollars and the development of training material will involve costs of 3100 to 4700 per registrant (USEPA, 2016c).

The concentration reduction of the product to 5% was one of the measures suggested to USEPA (USEPA, 2014). Presently, the Paraquat-based products have a concentration of about 30% and the ingestion of only 15 mL is enough to kill an adult man of 70kg. If the products had a concentration of 5%, the ingestion of 30 mL can theoretically be considered non-lethal. The company argued that the production, storage, transport and application of the volume of product necessary to achieve same application dose used presently would be impossible. Besides the economic unfeasibility, the company argued that this measure may not represent an increase in the survival rates after intoxication due to the high toxicity of the product (Dixon and Wells, 2014).

Nagami and collaborators (2013) also concluded that the adoption of the concentration reduction of the products is not a very significant strategy because they discovered that, although Japan had reduced the concentration from 24% to 5% of Paraquat



in the products, the mortality rate by suicide remained the same at about 80%. In view of these justifications, USEPA did not adopt this measure in its proposal. On the other hand, the Canadian regulatory agency PMRA (Health Canada's Pest Management Regulatory Agency) considered that the results in Japan showed an improvement in the death scenario by Paraquat intoxication and decided that in Canada the concentration of Paraquat in the final product will be reduced to a concentration lower than the lethal concentration or lower than the concentration that causes serious damages to the eyes or skin. The application rate will continue to be the same, thus there will be no increase in exposure (PMRA, 2015).

This measure adopted by PMRA is one of the various actions that determined by the Canadian agency after the reassessment of Paraquat in the country. In the reassessment conducted by PMRA, all the chronic risk information was reviewed and the points considered relevant were the potential risk of effects in health resulting from accidental exposure during occupational use and the occupational risk of back application. Among the measures proposed by PMRA are:

- a) Inclusion of Paraquat in the restricted class: use exclusively by certified/licensed applicators;
- b) Changes in the label including additional IPEs, warnings of danger and treatment;
- c) Prohibition of mixing in tanks used for other pesticides;
- d) Reduction of the concentration of Paraquat in the end-use product;
- e) Change of the packaging with the addition of a mechanism to measure the volume required for application;
- f) Education and handling program that includes information to vendors and users on the new risk mitigation measures and on the importance of complying with the recommendations of the label, as well as on the prevention of accidental exposure.

1.2 Occupational Exposure

Occupational exposure was discussed in the Technical Reassessment Panel for Paraquat and in some contributions of CP no. 94 of 2015, especially by health professionals and by the Task Force. In her contributions to the CP and panel, Dr. Ligia Fruchtgarten emphasized that in dermal or eye effects or lesions may occur in occupational exposure when the individual protection precautions appropriate for application of the product are not observed and that only skin lesion and significant increase of the absorption of Paraquat are expected when there is prolonged skin contact.

The Task Force and some other participants from CO also claimed that acute intoxication with Paraquat through dermal administration is not relevant and the cases are restricted to inadequate use of the products. However, it is important to point out that there is no consensus about the importance of cutaneous absorption during occupational exposure to Paraquat. Both USEPA and PMRA have already confirmed that although the studies in animals show low dermal absorption, the data in humans indicate that they are more sensitive to Paraquat in chronic exposure, resulting in bigger damage to skin and higher systemic absorption of this herbicide (PMRA, 2015; USEPA, 2016b). I.e., the studies of cutaneous absorption in animals do not characterize the toxic potential of Paraquat for human health adequately and thus, occupational exposure shall be a concern of the regulatory agencies. That is why, PMRA and USEPA have sought strategies to minimize the exposure of workers. PMRA, despite of reporting higher sensitivity of the humans, concluded that the daily exposure of the workers is minimum in Canada, because the use of IPE [*Individual Protection Equipment*] would be sufficient to protect them even during manual spraying for the treatment of oat (the only culture, for which use of Paraquat-based products in this modality is allowed). USEPA, as detailed above, intends to keep the product register, but has proposed quite restrictive measures.

Paraquat Technical Reassessment Opinion No. 1 dated 2014, supported by discussions either in the scientific literature or of other regulatory agencies, has already concluded the same about the intoxication of workers exposed to Paraquat, and even after the contributions received in CP No. 94, dated 2015, it is considered that occupational exposure cannot be based on damages to the skin or dermal absorption of Paraquat obtained from studies in animals. Thus, the discussion of the toxicological aspects and the proposals for mitigation measures shall take the evidences of higher human sensitivity to dermal exposure Paraquat with possibility of relevant systemic absorption through this administration route into consideration.



Initially, the Task Force presented some proposals to mitigate the risk of occupational exposure to Paraquat, such as:

- a) Intoxication monitoring program with expansion of the number of CITs;
- b) Maintenance of the safety elements in the products (emetic, drying and odorizing agents);
- c) Training of agricultural workers with emphasis on prevention from occupational exposure, storage and recommendations in case of ingestion;
- d) Training of community agents for monitoring of psychiatric diseases;
- e) Medical seminars prioritizing regions, where there are records of death due to intentional intoxication;
- f) Distribution of activated charcoal, Dithionite test and treatment protocol folder kits in reference hospitals;
- g) Educational programs in partnership with rural development program services;
- h) Adoption of the Paraquat Safety Plan (detailed hereunder);
- i) Use restrictions;
- j) Using 0.5% of the funds for educational and handling programs.

Many of these measures can be efficient and necessary for the reduction of the cases of intoxication; however, it is essential to present more detailed planning containing clear indicators of effectiveness for the follow-up of such measures, in case it is concluded to keep the Paraquat-based products on the market.

One of the concerns regarding the occupational exposure pointed out in Technical Reassessment Report No. 01 was the results observed in the models for prediction of the exposure to the Paraquat-based products, because the exposure to this active substance would extrapolate the proposed AOELs (acceptable occupational exposure levels) (Pesticide Safety Directorate, 2002) and this would be unacceptable considering the severe toxic effects of Paraquat and the signs of induction of Parkinsonism, discussed in more details in item 3 herein. The Task Force presented some relevant arguments both in its contribution to CP and in the Technical Reassessment Panel and therefore, it was necessary to discuss this aspect more deeply.

During the reassessment of Paraquat in the EU, it was verified that there was AOEL extrapolation using several models of exposure prediction, as already discussed in Reassessment Opinion No. 01. However, EFSA committee responsible to reassess Paraquat concluded that the results of the field monitoring studies indicated that the prediction models overestimated the actual exposure of the workers exposed to Paraquat and that there was no significant risk to the workers' health when the products were used as recommended and according to the good agricultural practices (Pesticide Safety Directorate, 2002).

After the EFSA decision to maintain the registers of Paraquat-based products, some countries from the EU protested against this opinion, indicating several studies, in which it would be possible to identify worrying exposure of works and confirming the results of the exposure prediction models. The discussion made by Sweden against the decision of the EFSA committee was considered relevant and was used as grounds for the position adopted in Paraquat Technical Reassessment Opinion No. 1; however, the Task Force presented contra-arguments and therefore, the monitoring studies were reviewed as it can be seen in chart 2 (EU, 2007).

During the reassessment of Paraquat in the EU, the disagreement of some countries in relation to the EFSA conclusion about the occupational exposure was based mainly on observation that the doses of Paraquat obtained in these monitoring studies exceeded the proposed AOEL. However, it was later verified, by conducting new studies, that the initially proposed value for AOEL could be increased based on the toxicological evidences. Thus, from the analysis of the monitoring studies, it can be concluded that they actually do not reveal exposure above the AOEL; however, it is evident that there is potentially relevant exposure to Paraquat, especially when the IPE is not used correctly.

Chart 2. Studies for monitoring exposure to Paraquat

SWAN, 1969	
- Malaysia (1965) - Backpack application in rubber tree plantations. - Gowning: shirt or T-shirt, long trousers, socks and closed shoes. Washing of the body before and after application. - Period of exposure 12 weeks (6 times a week): approximately 2600 gallons. - Paraquat detected in all workers' urine at least at some period along the exposure (78/134 urine samples). - Symptoms: testicle dermatitis, scaling on the hands and the ears, severe epistaxis. - The highest concentration of Paraquat detected was 0.32 ppm, but the bigger part was lower than 0.1 ppm.	- Detection of Paraquat in the urine of exposed workers, but not using IPE.



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SWAN, 1969 - Malaysia (1967) - Backpack application in rubber tree plantations. - Gowning: a group with common clothes (shirt or T-shirt, long trousers, socks and closed shoes, washing the body before and after application) and a group with IPE (boots, gloves and face mask). - Period of exposure 12 weeks (6 times a week): approximately 1800 gallons. - Paraquat detected in quite lower number of samples than in the study dated 1965 (53/394), but it was applied at lower quantity. - Symptoms: testicle dermatitis, scaling on the hands and the ears, nose bleeding. - The highest concentration of Paraquat detected was 0.15 ppm. - Higher percentage of the urine samples positive for Paraquat was found in the workers, who used common clothes (18 to 50%). The use of IPE reduced the percentage of positive samples to 7 to 14%. - Symptoms: dermal irritation, urticaria, genitals dermatitis, dermal lesion, epistaxis.	- It is possible to detect Paraquat in the exposed workers' urine, even when they use IPE. - There is an expressive reduction of the detection of Paraquat in urine, when IPE is used.
STAIF et al.; 1975 - USA - Spraying in orchards by tractor or backpack spraying in gardens. - Body exposure determined by means of absorbing paper and washing the hands after the application. - Inhalation measures by specific filters in the breathers. - Gowning: without IPE (tractor). - Urine samples taken during and after exposure. - Low dermal and respiratory exposure. - Higher exposure of the hands. - The dermal exposure varied from 0.01 to 3.4 mg/h for tractor application and from 0.01 to 0.57 mg/h for application in gardens. - No Paraquat was found in the urine samples. - Limit of detection 0.02 ppm.	- The assessed scenarios are not compatible to the backpack application in field culture, which enables detection of Paraquat in other studies; - Higher limit of detection than the limit of other studies, which verified Paraquat in the exposed workers' urine. - Lack of sensitivity of the analytical method.
WOJECK et al., 1983 - USA - Tractor application of Paraquat in tomato and citrus cultures. - Gowning: long or short sleeved shirts, long trousers, socks, closed shoes or boots. Many workers used hats. - The use of closed cabin or high clearance tractor reduced the exposure by about 85%. - Paraquat was found in the urine of one worker only (0.033 ppm). - Limit of Detection: 0.012-0.041 ppm.	- Limit of detection higher than the limit in the studies, which verified Paraquat in the exposed workers' urine. - Lack of sensitivity of the analytical method.
CHESTER et al. 1993 - Sri Lanka. - Broth preparation, tank supply, manual backpack sprayer application in tea plantation. - Paraquat quantification in urine at every 24 hours (broth preparation and spraying) by radioimmunoassay. - Gowning: short-sleeved shirt, pants or shorts, no socks and no closed shoes. Frequent washing habit. - Urine collected one day before the first application up to seven days after the last application (total of thirteen days). - Exposure quantification measured by the recovery of Paraquat in overalls with hood, gloves and socks. - Amount and concentration of Paraquat in spraying solution lower than usual backpack sprayer applications. - Paraquat detection limit in urine is 0.03 µg/ml. - Paraquat was not detected in any urine sample.	- Detection limit higher than the limit of studies, which found Paraquat in the urine of exposed workers. - Lack of sensitivity of the analytical method.
MEIER, 1995 - USA. - Application of Paraquat using an open cabin tractor in pecan orchards. - Gowning: Long clothes, boots and gloves. - Paraquat detected in the urine of 6/17 workers on the exposure day. - Paraquat excreted quickly (2 to 3 days). - Paraquat detected in urine at concentrations of 6 ng to 26 ng/mL. - The average exposure to Paraquat was 0.0002 mg/kg bw/day. - Limit of detection: 0.005 µg/mL. - Higher absorbed amount: 0.00044 mg/kg bw/day, in a worker who did not use the IPE properly.	- Nine operators did not use IPE or gloves. - Paraquat values in blood and in urine indicate doses with values between 0 and 60% of AOEL. - Average exposure corresponds to 28% of AOEL. - Only workers who did not use the IPE properly had values of Paraquat in urine close to AOEL.
BERNA et al., 1996 - Costa Rica - Backpack sprayer application in banana plantations. - Dermal exposure measured by means of absorbent papers, respiratory exposure by personal air samples and systemic exposure by urine samples - Gowning: some used long clothes and gloves. Everyone used boots. - Symptoms: bubbles on the skin; burns on the hands, thighs, back, testicles and legs; redness, irritation and burning sensation in the eyes, problems with nails and epistaxis. - Dermal exposure varied from 0.2 to 5.7 mg/h. - Areas with higher levels of dermal exposure: wrists, back and legs. - Inhalational exposure varied according to the day and dermal exposure varied according to the type of plantation.	Paraquat was detected in the urine of two out of the ten monitored employees, no use of IPE.



- Paraquat limit of detection in urine is 0.03 µg / mL. - Paraquat detection only in two urine samples from two workers (higher level 0.24 µg / L)	
FINDLAY et al., 1998 - Spain (1997). - Backpack sprayer application in citrus orchards. - Broth preparation, supply and manual backpack sprayer application of 15L Paraquat during 6 hours (one day). - Broth preparation and supply gowning: long clothes, boots, rubber gloves and facial visor. - Broth and preparation supply gowning: simple long overall and boots. - Urine collected at every 24 hours for seven days, the collection started on the day before the first application. - Limit of detection: 0.00075 µg/mL - Paraquat detected in the urine of 18/20 of workers after application for 4 or 5 days. - The paraquat values in urine ranged from 7.90 µg/mL to 262 µg/mL. - Average absorbed dose: 0.00015 mg/kg bw/day. The highest dose absorbed was 0.00041 mg/kg bw/day - Total elimination of Paraquat within 72 hours maximum, only one worker showed Paraquat in urine until the 5th day.	- The use of gloves could reduce the number of samples with Paraquat as well as the detected levels. - Absorbed dose was lower than AOEL for all workers, even using less IPE than recommended. - The rate of dermal exposure was also similar to that found in other occupational studies. - The highest amount of Paraquat detected in urine corresponds to absorption of 56% of AOEL.
LEE et al., 2009 - Costa Rica (2001). - Urine was collected from 119 Paraquat handlers (backpack sprayer application) and 59 non-manipulators, who also worked on the cultures. - Coffee culture gowning: long clothes and boots, 50% were using overalls and aprons. - Gowning for the cultures banana and palm: overalls, gloves, respirator and apron. - Paraquat quantification limit in urine: 2 ng/mL. - For the manipulators, 83%, 47% and 64% of the urine samples were lower than the quantification limit before, during and after spraying, respectively. - The culture, for which workers had higher levels of Paraquat in urine samples was the banana, followed by coffee and finally, palm. - The average quantity of Paraquat in urine for exposed workers was 6.3 µg / 24 h. - There was no variation in the level of Paraquat regarding the type of activity performed (mixing, loading, application or maintenance of equipment).	Paraquat detection in urine, even when using IPE.

Based on the new AOEL, Syngenta presented risk assessment for the use of Paraquat-based products, using the American database – PHED (The Pesticide Handler Exposure Database) to estimate the exposure of the exposed workers. The scenarios were selected according to the Brazilian reality, seeking data in Brazil, which could also ground this risk assessment. The simulations were carried out using less IPE than the recommended on the package inserts of the products in Brazil. To determine the non-observed adverse effect level (NOAEL = 0.56 mg/kgbw/day), a 90-day study in dogs (more sensitive species) was selected, in which lesions in the lungs and the kidneys were observed. The company pointed out that this period can be considered conservative, because the time of exposure of the workers in field would be much shorter, because the Paraquat-based products are not applied continuously. As the oral absorption of Paraquat is lower than 80%, the company carried out a bioavailability study, in order to determine the oral absorption factor for the correction of the systemic NOAEL, resulting in NOAEL = 0.073 mg/kg bw/day. Thus, there was increase in the AOEL value to 0.00073 mg/kg bw/day. Using this AOEL for risk assessment, there was no extrapolation of the scenarios by the PHED prediction model as it can be noted in the chart below (chart 3). All exposure scenarios, according to a study presented by Syngenta, presented values below AOEL. It shall be pointed out that the simulation was made with less IPE than the recommended in a package insert in Brazil.

Chart 3. Percentage of daily dermal exposure to Paraquat in relation to AOEL for the PHED scenarios

PHED Scenario	IPE	% AOEL
Liquids, open system, broth preparation, tank supply, air application.	Simple clothing (long shirts, long pants made of simple cotton, socks and shoes) WITH gloves resistant to chemical products.	59.8
Liquids, open system, broth preparation and tank supply, application by closed cabin tractor with bar.	Simple clothing WITH gloves resistant to chemical products.	7.2
Liquids, open system, broth	Simple clothing WITH gloves	14.5



preparation and tank supply, application by closed cabin tractor with bar.	resistant to chemical products.	
Liquids air application (fixed wing/closed cabin)	Simple clothing WITHOUT gloves resistant to chemical products.	13.0
Application by open cabin tractor with bar.	Simple clothing WITH gloves resistant to chemical products.	4.4
Application by closed cabin tractor with bar.	Simple clothing WITHOUT gloves resistant to chemical products.	3.1
Liquids, broth preparation and tank supply, application by backpack sprayer.	Simple clothing WITH gloves resistant to chemical products.	17.9

From the results from exposure prediction and the results from the monitoring studies, Syngenta concluded that the occupational exposure does not present unacceptable risk to the workers. However, the regulatory agencies have adopted measures to reduce exposure to Paraquat even more, because they do not consider that the initial register already contemplates all measures necessary for the mitigation of the risks of Paraquat to health. For example, USEPA and PMRA recently proposed the adoption of more restrictive measures to reduce exposure to Paraquat even more, because there are cases of occupational intoxication, which shall be mitigated and the exposure of human beings seems to be more relevant than that of animals used as models in the toxicological studies.

Furthermore, the monitoring studies reveal that Paraquat can be absorbed in doses, which, despite of not exceeding AOEL, are worrying due to the evidences associating this active substance to Parkinsonism, and it is difficult to affirm that the currently accepted AOEL is a safe limit in relation to the effects of Paraquat on the dopaminergic neurons pursuant to exposure along the workers' life. This subject will be deeper discussed later in a specific topic.

In many monitoring studies, additional clinical exams were made, including hematological and biochemical exams, to assess the hepatic and the renal function, as well as pulmonary functional tests, concluding that the use of Paraquat under normal conditions (including without the use of IPE) does not represent a serious risk to the workers' health in relation to these analyzed aspects. Therefore, it is not considered that Paraquat might cause severe intoxication in workers from the analysis of these monitoring studies, as well as from the case studies and the Brazilian data from the intoxication centers, as well as the data from Agrotoline reported by the Task Force. However, it shall be pointed out that Parkinsonism will be further detailed and shall be assessed separately from the other toxic aspects of occupational exposure, because depending on the evidences, it might represent a higher risk to the workers' health and, as precaution, it might need much more restrictive measures, although the studies currently demonstrate that exposure to Paraquat is below the level stipulated as safe.

1.3. Paraquat Product Safety Program (PPSP)

PPSP proposed by the Task Force includes different actions, which together could be used for the reduction of the cases of intoxication by Paraquat. For the execution of such actions, the Task Force proposed using 0.5% of the sales of the Paraquat-based products.

One of the proposed actions within this plan is monitoring of the intoxications, in order to determine measures to reduce the currently observed indexes. The proposal of the Task Force is to use Agrotoline, an intoxication notification system exclusively for pesticides implemented by Toxiclin in July 2012 for monitoring of intoxications; five public toxicology centers are part of this system: Centro de Controle de Intoxicações de Londrina, Centro de Informações Toxicológicas do Rio Grande do Sul (Porto Alegre), Centro de Controle de Intoxicações de São Paulo (Jabaquara), Centro de Informações Anti-Veneno da Bahia – CIAVE (Salvador) and Centro de Informações e Assistência Toxicológica (Brasília).

In this system, the intoxication survey is done by the trade name and by the active substance of the herbicide. From June 2012 to May 2015, 46 out of the 2,285 cases of intoxication were results from intoxication by Paraquat. 21 of these 46 cases were pursuant to occupational exposure (45.65%), 18 to attempts of suicide (39.13%) and 4 to accidents (8.7%). All severe intoxications and deaths were suicide cases. The cases of occupational exposure included only the intoxications considered moderate or slight. It was also perceived that there is a delay in diagnosis, lack of basic resources in the health units, and little training of the health professionals.



PPSP proposed by the Task Force is focused on monitoring (by means of the Agrotoline notification system) and training as an attempt to unify the programs of the different manufacturers. Monitoring is essential to allow the determination of actions and training to be ministered to the users (focused on the correct and safe use, storage, recommendations not to transfer package, use of IPE and hygiene fundamentals), people who recommend (focused on the molecule characteristics, use indications and necessary protection measures) and physicians (focused on diagnosis and treatment).

The trainings proposed by the Task Force are divided in external and internal training. The internal training would be aimed at the people involved in production, commercialization, technical recommendation, marketing and regulatory areas. The external training would involve users, buyers and appliers by means of obligatory online training at the time of purchase and with validity period, as well as training in person, where necessary, in addition to training for the medical class.

The Task Force reports that other measures were taken to reduce the exposure of small producers to Paraquat, such as exclusion of the cultures avocado, asparagus, beetroot, cocoa, tea, coco, pear, peach, sorghum, grapes and pastures in November 2006. In July 2008, the exclusion of the herbicide for desiccation of rice and the dose reduction from 1.5-3.0 L/ha to 1.5-2.0 L/ha were approved, in addition to the exclusion of the application in desiccation of corn and cotton. However, these restrictions were made for the Syngenta products, and the other registering companies should become adequate and the monograph should be updated.

It shall be pointed out that according to contributions made in the documents delivered by the proper Task Force (represented by Syngenta), the Paraquat registration for tea and cotton would be essential. However, Syngenta reports that they excluded these cultures, because they considered that this action is a way to reduce the exposure of the small agricultural producers. According to the proposal of the Task Force, Paraquat will remain authorized only the cultures cotton, rice, banana, potato, coffee, sugarcane, citric fruits, beans, corn, soybean, wheat and as desiccant in the cultures sugarcane, soybean and potato. Furthermore, many contributions from professionals from the agronomy area pointed out the importance to use Paraquat in fallow land, pre-cultivation areas, between the culture rows, picking, track cleaning and desiccation, especially for control of weeds in cultures, such as banana and corn.

Syngenta, representing the Task Force, proposed other regulatory actions as well: exclusion of the application on the cultures pineapple, kale, apple, rubber tree and grapes and elimination of the 1 L package. In the CP contributions, elimination of the 1 L packages and exclusive use on grain and fiber were also suggested. Although the Task Force has proposed the exclusion of the application on apples, in the public consultation, it was mentioned that Paraquat is the only product registered for this culture.

1.4. Education and handling program: Center-South Project for Beans and Corn

- g) Place: Paraná
- h) Target population: 120 thousand small producers of corn and beans (culture rotation).
- i) Period: from 1989 to the present date.
- J) Periodicity: Annual
- k) Diagnosis: low productivity and yield; soil degradation; inadequate handling of weeds, diseases and plague; low access and incorrect and unsafe use of the agricultural technologies, among which the pesticide.
- l) Participants: Syngenta, Emater-PR, Embrapa Arroz e Feijão and Instituto Agronômico do Paraná (Iapar).
- m) Participation: 3,000 producers participated in the administrative units and it is estimated that more than 10,000 visit at least one of the more than 200 events held during the year.
- n) Description: The project guides the producers about the good practices in all cultivation stages, including about the correct and safe use of pesticides.
- o) Results: The project helped increasing the production level of the region above the average for the State, improve the yield for the producer and promote higher quality of life in the region, in addition to reduction of the cases of intoxication by pesticides. Initially, the use of IPE was about 30% and increasing gradually to levels higher than 60%.

Education and handling programs, such as Center-South Project for Beans and Corn, are essential for training of workers involved in the use of pesticides and consequently, for the mitigation of the risks to health associated to the manipulation of these substances. Therefore, considering the high toxicity inherent to Paraquat, the maintenance of the register of this active substance shall be conditioned to expansion and better structuring of projects of this kind, with follow-up of their results by Anvisa.

2. Genotoxicity and mutagenicity of Paraquat



During the consolidation of CP No. 94 dated 2015, the limitations pointed out in the contributions regarding the studies used in TECHNICAL REASSESSMENT OPINION No. 01 dated 2015/GGTOX/ANVISA were assessed and those considered relevant were included in chart 4 and used for the final assessment of the mutagenic potential of Paraquat.

It shall be pointed out that some of the assessed studies present limitations, which do not make them inconclusive, and it is possible to use their results. On the other hand, some assessed studies presented substantial limitations, which impeded the use of their results in the assessment of the weight of the evidence.

It was verified that *in vitro* study considered valid for point mutation in mammal's cell and the prompt genic mutation studies in bacteria strains are consistently negative, and there was only one positive result for *in vitro* tests in unicellular eukaryote cell. This single result *in vitro* in fungus is not sufficient to conclude on the induction of point mutation by Paraquat.

Four *in vivo* and *in vitro* results in mammal's cells were negative; however, in fourteen tests, it was possible to see clearly occurrence of mutation of the chromosome aberration type. Some of the studies for assessment of chromosome aberrations in *Drosophila melanogaster in vivo* and in mammal's cells *in vitro* (test with mouse lymphoma) could indicate occurrence of point mutations; however, they were not carried out in a way to differentiate the parameters, which determine the type of mutation and, as there are no other evidences of point mutation occurrence, they can be used only to give weight of evidence to the induction of chromosome aberration by Paraquat.

Studies in germinative cells were considered inconclusive due to their limitations. However, although currently, there is no evidence proving the potential of Paraquat to induce mutation in germinative cells, it was found that the exposure to Paraquat reaches the testicles affecting the spermatozooids (Rios et al., 1995; D'Souza et al.; 2006). Furthermore, all positive tests presented in chart 4 related to the assessment of genotoxicity grant strong weight of evidence to the observation of induction of chromosome aberrations in somatic cells by exposure to Paraquat.

Chart 4. Summary of the results from the mutagenicity studies for Paraquat.

PARAQUAT MUTAGENICITY					
Reference	Concentration	Type of test	Limitations	Assessed parameter	Result
Report No. CTL/P/243	99%	<i>In vitro</i> Ames	-	Point mutation	NEGATIVE
Report No. 20725-2002	45%	<i>In vitro</i> Ames	-	Point mutation	NEGATIVE
Benigni et al., 1979	NI	<i>In vitro</i> Ames	-	Point mutation	NEGATIVE
Speit et al., 1998	NI	<i>In vitro</i> HPRT in lung fibroblasts of Chinese hamster	High cytotoxicity in the higher doses.	Point mutation	NEGATIVE
Report No. CTL/P/442 - 1979	100%	<i>In vitro</i> Cytogenetic in rat cells	Paraquat associated to the Giensa stain provoked alterations in the morphological appearance of the chromosomes, which can be confused with chromosome aberrations. This artifact was prevented upon five washings in the cells. No mitotic index was determined.	Chromosome aberration	NEGATIVE
Tanaka and Amano 1989	NI	<i>In vitro</i> Chromosome aberration in lung cells of Chinese hamster	Cellular viability was not assessed, but the concentration used was compatible to that of other studies. Gaps were accounted for. Lack of positive control.	Chromosome aberration	NEGATIVE
Report No. CTL/P/442 - 1979	100%	<i>In vivo</i> Micro-nucleus in mice	Paraquat associated to the Giensa stain provoked alterations in the morphological appearance of the chromosomes, which can be confused with chromosome aberrations. This artifact was prevented upon five washings in the cells. No mitotic index was determined.	Chromosome aberration	NEGATIVE
Report No. CTL/P/1369	33.7% (ion)	<i>In vivo</i> Micro-nucleus in mice	The decrease of the PCE/NCE ratio was very small and it occurred only at the highest dose. I.e., it is not possible to be sure that the substance reached its target. Only two doses were used, and dose-response analysis	Chromosome aberration	NEGATIVE



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			was not possible. Lower doses would be perfect.		
Benigni et al., 1979	NI	<i>In vitro</i> Resistance to 8-azaguanine in <i>Salmonella typhimurium</i>	High toxicity in higher doses.	Point mutation	INCONCLUSIVE
Moody and Hassan, 1982	NI	<i>In vitro</i> Ames	Number of revertants expressed according to the survival (revertants/ 10 ⁸ survivors); it does not enable assessing whether the increase is due to mutagenic action or cell death at higher concentration.	Point mutation	INCONCLUSIVE
Study RF-3252.401.182.04 - 2005	50%	<i>In vitro</i> Ames	The correct doses were not used, and there were number of doses with cytotoxicity lower than the recommended in the OECD guidelines.	Point mutation	INCONCLUSIVE
Osch et al., 2010	NI	<i>In vivo</i> Point mutation in <i>harlequin</i> mice	Increase considered small by the author himself, without dose-response assessment.	Point mutation	INCONCLUSIVE
Cantavenera et al., 2007	NI	<i>In vitro</i> Ames	Carried out only with one strain. Without dose-response curve.	Point mutation	INCONCLUSIVE
Report No. CTL/P/1560 - 1987	33.7% (ion)	<i>In vivo</i> Cytogenetic in rats Alpk: AP	Negative history unacceptable, because the reference is from a poster presented in a conference. The aberration count data per animal was not presented, but the average percentage of cells, without standard deviation. Not all doses were tested in 12 h, only the highest dose. All doses were tested only in 24 h. The highest dose was tested in 48 h. The number of metaphases analyzed per animal (50) was lower than the recommended (200).	Chromosome aberration	INCONCLUSIVE
Report No. CTL/P/367 - 1978	100%	<i>In vivo</i> Cytogenetic in rats	The mitotic index was not determined. Morphological alterations in the chromosomes make analysis of the chromosome aberrations difficult. The number of metaphases analyzed per animal (50) was lower than the recommended (200).	Chromosome aberration	INCONCLUSIVE
Ribas et al., 1997	99%	<i>In vivo</i> Micro-nucleus in human lymphocytes	The lymphocyte donors' age is not described. The treatment period was 2 h, and the recommended period is from 3 to 6 h. Number of analyzed cells lower than the recommended.	Chromosome aberration	INCONCLUSIVE
Ribas et al., 1997	99%	<i>In vivo</i> Chromosome aberrations in human lymphocytes	The lymphocyte donors' age is not described. The treatment period was 2 h, and the recommended period is from 3 to 6 h. Number of analyzed cells lower than the recommended.	Chromosome aberration	INCONCLUSIVE
Nicotera, 1985	NI	<i>In vivo</i> Chromosome aberrations in fibroblasts of Chinese hamster	Reduction of the mitotic index in the highest concentrations. Inclusion of gaps in the analysis.	Chromosome aberration	INCONCLUSIVE
Cantavenera et al., 2007	NI	<i>In vivo</i> Chromosome aberrations in V79 cells	Cytotoxicity was not described. Increase not statistically significant.	Chromosome aberration	INCONCLUSIVE
Anderson et al., 1976	NI	<i>In vivo</i> Dominant lethal in mice	Interval between doses longer than the recommended	Point mutation in germinative cells	INCONCLUSIVE
Pasi et al., 1974	NI	<i>In vivo</i> Dominant lethal in mice	The study was carried out at one dose only, it cannot be classified in the that suggested by the OECD protocol. In addition, the number of animals used was small.	Point mutation in germinative cells	INCONCLUSIVE
IRI Report No. 145	23.9%	<i>In vivo</i> Dominant lethal	Without increase in embryonic or fetal death in	Point mutation in germinative cells	INCONCLUSIVE



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		in mice	mating from 4 to 8 weeks and without significant difference in the embryonic or fetal death in mating in weeks 1 and 2, but there was a big variation between the groups in the first 3 weeks.		
Report No. CTL/P/1351 - 1985	99.6%	<i>In vivo</i> Cytogenetic in humans	Highest dose with 68% toxicity (perfect between 50 and 60%)	Chromosome aberration	POSITIVE
Report No. CTL/P/1398 - 1985	45.66%	<i>In vitro</i> Mice lymphoma cells L5178Y	High cytotoxicity doses (less than 20% viability), in which there was increase in the mutation frequency, with impaired interpretation of the data, because it was not possible to assess the mutagenic potential. In test, in which no increase of mutation frequency was observed, perfect cytotoxicity was not achieved as well (quite below the recommended). Shorter exposure (2 h) than the recommended.	Point mutation and/or chromosome aberration	POSITIVE
Gaivão et al., 1999	NI	<i>In vivo</i> Eye stain in <i>Drosophila</i> <i>melanogaster</i> (SMART)	High mortality in the last dose (40 to 80%). Without valid OECD guideline.	Point mutation and/or chromosome aberration	POSITIVE
Gaivão et al., 1996	NI	<i>In vivo</i> Eye stain in <i>Drosophila</i> <i>melanogaster</i> (SMART)	High mortality in the last dose (40 to 80%). Without valid OECD guideline.	Point mutation and/or chromosome aberration	POSITIVE
Jovtchev et al., 2010	NI	<i>In vitro</i> Chromosome aberration in human lymphocytes	Donors aged between 41 and 52 years. No positive control data was presented. Treatment period 1 h.	Chromosome aberration	POSITIVE
Sofuni and Ichiadie Jr., 1988	NI	<i>In vitro</i> Chromosome aberrations in fibroblasts of Chinese hamster	No cytotoxicity and use of positive control are reported. Number of analyzed cells below the recommended	Chromosome aberration	POSITIVE
Lin et al., 1987	45%	<i>In vitro</i> Chromosome aberrations in ovarian cells of Chinese hamster	-	Chromosome aberration	POSITIVE
McGregor et al., 1988	NI	<i>In vitro</i> Mice lymphoma assay (MLA)	There was no differentiation between small and big colonies, not allowing differentiation of the type of mutation.	Point mutation AND/OR chromosome aberration	POSITIVE
Benigni et al., 1979	NI	<i>In vitro</i> Genetic mutation in <i>Aspergillus</i> <i>nidulans</i>	Time of exposure inadequate. No use of positive control.	Point mutation	POSITIVE
Torres et al., 1992	99%	<i>In vivo</i> Wing stain in <i>Drosophila</i> <i>melanogaster</i> (SMART)	Without valid OECD guideline. There was no differentiation between types of stains.	Point mutation and/or chromosome aberration	POSITIVE
Ortiz et al., 2000	> 98%	<i>In vivo</i> Micro-nucleus in mice	Intraperitoneal administration. Assessment of a single dose. Number of PCE assessed lower than the recommended.	Chromosome aberration	POSITIVE
Speit et al., 1998	NI	<i>In vitro</i> Chromosome aberration in lung fibroblasts of Chinese hamster	High cytotoxicity at the highest doses.	Chromosome aberration	POSITIVE
Melchiorri et al., 1991	98%	<i>In vivo</i>	Intraperitoneal administration.	Chromosome aberration	POSITIVE
Melchiorri et al., 1998	98%	<i>In vivo</i> Micro-nucleus in mice	Intraperitoneal administration	Chromosome aberration	POSITIVE
D'Souza et al., 2005	~ 100%	<i>In vivo</i> Micro-nucleus in rat	Dermal administration. Number of PCE analyzed lower than the recommended. No use of positive control.	Chromosome aberration	POSITIVE
Report No. CTL/P/1339 - 1985	99.6%	<i>In vitro</i> Non-scheduled synthesis in rat hepatocytes	Without valid OECD guideline.	Genotoxicity	NEGATIVE



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Report No. CTL/P/1550 - 1987	33.7% (ion)	<i>In vivo</i> Non-scheduled synthesis of DNA in rat hepatocytes	Number of animals and cells analyzed per animal lower than the minimum recommended number.	Genotoxicity	NEGATIVE
Speit et al., 1998	NI	<i>In vitro</i> Comet in lung fibroblasts of Chinese hamster	High cytotoxicity at the highest doses.	Genotoxicity	NEGATIVE
Wang et al., 1987	45%	<i>In vitro</i> Exchange of sister chromatids in ovarian cells of hamsters	Only two concentrations used, and it was not possible to verify the dose-response relation. Without valid OECD guideline.	Genotoxicity	NEGATIVE
Report No. CTL/P/1392 - 1985	99.4%	<i>In vitro</i> Exchange of sister chromatids	-	Genotoxicity	POSITIVE
Yamamoto et al., 2001	NI	<i>In vitro</i> DNA break	The result does not show action in an intact cell.	Genotoxicity	POSITIVE
Ribas et al., 1995	98%	<i>In vitro</i> Comet in human lymphocytes	There is no description of the positive and the run controls.	Genotoxicity	POSITIVE
Dusinská et al., 1998	NI	<i>In vitro</i> Comet in lung macrophages and pneumocytes of the type II	Cellular viability was not assessed, but the concentration used was compatible to that in other studies.	Genotoxicity	POSITIVE
Ross et al., 1979	NI	<i>In vitro</i> Alkaline elution	-	Genotoxicity	POSITIVE
Tanaka, 2000	NI	<i>In vitro</i> Exchange of sister chromatids in lung cells of hamsters	Only two concentrations used, and it was not possible to verify the dose-response relation. Cytotoxicity was not reported, but the concentration used was compatible to that in other studies. Without valid OECD guideline.	Genotoxicity	POSITIVE
Wang et al., 1987	45%	<i>In vitro</i> Exchange of sister chromatids in trachea epithelial cells of rat	Only two concentrations used, and it was not possible to verify the dose-response relation. Without valid OECD guideline.	Genotoxicity	POSITIVE
Tanaka and Amano, 1989	NI	<i>In vitro</i> Exchange of sister chromatids in lung cells of Chinese hamster	Cellular viability was not assessed, but the concentration used was compatible to that in other studies.	Genotoxicity	POSITIVE
Ribas et al., 1997	99%	<i>In vitro</i> Exchange of sister chromatids in human lymphocytes	The age of the lymphocyte donors was not described.	Genotoxicity	POSITIVE
Benigni et al., 1979	NI	<i>In vitro</i> Non-scheduled synthesis of DNA in human cells similar to epithelial	-	Genotoxicity	POSITIVE
Petrovska, 1999	NI	<i>In vitro</i> Comet assay in HeLa and Hep G2 cells	-	Genotoxicity	POSITIVE
Parry, 1977	PF	<i>In vitro</i> Mitotic recombination in <i>Saccharomyces cerevisiae</i>	Without valid OECD guideline.	Genotoxicity	POSITIVE
D'Souza et al., 2006	~ 100%	<i>In vivo</i> Morphological alterations in germinative cells of rat	Selection of maturation stages did not comprehend all phases.	Induction of morphological alterations in spermatogenesis	POSITIVE
Rios et al., 1995	PF	<i>In vivo</i> Morphological alterations in the spermatogenesis in mice	No positive control was used in the assay.	Induction of morphological alterations in spermatogenesis	POSITIVE
Rios et al., 1995	PF	<i>In vivo</i> Chromosome aberration in bone medulla cells	No positive control was used in the assay. Inclusion of the number of gaps in the analysis.	Chromosome aberration	INCONCLUSIVE
Salam et al., 1993	PF	<i>In vitro</i> Genic mutation <i>Saccharomyces cerevisiae</i>	Very low purity. Lack of positive control. The induction of genic conversion, reverse mutation	Point mutation and chromosome aberration	INCONCLUSIVE



			and mitotic recombination observed only at high concentrations, in which survival is very much reduced. Without valid OECD guideline.		
Nicotera, 1985	NI	<i>In vitro</i> Exchange of sister chromatids in fibroblasts of Chinese hamster	Reduction of the mitotic index at the highest concentrations, and only one analyzable concentration was left.	Genotoxicity	INCONCLUSIVE

Legend: PF: Formulated Product; NI: Not Informed; Blue: Negative results for mutagenesis; Red: Positive results for mutagenesis; Brown: Inconclusive results, which do not provide evidence for the mutagenicity classification of the active substance; Purple: genotoxicity tests and/or tests carried out with PF.

Thus, the results from the mutagenesis tests reveal that Paraquat is able to provoke chromosome aberrations either *in vitro* or *in vivo* in different species (*in vitro* in mammals' and human cells; *in vivo* in drosophila and mammals) and by different exposure routes (intraperitoneal and dermal). This conclusion, although with some small differences, is according to the interpretations of the mutagenesis studies carried out by other international agencies, such as the Food and Agriculture Organization of the United Nations – FAO), the Joint FAO/WHO Meeting on Pesticide Residues – JMPR, USEPA and EFSA.

USEPA reported that Paraquat is weakly positive in a test of mouse lymphoma L5178Y with metabolic activation, inducing mutation in the cells exposed to Paraquat for two hours. Paraquat also presented a positive result with chromosomal damage in human cells in a cytogenetic test with and without metabolic activation and it was positive in the sister chromatid exchange test with and without metabolic activation. On the other hand, Paraquat was not mutagenic in the Ames tests and it was negative for chromosome aberrations in rat bone medulla. It also considered that there was no evidence of mutagenicity in mice in the lethal dominating test and there was no induction of unscheduled DNA synthesis in hepatocytes *in vitro* and *in vivo* (USEPA, 1997).

Regarding *in vitro* genotoxicity of Paraquat, the EFSA Reassessment Committee concluded that it has mutagenic potential in L5178Y cells with metabolic activation, it does not induce unscheduled DNA synthesis in hepatocytes, but it is clearly clastogenic in pulmonary cell of Chinese hamster with and without metabolic activation. Despite of these positive results, the European Union considered Paraquat non-genotoxic *in vivo*, because this substance did not present clastogenic effects in rat and mouse bone medulla cells in a micronucleus test, it did not induce unscheduled DNA synthesis in hepatocytes and there is no evidence of induction of effects in fertility or increase of the incidence of lethal dominating mutations in doses up to 4 mg/kg p. c./day in rats (Pesticide Safety Directorate, 2002). Therefore, EFSA concluded that Paraquat does not present genotoxic risk *in vivo*, despite of the positive results *in vitro*.

However, it shall be pointed out that the assessments made by USEPA and EFSA are old and thus, they do not consider more recent genotoxicity and mutagenicity studies available in the literature.

FAO reported positive results *in vivo* for genotoxicity in a cytogenetic study with human lymphocytes and a sister chromatid exchange test in lung fibroblasts of hamsters, both carried out according to the OECD protocols (FAO, 2003). In addition, JMPR reviewed a big part of the genotoxicity studies and reported that many consistent positive results were obtained in studies for damage to DNA in mammals' cells (chromosome aberration, cytogenetic studies, chromosomal damages, sister chromatid exchange, unscheduled DNA synthesis and DNA break in comet tests) and concluded that these results indicate *in vitro* genotoxic potential of Paraquat (Marrs and Adjei, 2003). For JMPR, Paraquat is clastogenic in high concentrations *in vitro*. JMPR emphasizes that the action mechanism of paraquat includes the generation of free radicals and the oxidative stress and that the non-observation of genotoxic effects in some tests, especially those with low concentrations of the active ingredient, may result from the antioxidant defense mechanism of the body. However, in its report, JMPR makes an exception reinforcing that the *in vivo* tests for micronucleus formation has been positive in mice, one with oral administration and two with intraperitoneal administration and, although the dose used has been considered high, it is possible to conclude that Paraquat can also induce chromosomal damages *in vivo*; however, it has been considered unlikely for it to cause damages to human beings in dietetic concentrations (Marrs and Adjei, 2003).

JMPR is right when concluding that it is unlikely mutations to occur in humans in dietetic concentrations, because it is not expected that Paraquat will leave residues in the food (this discussion shall be approached more deeply later); however, occupational exposure is extremely relevant, because the workers are in direct contact with the product and, as discussed above, dermal exposure is considered a concern by the main international regulatory agencies (USEPA, 2016b and PMRA, 2015).



The chromosome mutation observed for Paraquat would not be a concern if it occurred only in somatic cells, once the potential to provoke chromosome aberrations does not seem to be associated to any severe chronic event (the results from the carcinogenicity studies did not reveal any significant alteration). On the other hand, the only study available for assessment of the mutagenic potential in germinative cells was considered inconclusive given the low concentration of the active substance (use of commercial product) and the inconsistency of the results (significant difference among some groups, but with big variety of responses). There are also studies assessing the Paraquat action under the morphology of the spermatozooids with clearly positive results. Although the morphological alterations do not enable a definite conclusion about the genotoxic action of Paraquat on the germinative cells, they provide sufficient evidence that the substance reaches the testicles (Yauk et al. 2015).

When there is no conclusive data about the mutagenic potential of a substance, as for Paraquat, it is considered that the effect observed in somatic cells is sufficient to characterize the mutagenic potential of the substance in germinative cells (EU, 2013). IPCS - International Program on Chemical Safety also harmonized its understanding about mutagenicity this way, considering that the substances shall be considered mutagenic for germinative cells, when there are positive results for mutagenic effects in somatic cells *in vivo*, with potential to affect germinative cells confirmed by evidences, which show occurrence of exposure of the germinative cells to the mutagen. In these cases, it shall be assumed that the substance has mutagenic potential for the germinative cells and represents risks to the future generations (Eastmond et al., 2009).

This definition of mutagenicity reflects the current scientific understanding and its normative adoption has been proposed in RDC to replace Ordinance No. 03, dated January 16, 2002 (CP No. 87 dated October 02, 2015). Thus, based on this understanding, it can be concluded with the currently available evidences that Paraquat is mutagenic also for germinative cells, because there is a proof of somatic mutation *in vivo* and the Paraquat action on spermatozooids and there is no consistently negative study proving the lack of mutagenic effect on the germinative cells.

Although Decree No. 4.074 dated January 4, 2002 defines that a pesticide shall be forbidden in Brazil when it is considered mutagenic, able to induce mutation in at least two tests, one of them to detect genic mutations and the other to detect chromosomal mutations (Item V, Art. 31), Law No. 7.802 dated July 11, 1989 determines that it is forbidden to register pesticides, which reveal mutagenic characteristics, according to the results from **up-to-date experiences in the scientific community**. Currently, the concern regarding the mutagenicity of a substance is not restricted to assessment of occurrence of point or chromosomal mutations, because it is important to assess the consequence from the mutation in the somatic and the germinative cells, i.e., its potential effects on the exposed population's health. In somatic cells, as explained above, there is no great concern about the mutagenic potential of Paraquat, once no important effects have been observed in the chronic carcinogenicity studies. However, it was not possible to discard the mutagenic potential of Paraquat in germinative cells based on the available studies. Thus, severe effects resulting from induction of mutation in germinative cells could place the exposed population's health at risk. It shall be pointed out that the population considered exposed would be only the rural workers, because it is not expected for Paraquat to leave residues in food.

The relevance of the mutations in germinative cells is evident considering the number of cases of dominating, recessive autosomal genetic diseases related to sex. Many genetic disorders are caused by recessive mutations (cystic fibrosis, phenylketonuria, Tay-Sachs disease, among others). Most of these mutations are inherited and expressed when the recessive genes are received from both parents. New mutations have higher importance in incidence of dominating diseases rather than recessive, because a single mutation is sufficient to express the diseases already in the first generation. For dominating diseases with mild effect, reduced penetrance or manifestations at more advanced age, the contribution of the parental generation seems higher than for new mutations. In addition to the occurrence of inherited diseases, genic mutations in germinative cells contribute to alterations in fertility, including multi-factor disorders with delayed manifestation, such as heart disease, high blood pressure and diabetes. As a difference from point mutations, which are generally inherited from past generations, a big part of the chromosomal abnormalities looks like new mutations in the parents' germinative cells (Cassarett and Doull's, 2001). Unlike the somatic mutation, which is generally associated to carcinogenesis, a single mutation in a germinative cell may lead to a big variety of diseases, in addition to being the primary cause of embryonic and fetal death. It is recognized that new mutations in germinative cells contribute to neurological disorders, cancer and many other diseases (Yauk et al.; 2015).

Considering the expose de above, Paraquat shall be considered mutagenic although it induces only chromosomal mutation, because:

I – The analysis of the mutagenicity studies both in the scientific literature and in the tests carried out by the registering companies showed that there is sufficient



evidence to prove the potential of Paraquat to induce chromosome aberrations in somatic cells *in vitro*, *in vivo*, in different species and by different exposure routes (including dermal).

II – When there is no conclusive data about the mutagenic potential of a substance in germinative cells, as for Paraquat, it shall be considered that the effect observed in somatic cells is sufficient to characterize the mutagenic potential in germinative cells.

III - It was not possible to prove the inexistence of mutation in germinative cells.

IV - Exposure to Paraquat reaches the testicles, affecting the spermatozooids, and provokes somatic mutation *in vivo*, therefore, it is considered that Paraquat is mutagenic for germinative cells.

V - Mutagenic effects in germinative cells lead to severe consequences (alterations in fertility, malformations, embryonic death, neurological disorders, congenital abnormalities, including multi-factor disorders with delayed manifestation, such as heart disease, cancer, high blood pressure and diabetes), representing high risk to the exposed population's health.

VI - The main international regulatory agencies and the present opinion concluded that dermal exposure of human beings is relevant and most important than animal exposure, therefore, rural workers (exposed population) are susceptible to the mutagenic effects of Paraquat.

VII - Law 7.802 dated 1989 determines that it is forbidden to register pesticides, which reveal mutagenic characteristics, according to the results from **up-to-date experiences in the scientific community**.

It shall be pointed out that the potential to provoke somatic chromosome aberrations does not seem to be associated to any severe chronic event, because the results from the carcinogenicity studies do not reveal any significant alteration. However, as there is no evidence of lack of severe effects for chromosomal mutations in germinative cells, the only alternative not to propose prohibition of the active substance Paraquat, based on its mutagenic potential, would be obtaining of clearly negative results in tests for assessment of the mutagenic potential of Paraquat in germinative cells, carried out according accepted international protocols.

3. Neurotoxicity

TECHNICAL REASSESSMENT OPINION No. 01 DATED 2015/GGTOX/ANVISA concluded that the studies in animals reveal causality relation between the occurrence of effects typically associated to Parkinsonism and the exposure of laboratory animals to Paraquat based on the studies from the literature and the studies presented by the registering company.

However, the Task Force considers that there were no effects related to Parkinsonism after oral administration of Paraquat, even when using doses, which are significantly higher than the doses currently used to determine occupational and dietetic exposure. It also determined that Technical Reassessment Opinion No. 01 dated 2015/GGTOX/ANVISA emphasized a lot the loss of neuronal cells observed in one of the studies with stereological assessment and did not emphasize the other important toxicological parameters (neurochemical and neuropathological). Furthermore, the company argued that in order to be considered important, the loss of neurons should be reproducible. In relation to all these arguments of the Task Force, it shall be clarified that many studies were assessed either in the dossier or in the scientific literature and a big part of them showed clear effects in different parameters related to Parkinsonism and not only neuronal depletion as already discussed in TECHNICAL REASSESSMENT OPINION No. 01 DATED 2015/GGTOX/ANVISA.

The Task Force also argues that only results, which are statistically significant, shall be considered in the analysis, because the effect can only be considered when statistical difference is achieved. Regarding this aspect, it is important to highlight that the assessment of the result from the studies shall take all data available into consideration, in addition to the statistical differences: results from other studies, biological significance, response dose and reproducibility. Actually, the exclusive assessment of these studies would not clearly show a statistical difference and would not be sufficient to conclude about the relation between Paraquat and Parkinsonism. However, statistical trends were observed for some effects in the studies of the registering companies, which, when assessed together with the several results from studies in the scientific literature, provide sufficient evidence to conclude that exposure to Paraquat would lead to characteristic Parkinsonism effects in animal models.

The Task Force also discussed about other aspects they consider insufficient to conclude about the alterations provoked by Paraquat in the nervous system, such as irrelevant exposure route, time of



exposure, sensitivity of the animal models, dopamine levels found, administered doses. All these aspects have already been assessed and deeply discussed in TECHNICAL REASSESSMENT OPINION No. 01 DATED 2015/GGTOX/ANVISA and therefore, none of the presented arguments change the initial conclusion of the assessment regarding the relation between Parkinsonism and Paraquat in the studies carried out in laboratory animals.

Unlike the affirmed by the Task Force, it cannot be considered that the animal models are poor, according to discussion made by Dr. Estefânia Moreira in the technical panel for assessment of Paraquat and already presented in Technical Reassessment Opinion No. 01 dated 2015/GGTOX/Anvisa, because different effects related to Parkinsonism have been observed by independent research groups.

Among the already observed effects, which relate Paraquat to Parkinsonism, the following can be mentioned: statistically significant decrease of neurons immune-marked for tyrosine hydroxylases (TH) in the substantia nigra (SN) and considerable loss of dopaminergic neurons (25-65%); statistically significant alterations of the motor function; significant induction of oxidizing stress in the brain (activation of microglia, decrease of anti-oxidizing agents, increase of pro-oxidizing markers); reduction of dopamine or alteration in its metabolism; neuronal accumulation of α -synuclein; decrease of proteasome activity (Brooks et al., 1999; Manning-Bog et al., 2002; McCormack et al., 2002; Manning-Bog et al., 2003; McCormack & Di Monte, 2003; McCormack et al., 2005; Ossowska et al., 2005a; Ossowska et al., 2005b; Ossowska et al., 2006; Choi et al., 2006; Fernagut et al., 2007; Purisai et al., 2007; Kang et al., 2009; Somayajulu-Nitu et al., 2009; Kang et al., 2010; Wills et al., 2012). Furthermore, several *in vivo* studies with non-mammals, such as nematodes, fish and drosophila (Jimenez-Del-Rio et al., 2008; Inamdar et al., 2012; Bortolotto et al., 2014; González-Hunt et al., 2014; Cassar et al., 2015) and *in vitro* and *in situ* studies (Choi et al., 2016) provided evidence of effects associated to Parkinsonism resulting from exposure to Paraquat observed in mammals *in vivo*.

It shall be pointed out that although the Task Force insists in arguing that the studies carried out by the group, which the researcher Mona Thiruchelvan took part in, shall be disregarded from the analysis as it has been proven that this researcher changed its test data about Parkinson's with paraquat; all results of the other researchers from this research group cannot be invalidated. The data corrupted by Mona Thiruchelvan were identified and did not ground the conclusion of the reassessment opinion. It would not be correct to disregard the results of the other researchers for assuming that all of them had been falsified as well. In addition, it is necessary to point out that other independent research groups presented similar results.

From the observations of the experimental studies, it is possible to conclude that the evidenced effects are similar to the alterations observed in patients with Parkinson's disease, which reinforced the idea that Paraquat is a risk factor for this disease. It shall be emphasized that the design of the experimental studies does not allow establishment of NOAEL for degeneration of dopaminergic neurons and that the brain elimination half-life of Paraquat makes its concentration higher in the brain after repeated administrations (even if in 1-week intervals). In addition, it was found that the brain concentrations achieved after oral and intraperitoneal administration are similar. However, the relevance of these experimental findings in the extrapolation for human beings depends on the analysis of several existing epidemiological studies.

According to the Task Force, the epidemiological studies do not allow concluding that studies with Parkinson's and Paraquat reveal unacceptable risk to support the recommendation for register cancellation, because it considers that the epidemiological studies are inconclusive. It was argued that TECHNICAL ASSESSMENT OPINION No. 01 DATED 2015/GGTOX/ANVISA did not take important studies into consideration for epidemiological analysis, such as Brent and Schaeffe (2011) and Tomenson and Campbell (2011). It was also argued that the Mandiel's and collaborators' study (2012) indicating the faulty issues of the epidemiological studies FAME (Farming and Movement Evaluation Study) by Tanner and collaborators (2011) in relation to the methodological aspects, the different criteria of eligibility for selection of controls, the execution of controls for developing Parkinson's, the low participation rate, the much higher proportion of interviewed people replaced among cases than in the controls, the substantial differences in the participation rates between cases and controls and the lack of estimations based only on exposure reported at the time of enrolment, has not been assessed.

The Task Force also argues that no robust assessment of the exposure for most epidemiological studies has not been made; however, it mentioned two quite limited studies to prove that there is no relation between exposure to Paraquat and the Parkinson's disease (Brent and Chaeffer, 2011; Tomenson and Campbell, 2011).

Brent and Schaeffer (2011) assessed the occurrence of the Parkinson's disease in patients, who recovered or lived for up to thirty days after intoxication by Paraquat. Considering that Parkinson's is a neurodegenerative disease, with slow and progressive evolution, it cannot be expected that a study with such experimental design would provide any signs of relation between Paraquat and the Parkinson's disease, therefore, this study has been considered inadequate for discussion of this toxicological aspect in the reassessment of the active substance Paraquat.



Tomenson and Campbell (2011) carried a study out assessing the mortality of workers in a Paraquat factory by Parkinson's and other causes; Although the Task Force argues that the authors clearly show that exposure to Paraquat did not cause Parkinson's in the workers, this study is extremely fragile and was not considered relevant to assess the role of Paraquat in the Parkinson's pathogenesis. This study included assessment of workers, who worked between 1961 and 1995, and workers, who were followed up until 2009. Different workers with different functions in the factory were included. The study itself mentions that there is limited data about the exposure of the workers, and it is not sufficient to make a quantitative or qualitative assessment of the exposure. The article also reveals that it is likely for the workers to have been exposed to medium or high levels of Paraquat after 1980. Furthermore, the time and the level of exposure are not known. The causes of death have been assessed by means of death certificates, in which it is not expected Parkinson's to be indicated as cause of death, because the death, even when related to Parkinson's, could not have been declared as complication of the disease. In addition, many workers might have not had diagnosis of Parkinson's during their lives and obviously, this would not be reported in the death certificate, where the direct cause of death is stated.

In the contributions to CP, the need to include discussion about the Elbaz and collaborators (2009) study has also been argued; in this study, no association between the Parkinson's disease and exposure to Paraquat has been found. The authors of the study explain that the observation of this results, which is in conflict with the other epidemiological studies, may have occurred because in France, Paraquat is used mainly as non-selective herbicide to kill weeds around the plantations, resulting in low levels of exposure. In addition, if there is gene-environment interaction, paraquat can be associated to the Parkinson's disease only in more susceptible individuals. Opposite results have been found by Ntzani and collaborators (2013), who analyzed different epidemiological studies to assess the association of Paraquat to Parkinson's, and observed that these epidemiological studies indicated such association. Of course, it is inherent to any epidemiological assessment to have some limitations in the assessment made by Ntzani and collaborators (2013), which, although do not invalidate the analysis carried out, impede assertive conclusion about the causality between exposure to Paraquat and Parkinson's disease. Thus, it would be necessary to include carefully outlines epidemiological studies. For example, in the Ntzani study, it would be important to assess more complete epidemiological studies with detailed description of the use of IPE to assess the reduction of this association, the use of more grounded data than using of questionnaires and the reduction of confusion factors.

Recently, Furlong and collaborators (2015) assessed whether the use of gloves and hygiene habits modify the association between pesticides and the Parkinson's disease. FAME (Farming and Movement Evaluation) is a case-control study within the Agricultural Health Study (AHS). The use of IPE and hygiene practices were determined by a questionnaire and the use of gloves below 50% of the time and in 50% of the time was separated. High association between the Parkinson's disease and the use of gloves for time below 50% was found out, and this association did not occur when gloves were used 50% of the time. The study did not determine consistent relation between the adoption of hygiene practices and the reduction of the association between Parkinson's and exposure to Paraquat. The authors discussed that gloves are determining factor for dermal exposure, not using gloves exposes the worker at higher doses of the herbicide. Thus, it can be suggested that in the epidemiological studies, in which no association between Paraquat and the Parkinson's disease has been found, the results may have been masked by the fact that using IPE is common in the assessed community. The authors also discuss that this results may not be directly pursuant to the use of gloves, but to the adoption of another behavior, because people, who use gloves, tend to handle pesticides in a safer way, reducing their exposure. It shall be pointed out that as any epidemiological study, this study presents relevant limitations, such as the use of a small number of cases (although bigger than that in many studies) and broad confidence interval, among others. Despite of such limitations, the study is considered relevant for analysis in the reassessment of Paraquat.

Regarding the epidemiological results from AHS, among them FAME (Tanner et al., 2011), USEPA clarified at the beginning of its reassessment of Paraquat that these findings will be considered in the reassessment, as well as those, which demonstrate potential relation between exposure to Paraquat and the Parkinson's disease (USEPA, 2011). AHS is considered a high-quality prospective study with the following strong points: big sample, high response obtained and the fact of being prospective. Among its weak points, the following can be mentioned: very homogeneous sample (white men), lack of knowledge about the frequency of exposure or application, among others.

The results of Tanner and collaborators (2011) and Kamel and collaborators (2007), coming from AHS, reveal that there was an increase in the probability of occurrence of Parkinson's disease among the workers, who used Paraquat, which supports the experimental toxicological findings (USEPA, 2012). This assessment is still in progress at USEPA and the final conclusion has not been disclosed yet. However, in the public consultation dated March 2, 2016 (USEPA, 2016b), USEPA clarified that for the time being, only cases related to incidents have been assessed. However,



USEPA pointed out the results from the AHS studies again, explaining that a modest association in relation to asthma or chronic bronchitis had been verified in both AHS studies, considering that the data did not provide strong evidence of such relation; but in relation to Parkinson's, the probability ratio had been considered high.

Thus, it can be observed that Paraquat has been associated to the Parkinson's disease in epidemiological studies developed by different research groups and in different populations (Liou et al., 1997; Costello et al., 2009; Tanner, 2011; Kamel, 2007; Furlong et al., 2015) and in recent meta-analyses (Pezzoli and Cereda 2013; Ntzani et al., 2013), although some studies have observed lack of such association (Engel et al., 2001; Hertzman et al., 1994). A conclusive analysis about the causality of exposure to Paraquat in the pathogenesis of the Parkinson's disease would be possible only by means of development of better outlined epidemiological studies, which may demand a long time. It is important reminding that the limitations of the epidemiological studies may make interpretation of the results difficult due to the possible effects of confusing factors related either to interest exposure or risk of disease, which limits the capacity of the epidemiological studies to provide convincing evidence of causality (USEPA, 1994).

It is considered that causality is suspected if different studies show consistent findings; if the association between the agent and the risk of disease is strong (i.e., there is high ratio of chances) and if the association is in accordance with the biological theory. Proving animal data helps defending causality, especially upon establishment of biological reasonability and potential mechanisms. However, failure to detect association in an epidemiological study is not sufficient to disregard it, especially if the studies have limited statistical power due to the reduced size of the sample or the reduced follow-up time. Thus, in assessment of the weight of the evidence of epidemiological studies related to exposure to a substance, it is necessary to make a subjective judgment of the weight of the data from several studies and their conflicting conclusions. For Paraquat, the results from the epidemiological studies are conflicting and there are confusion factors either in the studies showing causality or in those that don't. However, there are several studies and meta-analyses, which demonstrate a relation between Paraquat and the Parkinson's Disease and the assessment of the inconsistencies does not allow to disregard such association. The confidence in epidemiological studies, when there is no direct measurement of exposure, is challenging; however, when the fact that the studies carried out in animals by different research groups are consistently positive and the fact that there is a big number of publications about the Paraquat action mechanism in the Central Nervous System (Choi et al., 2016) are added, it can be considered that, based on the currently available data, there is bigger evidence of causality relation between Paraquat and the Parkinson's Disease rather than evidence of lack of relation.

This conclusion can be proven by a study delivered to Anvisa by Syngenta (Breckenridge et al., 2016) with the purpose to assess the weight of the evidence of the association between the Parkinson's Disease and different aspects related to life in field, including exposure to Paraquat. In the results from the meta-analysis, a positive relation of association between Parkinson's and Paraquat was observed, with significant difference in the risk ratio. The authors concluded that there might be a risk factor associated to the rural life style, the use of pesticides (Paraquat was one of the assessed) and the consumption of water from wells, but that the results until today have not identified such factors conclusively, proposing the development of more studies, in order to characterize such relations.

Breckenridge and collaborators (2016) observed risk ratios (RR) for the Parkinson's Disease significantly higher than 1.0 for exposure to Paraquat when compared to lack of exposure (RR=1.69 and RR=1.47; IC=1.01-2.13; I²=69.9%, p=0.0001 with fixed and random assessment of the effects, respectively). When in the assessment the high use of Paraquat was compared to lack of use, this ratio was even higher, 1.75 (IC=1.19-2.57, I²=78.9%, p=0.0026) and 1.99 (IC=0.84-4.71) for fixed and random effects, respectively. These results confirm the results from the meta-analyses, where relation between Parkinsonism and Paraquat was observed, so that the authors of this study did not affirm for sure that these results reveal non-association between Paraquat and Parkinson's; they only conclude that it is not possible to observe the same consistently negative relation of the association between smoking and the Parkinson's disease for Paraquat and other assessed factors.

In the assessment of the weight of the evidence, Breckenridge and collaborators (2016) used only one qualitative method (Bradford Hill), not considering all available literature about the subject, they did not use even the results from their own meta-analysis, because they considered a single study as relevant for the assessment of the evidence weight. In a meta-analysis, classification of the studies attributing different weights according to their quality is important. However, the classification of the studies by Breckenridge and collaborators (2016) was made exclusively based on the way of diagnosis (clinical by a specialist) and the availability of individual exposure data. Thus, only one study was classified as level 1 and used to grant strength to the assessment of the evidence weight. As discussed above, the studies cannot be disregarded based on a few limitations. The restriction made by Breckenridge and collaborators (2016) was disproportional and good-



quality and internationally recognized studies, such as the AHS, were disregarded in the attribute strength of assessment of the evidence weight. In chart 5, assessment of the evidence weight made by the authors of the study, using the same method, and the assessment made based on the discussions in this opinion and considering all results from the meta-analysis by Breckenridge and collaborators (2016), and not only a single epidemiological study, can be observed.

Chart 5. Comparison of the assessments of the evidence weight of the causality relation between the Parkinson's Disease and the exposure to Paraquat, according to the Bradford Hill qualitative method.

Attribute	Concept	Breckenridge et al., 2016	Anvisa
Strength	The bigger the risk ratio, more probable the causality relation.	RR = 0.90 Without statistical difference , because only one epidemiological study was used for the assessment of this attribute, disregarding relevant studies.	RR = 1.69 (use of Paraquat) RR = 1.75 (high use of Paraquat) With statistical significance obtained from the meta-analysis of Breckenridge et al., 2016.
Consistency	Consistency of the associations between exposure and disease observed in independent studies indicates causality.	Inconsistent. All epidemiological results with RR significantly higher than 1.0 were disregarded.	Consistent. (Costello et al., 2009; Liou et al., 1997; Tunner, 2011; Kamel, 2007; Furlong et al., 2015)
Specificity	A specific association for given group of individuals or for a disease.	Highly specific.	Highly specific.
Temporality	For a factor to be causal, it shall precede the disease. Single condition considered essential.	Not established. All results from studies from the literature were disregarded.	Confirmed by consistent studies in the literature, demonstrating that PQ promotes death of dopaminergic neurons (precursor effect of Parkinson's) *
Biological Gradient	If the risk to develop the disease increases with the dose (exposure), the association is more likely to be causal.	Not existing. There is no level 1 study with high use of Paraquat.	Existing. RR = 1.69 (use of Paraquat) RR = 1.75 (high use of Paraquat)
Reasonability	If it is reasonable that the agent causes the disease, based on the current knowledge, the association is more likely to be causal.	Uncertain. Arguments that there are results demonstrating that Paraquat has oxy-reduction potential and could provoke death of dopaminergic neurons, but that these are controversial.	High. Data from <i>in situ</i> , <i>in vitro</i> and <i>in vivo</i> studies of different research groups prove that Paraquat is able to provoke several effects related to parkinsonism with statistical significance. *
Coherence	Whether the data is consistent with other data, for example, the known distribution of the disease in the population.	Not existing.	Unavailability of data for such comparison.
Experimental evidence	Causal inference is more probable if it is supported by experimental data. For example, if the implementation of a preventive measure, such as the reduction of the exposure results in reduction of the risk.	Inexistent studies. The Furlong and collaborators (2015) study was disregarded.	There is an experimental evidence. Furlong and collaborators (2015) assessed whether the use of gloves and hygiene habits modified the association between pesticides and the Parkinson's Disease, finding out high association when gloves were used for less than 50% of the time, but the association did not exist, when gloves were used 50% of the time.
Analogy	Analogy between the current association with other exposure-disease relation proves causality inference.	Existing.	Existing. MPTP is a substance, which is structurally similar to Paraquat and causes Parkinson's.
CONCLUSION		No causality relation between the Parkinson's Disease and exposure to Paraquat.	There is causality relation between the Parkinson's Disease and exposure to Paraquat.

Legend: RR: Risk Ratio; PQ: Paraquat. In red: attributes associating PQ to the Parkinson's Disease. In green: attributes that do not associate PQ to the Parkinson's Disease. For RR, the data of the meta-analysis of Breckenridge et al., 2016 was used. Even higher values were found in the epidemiological studies and in other meta-analysis.
* Brooks et al., 1999; Manning-Bog et al., 2002; McCormack et al., 2002; Manning-Bog et al., 2003; McCormack & Di Monte, 2003; McCormack et al., 2005; Ossowska et al., 2005a; Ossowska et al., 2005b; Ossowska et al., 2006; Choi et al., 2006; Fernagut et al., 2007; Purisai et al., 2007; Kamel et al., 2009; Somayajulu-Nitu et al., 2009; Kang et al., 2010; Li et al., 2012; Wills et al., 2012.

It is possible to find out that the epidemiological studies provide strong sings of causality between the exposure to Paraquat and the Parkinson's disease. Although one study indicates that the use of IPE can be sufficient to mitigate this risk (Furlong et al., 2016), this result is unique and there is no sufficient knowledge in the scientific literature about the Parkinson's Disease and about the action mechanism of Paraquat in human beings to ground the determination of a safe level of exposure.



Therefore, it is not possible to affirm that use restrictions would be sufficient to protect the health of rural workers, who handle Paraquat.

Based on the currently available evidences, there is a strong evidence weight (studies in animals and epidemiological studies) indicating that Paraquat is associated to the development of the Parkinson's Disease in humans. Considering that the consequences from the exposure to Paraquat in human beings are more relevant than in animals, because Parkinson's is a severe, progressive disease, whose treatment only retards the symptoms, but which is not able to revert the condition, i.e., there is no cure, and in face of the recognized severity of this disease for human beings, it is possible to conclude that Paraquat may reveal to be more hazardous for human beings than the tests with laboratory animals can demonstrate and there is no antidote or efficient treatment. Therefore, there is classification of Paraquat in the prohibitive criteria for registration determined by items "b" and "e", §6, article 3, Law 7.802, dated 1989.

It is expected that exposure to Paraquat would be restricted to workers, who manipulate this pesticide, and the population in general would not be susceptible to exposure upon consumption of food, because there is no evidence that the use of Paraquat leaves residues. Thus, due to the fact that the risk related to Paraquat is restricted to a part of the population, specific measures for mitigation of exposure could be determined as an alternative to the prohibition of this active substance. However, the proposal of measures to mitigate the risk to the workers is extremely difficult and cannot be grounded technically, because there is no scientific evidence about which level of exposure would be sufficient to assure the safe use of this pesticide. Therefore, even if some measures may result in lower exposure of workers to Paraquat, protection against possible development of the Parkinson's disease cannot be fully guaranteed. The clarification of these aspects also demands scientific progress, which demonstrates the exact action mechanism of Paraquat in humans and whether there are safe levels of exposure for protection against this effect.

In face of the exposed above, for adoption of any measure, either register cancellation or proposal of restrictions, it is necessary to consider that:

- I. There is a strong evidence (studies in animals and epidemiological studies) indicating the causality relation between the Parkinson's Disease and the exposure to Paraquat;
- II. The Furlong and collaborators (2015) study indicates that the results, which do not show association between PD and Paraquat may have been masked by the use of IPE;
- III. The Furlong and collaborators (2015) study indicates that the use of gloves impedes the association between the use of Paraquat and the Parkinson's Disease observed initially;
- IV. The only exposure considered worrying from the health point of view is the occupational, because there is no evidence that Paraquat leaves residues in food;
- V. **The development of the Parkinson's Disease, according to the results from the studies in animals, probably demands continuous exposure and high doses. However, in the epidemiological studies, it is observed that the current exposure would already be sufficient, showing that the consequences for human beings may be more severe than the studies in animals can demonstrate;**
- VI. Recent assessment of the risk after occupational exposure to Paraquat using the American model PHED indicates that the exposure to Paraquat in the different assessed scenarios does not extrapolate AOEL;
- VII. The main international regulating agencies and the present opinion concluded that dermal exposure of human beings to Paraquat is relevant and more important than exposure of animals, therefore, rural workers (exposed population) are susceptible to the mutagenic effects of Paraquat;
- VIII. With the current scientific knowledge, it is not possible to determine AOEL for parkinsonism (and it is not expected that such knowledge will be available in short terms);
- IX. Parkinsonism is classified in two prohibitive criteria for registration of pesticides in Brazil: lack of antidote and efficient treatment and more severe consequence for human beings than the tests in animals can demonstrate;
- X. USEPA proposed adoption of extremely restrictive measures, such as use of closed-system and inviolable packages and exclusion of backpack spraying application, which may result in big reduction of the exposure of the users;
- XI. There are no scientific grounds to propose restrictive measures for exposure to Paraquat, because there is a single study indicating that the reduction of the exposure seems to be sufficient to limit the development of the disease.



III – CONCLUSION

1. Acute intoxication and exposure

The cases of acute severe occupational intoxication by agricultural use of Paraquat, as stipulated, are not predominating and tend to be moderate or slight, with antidotes and efficient treatment able to revert the damages to health pursuant to occupational exposure to products made based on this pesticide. Therefore, this toxicological aspect cannot be classified in any criteria for registration prohibition in Brazil. However, occupational exposure to Paraquat is relevant, mainly due to the evidences of higher human sensitivity to dermal exposure to this pesticide, with possibility for system absorption by this route of exposure. Furthermore, the cases of acute severe intoxication by accidental ingestion of Paraquat or attempt to suicide are worrying. Considering these facts, Canada and the United States proposed adoption of restrictive measures for reduction of the cases and the severity of the intoxications associated to Paraquat.

Considering all these aspects related to acute intoxication by Paraquat, even it was not concluded that this active substance was classified in the criteria considered prohibitive for registration, quite restrictive risk-mitigation measures would be indispensable.

2. Mutagenicity

With the currently available scientific evidences, it was verified that:

I – The analysis of the mutagenicity studies either in the scientific literature or in the tests carried out by the registering companies showed that there is sufficient evidence weight to prove the potential of Paraquat to induce chromosome aberrations in somatic cells *in vitro* and *in vivo* in different species and by different exposure routes (including dermal).

II – When there is no conclusive data about the mutagenic potential of a substance in germinative cells, as for Paraquat, it shall be considered that the effect observed in somatic cells is sufficient to characterize its mutagenic potential in germinative cells.

III – It was not possible to prove the inexistence of mutation in germinative cells.

IV – Exposure to Paraquat reaches the testicles, affecting the spermatozoids and provokes somatic mutation *in vivo*, therefore, it is considered that Paraquat is mutagenic for germinative cells.

V – Mutagenic effects in germinative cells lead to severe consequences (alterations in fertility, malformations, embryonic death, neurological disorders, congenital abnormalities, including multi-factor disorders with delayed manifestation, such as heart disease, cancer, high blood pressure and diabetes), representing high risk to the exposed population.

VI – The main international regulatory agencies and the present opinion concluded that dermal exposure of human beings is relevant and most important than animal exposure, therefore, rural workers (exposed population) are susceptible to the mutagenic effects of Paraquat.

VII - Law 7.802 dated 1989 determines that it is forbidden to register pesticides, which reveal mutagenic characteristics, according to the results from **up-to-date experiences in the scientific community**

Considering these aspects, despite that Decree 4.074 dated 2002 determines that a substance is mutagenic when there is a proof of point and chromosome mutation (Item V, Art. 31), it is possible to conclude that this scientific understanding is out-of-date and would allow pesticides with mutagenic potential in germinative cells, able to cause severe effects to health, to be kept on the market. Thus, according to the current scientific knowledge and Law No. 7.802 dated 1989, which prohibits registration of mutagenic pesticides (item c, §6, Art. 2), it shall be considered that Paraquat is mutagenic, representing risk to the population's health.

It is important to point out that the potential to provoke somatic chromosome aberrations does not seem to be associated to any severe chronic event, because the results from the carcinogenicity studies do not reveal any significant alteration; however, there is no evidence of the absence of severe for the chromosome mutations in germinative cells. Thus, the only alternative not to propose prohibition of the active substance Paraquat, based on its mutagenic potential, would be obtaining of clearly negative results in mutagenicity tests with germinative cells, carried out according to internationally accepted protocols.



3. Parkinsonism

Based on the currently available evidences, there is a strong evidence weight (studies in animals and epidemiological studies) indicating that Paraquat is associated to the development of the Parkinson's Disease in humans. Considering that the consequences from the exposure to Paraquat in human beings are more relevant than in animals, because Parkinson's is a severe, progressive disease, whose treatment only retards the symptoms, but which is not able to revert the condition, i.e., there is no cure, and in face of the recognized severity of this disease for human beings, it is possible to conclude that Paraquat may reveal to be more hazardous for human beings than the tests with laboratory animals can demonstrate and there is no antidote or efficient treatment. Therefore, there is classification of Paraquat in the prohibitive criteria for registration determined by items "b" and "e", §6, article 3, Law 7.802, dated 1989.

The monitoring studies actually do not reveal exposure above AOEL; however, it is observed that Paraquat can be absorbed in doses, which, despite of not exceeding AOEL, are worrying due to the evidences associating this active substance with parkinsonism, and it is difficult to affirm that the currently accepted AOEL is a safe limit in relation to the effects from Paraquat on the dopaminergic neurons pursuant to exposure along the rural workers' life.

It is expected that exposure to Paraquat would be restricted to workers, who manipulate this pesticide, and the population in general would not be susceptible to exposure upon consumption of food, because there is no evidence that the use of Paraquat leaves residues. Thus, due to the fact that the risk related to Paraquat is restricted to a part of the population, specific measures for mitigation of exposure could be determined as an alternative to the prohibition of this active substance. However, the proposal of measures to mitigate the risk to the workers is extremely difficult and cannot be grounded technically, because there is no scientific evidence about which level of exposure would be sufficient to assure the safe use of this pesticide. Therefore, even if some measures may result in lower exposure of workers to Paraquat, protection against possible development of the Parkinson's disease cannot be fully guaranteed. The clarification of these aspects also demands scientific progress, which demonstrates the exact action mechanism of Paraquat in humans and whether there are safe levels of exposure for protection against this effect.

In face of the exposed above, for adoption of any measure, either register cancelation or proposal of restrictions, it is necessary to consider that:

- I. There is a strong evidence (studies in animals and epidemiological studies) indicating the causality relation between the Parkinson's Disease and the exposure to Paraquat;
- II. The Furlong and collaborators (2015) study indicates that the results, which do not show association between PD and Paraquat may have been masked by the use of IPE;
- III. The Furlong and collaborators (2015) study indicates that the use of gloves impedes the association between the use of Paraquat and the Parkinson's Disease observed initially;
- IV. The only exposure considered worrying from the health point of view is the occupational, because there is no evidence that Paraquat leaves residues in food;
- V. The development of the Parkinson's Disease, according to the results from the studies in animals, probably demands continuous exposure and high doses. However, in the epidemiological studies, it is observed that the current exposure would already be sufficient, showing that the consequences for human beings may be more severe than the studies in animals can demonstrate;
- VI. Recent assessment of the risk after occupational exposure to Paraquat using the American model PHED indicates that the exposure to Paraquat in the different assessed scenarios does not extrapolate AOEL;
- VII. The main international regulating agencies and the present opinion concluded that dermal exposure of human beings to Paraquat is relevant and more important than exposure of animals, therefore, rural workers (exposed population) are susceptible to the mutagenic effects of Paraquat;
- VIII. With the current scientific knowledge, it is not possible to determine AOEL for parkinsonism (and it is not expected that such knowledge will be available in short terms);
- IX. Parkinsonism is classified in two prohibitive criteria for registration of pesticides in Brazil: lack of antidote and efficient treatment and more severe consequence for human beings than the tests in animals can demonstrate;



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- X. USEPA proposed adoption of extremely restrictive measures, such as use of closed-system and inviolable packages and exclusion of backpack spraying application, which may result in big reduction of the exposure of the users;
- XI. There are no scientific grounds to propose restrictive measures for exposure to Paraquat, because there is a single study indicating that the reduction of the exposure seems to be sufficient to limit the development of the disease.

Considering the current scientific knowledge, it was concluded that Paraquat is mutagenic and that it is risk factor for Parkinson's Disease, with evidence of causality between exposure and the disease. Therefore, the active substance Paraquat is classified in the following criteria that prohibit registration:

- a) Scientific evidences of mutagenicity according to the updated results from experiences in the scientific community (Item c, §6, Art. 3, Law No. 7.802 dated 1989).
- b) Lack of antidote or efficient treatment (Item II, Art. 31, Decree No. 4.074 dated 2002 and Item b, §6, Art. 3, Law No. 7.802 dated 1989).
- c) Evidences that the active substance is more hazardous for the man than demonstrated in laboratory tests with animals (Item VII, Art. 31, Decree No. 4.074 dated 2002 and Item c, §6, Art. 3, Law No. 7.802 dated 1989).

Despite of the classification of the active substance Paraquat in the criteria that prohibit registration, it is necessary to clarify some relevant aspects:

I – New studies about the mutagenic potential in germinative cells, carried out according to internationally accepted protocols and with consistently negative results may reveal the lack of mutagenicity of Paraquat, which would classify it in the register-prohibition criteria determined in Item c, §6, Art. 3, Law No. 7.802 dated 1989).

II. – Considering that Parkinson is a progressive disease and probably demands repeated exposure to Paraquat to be developed, knowing that exposure to Paraquat is possibly restricted to a part of the population (rural workers); and knowing that a recent epidemiological study has indicated that the limitation of the exposure, by means of gloves, showed to be sufficient to disassociate the exposure to Paraquat from the Parkinson's Disease, the possibility to propose restrictive measures to mitigate exposure could be considered. However, it cannot be concluded that it is possible to prevent the development of the Parkinson's Disease with reduction of the exposure to Paraquat based on one study only, because important aspects about Paraquat the action in human beings and the development of the Parkinson's Disease have not yet been clarified. Therefore, with the currently available knowledge, it is not possible to propose scientifically grounded restrictive measures for the mitigation of the risk associated to exposure to Paraquat, because it is not yet possible to determine which the safe levels of exposure are.

Based on the currently available scientific data and considering the classification of the toxicological aspects of Paraquat in the prohibitive registration criteria provided in the Brazilian legislation, I suggest production, export, import, trade and use of all products made based on this pesticide active substance to be forbidden in the country.

This is the opinion for superior consideration.

Brasília, June 13th, 2016

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Juliana Machado Braz

Expert on Regulation and Health Surveillance

(x) Approved

() Not Approved, according to document:

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Brasília, 08/26/2016

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