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Studies on the Persistence of a Commercial Formulation of Chlorpyrifos on an Agricultural Soil from Provincia de Buenos Aires, República Argentina

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Abstract Chlorpyrifos (O, O-diethyl-O-(3,5,6-trichloro-2-pyridyl) phosphorothioate) is a broad-spectrum organophosphate insecticide and acaricide, widely used in our country. Nowadays, it is the principal insecticide in the market employed for agricultural purposes. A number of studies tending to study the affinity of different pesticides with soil have been performed, but only a few refer to chlorpyrifos. Because of its intensive use, a wide range of terrestrial ecosystems may be contaminated with chlorpyrifos, and there is a need to evaluate its environmental behavior and effects. The aim of our work is to study the interaction and persistence of a commercial formulation of chlorpyrifos on an agricultural soil from Provincia de Buenos Aires, Argentina. In this case, recovery percentages increased with the increase of initial concentration of the pesticide until a concentration of about 25 ppm is reached, and then a decrease was observed. The half-time life was not affected by an increase in chlorpyrifos concentration.

Keywords Chlorpyrifos · Pesticides · Solid matrices · Agricultural intensification · Half-time · Pollution

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1 Introduction

Buenos Aires province has undergone, in the last 30 years, a remarkable expansion in agricultural activities based in the incorporation of new technologies; the improved specialization and reduction of diversity to meet the increasing food demand. The extension of the cultivated area brought, as a consequence, an increase in the use of agrochemicals, as the agricultural intensification requires an even more aggressive fight against plagues, parasites, and other harmful factors. Soybean is the crop accounting for the highest proportional percentage of the pesticide market. The main products sold for soybean are, in order of importance, herbicides and insecticides. The principal insecticides in the market are, in order of importance, chlorpyrifos, endosulfan, and cypermethrin (CASAFE 2012). A large amount of agricultural pesticide application never reaches its target organisms but is, instead, dispersed through the air, soil, and water (Moses et al. 1993).

Chlorpyrifos (O, O-diethyl-O-(3,5,6-trichloro-2-pyridyl) phosphorothioate) is a broad-spectrum organophosphate insecticide and acaricide, which is widely used to control insect pests on grain, cotton, fruit, nut, and vegetable crops, as well as lawns and ornamental plants, household pests, and aquatic larvae (Fang et al. 2006). It is also used on sheep and cattle for the control of ectoparasites.

Chlorpyrifos (molecular weight—350.62) has a melting point in the range of 41.5–44 °C (USEPA 1999), a low water solubility (139 mg/l), a high soil sorption coefficient (849 ml/g), and a vapor pressure of 2.49 MPa (25 °C). It has been suspected to accumulate in living tissues, and it may create a potential risk to humans and other organisms (Serrano et al. 1997; Wang et al. 2005). It has an intermediate toxicity to mammals and may affect the central nervous system, the cardiovascular system, and the respiratory system, as well as causing skin and eye irritation (Oliver et al. 2000). It has been added to Annex I to Directive 91/414/EEC (European Union 2005) and is under study by the US Environmental Protection Agency (USEPA 2011a, b).

A number of studies tending to study the affinity of different pesticides with soil have been performed, but only a few refer to chlorpyrifos. Because of its intensive use, a wide range of terrestrial ecosystems may be contaminated with chlorpyrifos, and there is a need to evaluate its environmental behavior and effects. It has been reported that chlorpyrifos is moderately persistent in soils with half-life from less than 1 day to more than 240 days, depending on soil types, soil moisture, soil pH, temperature, organic carbon content, pesticide formulation, soil microorganisms, climatic condition, and initial concentrations (Awasthi and Prakash 1997; Getzin 1981; Pandey and Singh 2004; Racke et al. 1990; 1996; Racke 1993; Singh et al. 2002; Singh et al. 2003). Products of biodegradation include 3,4,6-trichloro-2-pyridinol which subsequently breaks down to organochlorine compounds and carbon dioxide. In some cases, the half-life of chlorpyrifos on sediments, when considered from the perspective of complete mineralization, is 5–6 years (Gebremariam and Beutel 2010). Thus, the pesticide and/or its metabolites can persist in aquatic environments for decades before they completely dissipate. The structure of chlorpyrifos and its metabolite are shown in Fig. 1.

In Argentina, studies on the possible pollution caused by chlorpyrifos are scarce. There are several reports of the occurrence of chlorpyrifos in agricultural districts (Marino and Ronco 2005; Jergentz et al. 2004, 2005). As far as our knowledge, there are no studies on the affinity or half-time of commercial chlorpyrifos with soils of the region.

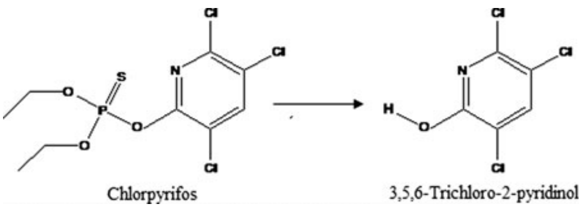


Fig. 1 Structures of chlorpyrifos and its degradation product

Because chlorpyrifos is widely used in our country, over US$ 46,769,236 sales annually, (CASAFE 2012) further research is needed to characterize chlorpyrifos mineralization on soils characteristic of our region. Due to the lack of systematic studies in this respect, its result is essential to perform studies on the interaction and persistence of commercial chlorpyrifos with Argentinean soils in order to assess the environmental impact caused by its application in crops. In this work, we analyze the behavior (interactions and persistence) of a commercial formulation of chlorpyrifos in a soil from a productive area (Navarro) of Provincia de Buenos Aires, Argentina.

2 Materials and Methods

Commercial chlorpyrifos (Pirfos Glex, 48 % Purity, from Punch Química S.A.), marked in the organophosphate insecticides group, was used. Pesticide grade dichloromethane, acetone, and hexane (Sintorgan) were employed in our experiments.

For studies of interaction and degradation, stock solutions of formulated chlorpyrifos (O, O-diethyl-O-(3,5,5-trichloro-2-pyridyl phosphorothioate) were prepared by dissolving 1 ml of commercial formulation in 99 ml of acetone/hexane (1:1). Calibration standards were prepared by dilution of stock solution with acetone/hexane (1:1) or dichloromethane.

Soil samples were obtained from Navarro, province of Buenos Aires, at 48 m above sea level and 35°00.519′S and 59°13.125′W. Navarro is located in the Salado River basin, in the Flooding Pampa, where the mean annual precipitation is between 900–1,000 mm. Soils samples were collected at a depth of 10 cm, were air dried, crushed and passed through a 2 mm sieve, and stored in closed containers at room temperature (20–25 °C). Before performing the experiments, soil was characterized (Table 1), and blanks were carried out with the matrices to assure absence of chlorpyrifos. Solid matrices were analyzed for extractable phosphorous (Olsen), pH (1:2.5 soil/water ratio), organic carbon (Walkley–Black), nitrogen (Micro–Kjeldahl), and clay content (Bouyoucous) by standardized methods (Sparks 1996; USDA 1993).

Table 1 Chemical characterization of soil

% Organic matter content	% Carbon	% Nitrogen	% Phosphorous	pH
1.82	0.91	0.360	12.57	7.05

Field capacity of soils was determined by the method describe by Mizuno et al. (1978). Field capacity is considered as the moisture retained by the profile of a uniformly wet soil that has virtually ceased to drain (Cavazza et al. 2007). To determine this parameter, a plastic container with holes on the bottom was used. Before adding the soil sample, a piece of cloth was used to cover the bottom, in order to avoid sample losses. The container was filled with the soil sample up to 5 cm of the top of the container. Water was added until the sample was wet, and water started to drain. The top of the container was then covered with a plastic bag. The container was checked daily; and when the drainage was no longer observed, two samples were taken from the middle of the container. Samples were dried in the oven, until reaching constant weight. Field capacity, determined by the equation: $H = Ph - Ps \times 100/Ps$ (H = percentage of moisture, Ph = weight of wet sample at field capacity, Ps = weight of dry sample), was 51.75 %.

2.1 Preparation of Solid Matrices for Recovery Experiments

Soil samples (50 g) were spiked with the commercial formulation of chlorpyrifos to obtain final concentrations of 6, 12, 24, and 48 mg/g. The spiked samples were allowed to rest for 3 days, and extraction was performed.

2.2 Preparation of Solid Matrices for Persistence Experiments

Soil samples (400 g) were spiked with the commercial formulation of chlorpyrifos to obtain final concentrations of 3 and 6 mg/kg. Dry samples (20 g) were weighed into 100 ml glass recipients, and then water was added in an amount predetermined by a field capacity experiment to reach 80 % field capacity. Eighty percent of field capacity was used as water content in all the experiences because the field capacity in soils with mean annual precipitation between 900–1,000 mm could remain as high as 80 % in the rain time. Soils were placed in a dark cabinet at 22–25 °C and in aerobic medium. The cabinet floor was covered with water in order to maintain the moisture of the soils. All the samples were weighed three times a week, and water was added if necessary to maintain the 80 % field capacity moisture level. Finally, extractions at 7, 14, 28, 35, 42, and 49 days were made. The tests were conducted in duplicate for each concentration.

2.3 Analysis and Quantification

EPA method 3540C was adapted for the extraction. The spiked samples (20 g) were extracted (Soxhlet) with 450 ml of acetone/hexane (50:50 v/v) mixture for 16 h. Extracts were passed through a drying column containing about 10 cm of anhydrous sodium sulfate. The extracts were concentrated in rotatory evaporator to 2 ml final volume. All the experiments were conducted in duplicates. Matrices without the addition of the pesticide were used as blanks.

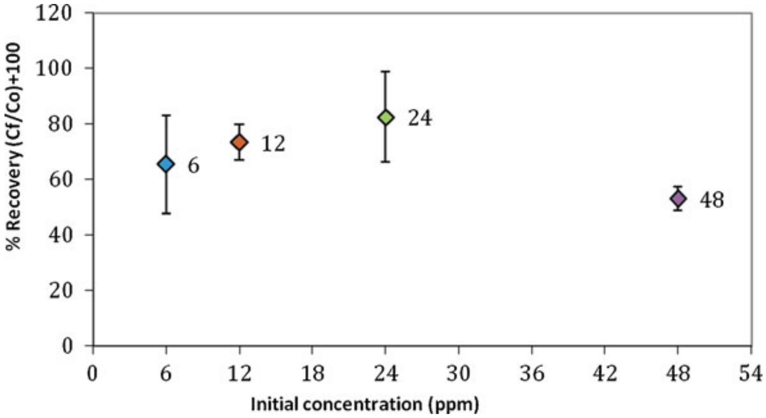
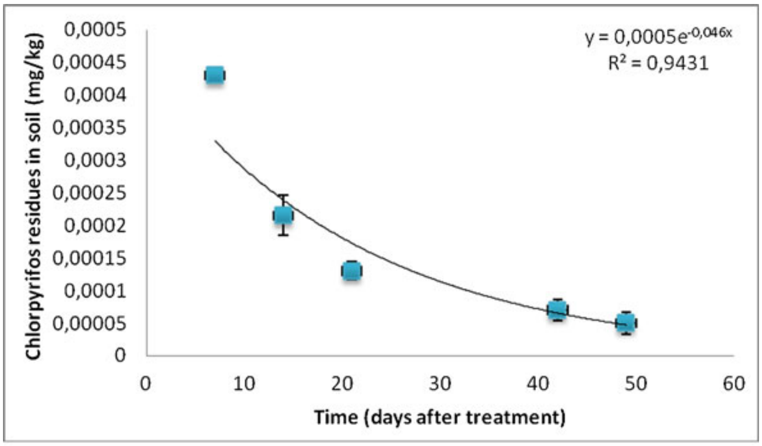
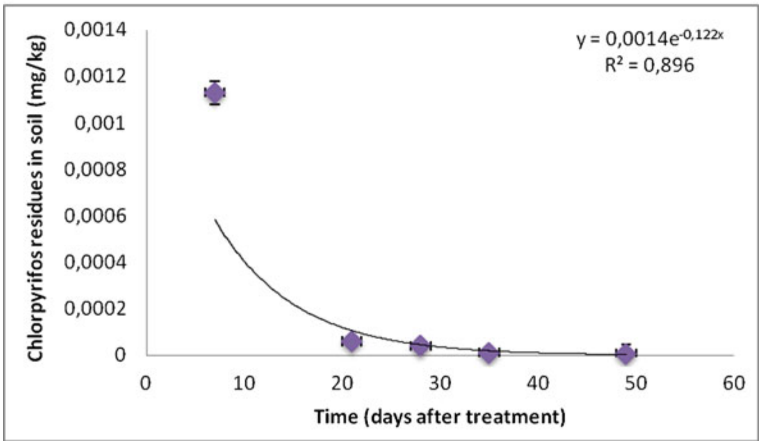


Fig. 2 Recovery (% of initial concentration) in Navarro's soil with commercial chlorpyrifos

Fig. 3 Degradation of chlorpyrifos at concentrations of 3 and 6 mg/kg



3ppm



6 ppm

EPA method 8080 was adapted for quantification. The gas chromatographic analyses were performed on a Perkin–Elmer AutoSystem XL instrument equipped with an electron capture detector and a capillary column Quadrex (phase: 007–5; large: 30 m; DI: 0.25; film: 0.25). Program: initial temperature 100 °C hold 2′, 15 °C per minute to 160 °C hold 0′, 5 °C per minute to 270 °C hold 10′. Injector: 225 °C; detector 300 °C. Standard solutions of 25, 50, 100, 125, 500, and 1,000 μg/l were used for calibration. The detection limit for chlorpyrifos was 5×10^5 mg/l.

3 Results and Discussion

3.1 Recovery of Commercial Chlorpyrifos from Soil

Soil samples were obtained from Navarro, province of Buenos Aires. Chemical characterization is shown in Table 1.

Soil samples were artificially contaminated with four different concentrations of the pesticide (6, 12, 24, and 48 ppm), extracted (Soxhlet) and analyzed by capillary gas chromatography using electron capture detection to determine the amount of extracted chlorpyrifos in each case. Recovery was estimated as

Table 2 Kinetics data of chlorpyrifos degradation in soil

Chlorpyrifos concentration (mg/kg)	Dynamic function	DT50* (days)	Coefficient (R^2)
3	$C=0{,}0005\ e^{-0.046\times}$	15.07	0.9431
6	$C=0{,}0014\ e^{-0.122\times}$	5.68	0.896

*The degradation of chlorpyrifos in soil was described by the first-order function ($C=C_0\times e^{-kt}$) where C is the concentration (mg kg^{-1}) and C_0 is the initial concentration. The degradation half-life of chlorpyrifos (DT50) in soil was obtained by the function DT50=ln2/k. Each value is a mean of two replicates percentage of initial concentration. Control experiments with uncontaminated samples were run in all cases to rule out any previous contamination. High reproducibility was observed for the extractions. Recoveries (% of initial concentration) are shown in Fig. 2.

In this case, recovery percentages increased with the increase of initial concentration of the pesticide until a concentration of about 25 ppm is reached, and then a decrease is observed. In previous work, we observed that when pure chlorpyrifos (98 % of purity) was used, a decrease in recovery percentages with the increase of initial concentration employed was observed.

3.2 Persistence of Commercial Formulations of Chlorpyrifos in Soil

The sorption and the persistence of a particular organic xenobiotic in the environment are essential to evaluate their potential contamination effect. The latter parameter is normally expressed as the degradation half-life. The half-life is the time, in days, when the initial concentration became half.

Degradation experiences were performed at two different concentrations of commercial chlorpyrifos (3 and 6 mg/kg). Extractions were performed every 7 days for 49 days, and chlorpyrifos was analyzed by gas chromatography with ECD detection. Chlorpyrifos concentrations (mg/kg, mean of two determinations), at each extraction time, are shown in Fig. 3. Degradation of chlorpyrifos in soil seems to be subjected to a first-order model, considering the fact that the determination coefficient (R^2) is more than 0.7 (Table 2) (Fang et al. 2009; Bin Liang et al. 2011). Kinetic data of chlorpyrifos degradation in Table 2 show that, in both cases, chlorpyrifos at initial concentrations of 3 and 6 mg/kg soil was degraded in 21 days by 99.9 % with half-life of 15.07 and 5.68 days after treatment for 42 days, respectively. Racke et al. (1988) reported half-life for chlorpyrifos from 10 to 120 days and Howard (1991) reported half-life of 2 weeks to over 1 year, depending on the soil type, climate, and other conditions. This large variation in half-life has been attributed to variation in factors such as pH, temperature, moisture content, organic carbon content, and pesticide formulation (Getzin 1981).

The half-lives were not significantly extended with increasing chlorpyrifos concentration, but they are consistent with half-lives reported previously (Chu et al. 2008; Bin et al. 2011; Fang et al. 2009). This could be explained by the fact that inhibition of soil microbial communities (bacteria, fungi, and actinomycetes) is reported to start at higher concentrations (10 mg/kg) (Shan et al. 2006).

4 Conclusions

The results reported in the present paper show that the recovery percentages increase exponentially with the increase of initial concentration of the pesticide until a concentration of about 25 ppm is reached, and the recovery starts to decrease. In fields where this pesticide is used several times in every crop, a greater application means a higher return to the surrounding water bodies due to the runoff and a possible contamination of the water bodies and the biota of the stream.

Some authors suggest that the half-life of chlorpyrifos had a positive correlation with chlorpyrifos concentration, and its inhibitory effect on soil microbial communities followed a positive dose-response pattern (Fang et al. 2009; Fogg et al. 2003 and Gan et al. 1996). No difference in the half-time was observed with the two concentrations assayed, maybe because the concentrations used were not high enough to modify soil microbial activity or diversity (Shan et al. 2006; Vischetti et al. 2007).

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References

Awasthi, M. D., & Prakash, N. B. (1997). Persistence of chlorpyrifos in soils under different moisture regimes. *Pesticide Science, 50*, 1–4.

Bin, L., Chengli, Y., Mingbo, G., Yanfu, Z., Jun, Z., Changxiong, Z., et al. (2011). Adsorption and degradation of triazophos, chlorpyrifos, and their main hydrolytic metabolites in paddy soil from Chaohu Lake, China. *Journal of Environmental Management, 92*(9), 2229–2234.

CASAFE (Cámara de Sanidad Agropecuaria y Fertilizantes, Buenos Aires, Argentina) (2012). Available from: <http://www.casafe.org>. Accessed October 2012.

Cavazza, L., Patruno, A., & Cirillo, E. (2007). Field capacity in soils with a yearly oscillating water table. *Biosystems Engineering, 98*(3), 364–370.

Chu, X., Fang, H., Pan, X., Wang, X., Shan, M., Feng, B., et al. (2008). Degradation of chlorpyrifos alone and in combination with chlorothalonil and their effects on soil microbial populations. *Journal of Environmental Sciences, 20*, 464–469.

European Union (2005). Amending Council Directive 91/414/EEC to include chlorpyrifos, chlorpyrifos –methyl, mancozeb, maneb, and metiram as active substances. Official J. Eur. Union, Comission Directive 2005/72/EC., p. 63.

Fang, H., Yu, Y. L., Wang, X., Shan, M., Wu, X. M., & Yu, J. Q. (2006). Dissipation of chlorpyrifos in pakchoi-vegetated soil in a greenhouse. *Journal of Environmental Sciences, 18*(4), 760–764.

Fang, H., Yu, Y., Chu, X., Wang, X., Yang, X., & Yu, J. (2009). Degradation of chlorpyrifos in laboratory soil and its impact on soil microbial functional diversity. *Journal of Environmental Sciences, 21*, 380–386.

Fogg, P., Boxall, A. B. A., & Walker, A. (2003). Degradation of pesticides in biobeds: the effect of concentration and pesticides mixtures. *Journal of Agricultural and Food Chemistry, 51*, 5344–5349.

Gan, J., Becker, R. L., Koskinen, W. C., & Buhler, D. D. (1996). Degradation of atrazine in two soils as a function of concentration. *Journal of Environmental Quality, 25*, 1064–1072.

Gebremariam, S., & Beutel, M. (2010). Effects of drain-fill cycling on chlorpyrifos mineralization in wetland sediment–water microcosms. *Chemosphere, 78*, 1337–1341.

Getzin, L. W. (1981). Degradation of chlorpyrifos in soil: influence of autoclaving, soil moisture, and temperature. *Journal of Economic Entomology, 74*, 158–162.

Howard, P. H. (Ed.). (1991). *Handbook of Environmental Fate and Exposure Data for Organic Chemicals, Pesticides, vol. 3* (pp. 5–13). Chelsea, MI: Lewis Publishers.

Jergentz, S., Pessacq, P., Mugni, H., Bonetto, C., & Schulz, R. (2004). Linking in situ bioassays and population dynamics of macroinvertebrates to assess agricultural contamination in streams of the Argentine pampa. *Ecotoxicology and Environmental Safety, 59*, 133–141.

Jergentz, S., Mugni, H., Bonetto, C., & Schulz, R. (2005). Assessment of insecticide contamination in runoff and stream water of small agricultural streams in the main soybean area of Argentina. *Chemosphere, 61*, 817–826.

Marino, D., & Ronco, A. (2005). Cypermethrin and chlorpiryfos concentration levels in surface water bodies of the Pampa Ondulada. *Argentina Bulletin Environment Contamination Toxicology, 75*(4), 820–826.

Moses, M., Johnston, E. S., Anger, W. K., Burse, V. W., Horstman, S. W., Kackson, R. J., et al. (1993). Environmental equity and pesticide exposure. *Toxicology and Industrial Health, 9*, 913–959.

Mizuno, I., Arrigo, N., & Svartz, H. (1978). *Método rápido para determinar Humedad Equivalente*. Buenos Aires: Actas de Reunión Argentina de la Ciencia del Suelo. Septiembre 1978.

Oliver, G. R., Bolles, H. G., & Shurdut, B. A. (2000). Chlorpyrifos: probabilistic assessment of exposure and risk. *Neurotoxicology, 21*, 203–208.

Pandey, S., & Singh, D. K. (2004). Total bacterial and fungal population after chlorpyrifos and quinalphos treatments in groundnut (*Arachis hypogaea* L.) soil. *Chemosphere, 55*(2), 197–205.

Racke, K. D., Coats, J. R., & Titus, K. R. (1988). Degradation of chlorpyrifos and its hydrolysis product, 3,5,6-trichloro-2-pyridinol, in soil. *Journal of Environmental Science and Health, B23*, 527–539.

Racke, K. D., Laskowski, D. A., & Schultz, M. R. (1990). Resistance of chlorpyrifos to enhanced biodegradation in soil. *Journal of Agricultural and Food Chemistry, 38*, 1430–1436.

Racke, K. D. (1993). Environmental fate of chlorpyrifos. *Reviews of Environmental Contamination and Toxicology, 131*, 1–154.

Racke, K. D., Steele, K. P., Yoder, R. N., Dick, W. A., & Avidov, E. (1996). Factors affecting the hydrolytic degradation of chlorpyrifos in soil. *Journal of Agricultural and Food Chemistry, 44*, 1582–1592.

Serrano, R., Lopez, F. J., Hernandez, F., & Pena, J. B. (1997). Bioconcentration of chlorpyrifos, chlorfenvinphos, and methidathion in *Mytilus galloprovincialis*. *Bulletin of Environmental Contamination and Toxicology, 59*, 968–975.

Shan, M., Fang, H., Wang, X., Feng, B., Chu, X., & Yu, Y. (2006). Effect of chlorpyrifos on soil microbial populations and enzyme activities. *Journal of Environmental Sciences, 18*, 4–5.

Singh, B. K., Walker, A., & Wright, D. J. (2002). Persistence of chlorpyrifos, fenamiphos, chlorothalonil, and pendimethalin in soil and their effects on soil microbial characteristics. *Bulletin of Environmental Contamination and Toxicology, 69*(2), 181–188.

Singh, B. K., Walker, A., Morgan, J. A., & Wright, D. J. (2003). Effects of soil pH on the biodegradation of chlorpyrifos and isolation of a chlorpyrifos-degrading bacterium. *Applied and Environmental Microbiology, 69*(9), 5198–5206.

Sparks, D. L. (1996). *Methods of Soil Analysis: Part 3 – Chemical Methods. SSSAV Book Series: 5*. Madison: ASA.

USDA (U.S. Department of Agriculture) (1993) Soil Survey Division Staff. Soil Conservation Service. Handbook 18.

USEPA (1999). Quantitative usage analysis, chlorpyrifos, US Environmental Protection Agency, 24 November 1999.

USEPA (2011a) Chlorpyrifos Preliminary Human Health Assessment for Registration Review Document ID: EPA-HQ-OPP-2008-0850-0025. June 2011.

USEPA (2011b) Revised Chlorpyrifos Preliminary Reg Review Drinking Water Assessment Document ID: EPA-HQ-OPP-2008-0850-0026. June 2011.

Vischetti, C., Coppola, L., Monaci, E., Cardinali, A., & Castillo, M. D. (2007). Microbial impact of the pesticide chlorpyrifos on Swedish and Italian biobeds. *Agronomy for Sustainable Development, 27*(3), 267–272.

Wang, L. G., Jiang, X., Mao, Y. M., Zhao, Z. H., & Bian, Y. R. (2005). Organophosphorus pesticide extraction and clean-up from soils and measurement using GC-NPD. *Pedosphere, 15*, 386–394.