

## **Human Health Risk Assessment of Pesticide Residues in Field Grown Yellow Peppers**

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**Abstract.** A field survey was carried out considering the application of 13 pesticides in normal and double doses on field grown yellow peppers, within the Phytosanitary Office Mureș (Romania). Seven fungicides (based on chlorothalonil, captan, folpet, tebuconazole, triadimenol, myclobutanil and metalaxyl-M), five insecticides (based on deltamethrin, alfa-cypermethrin, lambda-cyhalothrin, chlorpyrifos, biphenthrin) and one acaricide (based on propargite) were applied in three treatments considering the phenological growth stages of yellow peppers. The aim of our study was to assess the health risks associated with pesticides residues by yellow peppers consumption for both adults and children. Based on fruits consumption estimates released in 2015 for 2013, of 188.60 g/capita/day in EU-28, the human health risk assessment revealed that pesticides chlorothalonil and propargite can pose a threat to children health when applied in double doses. Health risks for both adults and children after consumption of yellow peppers treated with pesticides applied in normal dose may be considered negligible.

**Keywords:** Human Health Risk Assessment, Hazard Index, Maximum Residue Levels, Fruit Consumption, Pesticides

### **1. Introduction**

The application of pesticides in current agricultural practices has led to serious impacts to human health and the environment. Although, in the effort to prevent and control pests or eliminate yield losses and maintaining high quality products, the use of pesticides is strictly regulated, serious concerns are raised for human exposure to residues from fruits and vegetables [1, 2]. Fruits and vegetables are considered very important components of the human diet. The intake of 5 or more servings per day is considered essential for a good health and it is encouraged for vitamin deficiency prevention and also different diseases such as cancer or obesity [3]. Reports considering monitoring programs of pesticides residues in fruit and vegetable products in Europe, USA and Canada, have shown that most samples (including also fresh and processed foods), have amounts of residues between 6.7 and 58% [4]. Well-known effects of pesticides such as chronic neurotoxicity, endocrine disruption, immune impacts, genotoxicity, mutagenicity and carcinogenesis have been increasing public concern for food safety [4, 5]. Consequently, pesticides contamination of fruits, vegetables and grains has become a health issue across the entire world [6]. To meet the high request for fruits and vegetables, worldwide farmers apply large quantities of pesticides not only to prevent pests and diseases but also to boost their production, with continuous growth of the environmental impact and health risk consequences [7-10].

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In Europe and across the world, peppers are much appreciated. Pepper crops are attacked by a number of diseases and pests which can lead to great loss of production when pesticides are not used. Chemical protection of peppers is usually carried out by performing 2 or 3 treatments with different types of pesticides. Constant use of pesticides increases the possibility of finding multiple residues of these compounds in peppers, beyond the Maximum Residue Levels (MRLs), creating a significant risk to human health [11].

With respect to the above mentioned information, our primary objective was to assess the human health risk associated with the consumption of yellow peppers at harvest for which three treatments with 13 pesticides were applied in normal and double doses. We have considered both adults and children, as exposed population in the human health risk evaluation.

## 2. Materials and Methods

### 2.1. Reagents and analysis

Analytical standards were purchased from Chem Service (West Chester, SUA) and Sigma Aldrich Laborchemikalien GmbH (Seelze, Germany) with certified purity between 95.1% and 99.7%, while Dafcochim SRL (Tg. Mureş, Romania) and Chemark Rom SRL (Tg. Mureş, Romania) were the providers for pesticides applied in the field survey. The pesticide residues were analyzed by a gas chromatograph (Agilent 7890 type with 2 ovens) coupled with a mass spectrometer with flight time, CG\*GC-TOF-MS Pegasus 4.21 (LECO, SUA). The working conditions were considered as presented by Pogăcean et al. [12, 13].

### 2.2. Field survey

In the field survey developed at the Phytosanitary Office Mureş (Romania), the yellow pepper plants were transplanted to the field in late April 2013, in two rows, considering 0.8 m wide and 0.14 m distance between plants on the same row. We considered two types of experiments, and we applied a total of 3 treatments for each survey based on the normal recommended dose (ND) and on the double dose (DD). Treatments were carried out at 2 weeks time interval, from the moment of the first group of yellow peppers appearance and up to 80% of typical yellow peppers (fully ripe). We have ensured a buffer zone between the yellow peppers subjected to the experiments. The sprays with pesticides solution were carried out using a 1.5 L pump, in sunny days, without wind, in the morning, in compliance with Good Agricultural Practices (GAP). We used solutions containing seven fungicides (chlorothalonil, captan, folpet, tebuconazole, triadimenol, myclobutanil, metalaxyl-M), five insecticides (deltamethrin, alfa-cypermethrin, lambda-cyhalothrin, chlorpyrifos, biphenthrin) and one acaricide (propargite). Pesticides treatments were applied according to yellow peppers phenological growth phases considering the BBCH scale (Biologische Bundesanstalt, Bundessortenamt and CHEmical industry) [14, 15], as shown in Table 1.

Table 1: Phenological growth stages of yellow peppers.

No	BBCH scale	Fruit description	Time between treatments
1	701-702	Fruits with typical dimension	14 days
2	801-802	10% of fruits show typical size and color of a fully ripe fruit	14 days
3	807-808	80% of fruits show typical size and color of a fully ripe fruit	14 days
4	909	At harvest	25 days (from the last treatment)

### 2.3. Human health risk assessment

The human health risk was evaluated based on the concentration of pesticides residues in yellow peppers at harvest. The estimated lifetime exposure dose (mg/kg/day), food consumption (kg/person/day) and body weight (kg) were used to determine if there are any health risks to consumers posed by pesticide residues in yellow peppers. Based on latest edition of “Freshfel Consumption Monitor” in the EU-28 released in 2015, per capita fruit consumption in 2013 was estimated at 188.60 g/capita/day [16], while the average body weight of adults in Europe was estimated at 70.8 kg [17] and at 23.1 kg for children (age group, 3 to < 10 years) [18]. Based on food consumption rate for fruits in Europe, the estimated lifetime exposure dose (mg/kg/day) was calculated as indicated by Pogăcean et al. [13] and Bempah et al. [19]. The hazard indices

(HI) for adults and children were assessed based on the ratio between estimated pesticide exposure doses and the corresponding Reference Doses (RfDs) [20] or the analogous Acceptable Daily Intake (ADI) values, when RfDs were not available.

### 3. Results and Discussion

The field survey indicated that there are significant differences between the MRL and the final concentration of pesticides in yellow pepper samples. Food containing pesticide residues in higher levels than MRLs values can still be considered safe for consumption since the MRLs are always set far below levels considered to be safe for humans. Safety limits are evaluated by analogy with RfDs or ADI values [3].

The results indicated in Table 2 show that the only pesticide residues in yellow peppers at harvest, that are in conformity with European Union rules, by not exceeding the MRLs are: deltamethrin, alfa-cypermethrin and triadimenol. On the other side, pesticides myclobutanil and biphenthrin do not exceed the MRLs only when applied in normal dose. The other pesticides considered in our field survey exceed the MRLs, considering both types of treatments (ND and DD treatments). These findings have raised our concerns when it comes to human exposure to higher levels of pesticides than the acceptable limits set by European Union. By consuming yellow peppers treated with pesticides of which concentrations at harvest exceed the MRLs, the vulnerable population such as children may face exposure risks. These findings lead to our study fundamental objective: assessing the risk posed by consumption of yellow peppers treated with pesticides applied in normal and double doses, by adults and children.

Table 2: Pesticides concentration (mg/kg) in yellow peppers samples after treatments applied in normal dose (ND) and double dose (DD) according to BBCH scale.

Pesticides	BBCH scale								MRL (mg/kg)
	701-702		801-802		807-808		909		
	ND	DD	ND	DD	ND	DD	ND	DD	
Chlorothalonil	1.14	1.75	1.25	3.31	2.75	4.15	1.32	2.28	0.01*
Deltamethrin	0.1	0.18	0.15	0.29	0.11	0.38	0.08	0.12	0.2
Myclobutanil	0.3	0.45	0.22	0.36	0.45	0.8	0.21	0.54	0.5
Alfa-cypermethrin	0.31	0.62	0.22	0.47	0.389	0.55	0.12	0.32	0.5
Biphenthrin	0.301	0.5	0.34	0.76	0.44	0.88	0.34	0.79	0.5
Captan	0.8	2.07	0.96	1.96	1.02	3.73	0.43	1.27	0.1
Folpet	0.7	2.58	1.96	3.57	2.75	4.18	1.83	2.01	0.02*
Tebuconazole	0.69	2.01	0.65	1.02	0.82	2.72	0.62	1.41	0.6
Triadimenol	0.09	0.29	0.21	0.36	0.17	0.47	0.09	0.12	1
Metalaxyl-M	1.18	2.18	0.88	1.86	1.22	2.74	0.65	1.55	0.5
Chlorpyrifos	0.45	0.75	0.5	0.69	0.85	1.62	0.52	1.35	0.5
Lambda-cyhalothrin	0.47	0.61	0.09	0.29	0.17	0.48	0.09	0.14	0.01
Propargite	1.38	3.16	2.02	4.96	2.63	3.11	1.62	2.82	0.01*

MRL - Maximum Residue Level set by European Union legislation [21]

\* = Limit of determination

In our study, the U.S Environmental Protection Agency's guidelines for human health risk assessment have been taken into consideration. Therefore, we considered that the maximum absorption rate and the bioavailability rate are 100% [22]. The lifetime exposure dose was calculated for pesticides residues in yellow peppers at harvest considering the two types of experiments developed (for pesticides applied in both ND and DD). A comparison of lifetime exposure dose for the exposed population, both adults and children, with the RfDs is available in Table 3. When comparing the lifetime exposure dose with the available RfDs, it can be observed that chlorothalonil and propargite are the only pesticides that exceed the RfD values, if applied in double doses and when children are considered as exposed population. For the rest of 11 pesticides, the lifetime exposure dose value is under the value of RfD, although simultaneous exposure to multiple pesticides can lead to toxicological effects for both adults and children health.

The next step in our evaluation was to assess the health risk by considering the HI for adults and children. A concern can also be raised by considering the values of HI, as seen from Figs. 1 and 2. If HI is higher than 1, the pesticide residues in yellow peppers can be considered a risk to consumers, while for a HI lower than 1,

the pesticide residues are considered to be in an acceptable limit with no risk to human health [12, 13, 23]. When treatments with pesticides were applied in normal doses (Fig. 1) the HI do not exceed the value 1, so it can be considered that there are no risks involved for adults and children. The values of HI higher than 1 in the case of pesticides chlorothalonil and propargite, when applied in double doses (Fig. 2) indicate a risk to children health associated with the consumption of yellow peppers. Since children consume more calories of food per unit of body weight compared adults, their exposure to pesticide residues in foods is higher.

Table 3: Lifetime exposure dose calculated for pesticides residues in yellow peppers at harvest.

Pesticide	Reference dose (mg/kg-day)	Lifetime exposure dose (mg/kg/day)			
		ND		DD	
		Adults	Children	Adults	Children
Chlorothalonil	0.015	0.0035	0.0107	0.0060	0.0186
Deltamethrin	0.01*	0.0002	0.0006	0.0003	0.0009
Myclobutanil	0.31*	0.0005	0.0017	0.0014	0.0044
Alfa-cypermethrin	0.1	0.0003	0.0009	0.0008	0.0026
Biphenthrin	0.015	0.0009	0.0027	0.0021	0.0064
Captan	0.13	0.0011	0.0035	0.0033	0.0103
Folpet	0.1	0.0048	0.0149	0.0053	0.0164
Tebuconazole	0.03*	0.0016	0.0050	0.0037	0.0115
Triadimenol	0.05*	0.0002	0.0007	0.0003	0.0009
Metalaxyl-M	0.06	0.0017	0.0053	0.0041	0.0126
Chlorpyrifos	0.1*	0.0013	0.0042	0.0035	0.0110
Lambda-cyhalothrin	0.005	0.0002	0.0007	0.0003	0.0011
Propargite	0.02	0.0043	0.0132	0.0075	0.0230

\*ADI

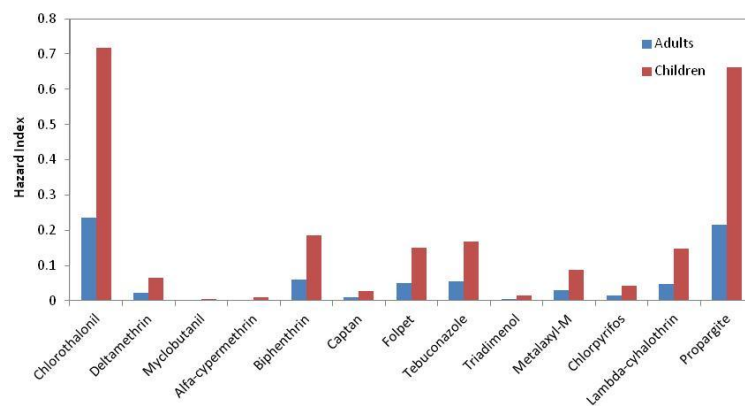


Fig. 1: Hazard Index calculated for pesticides residues in yellow peppers at harvest considering the ND treatments.

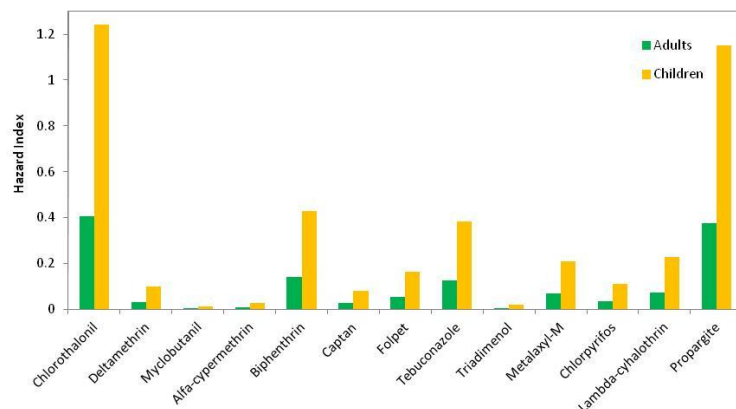


Fig. 2: Hazard Index calculated for pesticides residues in yellow peppers at harvest considering the DD treatments.

## 4. Conclusions

A number of 13 pesticides were applied in normal dose (recommended) and double dose treatments on field grown yellow peppers. Three treatments with pesticides were considered according to BBCH phenological growth stages of yellow peppers. Our findings have shown that although most of the pesticides residues in yellow peppers exceed the MRLs at harvest, only chlorothalonil and propargite are a threat to children health, if applied in double doses. The lifetime exposure dose which exceeds the RfDs, and the Hazard Index >1 for chlorothalonil and propargite applied in double dose also sustain these findings. Potential consumer risks should be taken into account, since continuous consumption of fruits and vegetables containing pesticides residues could result in higher toxicity, leading to deadly chronic effects.

## 5. Acknowledgements

This paper was elaborated with the support of a grant of the Romanian National Authority for Scientific Research, CNCS – UEFISCDI, project number PN-II-ID-PCE-2011-3-0559”, Contract 265/2011 and with the support of Phytosanitary Office Mureş, Romania.

## 6. References

- [1] C.A. Damalas and I.G. Eleftherohorinos. Pesticide Exposure, Safety Issues, and Risk Assessment Indicators. *Int. J. Environ. Res. Public Health* 2011, 8 (5): 1402-1419.
- [2] M.O. Pogăcean and M. Gavrilescu. Plant protection products and their sustainable and environmentally friendly use. *Environ. Eng. Manag. J.* 2014, 8 (3): 607-627.
- [3] B.M. Keikotlhaile and P. Spanoghe. Pesticide residues in fruits and vegetables. In: M. Stoytcheva (Ed.) Pesticides - formulations, effects, fate. InTech 2011, pp. 243-252.
- [4] M.A. Dalvie and L. London. Risk assessment of pesticide residues in South African raw wheat. *Crop Protection* 2009, 28 (10): 864–869.
- [5] S. Wanwimolruk, O. Kanchanamayoon, S. Boonpangrak, and V. Prachayasittikul. Food safety in Thailand 1: it is safe to eat watermelon and durian in Thailand. *Environ. Health. Prev. Med.* 2015, 20 (3): 204–215.
- [6] Y. Latif, S.T.H. Sherazi, and M.I. Bhangar. Monitoring of pesticide residues in commonly used fruits in Hyderabad Region, Pakistan. *American Journal of Analytical Chemistry* 2011, 2: 46-52.
- [7] C.K. Bempah and A.K. Donkor. Pesticide residues in fruits at the market level in Accra Metropolis, Ghana, a preliminary study. *Environ. Monit. Assess.* 2011, 175 (1-4): 551–561.
- [8] K.R. Everett, I.P.S. Pushparajah, J.T. Taylor, O.E. Timudo-Torrevilla, T.M. Spiers, A.A. Chee, P.W. Shaw, and D.R. Wallis. Evaluation of fungicides for control of bitter and sprinkler rots on apple fruit. *New Zealand Plant Protection* 2015, 68: 264-274.
- [9] B. Robu, O. Jitar, C. Teodosiu, S.-A. Strungaru, M. Nicoara, and G. Plavan. Environmental impact and risk assessment of the main pollution sources from the Romanian Black Sea coast. *Environ. Eng. Manag. J.* 2015, 14 (2): 331-340.
- [10] H. Wei, Z. Le, L. Shuxian, W. Dan, L. Xiaojun, J. Lan, and M. Xiping. Health risk assessment of heavy metals and polycyclic aromatic hydrocarbons in soil at coke oven gas plants. *Environ. Eng. Manag. J.* 2015, 14 (2): 487-496.
- [11] M. O. Pogăcean. *Studies on accumulation of some pesticides in vegetable products*. PhD thesis, Gheorghe Asachi Technical University of Iasi, Romania, 2013.
- [12] M. O. Pogăcean, R. M. Hlihor, C. Preda, and M. Gavrilescu. Humans in the environment: Comparative analysis and assessment of pesticide residues from field-grown tomatoes. *Eur. J. Sci. Theol.* 2013 9 (6): 79-95.
- [13] M. O. Pogăcean, R. M. Hlihor, and M. Gavrilescu. Monitoring pesticides degradation in apple fruits and potential effects of residues on human health. *J. Environ. Eng. Landsc.* 2014 22 (3): 171-182.
- [14] D. Gericke, J. Nekovar, and C. Hörd. Estimation of plant protection product application dates for environmental fate modeling based on phenological stages of crops. *J. Environ. Sci. Health B.* 2010 45 (7): 639-647.
- [15] U. Meier, H. Bleiholder, E. Weber, C. Feller, M. Hess, H. Wicke, T. van den Boom, P.D. Lancashire, L. Buhr, H. Hack, R. Klose, and R. Stauss. *BBCH Monograph - Growth stages of mono- and dicotyledonous plants, 2nd Edition*. Uwe Federal Biological Research Centre for Agriculture and Forestry, 2001.
- [16] FRESHFEL. *Freshfel Consumption Monitor* [online] 2015. Available from Internet: [http://www.freshfel.org/docs/2015/Press\\_Releases/20150610\\_-\\_Consumption\\_Monitor.pdf](http://www.freshfel.org/docs/2015/Press_Releases/20150610_-_Consumption_Monitor.pdf).

- [17] S. C. Walpole, D. Prieto-Merino, P. Edwards, J. Cleland, G. Stevens, and I. Roberts. The weight of nations: an estimation of adult human biomass. *BMC Public Health* 2012 12: 439.
- [18] EFSA. Guidance on selected default values to be used by the EFSA Scientific Committee, Scientific Panels and Units in the absence of actual measured data. *EFSA Journal* 2012 10 (3): 32 pp.
- [19] C. K. Bempah, A. Buah-Kwofie, D. Denutsui, J. Asomaning, and A. O. Tutu. Monitoring of pesticide residues in fruits and vegetables and related health risk assessment in Kumasi Metropolis, Ghana. *Res. J. Environ. Earth Sci.* 2011 3 (6): 761-771.
- [20] USEPA. *Integrated Risk Information System (IRIS)* [online] 2015. Available from Internet: <http://cfpub.epa.gov/ncea/iris/index.cfm?fuseaction=iris.showSubstanceList>.
- [21] HSE. *Maximum Residue Level (MRL) Database* [online] 2015. Available from Internet: <https://secure.pesticides.gov.uk/MRLs>.
- [22] USEPA. *Exposure Factors Handbook. 2011 Edition*. National Center for Environmental Assessment Office of Research and Development U.S. Environmental Protection Agency, Washington DC, 2011.
- [23] R. M. Hlihor, L. C. Apostol, C. Smaranda, L. V. Pavel, F. A. Căliman, B. M. Robu, and M. Gavrilescu. Bioavailability processes for contaminants in soils and their use in risk assessment. *Environ. Eng. Manag. J.* 2009, 8 (5): 1199-1206.