Inter and intraspecific variability of dieldrin accumulation in Cucurbita fruits: New perspectives for food safety and phytomanagement of contaminated soils

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Abstract

Due to past agricultural practices, it is common to identify arable soils contaminated with persistent and potentially toxic organochlorine pesticides (OCPs). Occurrence of OCPs, including dieldrin, in vegetables can lead to chronic exposure of the consumers. Some market vegetables, particularly the Cucurbitaceae, are known to accumulate high OCP concentrations. Dieldrin concentration in *Cucurbita* fruits can exceed the Maximal Residue Limit (MRL) resulting in cultivation and sale restrictions for market gardeners. To assess the intra- and interspecific variability of Cucurbitaceae species for low dieldrin concentration in fruits could be a solution.

Here, 24 varieties from seven Cucurbitaceae species were cultivated outdoors in large pots, until fruiting, in soils historically contaminated with dieldrin. More than 330 fruits were harvested and analyzed for determining the inter and intraspecific variability of dieldrin accumulation. Significant interspecific differences occurred with mean fruit concentration ranging between 4.2 ± 7.0 and 85.0 ± 19.4 µg dieldrin kg^{-1} fresh weigh (FW) in watermelons (*C. lanatus L.*) and cucumbers (*C. sativus L.*), respectively. Intraspecific differences only occurred for *Cucurbita pepo* L. with mean concentration ranging between 4.9 ± 1.1 and 70.3 ± 3.6 µg dieldrin kg^{-1} FW for the varieties Noire maraîchère and Orélia, respectively. For this plant species, the influence of soil concentration, plant exposure time and biomass on fruit dieldrin concentration depended mainly on varieties.

Keywords: Organochlorine pesticide; *Cucurbita pepo* L., soil contamination, plant exposure time, fruit biomass, soil - plant transfer, food safety

Introduction

Contamination of agricultural soils with organochlorine pesticides (OCPs) is frequently detected world-wide. Even though the use of OCPs is either banished or limited since decades, their persistence is nowadays a continuous challenge for farmers, farmland manager and policy makers (Gavrilescu, 2005; Jorgenson, 2001; Morillo and Villaverde, 2017; Widenfalk, 2002). Indeed, depending on plant species and cultivars, the impregnation of several edible plant parts by OCPs and other Persistent Organic Pollutants (POPs) can occur (Namiki et al., 2018; Otani et al., 2007; Singh and Singh, 2017). Plant species from the Cucurbitaceae family are known to absorb, translocate and accumulate OCPs (Donnarumma et al., 2009; Hashimoto, 2005; Mattina et al., 2004, 2000, 1999; Otani et al., 2007). Consequently, some plant products can contain OCP concentrations above the legal European Maximum Residue Limits (EU MRLs) (Official Journal of the European Union, 2008) and thus cannot be sold and must be destroyed. Finding strategies to decrease OCP bioavailability and/or uptake is therefore a main concern. However, fast, cheap and efficient processes allowing a bioavailability decrease are not yet available (Morillo and Villaverde, 2017). Consequently, short-term and ready to use solutions are needed to enable cultivation of cucurbits on agricultural soils contaminated with OCPs.

Several authors have highlighted inter and intraspecific variabilities of OCP accumulation in *Cucurbita* roots, stems, leaves, and fruits (Clostre et al., 2014; Otani et al., 2007; White, 2002; White et al., 2003b). Therefore, knowing the genotypic variability but also environmental factors influencing OCP accumulation may help identifying agronomical solutions to produce *Cucurbita* fruits with low OCP concentrations.

In addition, other factors can affect OCP concentration and distribution in plant parts for a same cucurbit variety. Previous studies evidenced the effect of soil contaminant concentration (Clostre et al., 2014; Mattina et al., 2004), plant exposure time (Donnarumma et al., 2009; Kelsey et al., 2006) and root to fruit distance (Whitfield Åslund et al., 2008, 2007) on POP concentration in different tissues from Cucurbits. For instance, an increase of chlordane concentration in the soil led to a higher chlordane concentration in *Cucurbita pepo* roots, stems, leaves and fruits. However, the increase in the fruit was lower than in the other plant parts (Mattina et al., 2004). A similar behavior was reported concerning the chlordecone concentration in pumpkin, cucumber, and christophine (chayote, *Sechium edule* (Jacq.) Sw.) fruits, for which the contaminant concentration increased in the fruit with the soil contamination level, although a generally poor relationship was found (Clostre et al., 2014). However, behavior and accumulation in plant may be contaminant dependent (Namiki et al., 2018), preventing extrapolation of the above results to other OCPs.

This in-situ study aimed at determining the inter and intraspecific variability of dieldrin accumulation in fruits of a wide range of *Cucurbita* species commercially available, cultivated at a French market garden field with a historically OCP-contaminated soil, located in a peri-urban perimeter, which supports and protects agricultural areas, near the Bordeaux agglomeration (France). Moreover, several plant and soil factors, which may influence the dieldrin concentration in fruits of *C. pepo* varieties were investigated, with a focus on soil contamination level, plant exposure time, and fruit biomass.

Experimental section

Plant selection and cultivation

Selection of plant species and varieties

Based on a survey conducted among French market gardeners, highlighting the most important Cucurbitaceae species locally used, 24 varieties included in seven plant species (*C. pepo* L., *Cucurbita moschata* Duchesne, *Cucurbita maxima* Duchesne, *Cucumis sativus* L., *Momordica charantia* L., *Melothria scabra* Naudin, and *Citrullus lanatus* (Thunb.) Matsum. & Nakai) were selected (Table 1). This selection especially focused on *C. pepo* (11 varieties), because this crop is one of the most important for local market gardeners, allowing investigation over a wide range of phenotypic characters. The seeds were obtained thanks to local suppliers and market gardeners.

Seedlings, transplantation in field, and crop maintenance

Crops were cultivated during spring/summer 2021. Following local farmer's practices, sowing was conducted in an uncontaminated substrate (Motte Bio 20, Proveen Substrates) and plantlets were grown in a greenhouse for a month before transplantation in situ. The field, located in the Bordeaux agglomeration presents mean climatic conditions (for the period 1981-2010) for the cultivation duration (June, July and August 2021) as follow: total precipitations of 62.2, 49.9 and 56.0 mm, mean minimal temperatures of 14.1, 15.8 and 15.7 °C, mean maximal temperatures of 24.5, 26.9 and 27.1 °C, mean temperatures of 19.3, 21.3 and 21.4 °C for June, July and August respectively. The field has a sandy soil (85 % sand, 5 % silt, and 10 % clay) classified as Umbrisol (Food and Agriculture Organization of the United Nations, 2015) with a neutral soil pH (6.83), a high organic matter content (9.7 %), and high cation exchange capacity (27 $cmol^{+}$ kg^{-1}). The soil dieldrin contamination (in average 123 ± 32 µg kg^{-1}) would mainly result from the past chronic use of insecticides against in particular the Colorado potato beetle (*Leptinotarsa decemlineata* Say).

Other OCPs (aldrin, hexachlorobenzene, and chlordanes) were detected in the soil with maximal concentrations of 6 µg kg^{-1}, leading to concentrations in the fruits below the limit of quantification (LQ): 0.5, 0.7, 0.4, and 0.4 µg kg^{-1} for hexachlorobenzene, aldrin and cis- and trans-chlordane, respectively. Transplantation into the field was performed as follows. For each plant, soil was sampled with a stainless spade in the 0-30 cm soil layer, sieved (1 cm), and homogenized before its introduction into a 25 L plastic pot. During this step, for each pot, soil samples (about 90 g) were collected for dieldrin quantification (results for each pot in SI 1). For each tested plant variety, four plant replicates were set up in individual pot, leading to a total of 112 plants. The 112 pots were placed at least 1 m apart in the field where the soil was sampled. They were buried at two third of their height to limit the increase of soil temperature in the pots and regularly inspected to cut off any roots that might emerge from the drainage holes. Depending on the climatic conditions, especially rainfall events, the crops were watered two or three times a week with local groundwater (dieldrin concentration in the well water was below 0.1 µg L^{-1}). Once a week, the crops were fertilized with an organic fertilizer (Bio-grow, Biobizz®), with NPK values of 4-3-6, diluted at 1:1000 (v:v) with groundwater.

Cucurbit fruit harvest, physiological data collection, and crop termination

The first fruits were harvested one month after the transplantation and then three times a week. They were harvested when they reached the commercial size or when no growth was observed for a few days. For each harvested fruit, the following parameters were recorded: pot number, harvest date, fresh weight (FW), distance to the root collar, length, and circumference. The crops were terminated 3.5 months after transplantation.

Dieldrin analysis in soil and fruits

Concerning the soil, dieldrin was analyzed after n-heptane (n-heptane, 99+ %, residues analysis, ACROS Organic, UK) extraction following the protocol described in Colin et al. (2022). This protocol was adapted to quantify dieldrin in fruits. Each fruit was analyzed individually, except for Mexican cucumbers, which were gathered and analyzed by plant and harvest date due to their small size (about 3 g FW per fruit). After harvest, each fruit was first washed with tap water for removing any soil particle and then wiped with paper towel. The entire fruit (flesh and skin together, even for fruits with non-edible peel) was then cut and stored at −18 °C. Frozen fruits were ground with a knife mill (Pulverisette 11, Fritsch) to obtain a homogenized puree. The following grinding cycle was applied: 5 s at 2000 rpm in reverse mode, 5 s at 2000 rpm, 10 s at 6000 rpm and 10 s at 10000 rpm. Liquid-liquid extractions were then performed with 20 g FW of this puree mixed with 10 mL of n-heptane into a 50 mL glass flask and shaken during 48 h (Roller 10, IKA™). Supernatant was recovered after 5 min of centrifugation at 1500 rpm (JOUAN™ C412) and placed in a 2 mL vial. Prior analysis, a few grains of sodium sulfate anhydrous (Fisher Scientific), used to remove residual water, and 50 ng of phenanthrene d-10 (Restek, France), used as internal standard, were added to the sample. Analyses were conducted as presented in Colin et al. (2022). Validation of this protocol for *Cucurbita* fruits is presented in SI 2

Statistical analysis

Calculations and statistical analyses were performed using the R open source software environment (version 4.2.1, (R Core Team, 2021)) and functions available in the 'stats', 'rstatix', 'outliers' and 'agricolae' packages (Alboukadel, 2021; De Mandiburu, 2021; Komsta, 2022; R Core Team, 2021). First, outliers were identified using Grubbs test and removed from the data set, then data normality was checked using the Shapiro-Wilk test. As data were not normal, mean comparisons were made with a Kruskal-Wallis test. Relationships between fruit dieldrin concentrations and other parameters such as soil dieldrin concentration, plant exposure time, and fruit biomass were determined using linear and quadratic models as well as Spearman's correlation tests. The later assessed monotonic relationships, which can be linear or not.

Results and discussion

Fruit biomass and harvest

In total, 334 fruits were harvested. Zucchinis were the most productive (Table 1) with an important discrepancy between varieties producing 1.5 (Adrielle) and 9.75 (Mirza) fruits per plant, in average. Harvest for cucumbers and watermelon was scarce, with small fruits due to the 2021 climatic conditions (rainfall higher than 1981-2010 seasonal standards in June, 141 mm vs. 62 mm, and mean temperatures 0.3 and 0.8 °C lower than mean seasonal values of 21.3 and 21.4 in July and August, respectively), promoting plant diseases and limiting growth for those plant species needing warm temperatures. Squash and pumpkin varieties produced in average only one fruit per plant, due to the limitation of the pot cultivation.

Intra and interspecific variability of dieldrin accumulation in fruits

Dieldrin concentrations in *Cucurbita* fruits are presented in Figure 1, with significant interspecific differences. Results are expressed on the fruit FW basis as water content variability in *Cucurbita* fruits is low (90 ± 5 %, Kusumiyati et al., 2021) but also to be able to compare with EU MRL. The mean concentrations ranged from 4.2 ± 7.0 in watermelons (*C. lanatus*) to 85.0 ± 19.4 µg dieldrin kg^{-1} FW in cucumbers (*C. sativus*). Dieldrin concentration ranked in the decreasing order: *C.*

sativus > *C. maxima* = *M. scabra* > *C. pepo* = *C. moschata* > *M. charantia* = *C. lanatus*. This differs from previous reports were zucchinis contained higher (Donnarumma et al., 2009) or similar (fruits from the market, Khandekar et al., 1982; historically contaminated soil, Saito et al., 2012) dieldrin concentrations than cucumbers. It may be explained by the use of different zucchini varieties, soil types, agricultural practices, root distribution in the soil profile, and dieldrin distribution in the soil bearing phases.

Momordica charantia and *C. lanatus* produced fruits with the lowest dieldrin concentrations. Khandekar et al. (1982) and Saito et al. (2012) reported similar results. Saito et al. (2012) grew two watermelon varieties on a historically contaminated soil (72 µg dieldrin kg^{-1} dry weigh (DW)) and analyzed the peeled fruits: for both varieties, dieldrin concentrations were lower than 1 µg kg^{-1} FW. However, extraction methods being different in Saito et al. (2012) and this study, it was difficult to compare the absolute concentrations. Nevertheless, as in this study, fruits from both watermelon varieties accumulated 4 to 50 times less dieldrin than the other *Cucurbita* species (Saito et al., 2012). The low accumulation level evidenced here would be a trait of the watermelon species rather than a bias due to the tested variety and the protocol used. Regarding *M. charantia*, to the authors knowledge, no other study reported pesticide residues in its fruits with known soil dieldrin concentration. A 3-year Indian survey analyzing market vegetables showed OCP concentration under the detection limit in seven bitter melons, while other vegetables contained some (Khandekar et al., 2009). However, soil and cultivation conditions can highly differ between the bitter melons and other vegetables analyzed.

Mean fruit dieldrin concentrations for all the *Cucurbita* varieties tested are presented in Figure 1. No significant intraspecific difference was found for *C. sativus* (four varieties), *C. maxima* (two varieties), and *C. moschata* (four varieties). For those plant species, all the tested varieties displayed mean dieldrin concentrations above the EU MRLs (20 µg kg^{-1} FW for cucurbits with edible peel except zucchinis, 30 µg kg^{-1} FW for cucurbits with non-edible peel). However, it cannot be ruled out that other varieties (untested here) with low accumulation pattern may exist for these three *Cucurbita* species.

Mean fruit dieldrin concentrations of *C. pepo* (11 varieties) showed significant differences. Intraspecific variability was important as the mean dieldrin concentration in zucchinis (in µg kg^{-1} FW) ranged from 4.9 ± 1.1 for Noire maraîchère to 70.3 ± 3.6 for Orélia. Several zucchini varieties accumulated, in average, lower dieldrin concentrations than the EU MRL (50 µg kg^{-1} FW). The percentage of fruits with dieldrin concentration below the EU MRL was 100 % for Noire maraîchère, more than 90 % for Mirza and Kopana, and between 65 % to 75 % for Anissa, Ola escaladora, and Gold Rush. The Noire maraîchère variety, which displayed lower dieldrin concentration in the fruits, as well as Mirza, which was the most productive variety out of the 11 zucchini tested, and presented dieldrin concentration mainly under the EU MRL, are relevant candidates for cultivation in dieldrin-contaminated soils.

It is worth mentioning the relationship between the fruit's skin color and fruit dieldrin concentration (SI 3). Among the zucchini varieties with mean dieldrin concentrations below the EU MRL, only one was not dark green, while the opposite was true for the varieties exceeding the EU MRL. The Orelia and Floridor varieties produced both yellow fruits, Adrielle pale green fruits, and Verte non coureuse d'Italie striped clear and dark green fruits. This difference of dieldrin content in fruits could thus be related to their carotenoids content. Indeed, previous studies showed that yellow and orange fruits contained more carotenoids than green ones (Houhou et al., 2021; Kim et al., 2012; Xu et al., 2021). Carotenoids, as well as other lipophilic compounds, are sequestrated into chromoplasts to prevent the detrimental effect of their excess on cellular functions (Egea et al., 2010). Houhou et al. (2021) highlighted that, triggering the transformation of chloroplasts to chromoplasts allowed to change zucchini peel color from green to yellow, suggesting that yellow fruits contained more chromoplasts than green ones. Dieldrin and other lipophilic organic contaminant might thus be sequestrated and therefore accumulated in chromoplasts with carotenoids. However, more investigations are needed to confirm this observation and hypothesis.

The bioconcentration factor (BCF), which is the ratio between dieldrin concentration in the fruit and that in the soil, was calculated to confirm that the intraspecific variability was not due to differences in soil contamination level (ranging between 58 and 192 µg dieldrin kg^{-1}), as the soil dieldrin concentration slightly differed for each pot (SI 2). The results showed a similar trend as the one evidenced for the mean values of fruit dieldrin concentration (SI 4). This suggested that intraspecific variability would be merely due to phenotypic differences, with possible genome x environment interaction, and not to small changes in the soil dieldrin concentration. Previous studies suggested that changes in POP accumulation in *Cucurbitaceae* plants can be related to differences in (i) contaminant solubilization from the soil involving the occurrence of citric acid in the root exudates (White et al., 2006b, 2003; White and Kottler, 2002; Yoshihara et al., 2014), and (ii) in the POP translocation, involving the major latex-like proteins (MLP) (Fujita and Inui, 2021). Differences in MLP gene expression or the affinity of the MLP produced for the POP (Inui et al., 2013; Iwabuchi et al., 2020), as well as root exudate composition and flux might be involved in the inter and intraspecific variabilities found in this study.

Based on Figure 2, dieldrin transfer from the soil to the zucchini fruit could be minimized by selecting zucchini varieties characterized by low fruit dieldrin concentration. As for metal(loid)s (Yang et al., 2010; Zeb et al., 2022), selection and breeding of cultivars adapted to the phytoavailable contaminant can be a suitable option for food safety, especially considering the persistent problems in line with OCP residues in arable lands (Tzanetou and Karasali, 2022). However, deeper investigations are needed to determine if low dieldrin accumulation character could be extrapolated to other OCPs. According to Namiki et al. (2018), the accumulation capacities in the shoots and roots of a plant species depend on the molecule considered.

For a same *C. pepo* variety, fruit dieldrin concentration varied (Figure 1), with, in some cases, maximal concentrations three to four times higher that minimal concentrations. To explain this observation, the influence of soil dieldrin concentration, plant exposure time, and fruit biomass on the dieldrin concentration of zucchini fruits was investigated.

Influence of soil dieldrin concentration on dieldrin concentration in zucchini fruits

The effect of soil dieldrin concentration on the fruit dieldrin concentration of *C. pepo* is presented in Figure 3. This was only carried out for this plant species, excluding the Adrielle variety, as the number of fruits harvested per pot, and therefore per soil dieldrin concentration, was too limited (< 3) to be relevant for the other plant species. The dieldrin concentration in potted soils cultivated with *C. pepo* plants varied from 60 to 180 $\mu g\ kg^{-1}$ DW. For the range of soil dieldrin concentrations considered, it had a low effect on the fruit dieldrin concentration. Only three zucchini varieties showed a significant positive linear relationship and a quadratic relationship could also be fitted for two of them (details of R^2, p-value, and equation in SI 5), although the slopes were weak (< 0.4) between dieldrin concentration in fruits and soil: Bélor (0.155), Floridor (0.362), and Gold Rush (0.244). For other zucchini varieties (Anissa, Noire maraîchère, Orélia, and Verte non coureuse d'Italie), dieldrin concentration in fruits did not show significant changes with increasing soil dieldrin concentrations, which could correspond to a plateau already reached. This is for instance a frequent plant response to excess of metal(loid)s (Affholder et al., 2020; Baker, 1981). For the last three zucchini varieties (Mirza, Kopana, and Ola escaladora), the range of soil dieldrin concentration was narrower, making it difficult to evidence any possible relationship. Determination of a threshold concentration of dieldrin in soil to obtain zucchini under the EU MRL was therefore not possible for most varieties. For the three zucchini varieties displaying a linear relationship, soil dieldrin concentrations lower than 60, 75 and 150 µg kg^{-1} DW (using the same extraction protocol) resulted in mean values of fruit dieldrin concentration under the EU MRL for Bélor, Floridor, and Gold Rush, respectively.

Influence of physiological factors on dieldrin concentration in zucchini fruits

Figure 4 presents the influence of plant exposure time on dieldrin concentration in *C. pepo* fruit. For five out of the 10 zucchini varieties tested, a significant linear relationship occurred between fruit dieldrin concentration and the plant exposure time. Significant quadratic relationships were also positive for two of these varieties (details of R^2, p-value, and equation in SI 5). Correlations were positive for Anissa, Floridor, Noire maraîchère, and Verte non coureuse d'Italie, and negative for Bélor. This agreed with previous studies highlighting significant differences in fruit dieldrin concentration of *C. pepo* collected at three development stages (Donnarumma et al., 2009). With the plant development, the root system increases, which could result in a more efficient nutrient but also contaminant uptake. Moreover, several plant species (rice, lupine, and maize) display changes in root exudation depending on the growth stage and the variety (Aulakh et al., 2001; Gransee and Wittenmayer, 2000; Lucas García et al., 2001). This may contribute to explain that *C. pepo* varieties were not behaving the same way in this study. Root exudation flux and composition could affect the desorption, uptake (White et al., 2006a, 2003; White and Kottler, 2002) and translocation of OCPs (Fujita and Inui, 2021). It could lead either to an increase or a decrease of OCP concentration in the fruit and other plant parts. Indeed, White et al. (2006) reported changes in p,p-DDE translocation as well as accumulation in roots, stems and leaves with plant exposure time for two *C. pepo* subspecies. In addition, the shoots are also growing, increasing the root-to-fruit distance, which was correlated to the plant exposure time, except for the climbing variety, Ola escaladora (SI 6). Whitfield Åslund et al. (2008, 2007) showed that polychlorinated biphenyl (PCB) concentration in stems and leaves decreased with increasing distance from the roots in pumpkin (*C. pepo spp. pepo*). As zucchini plants usually have a smaller internodal distance than pumpkin plants, it would probably minimize the root distance effect. However, the climbing variety Ola escaladora was assessed and no correlation was found between fruit dieldrin concentration and the root-to-fruit distance (SI 7), even with fruits harvested up to 2 m height. This suggested that dieldrin translocation to fruits differs from translocation to the above ground vegetative parts.

The fruit biomass significantly influenced the fruit dieldrin concentration only for the Floridor variety (Figure 5, SI 5). Nonetheless, this zucchini variety was the only tested one that produced yellow, round-shaped fruits. This could confirm that dieldrin concentration in zucchini was higher in skin than in flesh (Saito et al., 2012). Contrarily to the long-shaped fruits, when the biomass of the round-shaped fruit increases, the skin to flesh ratio decreases. Consequently, the whole fruit dieldrin concentration (determined here) decreased. For the other zucchini varieties, no dilution effect was observed with the increase of the fruit biomass.

Conclusion

To conclude, this study highlighted interspecific variability of dieldrin concentration in *Cucurbita* fruits, as well as intraspecific variability in zucchini fruits, some zucchini varieties producing fruits under the EU MRL (e.g. Noire maraîchère and Mirza). Further investigations would be needed to evidence the mechanisms leading to lower fruit dieldrin accumulation for these varieties such as i) the root exudate composition and flux and their impact on dieldrin bioavailability and uptake, ii) the MLP type and gene expression as *C. pepo* contains 21 MLP genes, which can have different affinity for POP (Fujita et al., 2022), and iii) pigment content such as carotenoids in fruits and its potential relationship with dieldrin concentration.

In addition, significant differences in the dieldrin concentration of zucchini fruits were evidenced for a same variety and even individual plant. However, out of the three soil and plant factors investigated, none had a ubiquitous effect on the varieties tested, suggesting that other parameters could be involved. For instance, MLP and carotenoid biosynthesis is regulated by environmental signals such as light, temperature, and water availability, which are changing during the cultivation period (Inui et al., 2013; Sun et al., 2010; Wang et al., 2016; Yuan et al., 2015).

Finally, considering the biomass that *Cucurbita* fruits represent and their dieldrin concentration, as well as the potential accumulation in the other plant parts (leaves, stems, and roots), the potential for dieldrin phytoextraction and dieldrin dissipation from the soil by the whole plant will be presented in a forthcoming paper.

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Table 1: number of fruits harvested by variety, plant species, and average biomass of fruits per variety. (mean value ± SE)

Plant species *Latin name* Common name	Variety	Morphological traits (color, shape, other)	Number of fruits harvested		Fruit biomass (g FW)
			Total/variety	Mean/plant	
Cucurbita pepo Zucchini	Adrielle	Clear green, long	3	1.50 ± 0.50	401 ±19
	Anissa	Dark green, long	32	8.00 ±2.04	349 ±27
	Bélor	Dark green, long	25	6.25 ±1.75	333 ±38
	Floridor	Yellow, round	16	4.00 ±0.91	382 ±31
	Gold Rush	Golden Yellow, long	26	6.25 ±1.38	318 ±21
	Kopana	Dark green, long	30	7.50 ±1.71	326 ±26
	Mirza	Dark green, long	39	9.75 ±0.48	303 ±17
	Noire maraîchère	Very dark green, long	21	5.25 ±1.38	377 ±27
	Ola escaladora	Dark green, climbing plant	12	3.25 ±1.31	339 ±34
	Orélia	Yellow, long	35	8.75 ±2.78	256 ±16
	Verte non coureuse d'Italie	Dark and clear green stripes, long	15	4.00 ±2.12	397 ±46
Cucumis sativus	Rocker	Green, short with spikes	11	2.75 ±0.63	189 ±18

Cucumber	Diapason	Green, long, smooth	6	1.50 ±0.65	182 ±22
	Le Généreux	Green, short with spikes	5	1.25 ±0.25	140 ±24
	Marketer	Green, short with spikes	8	2.25 ±0.48	215 ±30
Melothria scabra Mexican cucumber	Mexican	Very small fruits (2/3 cm), with dark green and clear green stripes	15	3.75 ±0.25	70 ±11
Momordica charantia Bitter melon	Moonlight	Clear green, bumpy skin	6	1.50 ±0.29	128 ±36
Cucurbita moschata Pumpkin	Longue de Nice	Dark green, long, old French variety	3	0.75 ±0.25	446 ±42
	Sucrine berry	Small, clear orange skin, dark orange flesh, old French variety	4	1.00 ±0.00	559 ±46
	Waltham Butternut	Camel skin	3	1.00 ±0.00	520 ±113
	Hercules butternut	Camel skin, fast growth	2	1.00 ±0.00	393 ±97
Cucurbita maxima	Orange summer	Small, dark orange skin and flesh	9	2.25 ±0.00	444 ±60

Squash	Rouge d'Etampes	Big, red skin, orange flesh	4	1.00 ±0.00	1922 ±213
Citrullus lanatus Watermelon	Sugar baby	Dark green skin, dark pink flesh, fast growing	4	1.00 ±0.00	368±108
TOTAL : 7	**24**		**334**	**3.56**	

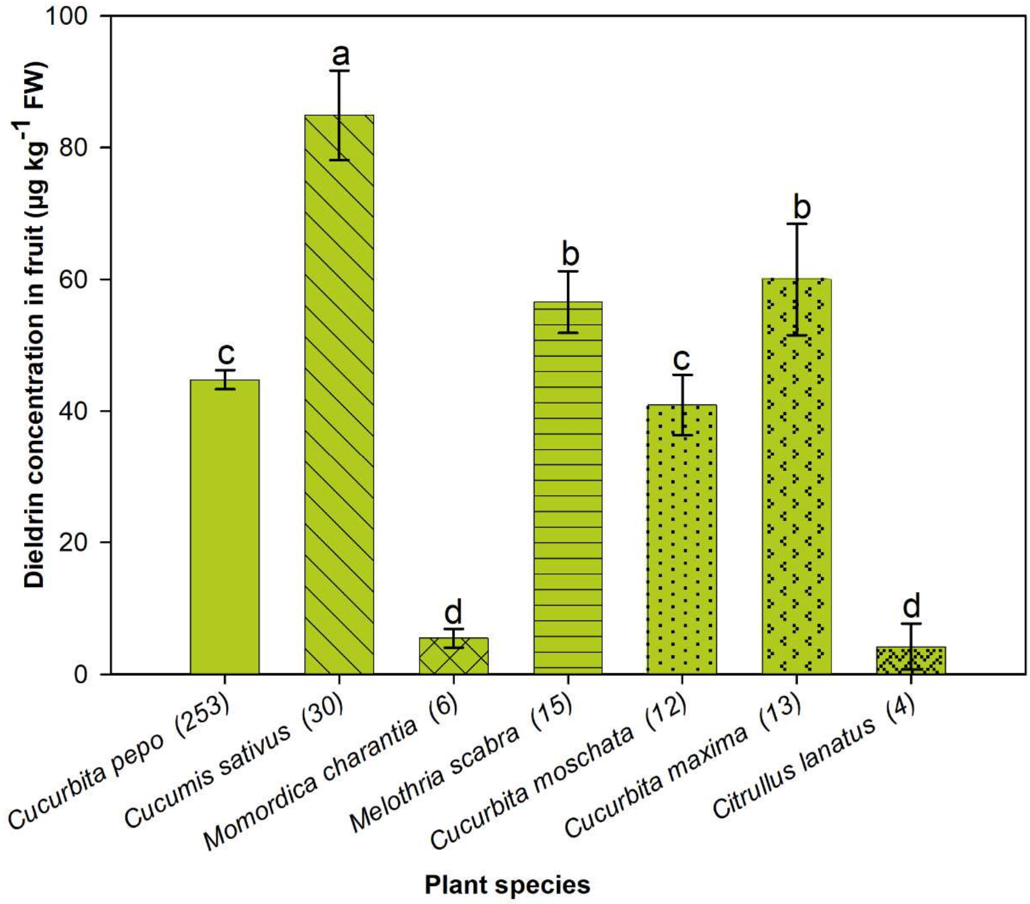


Figure 1: Mean values of fruit dieldrin concentration (µg kg^{-1} FW) depending on *Cucurbita* species (mean ± standard error (SE)). Mean values with different letters are significantly different (Kruskal-Wallis, $p < 0.05$). The number of fruits analyzed for each species is presented in brackets.

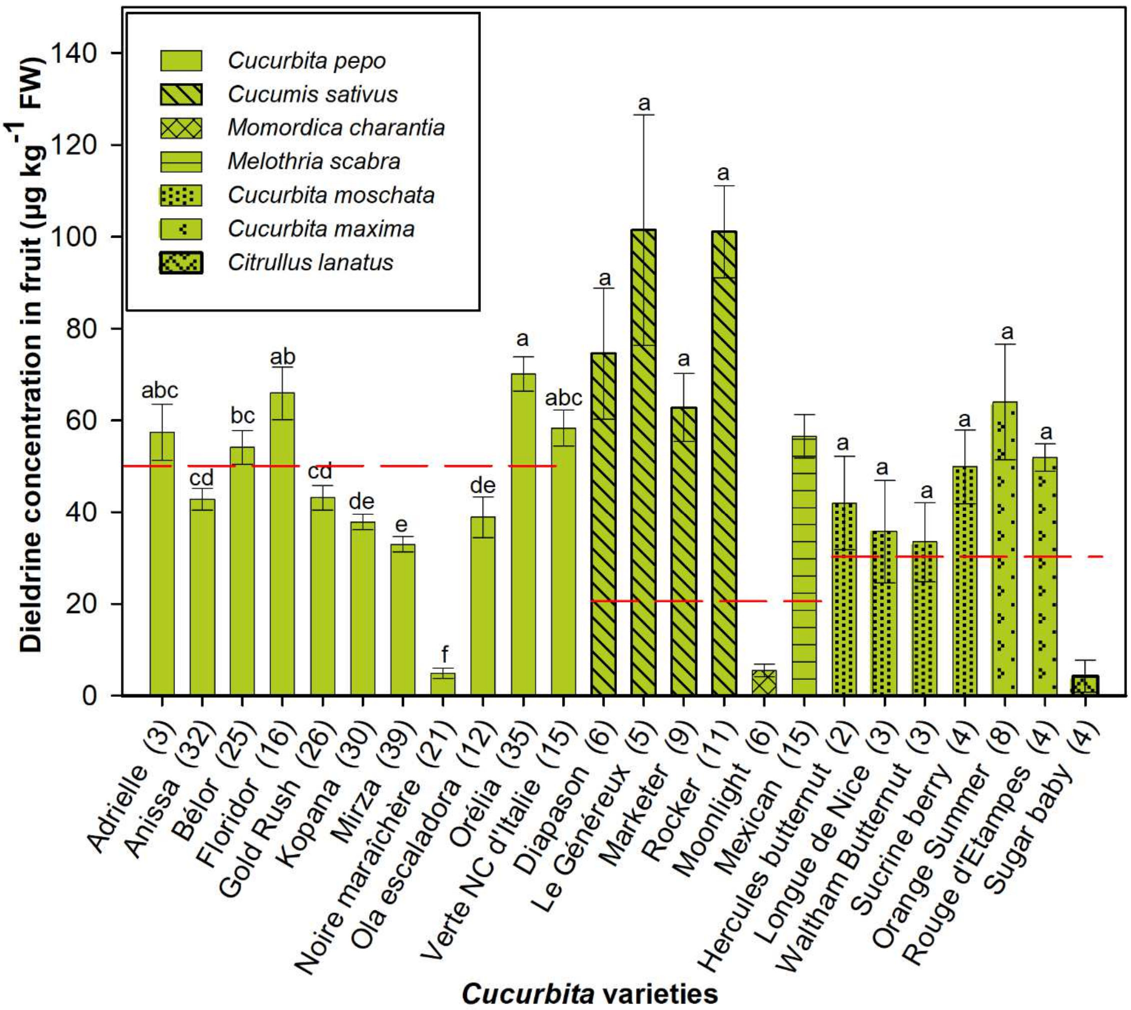
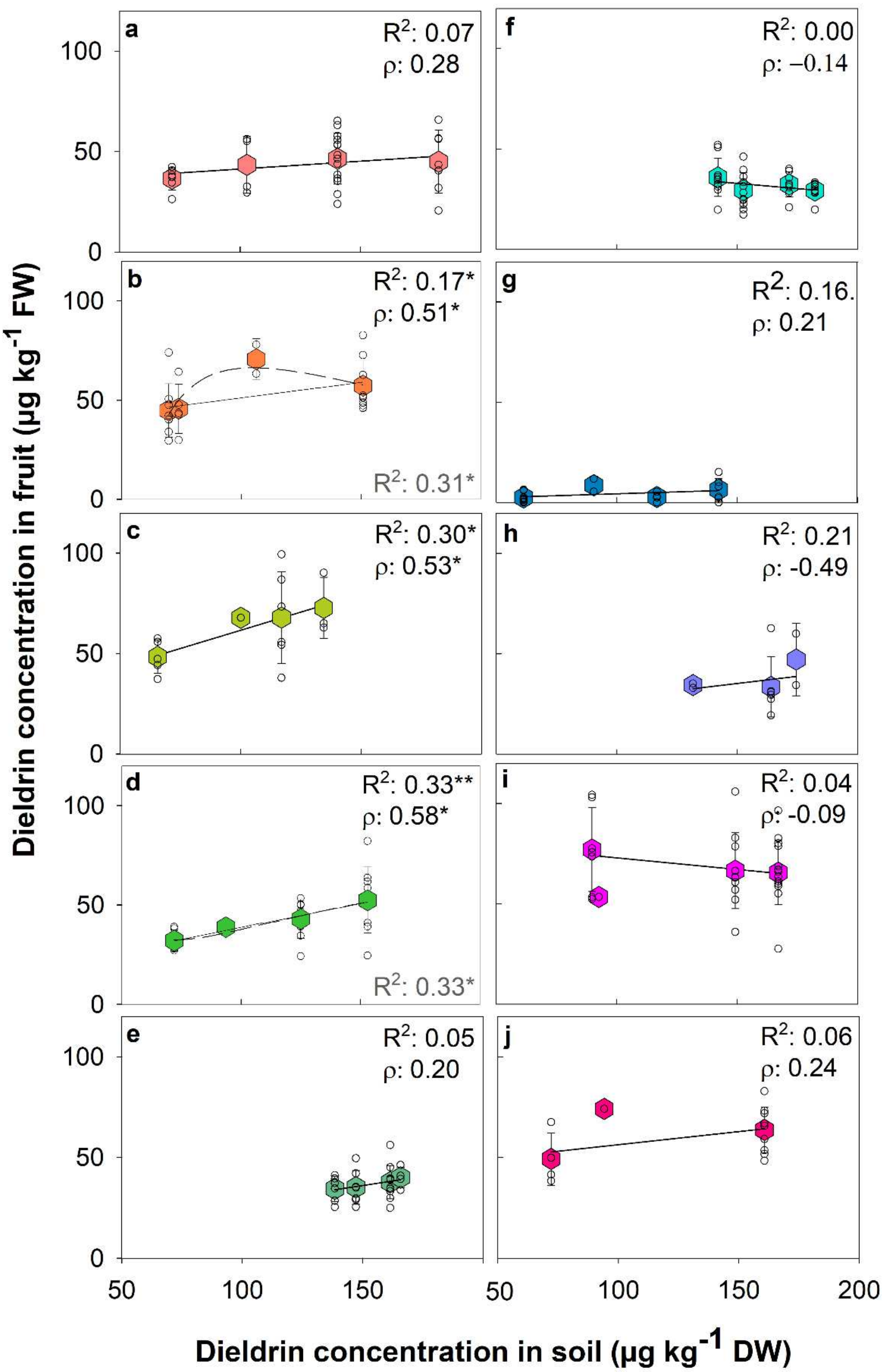
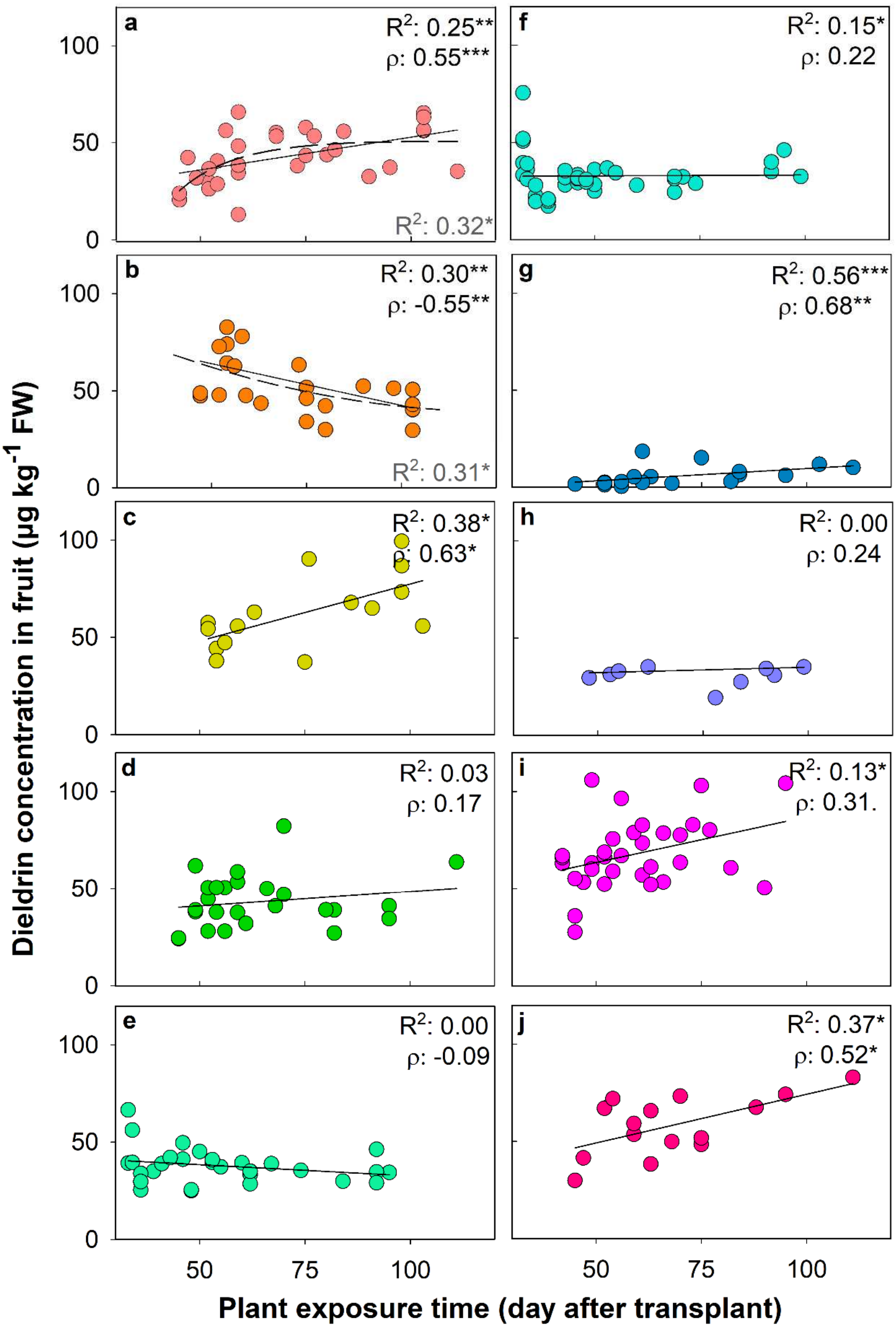


Figure 2: mean values (± SE) of fruit dieldrin concentrations (µg kg^{-1} FW) depending on *Cucurbita* varieties. For a same plant species, mean values with different letters significantly differed (Kruskal-Wallis, $p < 0.05$). Dashed red lines represent the EU MRLs for dieldrin corresponding to the various plant species: 50 µg kg^{-1} FW for *C. pepo*, 20 µg kg^{-1} FW for Cucurbits with edible peel, and 30 µg kg^{-1} FW for Cucurbits with non-edible peel. The number of fruits analyzed for each variety is presented in brackets.


a
R²: 0.07
ρ: 0.28
b
R²: 0.17*
ρ: 0.51*
R²: 0.31*
c
R²: 0.30*
ρ: 0.53*
d
R²: 0.33**
ρ: 0.58*
R²: 0.33*
e
R²: 0.05
ρ: 0.20
f
R²: 0.00
ρ: –0.14
g
R²: 0.16.
ρ: 0.21
h
R²: 0.21
ρ: -0.49
i
R²: 0.04
ρ: -0.09
j
R²: 0.06
ρ: 0.24
Dieldrin concentration in fruit (µg kg⁻¹ FW)
Dieldrin concentration in soil (µg kg⁻¹ DW)

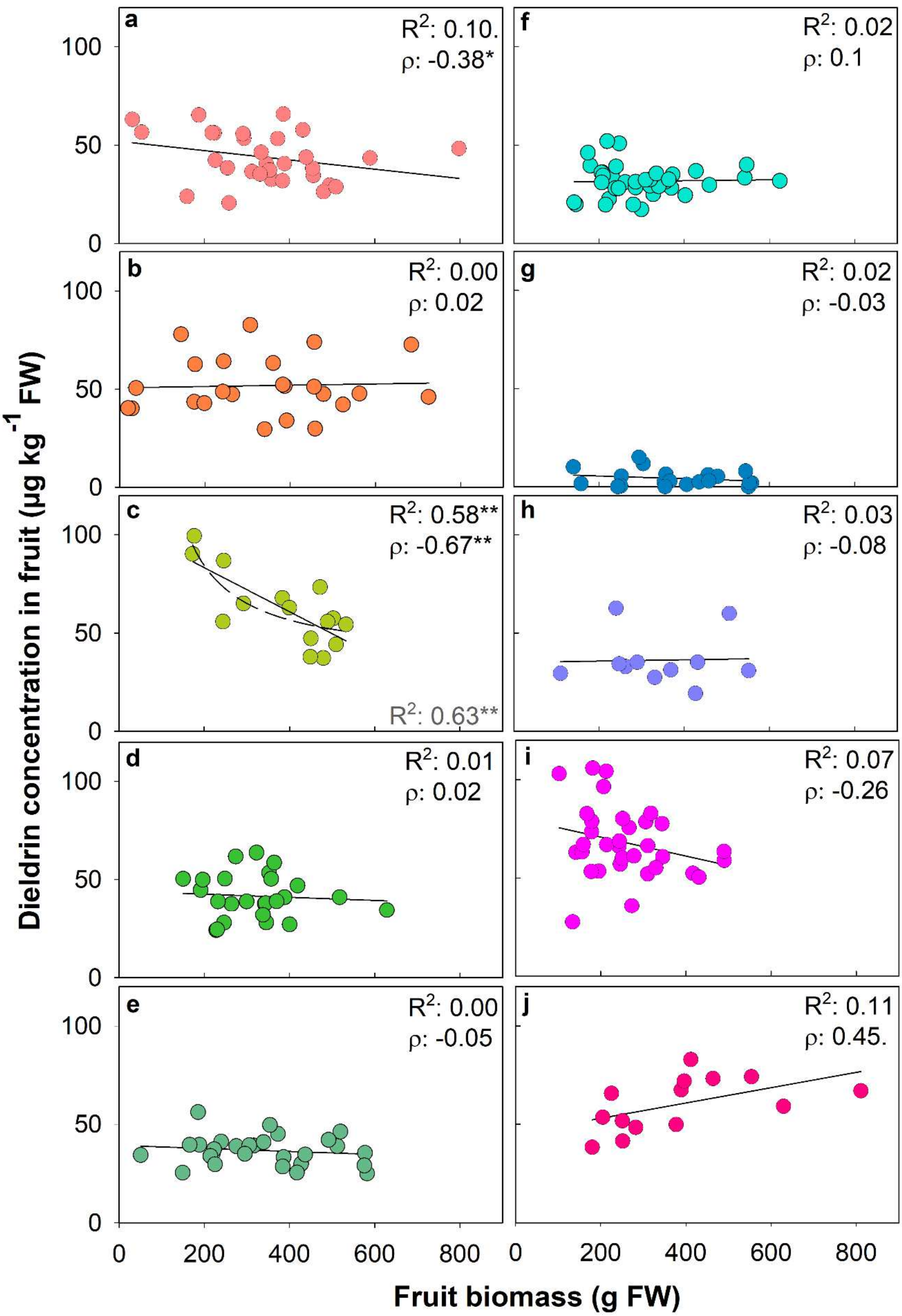
488

Figure 3: Fruit dieldrin concentration (µg kg^{-1} FW) depending on the soil dieldrin concentration (µg kg^{-1} DW),for various zucchini varieties: a) Anissa, b) Bélor, c) Floridor, d) Gold Rush, e) Kopana, f) Mirza, g) Noire maraîchère, h) Ola escaladora, i) Orélia, and j) Verte non coureuse d'Italie. Each hexagon corresponds to the mean dieldrin concentration of fruits (± SE) from one pot. Dots represent the dieldrin concentration for individual fruit harvested on the corresponding pot. R^2: coefficient of determination for linear model plotted as a black solid line, ρ: Spearman's rank correlation coefficient. When significant, quadratic regression is plotted as a dashed black line, the corresponding R^2 is presented in grey. Significance levels: $p < 0.001$: ***; $p < 0.01$: **; $p < 0.05$: *; and $p < 0.1$: .



a
R2: 0.25**
ρ: 0.55***
R2: 0.32*
b
R2: 0.30**
ρ: -0.55**
R2: 0.31*
c
R2: 0.38*
ρ: 0.63*
d
R2: 0.03
ρ: 0.17
e
R2: 0.00
ρ: -0.09
f
R2: 0.15*
ρ: 0.22
g
R2: 0.56***
ρ: 0.68**
h
R2: 0.00
ρ: 0.24
i
R2: 0.13*
ρ: 0.31.
j
R2: 0.37*
ρ: 0.52*
0
50
100
50
75
100
Dieldrin concentration in fruit (µg kg-1 FW)
Plant exposure time (day after transplant)

Figure 4 : Fruit dieldrin concentration (µg kg^{-1} FW) depending on the plant exposure time (days after transplant) for various varieties of *C. pepo*: a) Anissa, b) Bélor, c) Floridor, d) Gold Rush, e) Kopana, f) Mirza, g) Noire maraîchère, h) Ola escaladora, i) Orélia, and j) Verte non coureuse d'Italie. Each dot corresponds to one fruit. R^2: coefficient of determination for linear model plotted as a black solid line, ρ: Spearman's rank correlation coefficient. When significant, quadratic regression is plotted as a dashed black line, the corresponding R^2 is presented in grey. Significance levels: $p < 0.001$: ***; $p < 0.01$: **; $p < 0.05$: *; and $p < 0.1$: ..


a
R2: 0.10.
ρ: -0.38*
b
R2: 0.00
ρ: 0.02
c
R2: 0.58**
ρ: -0.67**
R2: 0.63**
d
R2: 0.01
ρ: 0.02
e
R2: 0.00
ρ: -0.05
f
R2: 0.02
ρ: 0.1
g
R2: 0.02
ρ: -0.03
h
R2: 0.03
ρ: -0.08
i
R2: 0.07
ρ: -0.26
j
R2: 0.11
ρ: 0.45.
Dieldrin concentration in fruit (μg kg-1 FW)
Fruit biomass (g FW)
0
50
100
0
200
400
600
800

509

Figure 5: Fruit dieldrin concentration ($\mu g\ kg^{-1}$ FW) depending on the fruit biomass (g FW) for various varieties of *C. pepo*: a) Anissa, b) Bélor, c) Floridor, d) Gold Rush, e) Kopana, f) Mirza, g) Noire maraîchère, h) Ola escaladora, i) Orélia, and j) Verte non coureuse d'Italie. Each dot corresponds to one fruit. R^2: coefficient of determination for linear model plotted as a black solid line, ρ: Spearman's rank correlation coefficient. When significant, quadratic regression is plotted as a dashed black line, the corresponding R^2 is presented in grey. Significance levels: $p < 0.001$: ***; $p < 0.01$: **; $p < 0.05$: *; and $p < 0.1$: ..


1. Inter and intraspecific differences of dieldrin concentration in Cucurbita fruits
Soil historically contaminated
Dieldrin concentration ~ 100 µg kg-1
Dieldrin concentration in fruits
Species
Varieties number
identification of intraspecific differences
Cucumis sativus
4
X
Cucurbita maxima
2
X
Melothria scabra
1
NA
Cucurbita pepo
11
✓
Cucurbita moschata
4
X
Momordica charantia
1
NA
Citrullus lanatus
1
NA
NA: not applicable
2. Effect of soil, plant exposure time and fruit biomass on dieldrin concentration in zucchinis
Varieties significantly affected
a. Soil dieldrin concentration
Dieldrin concentration in fruits
Gold Rush
Floridor
Bélor
b. Plant exposure time
Dieldrin concentration in fruits
Dieldrin concentration in fruits
Anissa
Floridor
Noire maraichère
Verte Italie
Bélor
c. Fruit biomass
Dieldrin concentration in fruits
Floridor