Exposure to PPPs by gender in farmers, neighbours and consumers

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List of abbreviations

C	Consumer
CH	Switzerland
CSS	Case study site
CZ	Czech Republic
DDE	Dichlorodiphenyl dichloroethylene
DDT	Dichlorodiphenyl trichloroethane
DK	Denmark
EU	European Union
F	farmer
FR	France
GI	Gastrointestinal
HCB	Hexachlorobenzene
HCH	hexachlorocyclohexane
HR	Croatia
IT	Italy
LOD	Limit of determination
LOQ	Limit of quantification
N	Neighbour
NHANES	National Health and Nutrition Examination Survey
NL	The Netherlands
PBPK	physiologically-based pharmacokinetic
PPE	Personal Protective Equipment
PPP	Plant Protection Product
PT	Portugal
SL	Slovenia
SP	Spain
SPRINT	Sustainable Plant Protection Transition
UK	United Kingdom

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Contents

Summary of main findings

Previous studies indicated that exposure to pesticides may depend on gender. For agriculture, this is well-established in occupational settings. The aim of this study is to assess the exposure to pesticides (active substances from plant protection products, PPPs) by gender using chemical analysis of samples from individual study participants collected during a population-based study in 10 EU countries during the growing season of 2021. Individual samples of in total 669 study participants (337 women and 332 men) were available from subgroups of farmers (N=235), neighbours (N=208) and consumers (N=236). In total 648 blood samples, 664 urine samples, 655 faeces samples and 641 silicon wristbands were analysed. These analyses covered 208 active pesticide ingredients one synergist, and 46 related metabolites. The numbers of reported analyses varied depending on the matrix and method capabilities. The gender analysis was done for detection frequencies above the limit of detection (LOD) and for concentrations based on results above the limit of quantification (LOQ). The effects of exposure on the individuals' health have not been studied. The exposures were very similar when compared by gender and subgroup for different sample matrices:

a. In blood the median number of observed pesticides and metabolites per sample was two. Frequency distributions, ranging from 0 to 10, were very similar across women and men, and across subgroups by gender. No significant gender-related differences were observed for blood concentrations except for bromoxynil in a very limited number of study participants.
b. In urine of men, the median number of pesticides and metabolites per sample was 8, 7, and 7 in farmers, neighbours and consumers, respectively, with a range from 0 to 20. In urine of women, the median was 7 with very similar frequency distributions across subgroups. Urinary metabolite concentrations were very similar across gender and subgroups with exceptions for imidacloprid olefin, pyrimethanil-OH and propamocarb.
c. In faeces the median number of pesticides and metabolites was 3 across gender in all subgroups and frequency distributions were similar ranging from 0 to 19. Concentrations of pesticides residues in faeces were very similar across gender and subgroups except for fenhexamid. Permethrin, the metabolite fipronil sulfone and the synergist piperonyl butoxide that are used as PPP but also have an authorisation as biocide and veterinary drug also showed some gender related deviations.
d. Farmers had the highest median number of pesticides per wrist band. The median number of pesticides per wristband shows a clear pattern in mean numbers that is repeated for across genders: farmers had the highest median number of pesticides per wristband (23 in women and 30 in men), followed a lower number in neighbours (21 in both women and men) and the lowest number in consumers (17 in women and 16 in men). A gender-dependent effect was observed in farmers but not statistically significant due to a high variability in this parameter. Concentrations in wristbands were very similar across gender for all subgroups.

The results of a standardised approach to pesticide exposure assessment across different crops, climate zones and farming practices indicates few gender-dependent differences. Frequency distributions were very similar by gender for the entire study population and

also within each of the subgroups by gender. No significant gender-related differences were observed with exceptions for a limited number of specific pesticides based on a very limited number of observations in each case. Some of these findings were consistent where others were inconsistent and should therefore be interpreted with caution. In future studies gender analyses are needed to confirm gender-related effects and explore underlying causes.

1. Introduction

This deliverable describes the exposure of residues found in humans, stratified by gender. In our databases we have collected information on biological sex (male/female). Gender also relates to social and behavioural characteristics that were studied only to a very limited extent. Most of the information on the gender distribution in the study population was already reported as one of the main characteristics of the study population as a whole in D2.3 (Vested et al., 2022). In that report some of the gender-based differences in concentrations in human samples were discussed but only related to the top-10 of the most frequently detected pesticides.

The main objective of this study is to compare these exposure data for the gender aspect. These data were collected from male and female participants of the case study sites (CSS) in a cross-sectional study in 10 EU countries during the growing season of 2021. More information about the data collection can be found in D2.3, and D2.4 and in the published protocol (Silva et al., 2021). The participating countries are also listed as part of the supplementary Table S1. The exposure data relate to the SPRINT list of pesticides approved as active ingredients for use as PPP. Some of these pesticides also have an approval for biocide and/or veterinary drug use. The same applies to the co-formulant piperonyl butoxide that is used as a synergist in pyrethroid-based products. For some active ingredients the authorisation was discontinued in the EU but exposure still occurs presumably from environmental sources and in some cases through imported agricultural products because residues of non-authorised pesticides are still tolerated in those imported goods up to a default maximum residue level. The exposure data used for this report were obtained through analysis of blood, urine and faeces (internal exposure), and from silicon wristbands (as a proxy for the individual's non-dietary exposure).

2. Methods

2.1. Data collection and chemical analyses

Gender-related exposure information was collected during the field campaign (Silva et al., 2021). Here we analysed the data at the individual level by wristbands and by human biomonitoring of blood, urine and faeces. Chemical analyses have been performed centrally by laboratories according to methods that were briefly described in D2.3 and that are currently in preparation for peer-reviewed publication.

2.2. Calculations and analysis

Analyses were done on individual matrices of the following subgroups: farmers (F), neighbours (N) and consumers (C) for all case study sites (CSS) in all of the ten participating countries from the EU. Gender differences were assessed based on two parameters: frequency of detects above the limit of detection (>LOD) and the concentrations of pesticides and/or their metabolites with quantifiable levels, i.e. with concentrations above the limit of quantification (>LOQ).

3. Results

The gender ratio in the study sample is presented by country in **Figure 3.1**. For this report we have collated the data from 10 EU countries. Overall, the study population is balanced by gender. Males were overrepresented in CH, CZ, DK and FR and females were overrepresented in HR, IT, NL, PT, SL and SP.

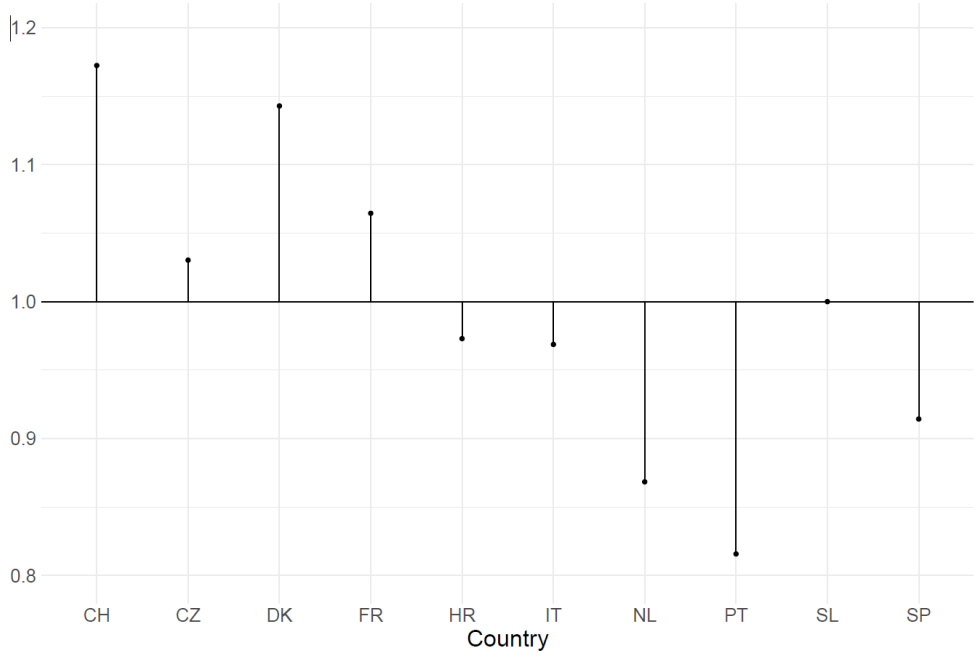

Figure 3.1. M/F ratio disaggregated by CSS country.

The characteristics of the analysed sample types that were collected from individual participants are shown in **Table 1**. These samples were available from females and males in three different subgroups: farmers, neighbours and consumers. Overall gender was balanced but males were somewhat overrepresented in the samples analysed from farmers. For neighbours females were overrepresented and for consumers even more. Male and female participants were comparable by age (see **Table S1** for specification by country).

Table 1. Numbers of analysed samples per matrix.

Matrix	Analy-sed	Farmers (N=225)		Neighbours (N=208)		Consumers (N= 236)	
		Female (N=101)	Male (N=124)	Female (N=107)	Male (N=101)	Female (N-=129)	Male (N=107)
Blood	648	99	122	106	100	115	106
Urine	664	101	123	105	99	129	107
Faeces	655	90	115	100	108	121	121
Wristbands	641	99	122	104	97	120	99

3.1. Blood pesticide frequency and concentration

In **Figure 3.2.** the median number of detected pesticides per blood sample is presented. This numbers ranged from 0 to 10. For the subgroups the median values are two per blood sample in all subgroups and genders. Within the subgroups the distribution of detected frequencies is very similar with the exception for male farmers that shows a slightly different distribution with a somewhat higher 25th and 75th percentile value.

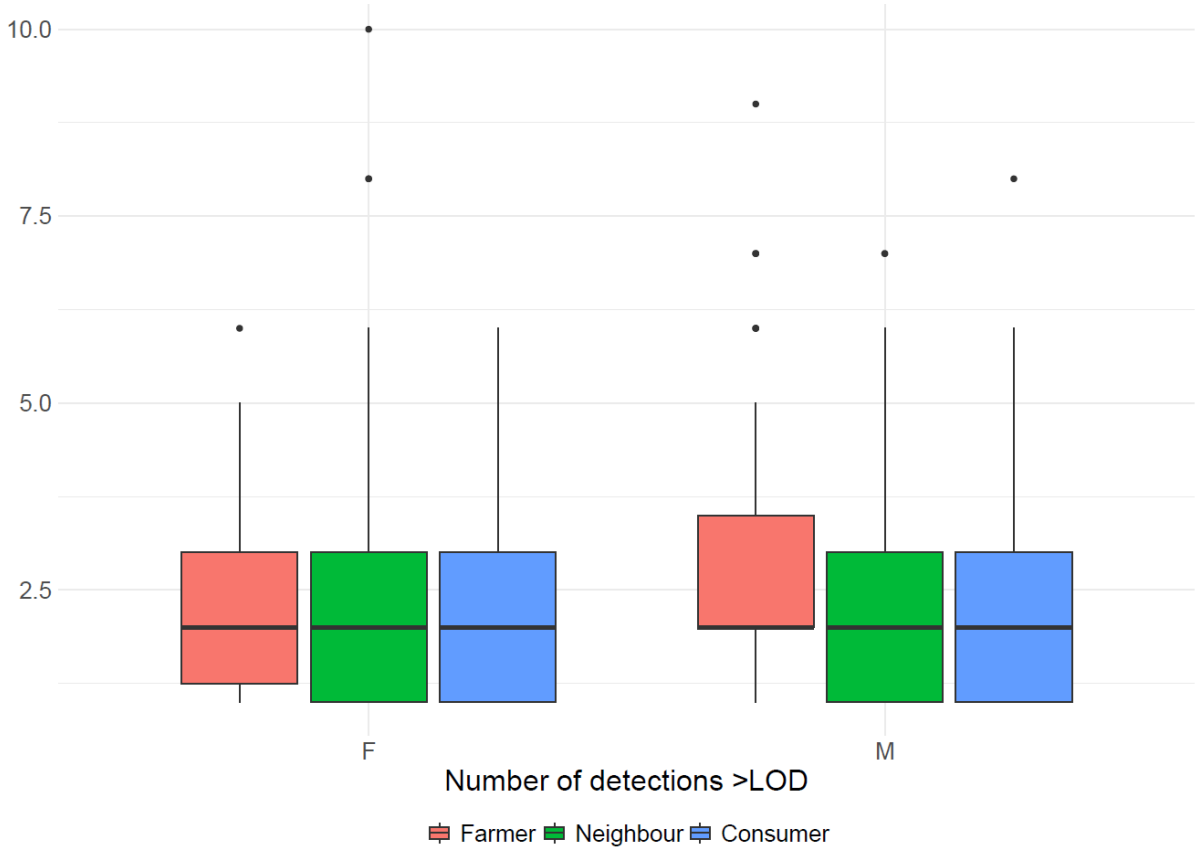

Figure 3.2. Number of detected pesticides (>LOD) per blood sample based on the analyses of 655 analysed samples stratified by gender and disaggregated by subgroup (farmer, neighbour, consumer). The boxes indicate P25, median and P75. The whiskers indicate the P5 and P95. The dots represent individual results outside the P5 - P95 range.

The concentrations of pesticides and their metabolites detected in blood plasma are all very low (< 0.1 µg/L) with only few exceptions, e.g. for the herbicide bromoxynil five male farmers had higher blood concentrations (**Figure 3.3.**). This was also the case for two female consumers and one male consumer. There is also a single high value (statistical outlier) for the fluopyram metabolite fluopyram benzamide for a female farmer and a single somewhat higher value for MCPA in a male farmer (see **Figure S1** in supplementary material for the complete dataset). Fipronil concentrations were higher in consumers but no difference by gender was observed.

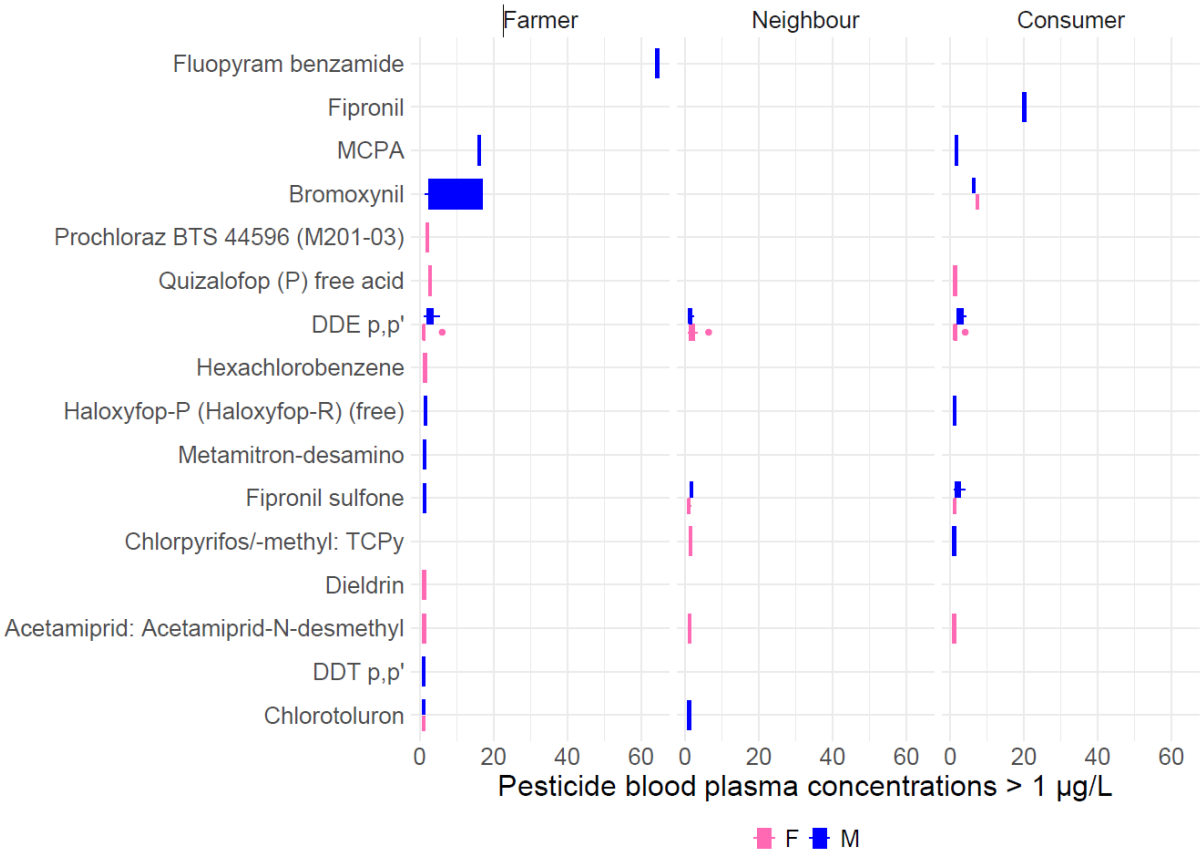

Figure 3.3. Blood plasma concentrations of pesticides in µg/L from high to low concentrations disaggregated by subgroups and gender (F = female, M = male). See the legend of **Figure 3.2.** for explanation of the symbols used in this boxplot and supplementary **Figure S1** for the complete data.

3.2. Urinary excretion of metabolites by frequency and concentration

In **Figure 3.4.** the median number of detected metabolites per urine sample is presented. This number ranges from 0 to 20 and is similar between males and females. Within females the distributions are the same for all subgroups. For males these medians were 8, 7, and 7 for farmers, neighbours and consumers, respectively. For farmers the P75 and P25 were higher than for male consumers and the frequency distribution in neighbours was intermediate. The distributions deviated from a normal distribution with the number in neighbours slightly skewed towards higher values and the distribution in consumers skewed towards lower values.

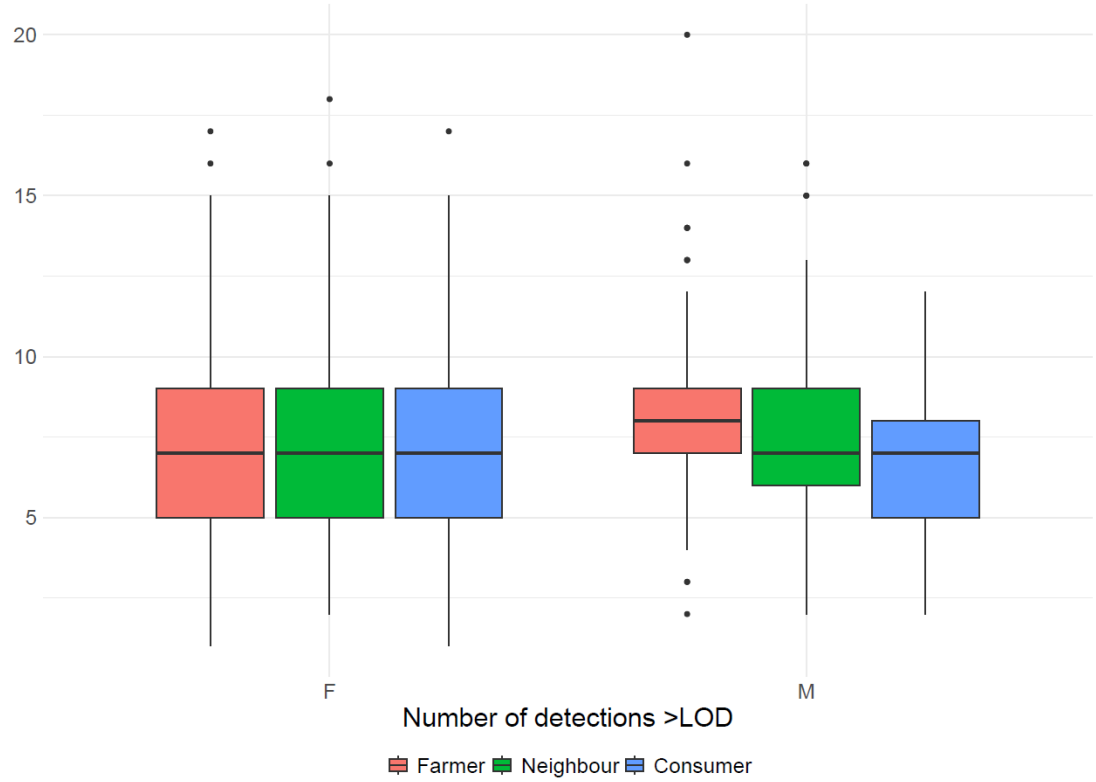

Figure 3.4. Number of detected pesticides (>LOD) per urine sample based on 664 analysed samples stratified by gender and disaggregated by subgroup (farmer, neighbour, consumer). For an explanation of the boxplot, see the legend of **Figure 3.2.**

The urinary metabolite excretions were overall low and not much different by subgroups and gender (**Figure 3.5.**). Outliers were observed for different metabolites but with no clear pattern that stands out for either of the subgroups. The pirimiphos-methyl metabolite DEAMPY and the tebuconazole metabolite tebuconazole-OH were scattered in all subgroups, not indicating a difference by gender. The neonicotinoid insecticide imidacloprid olefin was higher in males compared to females for both farmers and neighbours. Pyrimethanil-OH urinary concentrations were higher in females than males for farmers and neighbours. The fungicide propamocarb was also higher in females compared to males but only in neighbours.

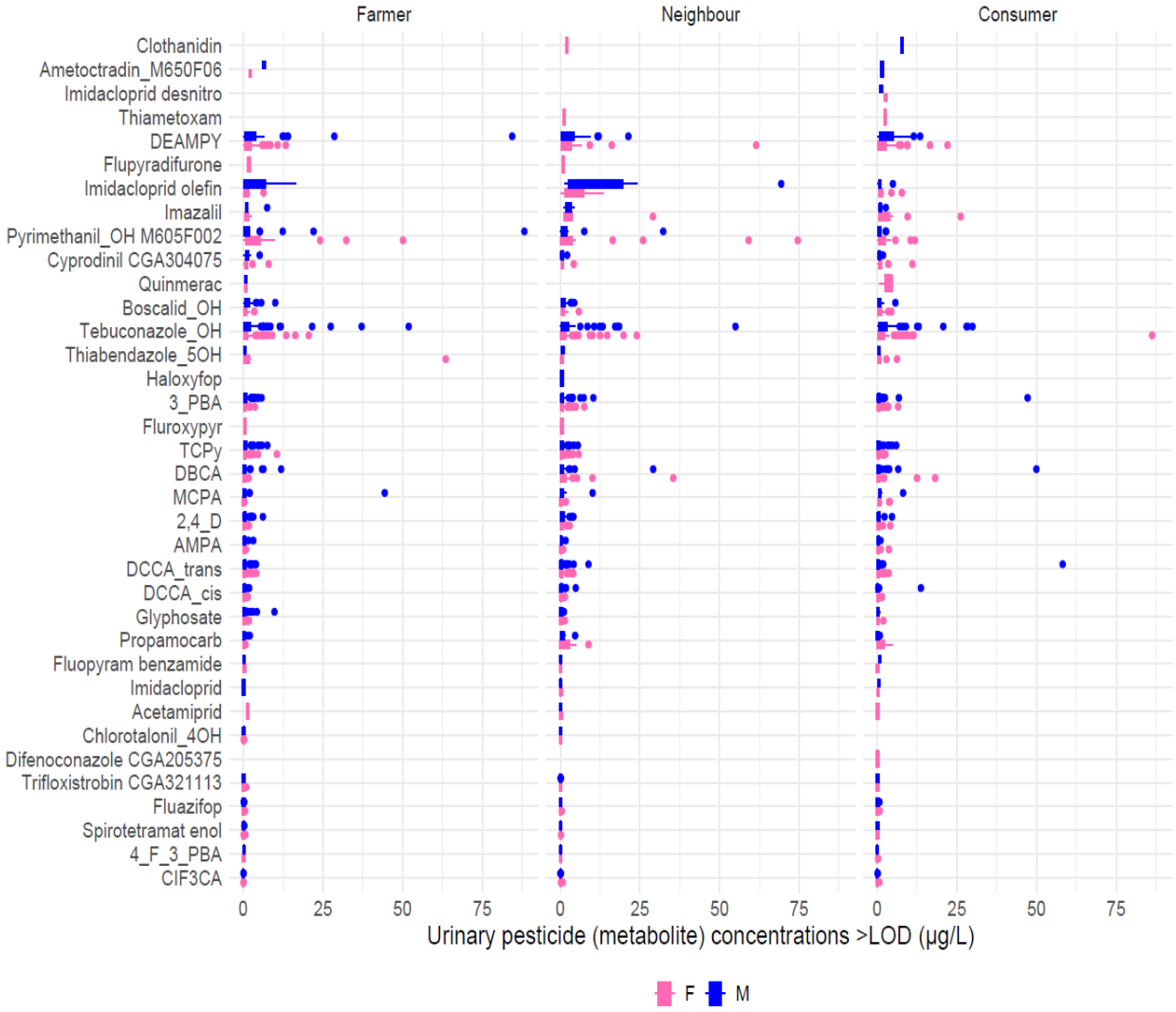

Figure 3.5. Urinary metabolite concentrations in µg/L disaggregated by subgroup. See the legend of **Figure 3.2.** for explanation of the symbols used in this boxplot.

3.3. Faeces pesticides frequency and concentration

In **Figure 3.6.** the median number of detected pesticides per faeces sample is presented. This number ranges from 0 to 19. For each of the subgroups the median is 3 in both genders and all subgroups. The distributions of detects per sample are also very similar.

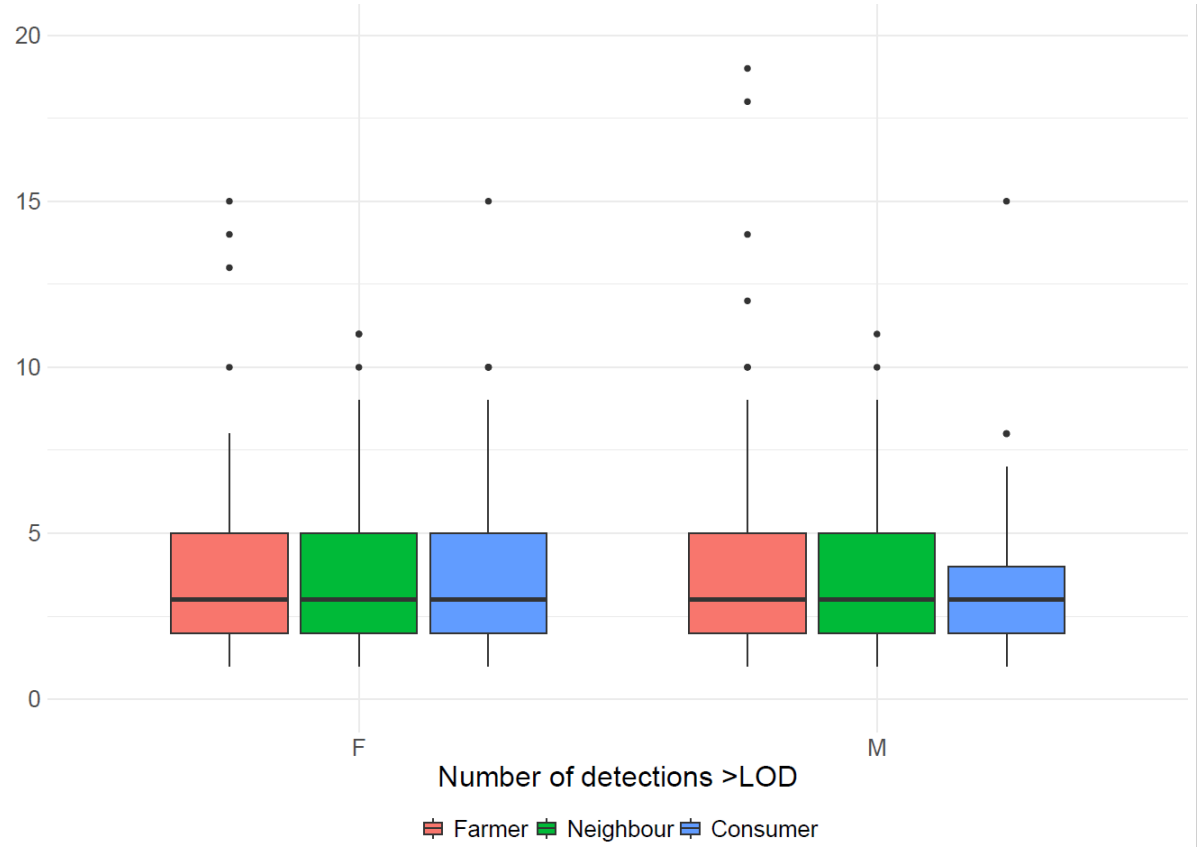

Figure 3.6. Number of detected pesticides (>LOD) per faeces sample based on 646 analysed samples stratified by gender and disaggregated by subgroup. See the legend of **Figure 3.2.** for more information about the symbols used in these boxplots.

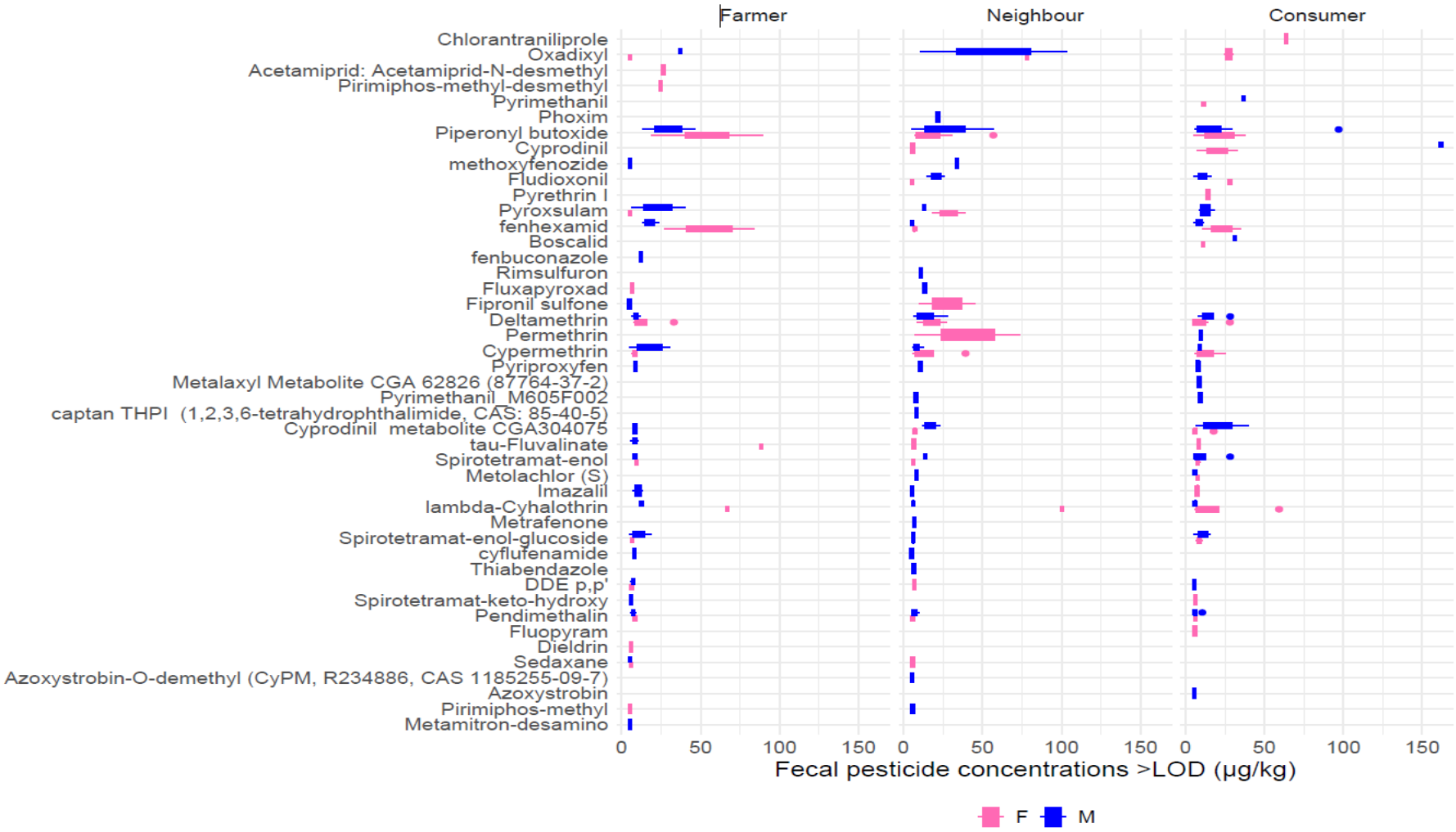

Figure 3.7. Pesticides and metabolites in µg/kg faeces disaggregated by subgroup (subset of compounds with elevated concentrations). The results of glyphosate are presented separately in **Figure 3.8** and the complete dataset is provided in supplementary **Figure S2**. See the legend of **Figure 3.2.** for more information about the symbols used in these boxplots.

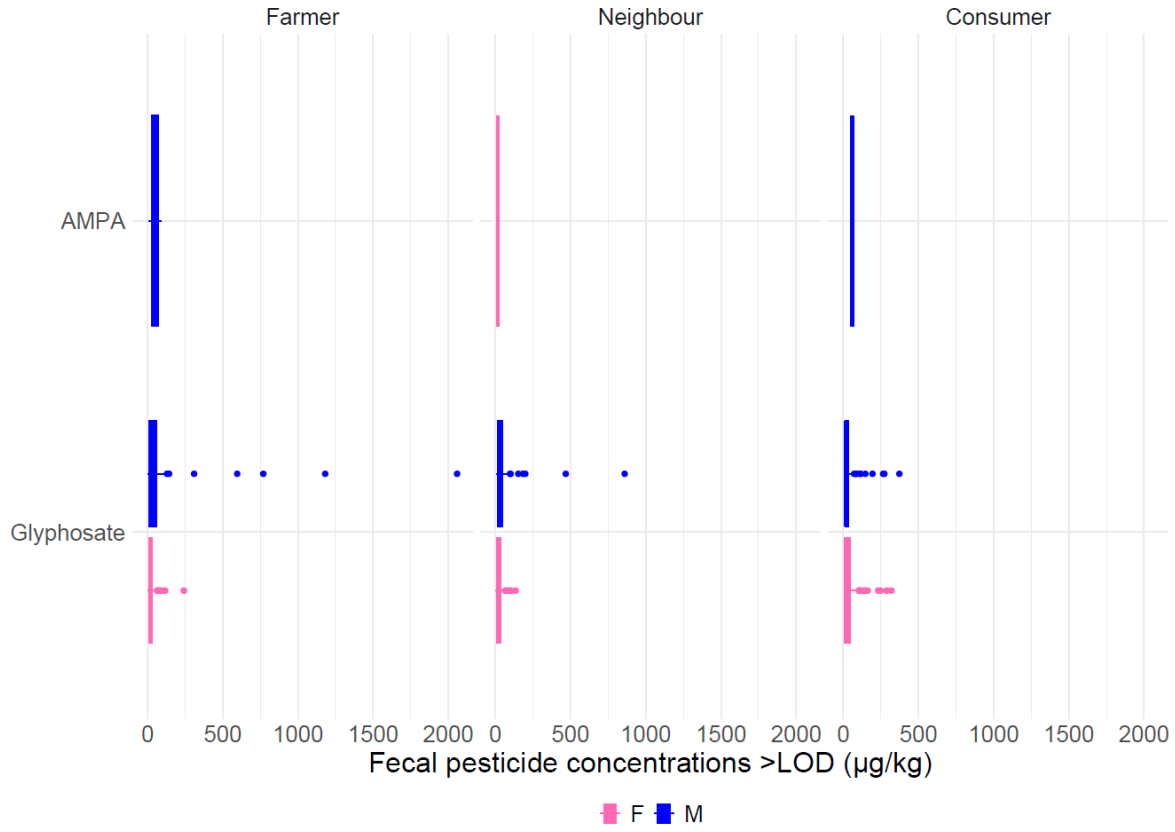

Figure 3.8. AMPA and glyphosate concentrations in µg/kg faeces. See the legend of **Figure 3.2** for more information about the symbols used in these boxplots.

In **Figure 3.7.** shows a subset of elevated pesticides and metabolite concentrations in faeces. See **Figure 3.8** for the results of glyphosate and AMPA and **Figure S2** for the complete dataset. For some components the box plots indicate a gender-based difference. In farmers the higher concentration range for the synergist piperonyl butoxide and the fungicide fenhexamid in female compared to male is remarkable. Female neighbours appear to be higher exposed to the biocides permethrin and the fipronil metabolite fipronil sulfone, compared to males. Both compounds are also authorised as biocides for companion animals. For other detected compounds the detection frequencies are too low to make any useful inferences. For glyphosate and AMPA the detection frequencies were higher than 70% but median concentrations were very low and similar by gender and subgroup with only some occasional higher values primarily in male farmers and neighbours compared to consumers (see **Figure 3.8.**).

3.4. Wristbands

In **Figures 3.10., 3.11. and 3.12.** the detection frequency of each pesticide in the wristbands by gender is presented, stratified by farmer, neighbour and consumer, respectively. For farmers, P95 are 59% and 68%, for females and males, respectively. For neighbours, P95 are 59% and 59%, for females and males, respectively. For consumers,

P95 are 57% and 54%, for females and males, respectively. For each of the subgroups the medians are low: 5%, 2% and 1% for female farmers, neighbours and consumers, respectively. For males these medians were 9%, 4%, and 2%, respectively. For farmers the detection frequencies for most pesticides are higher in males compared to females. For neighbours and consumers detection frequencies are similar by gender (**Figure 3.12.**).

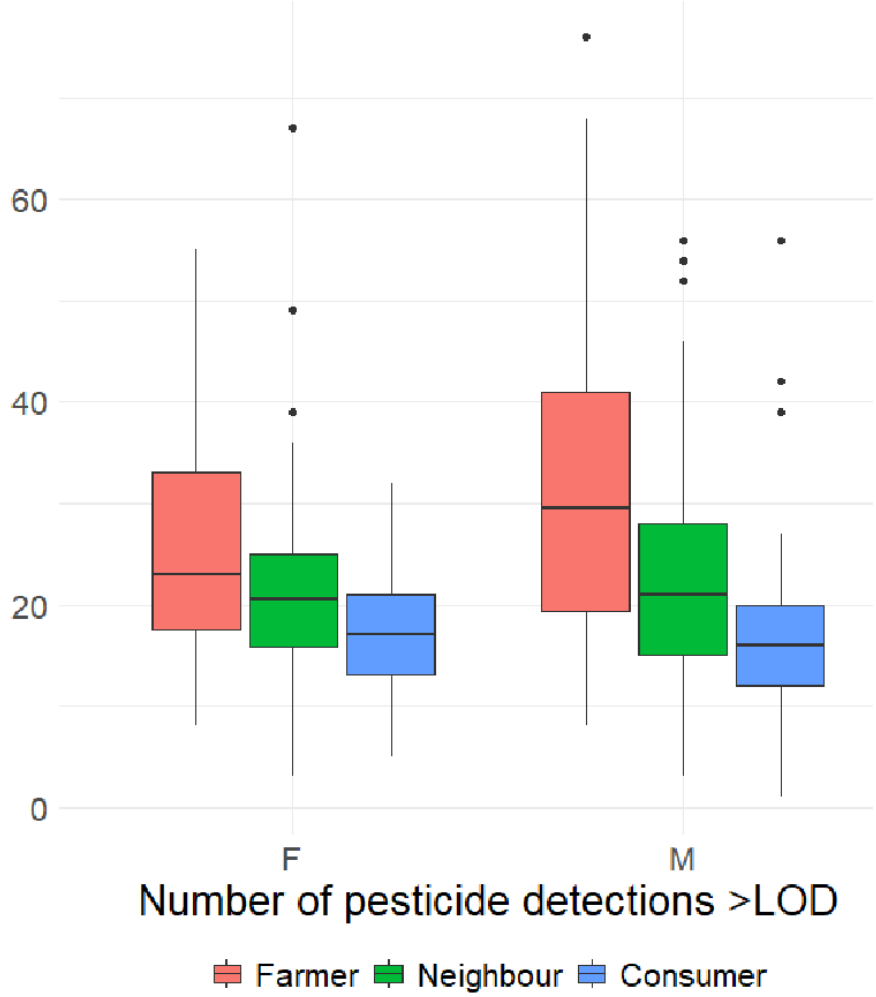

Figure 3.9. Number of detected pesticides (>LOD) per wristband based on 6641 analysed samples stratified by gender and disaggregated by subgroup. See the legend of **Figure 3.2.** for more information about the symbols used in these boxplots.

The number of pesticides per wristband shows a clear pattern in mean numbers that is repeated for across genders (**Figure 3.9**): farmers had the highest median number of pesticides per wristband (23 in women and 30 in men), followed by a lower number in neighbours (21 in both women and men) and the lowest in consumers (17 in women and 16 in man). The median number is higher considerably higher in male compared to female farmers but not statistically significant due to the high variability. In neighbours and consumers, the numbers by gender are very similar, with some more variability observed in consumers than neighbours.

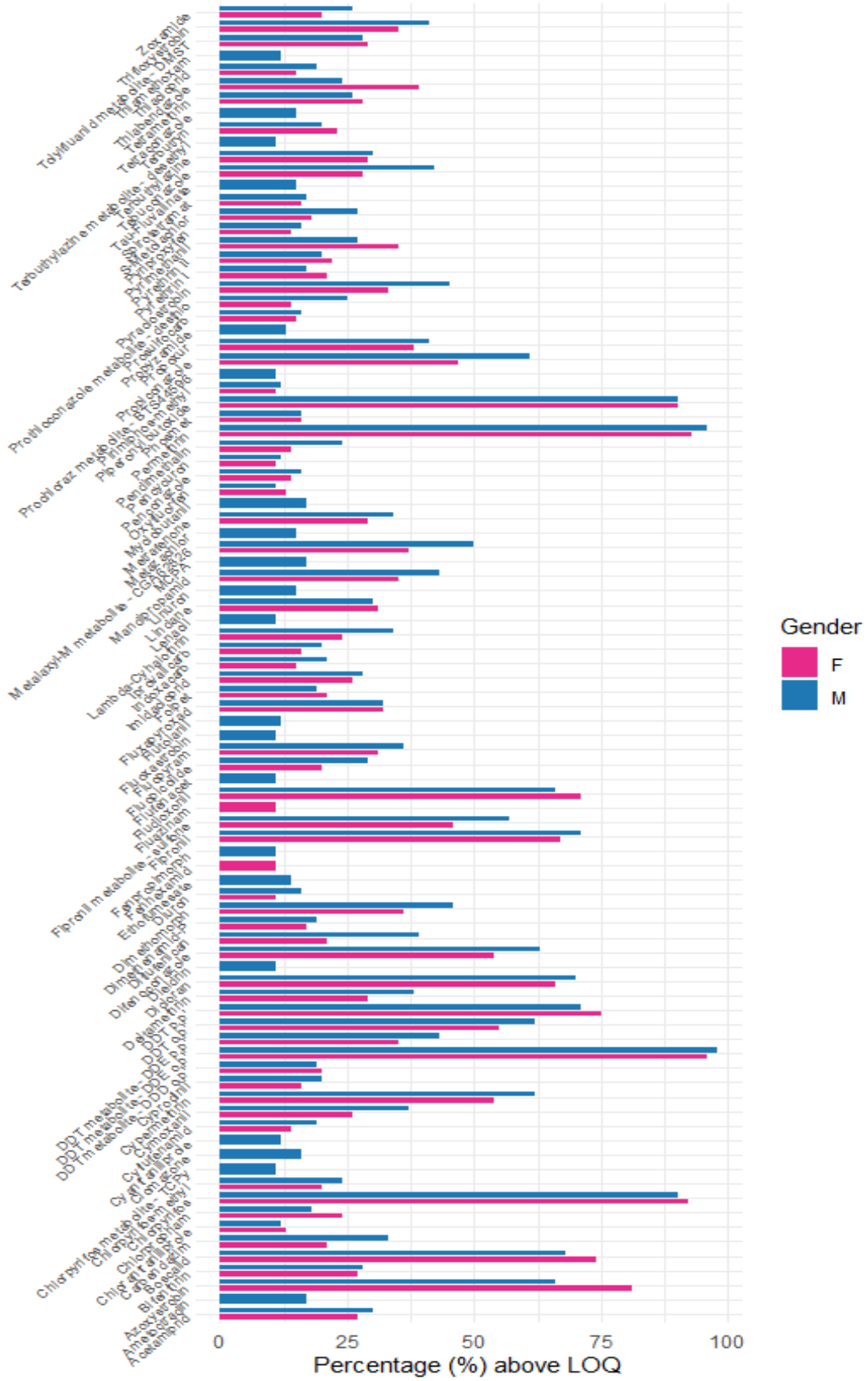

Figure 3.10. Detection frequency of each pesticide in 221 analysed farmer's wristbands stratified by gender. For visualization purposes only pesticides with more than 10% observations above LOQ are plotted.

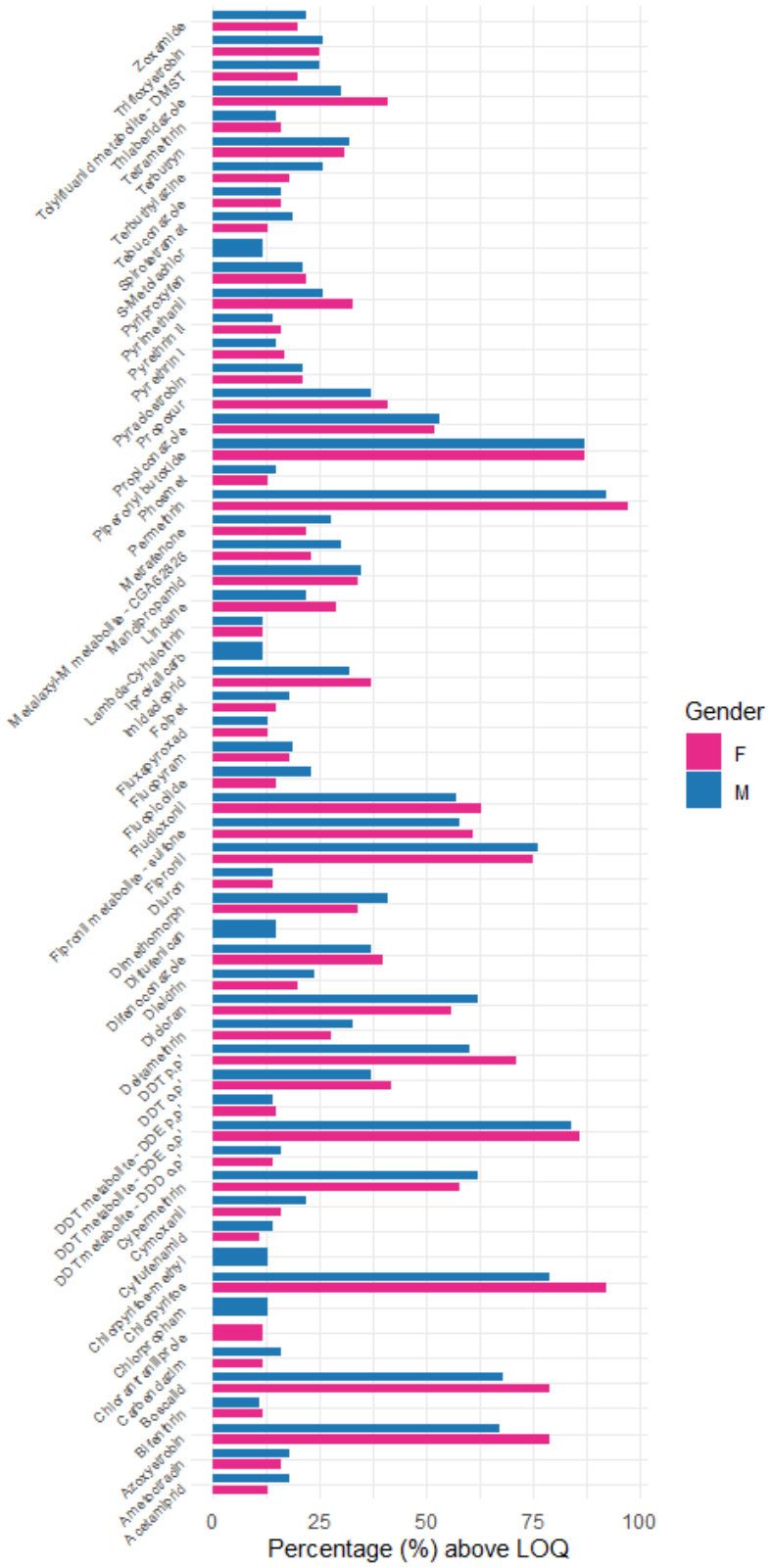

Figure 3.11. Percentage of wristband samples above LOQ. per pesticides based on 201 analysed neighbour's samples stratified by gender. For visualization purposes only pesticides with more than 10% observations above LOQ are plotted. Y-axis the pesticides names. X-axis the percentage of samples above LOQ.

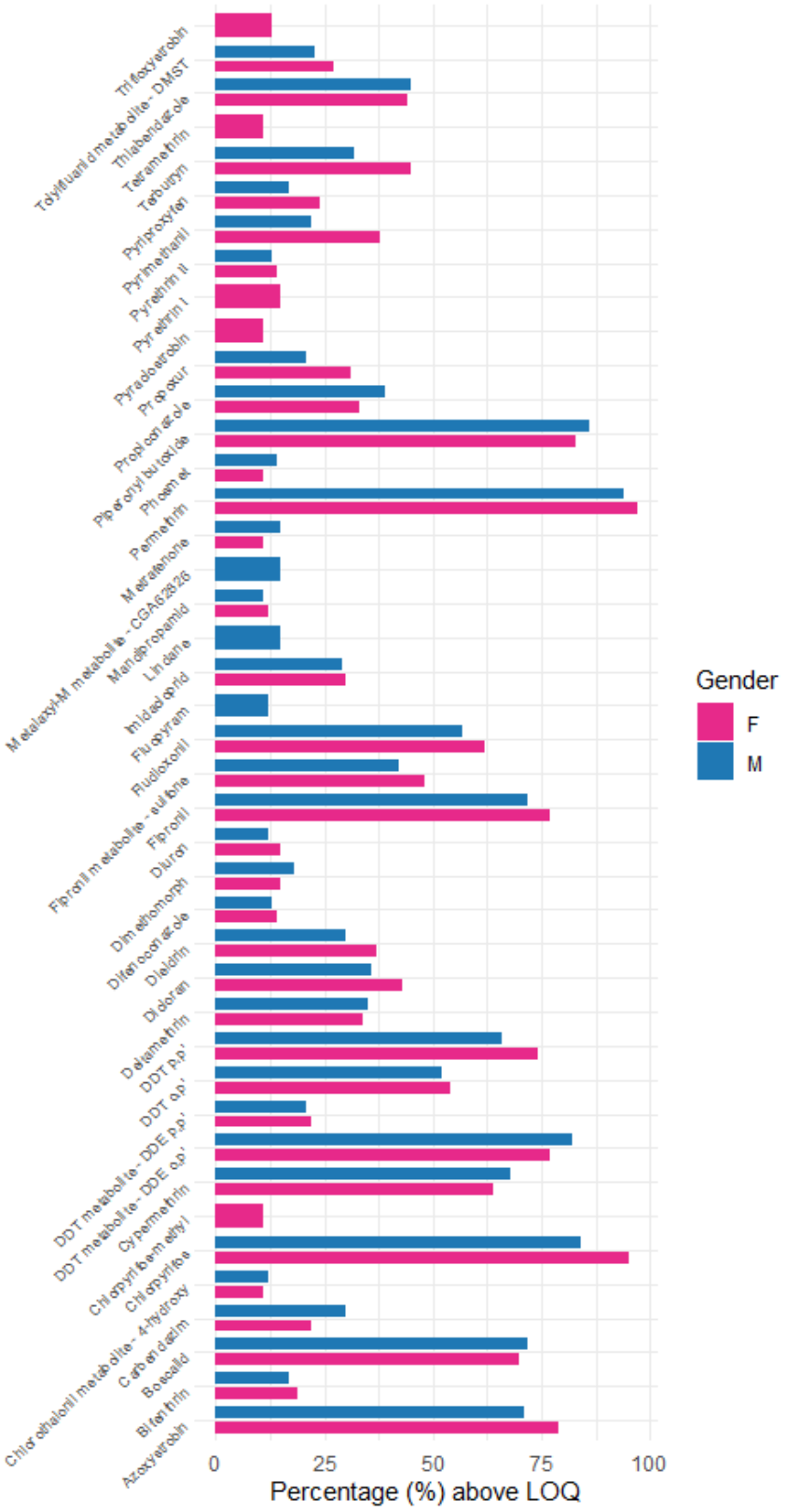

Figure 3.12. Percentage of wristband samples above LOQ per pesticide based on 219 analysed consumer's samples stratified by gender. For visualization purposes only pesticides with more than 10% observations above LOQ are plotted.

The wristband concentrations were overall not much different by subgroups and gender (**Figure 3.13.**). Many outliers were observed for different pesticides but with no clear pattern that stands out for either of the subgroups. Significant differences (at p-value < 0.05) were observed for chlorpyrifos, higher in consumer females when compared to consumer males. For o,p'-DDE, higher in males when compared to females, for both farmer and consumer populations. For o,p'-DDT, higher in males when compared to females, for farmers. The concentrations in wristbands vary substantially between several orders of magnitude (**Figure 3.13**), but with similar interquartile ranges between genders.

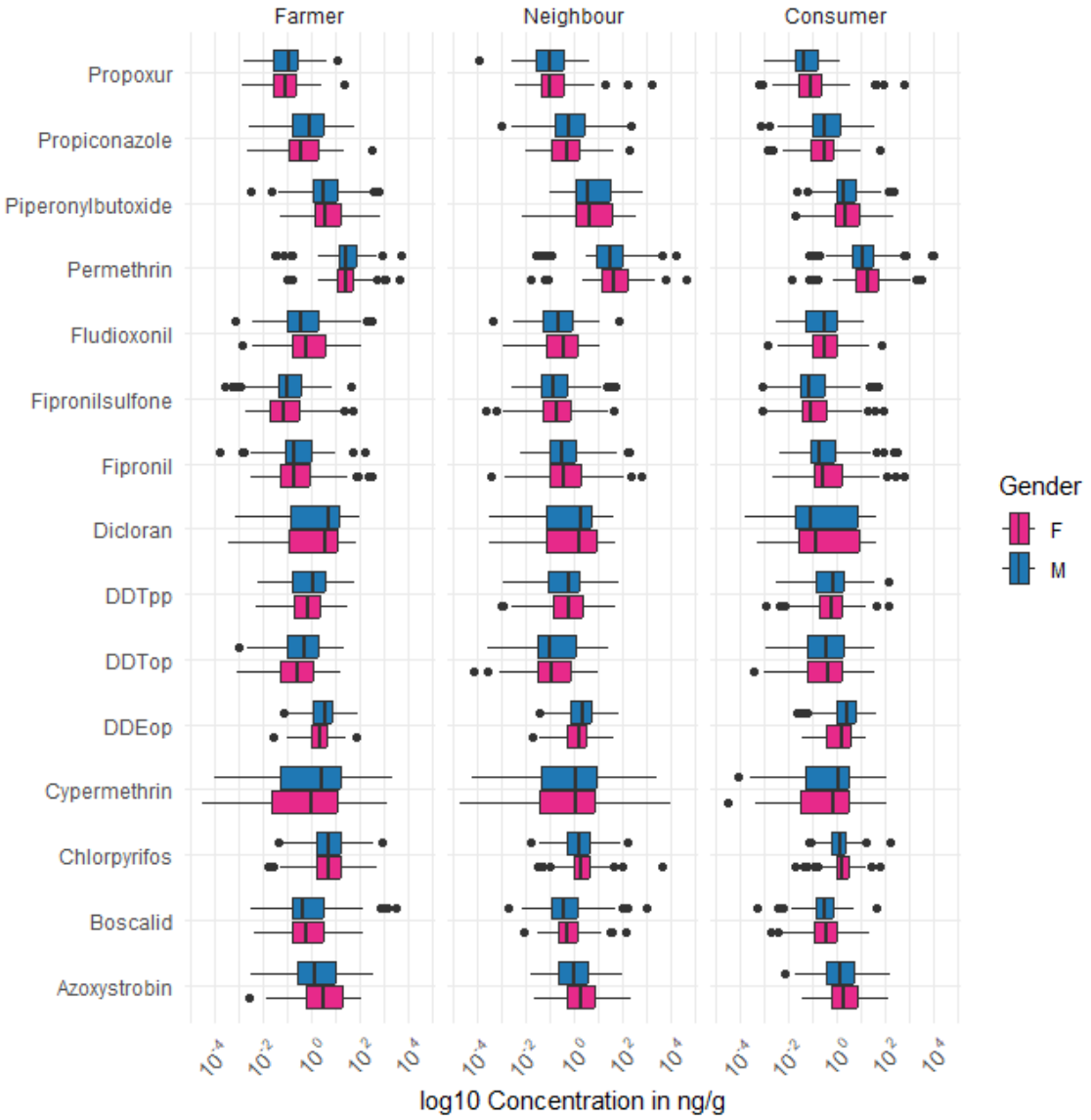

Figure 3.13. Wristband concentrations in ng/g disaggregated by subgroup. Only pesticides with more than 40% observations above LOQ are plotted. Values below LOQ were imputed as described in deliverable 3.3. See the legend of Figure 3.2. for explanation of the symbols used in this boxplot.

4. Discussion

For most pesticides exposure based on blood, urine and faeces was not different by gender. Differences were seen in urine and faeces for very few pesticides only. For instance, urinary excretion of imidacloprid olefin was higher in males compared to females in the group of farmers and neighbours but the rate of imidacloprid olefin detection >LOD was only 1.5% in the complete set of analysed samples. Urinary excretion of pyrimethanil OH was higher in females compared to males in both farmers and neighbours, based on a somewhat higher overall detection rate of pyrimethanil OH in the total number of analysed samples of 18.1%. For the parent propamocarb with an overall rate of detection >LOD of 7.4%, a higher concentration was observed in females than males in the subgroup neighbours. For some other more frequently detected pesticide metabolites in urine the patterns of scattered concentrations were somewhat different.

In female farmers concentrations of faeces residues of the fungicide fenhexamid were higher compared to male farmers. The fipronil metabolite fipronil sulfone as well as the pyrethroid insecticide permethrin were both higher in faeces samples from female neighbours compared to male neighbours. Both findings may be derived from agricultural use but are more likely derived from domestic applications as biocide/veterinary drug as flee-repellent in companion animals. The latter finding is not consistent with the finding of piperonyl butoxide often applied as a synergist in the same pyrethroid-based products for which the gender ratio was reverse (higher in male than female neighbours). Such inconsistency may be related to specific uses and may also be due to substance properties or the specific kinetics of excretion of these substances in faeces for which there is not yet much experience and no previous reports in published literature.

In wristbands wide ranges in pesticide detection rates and concentrations in all subgroups were observed and for most pesticides the detection rates and recovered pesticide concentrations were similar by gender. For a minority of the pesticides frequencies or concentrations were somewhat different by gender but in the observed patterns there was no consistent disbalance in either direction (female or male).

Below potential contributors to explaining differences of pesticide exposure by gender are introduced based on published literature. Suggestions have been made how to explain exposure by gender for the study population and for specific subpopulations studied in the SPRINT project.

4.1. Exposure determinants by gender

Most studies report on the gender aspect reporting on both males or female merely as a study population characteristic. Some studies selectively reported on mother-child and/or father-child cohorts (Hall et al., 2022; Manley et al., 2022; Limon-Miro et al., 2017; Ye et al., 2009). When both genders are studied, these are often gender balanced but do not aim to specifically study the gender aspect. Often gender is used for adjustment in statistical modelling, next to age. In our study population the subgroups of women and men were comparable by mean age (**Table S1**).

4.1.1. Farmers

In occupational exposures the gender issue is of significant interest but there a knowledge gap. Only few studies addressed this issue related to occupational settings (Schlünssen and Jones, 2023). Some studies reported a higher susceptibility of female workers related to chemical exposures (Hursidić-Radulović et al., 2002). In the subgroup of farmers, we have not observed a clear gender-related effect on our exposure for most pesticides. According to previously published studies males are more likely than females to be exposed due to their involvement in pesticide applications such as spraying, mixing and loading. Female farmers are reported to be more often involved in pre- or post-harvest processes and likely exposed due to re-entry work such as in glasshouse farming (Bretveld et al., 2006). To some extent occupational exposures have covered the gender aspect, e.g. growing winter wheat, scallions, garlic and peanuts in China (Wang et al., 2017). The authors of this study conclude that gender differences are associated with aspects like awareness and knowledge of health impacts, best use practices and worker protection. Similar results were reported by Jørs and co-workers (2013) who found that female farmers are at a higher risk for pesticide intoxications than male farmers in Bolivia. Similar findings showed a higher risk of female farmer's involvement in occupational illness and injury cases in the US and Nepal (Kasner et al., 2012; Atreya et al., 2007). In tobacco farming in Malaysia more male than female workers used pesticide in an environmentally-friendly way. However, fewer males than females applied self-protective measures. Male workers reported to be using more PPE compared to their female colleagues but compliance with proper handling of pesticide and the level of maintenance of pesticide spray equipment was reported to be higher in female workers than males (Bin Nordin et al., 2001). It is important to acknowledge that spray activities may also lead to potential inhalation of spray drift and skin contamination due to direct contact with the product during preparation of the sprayed solution before and cleaning of the spray equipment afterwards. Work related to the harvest may be related to direct skin contact with residues on the crop.

4.1.2. General population

Exposure to pesticides from plant protection products (PPPs) originates from both dietary and non-dietary sources. Diet as a source of uptake is well known and some studies have reported gender-related aspects, mainly in children (Cook and Wardle, 2005; Petrik et al., 2006) or report specifically on mother-child pairs (Hall et al., 2022). Non-dietary sources of exposure are less studied for their effects and not much is known about their heterogeneity regarding the gender aspect. In the general population there were only few reports available e.g. reporting blood levels of organochlorines and gender. In two regions in the east of Slovakia consistent differences between genders indicated levels of serum alpha-, beta-, gamma-HCH, HCB, p,p'-DDT and p,p'-DDE in males to be about 20% higher than females (Petrik et al., 2006). In Tunesia, levels of p,p'-DDE and the sum of DDTs were significantly higher in females than in males (Ben Hassine, 2014). For hexachlorobenzene (HCB), β-hexachlorocyclohexane (β-HCH), γ -hexachlorocyclohexane (lindane) (γ-HCH), oxy-chlordane, trans-nonachlor, p,p'-DDE, o,p'-DDT, p,p'-DDT and Mirex no differences were observed by gender in a large study in samples pooled by gender and age from 12,175 individuals in the general population of Australia in the period 2022-2013 (Thomas et al. 2016). In a follow-up study, Thomas and co-workers (2019) reported that gender did not have a strong effect on the serum β-HCH concentrations which were modelled as a function of the interaction between age group and time. For p,p'-DDT the

interactions between age and gender showed an effect of age group, gender and time on the change in concentrations over time. Two recent studies report on urine biomonitoring in adults and children in several European countries for current use pesticides. Although the study populations were balanced by gender the authors did not report on the gender aspect, other than using it for adjustment in their statistical models (Ottenbros et al., 2023a, 2023b).

4.1.2. Physiology, biometrics and biokinetics

For males an average bodyweight of 70 kg has been used long-term and a much lower average bodyweight of 58 for women (ICRP, 1975). There is also a difference in total fat mass between male and female. For males the average fat mass is 21.3% and in females it is 32.7% of the body weight (Brown et al., 1997). This gender-related variation in body composition could result in differences in the relative distribution of cardiac output among the various organs such as the brain and myocardial tissue and other tissues. The GI tract and adipose tissue may represent a larger portion of total body mass in females than in males (ICRP, 1975). Brown and co-workers suggested that these tissues receive a greater portion of cardiac output in females than in males. They also indicated that it is likely that blood flow rates to reproductive tissues are different when expressed per unit mass or as % cardiac output. Beaudouin (2010) developed new mathematical functions to describe gender-related parameters in PBPK-models using data from the NHANES study. He assumed metabolism to be dependent on age and not gender. In women he accounted for body changes post-pregnancy. And in this lifetime model, a different starting age for puberty was set for women compared to men.

4.2. Dietary and non-dietary patterns

4.2.1 Dietary patterns

Overall, it can be concluded that our urine data show a gender balance of urinary pesticide residue excretion values with very similar patterns in subgroups with only few exceptions. The pattern of somewhat higher excretion of imidacloprid olefin in males compared to females was found in both farmers and neighbours. Pyrimethanil-OH urinary concentrations were higher in females than males for farmers and neighbours. Propamocarb was also higher in females compared to males but only in neighbours. The question is to what extent this can be explained by differences in the contribution of non-dietary exposure attributed to differences in dietary routes of exposure between females and males in these subgroups. It is tempting to suggest that this may relate to non-dietary factors as aforementioned differences were not observed in consumers. For consumers it is assumed that food is the primary source of exposure to pesticides. In a study by Rempelos and co-workers (2022) the study population was quite small and included students from Greece and the UK (N=27). Overall urinary pesticide residue excretion was 60% lower for women compared to men. The only significant finding in reported food consumption was related to 1.7 times higher refined flour bread and 28% higher egg consumption by men compared to women. In this study environmental, i.e. non-dietary was treated as a confounder. An indication of environmental exposure as the detection of 4-nitrophenol, a metabolite of parathion but this organophosphate pesticide was not detected as a food residue.

4.2.2. Other gender-related underlying social/cultural/lifestyle factors

In wristbands, there was not a distinguished difference in pesticides concentrations between genders. The major observed differences were observed for pesticides detection frequency. Other factors, as discussed in deliverable 3.3, have shown to influence wristband concentrations for certain pesticides. Such factors include: smoking status, time spent indoors, owning pets and cleaning frequency. However, there was no major variability in any of these factors by gender with exception for smoking status (smoker vs non-smoker), with active smoking being somewhat more often reported by males than females (**Table 2**).

Table 2. Self-reported active smoking of tobacco products stratified by subgroup and gender.

Smoking	Farmers		Neighbours		Consumers	
	Male	Female	Male	Female	Male	Female
Yes	28 (20%)	12 (11%)	26 (22%)	22 (18%)	22 (19%)	12 (9%)
No	108 (78%)	92 (86%)	84 (78%)	100 (81%)	95 (81%)	130 (90%)
Missing	3 (2%)		1 (1%)		1 (1%)	

Differences in smoking habits by gender might by itself have an influence on biomarkers of exposure to pesticides e.g. by interaction liver enzyme activity (Kiyohara et al. 2010). We did not collect the data to test this hypothesis. Moreover, the differences in smoking were probably too small to study this effect in our population.

In summary, only few studies have previously reported about gender-related effects on pesticide exposure, mostly based on interviews and self-reports in questionnaires. It is possible that in published literature there is some selective reporting on the issue of observing a gender effect (i.e. under representing reports of not finding such differences). Studies that have applied other methods of exposure assessment often have not explicitly reported on the gender-aspect such as the previous HBM4EU studies (Ottenbros et al., 2023) and studies on effect biomarkers (Ramirez et al., 2018). In the current study exposures were studied based on human biomonitoring of three different biological matrices, integrating multiple sources and routes of exposure. We cannot rule out the possibility that there may be underlying gender-related determinants of exposure in our population. However, we have not been able to find consistent indications. Small contributions of gender-related factors in residential or work-related pesticide applications leading to non-dietary exposures may have been overwhelmed by uptake of residues from the diet. We were not able to find examples of pesticides exposure biomarkers for which we are confident that gender is driving the exposure to PPP.

5. Conclusions

The following main messages best describe the results from this study:

5.1. Approach

- Samples of blood, urine and faeces as well as from wristbands were analysed from 669 study participants across different crops, climate zones and countries
- Individual samples of in total 669 study participants (337 females and 332 males) were available from subgroups of farmers (N=235), neighbours (N=208) and consumers (N=236).
- In total 648 blood samples, 664 urine samples, 655 faeces samples and 641 silicon wristbands were analysed for 208 pesticides/metabolites and one synergist. In urine 46 pesticide biomarkers were also analysed, and some additionally as metabolites from the list of 208 pesticides.
- Overall gender was well balanced but there were some deviations by subgroup and country.
- Exposure to 208 pesticide related exposure biomarkers was compared by detection frequencies of results above the limit of detection (LOD) and by concentrations from results above the limit of quantification (LOQ).
- Gender effects on exposure were disaggregated by subgroups: farmers, neighbours and consumers.

5.2. Results

- In blood the median number of observed pesticides and metabolites per sample was two. Frequency distributions were similar across female and male and subgroups and genders (ranging from 0 to 10). No gender-related differences were observed for blood concentrations except for bromoxynil. Five male farmers two female consumers and one male consumer had higher blood concentrations for this herbicide.
- In urine of men, the median number of pesticides and metabolites per sample was 8, 7, and 7 in farmers, neighbours and consumers, respectively, and with a range from 0 to 20. In urine of women the median was 7 with very similar frequency distributions across subgroups. Urinary metabolite concentrations were very similar across gender and subgroups. Imidacloprid olefin was higher for men compared to women in the group of farmers and neighbours. Urinary excretion of two fungicides were higher for women compared to men: pyrimethanil_OH in farmers and neighbours and propamocarb only in neighbours.
- In faeces the median number of pesticides and metabolites was 3 across gender in all subgroups and frequency distributions were similar with a ranging from 0 to 20. Concentrations of pesticides residues in faeces were very similar across gender and subgroups except for the fungicide fenhexamid and the synergist piperonyl butoxide. Female neighbours had higher faeces residue levels compared to males. Female neighbours were also higher exposed to the biocides permethrin and the fipronil metabolite fipronil sulfone than to males. Both pesticides and the synergist piperonyl butoxide are also used as biocides for companion animals.
- Farmers had the highest median number of pesticides per wrist band (23 in women and 30 in men), followed a lower number in neighbours (21 in both women and

men) and the lowest number in consumers (17 in women and 16 in men). A gender-dependent effect was observed in farmers but not statistically significant due to a high variability in this parameter. Concentrations in wristbands were very similar across gender for all subgroups.

5.3. Interpretation

- In blood, urine and faeces for most pesticides findings stratified by gender were similar. For a very limited number of pesticides deviations by gender were observed in both directions but each time only in small numbers of study participants at a time and only in one specific subgroup at a time. A gender effect should therefore be interpreted with caution.
- The median numbers of detected pesticides were much higher in wristbands compared to blood, urine and faeces. Differences across gender were observed in farmers but high variability resulted no statistically significance for this difference. In neighbours and consumers, the median numbers of detects per wristbands by gender were almost the same.
- Observed differences in exposure by gender was difficult to interpret regarding the small differences and high variability observed, combined with a low number of observations per substance. Many potential underlying factors could explain patterns in exposure such as e.g. work-related exposure and domestic use. Co-occurrence of analysed active ingredients in PPP, biocide and veterinary drug applications could be indicative of uses in both agricultural and domestic settings.
- Gender does not directly explain variability in pesticides wristband concentrations, but can indirectly affect these through behavioural differences and activity patterns.
- The assessment of exposure by gender was based on measured samples of blood, urine and faeces to detect the frequency and quantity of PPP residues of exposure but not their effect on the individuals' health.

5.4. Suggestions for future studies

It is recommended to address the gender aspect in future exposure studies, e.g. when recruiting study populations and also when reporting results, even in the case when gender is not considered a significant factor to explain observed patterns of exposure in the population studied. In future studies gender analyses are needed to confirm gender-related effects and explore underlying causes.

6. References

- Atreya K. Pesticide use knowledge and practices: a gender differences in Nepal. Environ Res. 2007 Jun;104(2):305-11.
- Ben Hassine S, Hammami B, Ben Ameur W, El Megdiche Y, Barhoumi B, El Abidi R, Driss MR. Concentrations of organochlorine pesticides and polychlorinated biphenyls in human serum and their relation with age, gender, and BMI for the general population of Bizerte, Tunisia. Environ Sci Pollut Res Int. 2014 May;21(10):6303-13.
- Bin Nordin R, Araki S, Sato H, Yokoyama K, Bin Wan Muda WA, Win Kyi D. Gender difference in safe and unsafe practice of pesticide handling in tobacco farmers of 22alaysia. Malays J Med Sci. 2001 Jan;8(1):1-10.
- Bretveld R, Zielhuis GA, Roeleveld N. Time to pregnancy among female greenhouse workers. Scand J Work Environ Health. 2006 Oct;32(5):359-67.
- Brown RP, Delp MD, Lindstedt SL, Rhomberg LR, Beliles RP. Physiological parameter values for physiologically based pharmacokinetic models. Toxicol Ind Health. 1997 Jul-Aug;13(4):407-84.
- Cooke LJ, Wardle J. Age and gender differences in children's food preferences. Br J Nutr. 2005 May;93(5):741-6.
- Hall AM, Thistle JE, Manley CK, Roell KR, Ramos AM, Villanger GD, Reichborn-Kjennerud T, Zeiner P, Cequier E, Sakhi AK, Thomsen C, Aase H, Engel SM. Organophosphorus Pesticide Exposure at 17 Weeks' Gestation and Odds of Offspring Attention-Deficit/Hyperactivity Disorder Diagnosis in the Norwegian Mother, Father, and Child Cohort Study. Int J Environ Res Public Health. 2022 Dec 15;19(24):16851.
- Hursidić-Radulović A, Mustajbegović J, Zuskin E, Ivanković D, Schachter EN. Gender related differences of low-level exposure to occupational irritants–a three-year follow-up of chemical industry workers. Coll Antropol. 2002 Dec;26 Suppl:109-18.
- Jørs E, Hay-Younes J, Condarco MA, Condarco G, Cervantes R, Huici O, Bælum J. Is gender a risk factor for pesticide intoxications among farmers in Bolivia? A cross-sectional study. J Agromedicine. 2013;18(2):132-9.
- Kasner EJ, Keralis JM, Mehler L, Beckman J, Bonnar-Prado J, Lee SJ, Diebolt-Brown B, Mulay P, Lackovic M, Waltz J, Schwartz A, Mitchell Y, Moraga-McHaley S, Roisman R, Gergely R, Calvert GM. Gender differences in acute pesticide-related illnesses and injuries among farmworkers in the United States, 1998-2007. Am J Ind Med. 2012 Jul;55(7):571-83.
- Kiyohara C, Miyake Y, Koyanagi M, Fujimoto T, Shirasawa S, Tanaka K, Fukushima W, Sasaki S, Tsuboi Y, Yamada T, Oeda T, Miki T, Kawamura N, Sakae N, Fukuyama H, Hirota Y, Nagai M; Fukuoka Kinki Parkinson's Disease Study Group. GST polymorphisms, interaction with smoking and pesticide use, and risk for Parkinson's disease in a Japanese population. Parkinsonism Relat Disord. 2010 Aug;16(7):447-52. doi: 10.1016/j.parkreldis.2010.04.009. Epub 2010 May 15. PMID: 20472488.
- Limon-Miro AT, Aldana-Madrid ML, Alvarez-Hernandez G, Antunez-Roman LE, Rodriguez-Olibarria G, Valencia Juillerat ME. Breast milk intake and mother to infant pesticide transfer measured by deuterium oxide dilution in agricultural and urban areas of Mexico. Chemosphere. 2017 Aug;181:682-689.
- Manley CK, Villanger GD, Thomsen C, Cequier E, Sakhi AK, Reichborn-Kjennerud T, Herring AH, Øvergaard KR, Zeiner P, Roell KR, Engel LS, Kamai EM, Thistle J, Hall A, Aase H, Engel SM. Prenatal Exposure to Organophosphorus Pesticides and Preschool

ADHD in the Norwegian Mother, Father and Child Cohort Study. Int J Environ Res Public Health. 2022 Jul 2;19(13):8148.

- Ottenbros I, Lebret E, Huber C, Lommen A, Antignac JP, Čupr P, Šulc L, Mikeš O, Szigeti T, Középesy S, Martinsone I, Martinsone Z, Akulova L, Pardo O, Fernández SF, Coscollá C, Pedraza-Diaz S, Krauss M, Debrauwer L, Wagner K, Nijssen R, Mol H, Vitale CM, Klanova J, Molina BG, León N, Vermeulen R, Luijten M, Vlaanderen J. Assessment of exposure to pesticide mixtures in five European countries by a harmonized urinary suspect screening approach. Int J Hyg Environ Health. 2023a Mar;248:114105.
- Ottenbros IB, Ammann P, Imboden M, Fuhrimann S, Zock JP, Lebret E, Vermeulen RCH, Nijssen R, Lommen A, Mol H, Vlaanderen JJ, Probst-Hensch N. Urinary pesticide mixture patterns and exposure determinants in the adult population from the Netherlands and Switzerland: Application of a suspect screening approach. Environ Res. 2023 Dec 15;239(Pt 1):117216.
- Petrik J, Drobna B, Pavuk M, Jursa S, Wimmerova S, Chovancova J. Serum PCBs and organochlorine pesticides in Slovakia: age, gender, and residence as determinants of organochlorine concentrations. Chemosphere. 2006 Oct;65(3):410-8.
- Rempelos L, Wang J, Barański M, Watson A, Volakakis N, Hoppe HW, Kühn-Velten WN, Hadall C, Hasanaliyeva G, Chatzidimitriou E, Magistrali A, Davis H, Vigar V, Średnicka-Tober D, Rushton S, Iversen PO, Seal CJ, Leifert C. Diet and food type affect urinary pesticide residue excretion profiles in healthy individuals: results of a randomized controlled dietary intervention trial. Am J Clin Nutr. 2022 Feb 9;115(2):364-377.
- Schlünssen V, Jones RM. Gender aspects in occupational exposure and health studies. Ann Work Expo Health. 2023 Nov 28;67(9):1023-1026.
- Thomas A, Toms LL, Harden FA, Hobson P, White NM, Mengersen KL, Mueller JF. Concentrations of organochlorine pesticides in pooled human serum by age and gender. Environ Res. 2017 Apr;154:10-18.
- Thomas A, White NM, Leontjew Toms LM, Mengersen K. Application of ensemble methods to analyse the decline of organochlorine pesticides in relation to the interactions between age, gender and time. PloS One. 2019 Nov 13;14(11): e0223956.
- Vested A, Gai L, Schlünssen V, Alaoui, A and Crist, F (2022) D2.3 – Inventory of PPP distribution and accumulation in EPAH, including gender-mediated differences.
- Wang W, Jin J, He R, Gong H. Gender differences in pesticide use knowledge, risk awareness and practices in Chinese farmers. Sci Total Environ. 2017 Jul 15;590-591:22-28.
- Ye X, Pierik FH, Angerer J, Meltzer HM, Jaddoe VW, Tiemeier H, Hoppin JA, Longnecker MP. Levels of metabolites of organophosphate pesticides, phthalates, and bisphenol A in pooled urine specimens from pregnant women participating in the Norwegian Mother and Child Cohort Study (MoBa). Int J Hyg Environ Health. 2009 Sep;212(5):481-91.

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Supplementary Table and Figures

Table S1. Age by gender (mean ± sd)

Country	**Mean age in years (mean ± sd)**	
	Women	**Men**
Croatia	47.5 ± 12.0	45.4 ± 10.1
Czech Republic	42.6 ± 10.2	44.8 ± 9.41
Denmark	49.2 ± 9.30	53.0 ± 10.9
France	44.2 ± 12.8	42.7 ± 14.0
Slovenia	43.0 ± 13.7	41.0 ± 13.2
Italy	51.6 ± 14.2	50.6 ± 15.7
Spain	46.3 ± 12.7	39.1 ± 11.7
Switzerland	47.8 ± 15.1	47.8 ± 16.7
Portugal	50.6 ± 11.7	48.8 ± 14.7
The Netherlands	48.1 ± 16.4	52.5 ± 16.2
Total	47.1 ± 13.1	46.6 ± 13.8

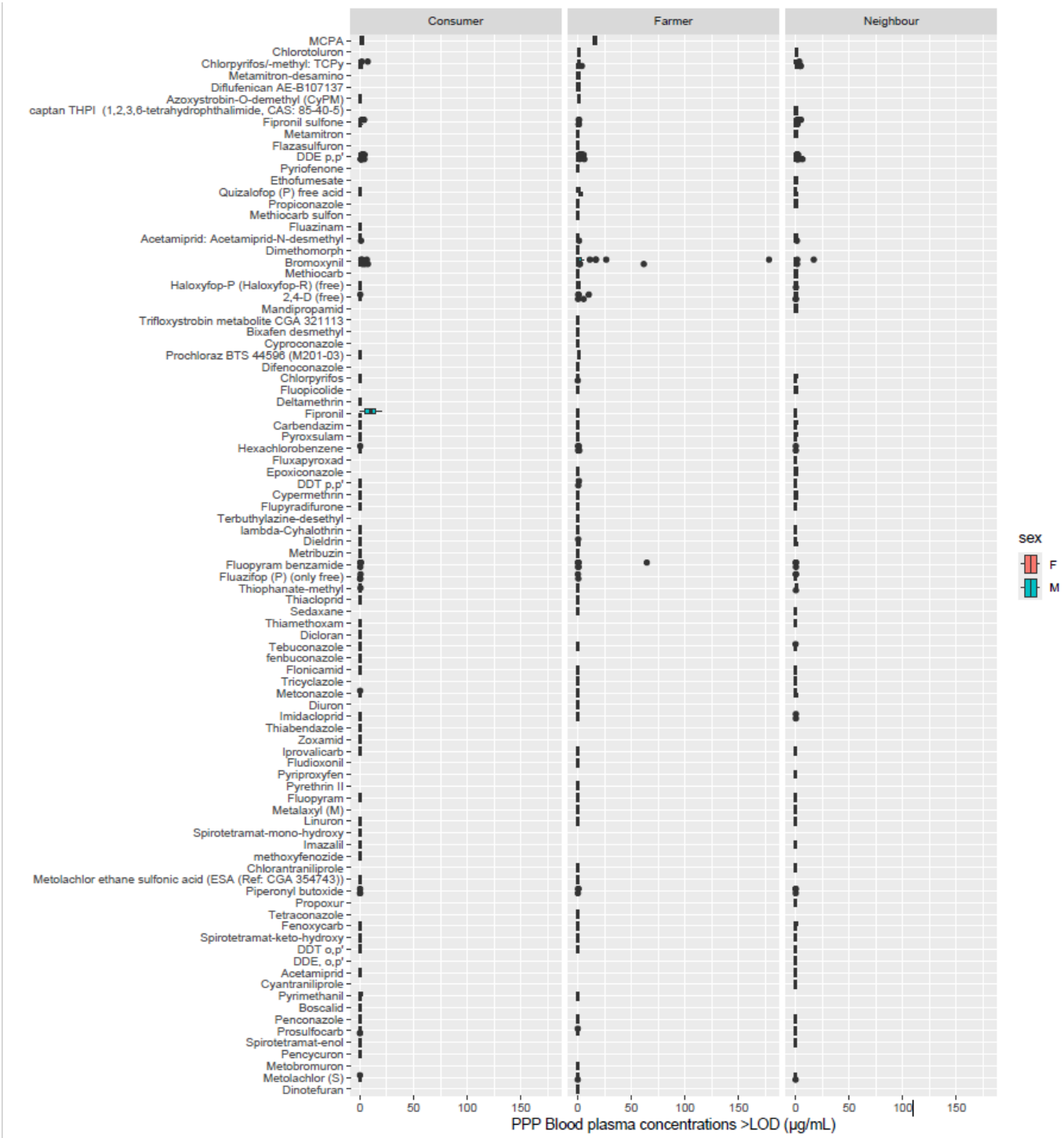

Figure S1. Blood plasma concentrations in µg/L of pesticides disaggregated by subgroup and gender (F = female, M = male).

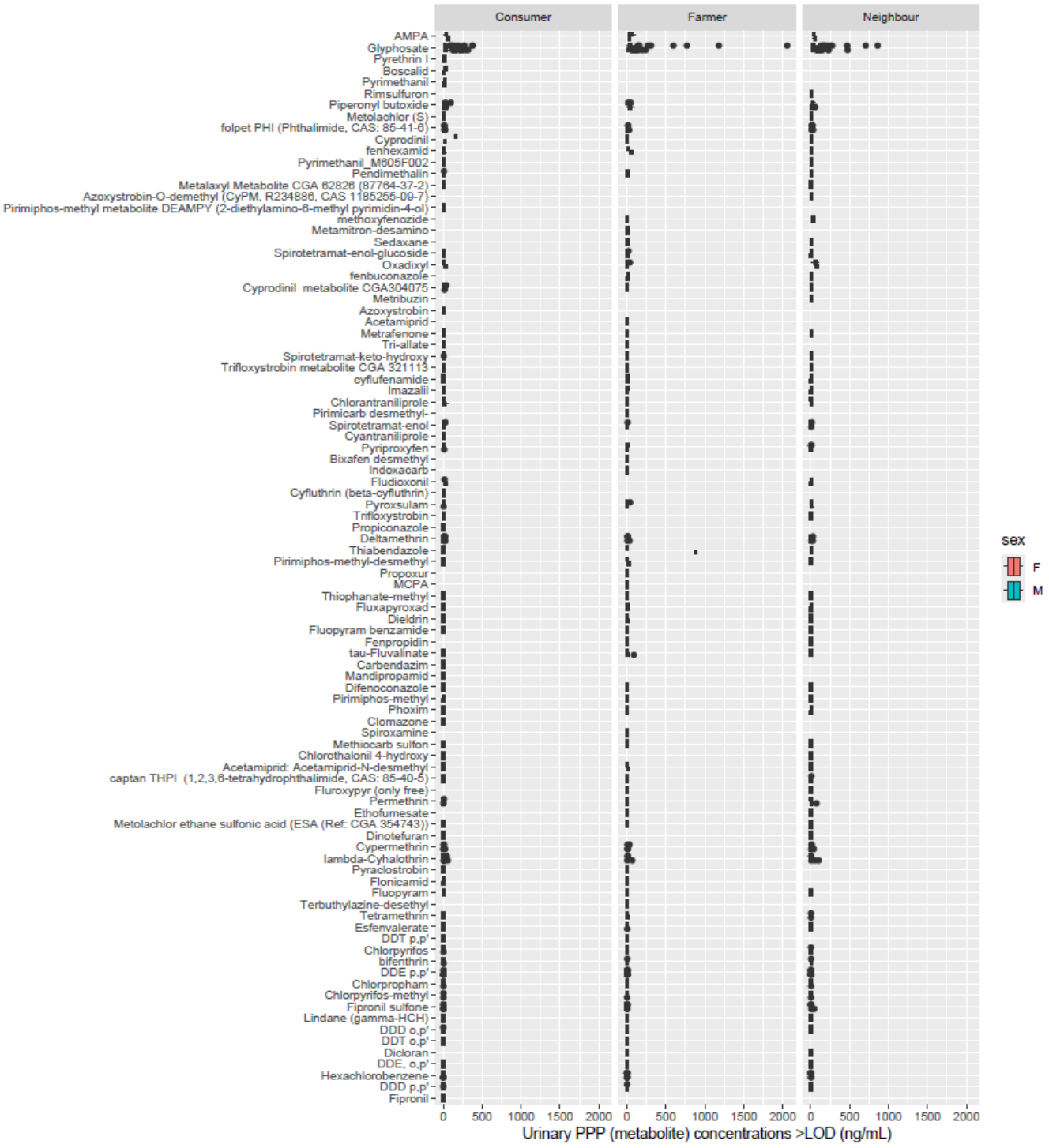

Figure S2. Faecal concentrations of pesticides and metabolites (µg/L) disaggregated by subgroup and gender (F = female and M = male).