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Analysis of Pesticides Residues in Breast Milk of Primiparous and Multiparous Women in Gilgit

Original Article

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Milk contains all the essential nutrients like fats, proteins, and minerals. The utilization of contaminated food can induce a proportion of pesticides in the body. The main purpose of the study was to determine the pesticide residue and current status of Breast milk in primiparous and multiparous mothers. In a current study, a total of 50 samples were collected from different areas of Gilgit and Astore. The pesticides cypermethrin, deltamethrin, and chlorpyrifos were analyzed using Gas Chromatography (GC). The present study shows analysis of the variations among the sample. The presence of cypermethrin in 10 samples ranged between 0.00 – 0.012 mg/kg, while the detection of Deltamethrin in 07 with variation from 0.000.12mg/kg. Whereas chlorpyrifos was found in 05 samples within the ranges of 0.00-0.0062 mg/kg respectively. Residue level was quite higher in urban areas than rural areas. The multiparous women had prominent residues level than primiparas and the concentration of Deltamethrin was higher than other pesticides. All the pesticides residues levels in the breast milk of primiparous and multiparous mothers were within the limits of WHO. Yet the women of these areas are not vulnerable but prolong exposure may pose a serious threat to neonatal and maternal health and other relevant reproductive issues. To manage the risk of milk contamination in the future, the demand for public awareness campaigns and the adoption of alternative clean approaches to control pests and other disease-spreading vectors in the best interests of public health seems reasonable.

Keywords: Gas Chromatography, Milk, Multiparous , Pesticides, Primiparous.

Author's Contribution
This study was designed and directed by S. urooj and M.A.NAfees as principal investigators. Saif-Ud-Din provided all the technical and experimental Guidance. S.Ali and Saif-Ud-Din performed and participated in Statistical analysis and preparation of the manuscript draft.

Conflict of interest
The authors declare no conflict of interest in publishing this manuscript in IJIST.

Project details. NIL

Introduction

Pesticides are used to protect crops, control damage, and maximize crop production. Different pesticide groups were in use including Organochlorine, Organophosphorus pesticides, pyrethroids, and carbamate, pesticides which contains toxic compounds that are banned [1]. Breast milk is the only source of diet for growing children it contains all the essential nutrients required for the development of newborns such as proteins, fats, and carbohydrates. Unluckily nowadays breast milk is not pure it is contaminated with toxic substances [2,3].

Pesticides are extensively used throughout the world for enhancing food production, controlling pests, and insects, and destroying the vectors of human and animal diseases like malaria, dengue, encephalitis [4]. Consumption of toxic chemicals through the field, diet, or through inhalation and dermal contact, may cause instability in daily life. Organophosphorus pesticides are lipophilic and are more persistent in breast milk [5, 6].In Pakistan farmers use different types of pesticides for the betterment of crops, it includes 39 types of herbicides, 108 insecticides, and 30 fungicides. Farmer's usually used Organochlorine, Organophosphorus, and alternate of urea. Residues are extensively found in different areas of Pakistan due to their unrelenting nature. When mothers consume contaminated food, level of residue rises in the body [7, 8, 9]. According to an economic survey 2012-13 of Pakistan locally 30,000 tons of pesticides were manufactured and 12665 tons were imported. In Pakistan mostly pesticides are used in fields which is an alarming situation for the presence of residues in food [10].

Human milk is thought to be the natural superior food for infants to meet their nutritional needs as they grow. Human milk, on the other hand, is an ideal matrix for the accumulation of pollutants. Chlorinated pesticides were common chemicals in the previous century's environment. They were widely used to control agricultural pests all over the world, and they are still used in some countries to control the malaria disease insect vector. When newborns and infants are exposed to these pollutants through feeding, they may suffer from lower birth weight [11], neuro-developmental delay [12].

Pregnant women are primarily exposed to these compounds through their diet [13], particularly through fish, meat, and milk [14, 15]. Despite a decades-long global ban on the use of OCPs, residues have been discovered in breast milk all over the world [16, 17, 18, 19].

Human breast milk is an ideal marker for OCPs because it provides information on the toxic effects of these substances on mothers and newborns. Pesticide accumulation in breast milk can be influenced by a variety of factors, including diet, place of residence, smoking, maternal age and weight, and previous lactation duration; however, the literature provides conflicting information [20].

Material and Methods.

Study Area.

Samples have been collected from five areas of District Gilgit (Danyore, Chilmis, Nomal, Gilgit City, and Oshikhandas) and five areas District Astore (Eidgah, Nowgam, Fina, Patipora, and Pakora).

Sample collections.

50 Samples were collected from volunteer mothers primiparas and multiparas between the ages of 20-40 before they are enrolled in the study all women signed authorization. Around 40 ml milk was collected in 100ml sterilized glass bottles with identification codes and placed in an icebox during the collection period and stored in a freezer at -20 C until laboratory analysis.

Samples have been collected during a period of two months from November to October 2019 from mothers who were either native to or lived there for the last five years. A questionnaire has been designed to access the socio-demographic data, food habits during and after pregnancy, age, and residence. The selective criteria in the current study were followed in previous similar studies. A self-administered questionnaire was also employed to acquire information about the subject's age, parity, body mass index,

eating habits. Questions regarding demographic characteristics and feeding habits were modified by Environmental Protection Agency EPA (21).

Laboratory Analysis

1ml of milk sample and 15ml of acetonitrile containing 0.01% acetic acid were mixed in a polytetrafluoroethylene (PTFE) centrifuge tube. Shaken the samples vigorously for 02 minutes and sonicate samples for 10 minutes. 6g of anhydrous MgSO4, 1g of NaCl, and 1g of sodium acetate trihydric were added to the sample. Again shaken the sample for 02 minutes and centrifuged for 05 minutes at 400rpm. The supernatant of 8ml was transferred into another PTFE tube and added 1g of MgSO4 and Florisil. Again centrifuged the samples and transferred the upper layer into a round bottom flask and left to evaporate until complete dehydration using a Rotavapor. Then reconstituted in 1ml of n-hexane containing 10-15% acetone and finally analyzed the sample through GC.

Statistical Analysis:

Obtained data were précised as mean. ANOVA and LSD were applied to check to mean the difference among samples by using SPSS 21 and Statitix 8.1 Capital letter represents LSD and Significance difference among means in column-wise.

RESULTS AND DISCUSSIONS.

Table 1. Mean value of pesticides residue area wise in mg/kg

Locations		Cypermethrin	Deltamethrin	Chlorpyrifos
Astore	Pakora	0.012^{A}	ND^{A}	0.005^{A}
	Nowgam	0.001^{B}	0.01^{B}	ND^{B}
	Patipora	ND^{C}	ND^{AC}	ND^{BC}
	Fina	ND^{CD}	ND^{ACD}	0.002^{D}
	Eidgah	0.011^{AE}	0.002^{E}	ND^{BCE}
Gilgit	Oshikhandas	ND^{CDF}	ND^{ACDF}	ND^{BCEF}
	Chilmis Das	ND^{CDFG}	ND^{ACDFG}	ND^{BCEFG}
	Nomal	0.0012^{H}	0.015^{H}	ND^{BCEFGH}
	Danyore	0.00574^{I}	0.00528^{I}	0.0062^{I}
	Gilgit City	0.0005^{J}	0.12^{J}	ND^{BCEFGJ}

Note: ND: Not Detected

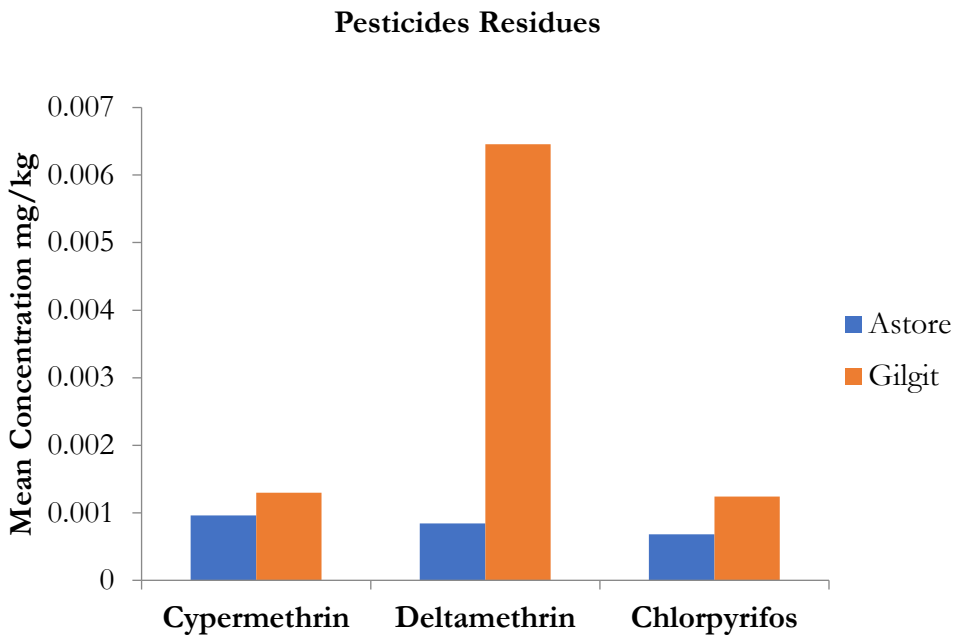

Figure1. Mean concentration of pesticide residues in District Astore and Gilgit

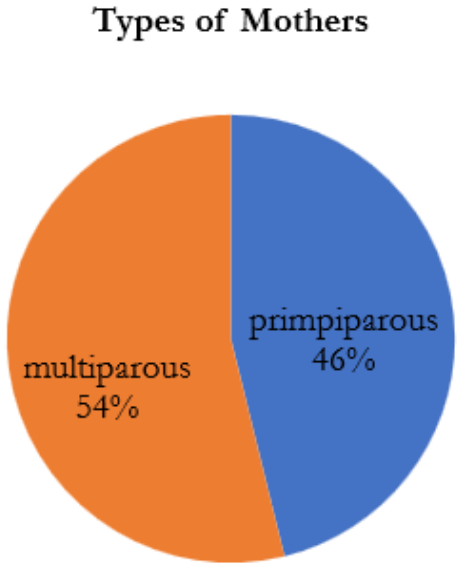

Figure 2. percentage of primiparous and multiparous mothers

In a current study, three pesticides were under observation named Cypermethrin, Deltamethrin and Chlorpyrifos from breast milk, 54% of mothers were multiparous and 46% were primiparous among selected mothers for sampling shown in Figure 2. Out of 50 samples, 13were detected with pesticide residues. The residues of Cypermethrin were found in 10 samples maximum value was found in Danyore 0.014mg/kg and the minimum value was obtained in Nowgam 0.001mg/kg. Deltamethrin in 09 samples maximum value of 0.12mg/kg was found in Gilgit and lowest 0.001mg/kg in Eidgah. Chlorpyrifos was found in 05 samples highest value in Danyore 0.019mg/kg and lowest in Fina 0.002mg/kg. The result shows that all the samples have residues of pesticide among them Cypermethrin was the major contaminant in milk samples mean values are shown in table 1 and Figure 1. Residue level in all samples was lower than the MRLs set by FAO/WHO.

In the current study, Cypermethrin residues were found in 7 samples of District Gilgit highest residue level was obtained in the sample of Danyore and the value was 0.014mg/kg and the lowest was found in Nomal and Gilgit with the residual level of 0.0012 mg/kg and 3 samples from Astore have the residues of cypermethrin maximum value was present in the sample of Pakora 0.012mg/kg minimum value in Nowgam 0.001mg/kg. To analyze the presence of Pyrethroids in human milk a study was conducted in Columbia. The residues of Cypermethrin, permethrin, and fenvalerate were present in the samples. Pyrethroids were useful to control Dengue and the level reach 28ngg-1/W. Evaluation of daily intake in nursing infants was calculated and compared to an adequate WHO level [22]. A similar study was conducted in urban and agricultural areas in Mexico on lactating women. The concentration of pp-DDT and cypermethrin in breast milk was higher in those women who are living in urban areas than in agricultural areas ($p < 0.05$and $p = 0,001$). Since pesticide levels do not exceed the ADI according to EPA and FAO/WHO [23]. The study investigates the residues of chlorpyrifos and other pesticides in cow's milk from Punjab India show CPS, DDT, and HCH as the major contaminants. Also the presence of cypermethrin, cyhalothrin, fenvalerate, deltamethrin, Malathion, profenofs, andehion was reported. 12 samples exceed the maximum residue limits (MRLs) for lindane, 18 for DDT, 1 sample for chlorpyrifos, profenophos and cypermethrin [24].

The current study shows that the residue level of Deltamethrin was presented in 09 samples among these 6 samples are from different areas of District Gilgit and the maximum value 0.12mg/kg was found in Gilgit-city and the minimum value detected in Danyore was 0.0011mg/kg and 3 samples are from District Astore the maximal value is obtained in Nowgam and Eidgah 0.01mg/kg and minimal value in Eidgah 0.01mg/kg. A similar study was conducted for the

determination of nine pesticides. The residues of Deltamethrin, trifluralin, cypermethrin, endosulfan, HGH, p'p'DDE and p'p-DDT were found in breast milk. Still, mothers were subjected to feeding infants because breast milk is considered as nutritive food during infancy [25]. A study was conducted in Punjab to monitor the residue level of various pesticides (DDT, DDE, cypermethrin, deltamethrin, permethrin, aldrin, and bifenthrin) 70% of samples were contaminated. DDE, DDT and endosulfan were present in minor proportions. Bifenthin residues are higher among Pyrethroids with a mean concentration of 1.68µg/ml, cypermethrin 0.23µg/ml and deltamethrin 0.21µg/ml mostly samples were polluted with cypermethrin, deltamethrin, permethrin and bifenthrin [26]. In the present study chlorpyrios residue was found in 3 samples of District Astore the highest residue level was 0.01mg/kg found in Eidgah4 and the lowest is 0.002mg/kg in Fina. Only 2 samples from Gilgit were detected with the value of 0.019 mg/kg and 0.012mg/kg in Danyore. A study was conducted to investigate OCPs, OPs, Pyrethroids, and carbamate residues in human and cow milk. Hexachlorobenzene, p, p'-dicofol, and chlorpyrifos were found in all samples [27]. Shangi *et al.*, [28] studied the presence of isomers of HGH, chlorpyrifos, Malathion and methyl-parathion was monitored in human milk from Bhopal, Madhya Pradesh. The concentration of endosulfan was highest and the concentrations of chlorpyrifos and Malathion were 3.5, 1.5, and 8.4 fold.

According to the study was conducted to investigate the relation between pesticides contamination of feedstuffs and residues in bovine milk, chlorpyrifos is the main contaminant with a residue level of 6.01 in feedstuff and 2.58ng/g in milk samples. Other pesticide residues observed in feed and milk samples endosulphan sulphate, cypermethrin, DDE, lindane, Malathion and fenvalerate. The main source of pesticide residue in milk is the feedstuff on that animal's feed [29]. The milk samples were analyzed for pesticide residues of chlorpyrios, endosulfan, profenofs, and bifenthrin. The residues in raw and ultra-heated milk were determined between the range of 0.1-30µg/g. Residues in heat-treated samples were within the range of 0.1-30µg/L. All UTH processed samples contain pesticide residue within the acceptable limit set by the (WHO) on the other hand raw milk samples, chlorpyrios and endosulfan were found above the maximum residue limit (MRL), raw milk samples show a higher prevalence than heat-treated samples [30].

Conclusion.

It is concluded that the pesticide residues were found magnificently in the mother milk of Danyore, Gilgit and Eidgah, Astore. Mostly detected pesticides in mothers milk were found from fruits and vegetables exported from urban areas of Pakistan, and use of pesticides and synthetic fertilizers to their fields extensively. The multiparous women had prominent pesticide residues than primiparas. Among the analyzed pesticides the residue of cypermethrin, Deltamethrin and Chlorpyrifos was present in some of the samples. The lactating mothers were mainly consuming vegetables and fruits imported from urban areas during the gestation period and after delivery. Although the residue level was low in all samples if preventive measures are not taken by people and the government it would be a threat to human health in future.

References

[1] Nida, M, S., Ahmad, R., and Estaitieh, H. Organochlorine pesticide residues in dairy products in Jordan. Chemosphere, (2009). 77(5), 673-678.

[2] Landrigan, P. J., B. Sonawane, D. Mattison, M. McCally and A. Garg. Chemical contaminants in breast milk and their impacts on children's health: an overview. Environmental health perspectives, (2002). 110(6): A313-A315.

[3] Siddiqui, M., Srivastava, S., Mehrotra, P., Mathur, N., & Tandon, I. Persistent chlorinated pesticides and intra-uterine foetal growth retardation: a possible association. International archives of occupational and environmental health, (2003). 76(1), 75-80.

[4] Sharma, A., Gill, J. P. S., Bedi, J. S., & Pooni, P. A. Monitoring of pesticide residues in human breast milk from Punjab, India and its correlation with health associated parameters. Bulletin of environmental contamination and toxicology, (2014). 93(4), 465-471.

[5] Kumar, A., A. Baroth, I. Soni, P. Bhatnagar, P.J. John. Organochlorine Pesticide Residues in Milk and Blood of Women from Anupgarh, Rajasthan, India. Environmental Monitoring and Assessment, (2006). 116(1-3):1-7.

[6] Wong C.K.C., K.M. Leung, B.H.T. Poon, C.Y. Lan and M.H. Wong. 2002. Organochlorine hydrocarbons in human breast milk collected in Hong Kong and Guangzhou. Arch Environ Contam Toxicol 43:364–372.

[7] Khan, D. A., Shabbir, S., Majid, M., Naqvi, T. A., & Khan, F. A. (2010). Risk assessment of pesticide exposure on health of Pakistani tobacco farmers. Journal of exposure science & environmental epidemiology, 20(2), 196-204.

[8] Ishaq, Z. and M.A. Nawaz. 2018. Analysis of contaminated milk with organochlorine pesticide residues using gas chromatography. International journal of food properties, 21(1), 879-891.

[9] Damalas, C. A., & Eleftherohorinos, I. G. (2011). Pesticide exposure, safety issues, and risk assessment indicators. International journal of environmental research and public health, 8(5), 1402-1419.

[10] Tariq, M.I., S. Afzal, I. Hussain and N. Sultan. Pesticide Exposure in Pakistan: A review. Environ. Inter. (2007). 33:1107-1122.

[11] Siddiqui, M., Srivastava, S., Mehrotra, P., Mathur, N., and Tandon, I. (2003). Persistent chlorinated pesticides and intra-uterine foetal growth retardation: a possible association. International archives of occupational and environmental health, 76(1), 75-80.

[12] Ribas-Fitó, N., Cardo, E., Sala, M., De Muga, M. E., Mazón, C., Verdu, A., and Sunyer, J. (2003). Breastfeeding, exposure to organochlorine compounds, and neurodevelopment in infants. Pediatrics, 111(5), e580-e585.

[13] Hassine, S. B., Ameur, W. B., Gandoura, N., and Driss, M. R. (2012). Determination of chlorinated pesticides, polychlorinated biphenyls, and polybrominated diphenyl ethers in human milk from Bizerte (Tunisia) in 2010. Chemosphere, 89(4), 369-377.

[14] Chao, H. R., Wang, S. L., Lee, C. C., Yu, H. Y., Lu, Y. K., and Päpke, O. (2004). Level of polychlorinated dibenzo-p-dioxins, dibenzofurans and biphenyls (PCDD/Fs, PCBs) in human milk and the input to infant body burden. Food and chemical toxicology, 42(8), 1299-1308.

[15] Barr, D. B., Wang, R. Y., and Needham, L. L. (2005). Biologic monitoring of exposure to environmental chemicals throughout the life stages: requirements and issues for consideration for the National Children's Study. Environmental Health Perspectives, 113(8), 1083-1091.

[16] Rodríguez, Á. G. P., López, M. I. R., Casillas, T. A. D., León, J. A. A., Prusty, B. A. K., and Cervera, F. J. Á. (2017). Levels of persistent organic pollutants in breast milk of Maya women in Yucatan, Mexico. Environmental monitoring and assessment, 189(2), 59.

[17] Mannetje, A. T., Coakley, J., Bridgen, P., Brooks, C., Harrad, S., Smith, A. H., ... and Douwes, J. (2013). Current concentrations, temporal trends and determinants of persistent organic pollutants in breast milk of New Zealand women. Science of the total environment, 458, 399-407.

[18] Çok, I., Mazmanci, B., Mazmanci, M. A., Turgut, C., Henkelmann, B., and Schramm, K. W. (2012). Analysis of human milk to assess exposure to PAHs, PCBs and organochlorine pesticides in the vicinity Mediterranean city Mersin, Turkey. Environment international, 40, 63-69.

[19] Bedi, J. S., Gill, J. P. S., Aulakh, R. S., Kaur, P., Sharma, A., and Pooni, P. A. (2013). Pesticide residues in human breast milk: Risk assessment for infants from Punjab, India. Science of the total environment, 463, 720-726.

[20] Stockholm Convention. Available online:http:// www.pops.int/ TheConvention/ Overview/ TextoftheConvention/tabid/2232/Default.aspx (accessed on 14 January 2017).

[21] Naqvi, A., Qadir, A., Mahmood, A., Baqar, M., Aslam, I., Jamil, N., ... & Zhang, G. Screening of human health risk to infants associated with the polychlorinated biphenyl (PCB) levels in human milk from Punjab Province, Pakistan. Environmental Science and Pollution Research, (2020). 27(7), 6837-6850.

[22] Corcellas, C., M. L. Feo, J.P. Torres, O. Malm, W. Ocampo-Duque, E. Eljarrat and D. Barceló. Pyrethroids in human breast milk: occurrence and nursing daily intake estimation, Environment international, (2012). 47, 17-22.

[23] Limon-Miro, A. T., M.L. Aldana-Madrid, G. Alvarez-Hernandez, L.E. Antunez-Roman, G. Rodriguez-Olibarria and M.E.V. Juillerat. Breast milk intake and mother to infant pesticide transfer measured by deuterium oxide dilution in agricultural and urban areas of Mexico. Chemosphere, (2017). 181:682-689.

[24] Bedi, J. S., J.P.S. Gill, R.S. Aulakh and P. Kaur. Pesticide residues in bovine milk in Punjab, India: spatial variation and risk assessment to human health. Archives of environmental contamination and toxicology, (2015). 69(2), 230-240.

[25] Palma, D. C., C. Lourencetti, M.E. Uecker, P.R. Mello, W.A. Pignati and E.F. Dores. Simultaneous determination of different classes of pesticides in breast milk by solid-phase dispersion and GC/ECD. Journal of the Brazilian Chemical Society, (2014). 25(8), 1419-1430.

[26] ul Hassan, A., A.B. Tabinda, M. Abbas and A.M. Khan. Organochlorine and pyrethroid pesticides analysis in dairy milk samples collected from cotton growing belt of Punjab, Pakistan. Pak. J. Agri. Sci., (2014). 51(2), 331-335.

[27] Chen, X., P. Panuwet, R.E. Hunter, A.M. Riederer, G.C. Bernoudy, D.B. Barr and P.B. Ryan, P. B. 2014. Method for the quantification of current use and persistent pesticides in cow milk, human milk and baby formula using gas chromatography-tandem mass spectrometry. Journal of Chromatography B,(2014). 970, 121-130.

[28] Sanghi, R., M. K. Pillai, T. R. Jayalekshmi and A. Nair. 2003. Organochlorine and organophosphorus pesticide residues in breast milk from Bhopal, Madhya Pradesh, India. Human & experimental toxicology, 22(2), 73.

[29] Bedi, J. S., Gill, J. P. S., Kaur, P., & Aulakh, R. S. Pesticide residues in milk and their relationship with pesticide contamination of feedstuffs supplied to dairy cattle in Punjab (India). Journal of Animal and Feed Sciences, (2018). 27(1), 18-25.

[30] Jawaid, S., F.N. Talpur, S.M. Nizamani, A.A. Khaskheli and H.I. Afridi. 2016. Multipesticide residue levels in UHT and raw milk samples by GC-μECD after the QuEChER extraction method. Environmental monitoring and assessment, 188(4), 230.