Journal of Applied and Natural Science 9 (2): 1012 - 1016 (2017)

JANS

Monitoring of organochlorine pesticide residues from bovine milk in Patna (Bihar), India

S. B. Sah[1], R. N. Gupta[2], T. Saha[2*] and S. P. Singh[3]

[1]Department of Entomology Bhola Paswan Shastri Agricultural College, Purnea (Bihar), INDIA
[2]Bihar Agricultural College, Sabour, Bhagalpur-813 210 (Bihar), INDIA
[3]Rajendra Agricultural University, Pusa, Samastipur (Bihar), INDIA
*Corresponding author. E-mail: tamoghnasaha1984@gmail.com

Received: December 12, 2015; Revised received: December 12, 2016; Accepted: May 6, 2017

Abstract: The milk has high nutritional properties and is widely used as baby food in different forms. The present study was undertaken to evaluate pesticide contamination in bovine milk samples from Patna district of Bihar. Out of 24 samples analyzed during 2012, 18 samples (75 %) were found to be contaminated with HCH residues varying from ND-0.178 mg kg^{-1} (mean value 0.135 mg kg^{-1}). Seven samples (29.2 %) had HCH exceeding MRL of 0.01 mg kg^{-1}. DDT residues were detected in 20 samples (83.3 %) and ranged from ND-0.132 mg kg^{-1} (mean value 0.122 mg kg^{-1}). Five samples (20.8 %) contained DDT residues above the prescribed MRL of 0.05 mg kg^{-1}. Out of 24 samples of bovine milk analyzed during 2013, 16 samples (66.7 %) were found to be contaminated with HCH and 15 samples (62.5%) with DDT residues. The residues of HCH varied from ND-0.154 mg kg^{-1} (mean value 0.053 mg kg^{-1}) and DDT from ND-0.120 mg kg^{-1} (mean value 0.122 mg kg^{-1}). The residues of HCH and DDT were above MRL in four samples (16.7 %) and three samples (12.5 %) respectively. The management practices of animals and legal punishment on using banned pesticides are the alternatives to reduce pesticide contamination incidences in milk.

Keywords: Bovine milk, DDT, HCH, Monitoring, Pesticide, Residue

INTRODUCTION

Milk has been recognized as the most wholesome and complete single food available in nature for health and as a medicine, both preventive as well as curative. It also acts as a raw material in the manufacture of a wide variety of products. Milk has also unique nutritional properties that make it an especially important food particularly for the young. A continuous use of persistent organochlorine pesticides (OCPs) like DDT and HCH during the last decades has resulted in wide spread contamination of milk. Although the use of these pesticides in agriculture in India has been banned, but still they are being used for the public health programme for control of vectors of malaria, filaria and kalazar etc (Agnihotri, 1999). Contamination of bovine milk with organ chlorine pesticides particularly DDT and HCH was first reported in 60`s and 70`s from Delhi and Punjab (Agnihotri *et al.,* 1974; Dhaliwal and Kalra, 1977). The presence of OCPs residues in dairy milk (John *et al*., 2001; Mukherjee and Gopal, 1995; Nicholas., 2008) have been reported in earlier studies. Likewise various workers (Kathpal *et al*., 1992; Kumar and Nath, 1996; Agnihotri, 1999; Vasanthi *et al.,* 2003) revealed wide spread contamination of bovine milk from different regions of the country. According to ICMR (1993), out of 2200 milk samples analyzed all over India, 37 per cent contained DDT and 42 per cent HCH above MRL values. Studies conducted (Anonymous, 2001) under All India Coordinated Research Project (AICRP) on Pesticide Residue during 1999-2000 revealed that out of 537 samples, 52 per cent were found to be contaminated with DDT and 94 per cent with HCH. Among which 14 per cent contained DDT and 8 per cent contained HCH residues above MRL values. The residues of these persistent pesticides, if present, could act as a potential health hazard to the consumers and may cause many chronic diseases. Hence, the contamination of bovine milk with residues of persistent organ chlorine insecticides is a matter of great concern. Lemesm *et al*. (2004) assessed the levels of pesticide residues and metabolites in seventy-three milk samples collected in 2001 and 2002 by Brazilian sanitary surveillance authorities from different commercial establishment in SaPaulo, Brazil and none of the pesticides were detected in the milk sample analyzed. In India, the largest pesticide consumption state was Uttar Pradesh, according to the data of 1995-1996 and 1999-2000, demonstrated by Central Insecticide Board and Registration Committee, India (Srivastava *et al*., 2008). Thus, the present study was carried out to assess bovine milk samples from Patna District of Bihar.

MATERIALS AND METHODS

Twenty four samples of bovine milk were collected

from vendors and milk dairies from Patna district in 2012 and 2013. Each sample consists of 250 ml of milk. The samples were brought into laboratory and analyzed for organochlorine pesticides. Analysis of milk for pesticide residues is in consonance with Development of an Analytical Method for Determination of Antiparasitics Residues in milk using QuEChERS (Quick Easy Cheap Effective Rugged Safe) method for analysis (Agilent technologies, R. Furlani et al Food technology institute-ITAL, Campnas-SP, Brazil, B.Ramos UNIMEP Brazil, Daniela Daniel Agilent Technologies, Inc.Brazil.)

Extraction: 10 ml of milk sample taken in a clean 50 ml centrifuge tube and added 10 ml of 1 % acetonitrile, 1 gram of NaCl and 4 gram of $MgSo_4$ and shake well for two minutes. Centrifuge the sample containing tube at 4000 RPM for 5 min at 10 ^{0}C temperature.

Cleanup: For cleaning we have to take 6 ml supernatant in a 15 ml centrifuge tube and add 150 mg PSA and 1 g MgSo4 then vertex for 30 minute. Then centrifuge the content at 4000 RPM for 5 minute.2 ml supernatant is taken in a vial and evaporate using nitrogen concentrator. Again reconstitute the residue with 2 ml GC compatible solvent and vertex the mixture for 30 sec to dissolve the residues. Finally filtered the residue sample through 0.2µ PTFE membrane filter and analyzed in Gas Chromatograph-Mass Spectrometer (GC MS), Chemito GC 1000

Detector: ECD-Ni ^{63}for organochlorines and synthetic pyrethroids.

Column: Glass column (2m) packed with 3 % OV-101 on 80-100 mesh CHW (HP).

Glass column: (2m) packed with 1.5 % OV-17+1.95 % QF-1 on 80-100 mesh CHW (HP) (For organochlorines only).

RESULTS AND DISCUSSION

The data on the residues analysis of organochlorine pesticides in bovine milk samples collected from milk vendors and dairies of Patna (Bihar) during 2012 and 2013 are presented in Tables 1 to 2 and 3 to 4 respectively.

The data revealed that out of 24 milk samples analyzed during 2012, 18 samples were found to be contaminated with residues of HCH which existed mainly in the form α, β and γ-isomers with α and γ-isomers as the major contaminant. The total HCH residues varied from ND-0.178 mg kg-1. Seven samples (29.2 %) data

Table 1. Residues (mg kg-^{1}) of organochlorine pesticides in bovine milk at Patna district during the year 2012.

Sample No.	α- HCH	β- HCH	γ- HCH	δ- HCH	∑ HCH	p,p-DDE	p,p-DDD	p,p-DDT	∑DDT
1	0.024	0.017	0.033	ND	0.074	0.018	0.018	0.027	0.068
2	ND	ND	0.108	ND	0.108	0.013	ND	ND	0.013
3	ND	ND	ND	ND	ND	ND	ND	ND	ND
4	0.019	0.051	0.028	0.012	0.110	0.015	ND	0.014	0.029
5	0.023	0.034	0.025	ND	0.082	0.023	0.009	ND	0.032
6	ND	ND	ND	ND	ND	0.011	ND	ND	0.011
7	0.023	0.013	0.030	ND	0.064	0.013	0.006	ND	0.019
8	ND	ND	0.057	ND	0.057	0.036	0.020	ND	0.056
9	ND	ND	ND	ND	ND	0.030	0.007	ND	0.037
10	0.037	0.024	0.041	0.016	0.118	ND	ND	ND	ND
11	0.028	ND	0.041	ND	0.029	0.022	0.010	ND	0.032
12	0.061	0.032	0.026	ND	0.119	ND	ND	ND	ND
13	ND	ND	ND	ND	ND	0.014	0.006	ND	0.020
14	0.018	0.047	0.035	0.020	0.120	0.035	0.019	0.076	0.132
15	ND	ND	ND	ND	ND	ND	ND	ND	ND
16	0.012	0.020	0.010	ND	0.042	0.022	0.017	0.019	0.058
17	0.015	0.019	0.010	ND	0.044	0.013	ND	ND	0.013
18	ND	ND	ND	ND	ND	0.012	ND	ND	0.012
19	0.058	0.024	0.039	ND	0.121	0.018	ND	ND	0.018
20	0.015	0.032	0.024	0.008	0.079	0.022	0.010	ND	0.032
21	0.020	0.024	0.030	ND	0.074	0.024	0.008	0.012	0.044
22	0.038	0.059	0.081	ND	0.178	0.029	0.014	0.021	0.064
23	0.015	ND	0.024	ND	0.039	0.027	0.020	ND	0.047
24	0.016	0.026	0.022	0.010	0.074	0.017	0.014	ND	0.031

ND-Not detected

Table 2. Organochlorine pesticides residues (mg kg-^{1}) in bovine milk at Patna district during the year 2012.

Pesticides	Number of samples			Range of residues (Mean)
	Analyzed	Contaminated (%)	>MRL (%)	
∑ HCH	24	18 (75.0)	7 (29.2)	ND-0.178 (0.135)
∑DDT	24	20 (83.3)	5 (20.8)	ND-0.132 (0.122)
Total samples	24			

ND:Not detected

Table 3. Residues (mg kg-[1]) on organochlorine pesticides in bovine milk at Patna district during the year 2013.

Sample No.	α- HCH	β- HCH	γ- HCH	δ- HCH	∑ HCH	p,p-DDE	p,p-DDD	p,p-DDT	∑DDT
1	0.027	0.010	0.014	ND	0.051	0.026	0.008	0.021	0.055
2	0.018	0.029	0.012	ND	0.059	ND	ND	ND	ND
3	ND	ND	ND	ND	ND	0.022	ND	ND	0.022
4	ND	ND	ND	ND	ND	0.013	ND	ND	0.013
5	0.021	0.045	0.030	0.020	0.116	ND	ND	ND	ND
6	ND	0.008	0.012	ND	0.020	0.011	ND	ND	0.011
7	0.026	0.022	0.027	ND	0.075	0.021	0.013	ND	0.034
8	0.016	0.034	0.026	0.009	0.085	ND	ND	ND	ND
9	ND	ND	ND	ND	ND	0.019	0.013	ND	0.032
10	0.010	ND	0.012	ND	0.022	ND	ND	ND	ND
11	ND	ND	ND	ND	ND	0.012	0.008	ND	0.020
12	0.020	ND	0.043	ND	0.063	ND	ND	ND	ND
13	0.035	0.030	0.055	ND	0.120	0.018	0.023	0.028	0.069
14	ND	ND	ND	ND	ND	0.034	0.011	ND	0.045
15	0.027	0.033	0.024	0.018	0.102	ND	ND	ND	ND
16	0.030	0.024	0.034	ND	0.088	ND	ND	ND	ND
17	ND	ND	ND	ND	ND	0.019	0.012	ND	0.031
18	ND	ND	ND	ND	ND	0.022	0.015	ND	0.037
19	0.046	0.010	0.030	ND	0.086	ND	ND	ND	ND
20	0.018	ND	0.033	ND	0.051	0.020	0.011	0.018	0.049
21	0.016	0.056	0.019	ND	0.091	0.026	ND	ND	ND
22	0.032	0.051	0.073	0.021	0.177	0.066	0.024	0.030	0.120
23	0.019	0.024	0.020	ND	0.063	0.020	0.009	0.010	0.037
24	ND	ND	ND	ND	ND	ND	ND	ND	ND

ND:Not detected

Table 4. Organochlorine pesticides residues (mg kg-[1]) in bovine milk at Patna district during the year 2013.

Pesticides	Number of samples			Range of residues (Mean)
	Analyzed	Contaminated (%)	>MRL (%)	
∑ HCH	24	16 (66.7)	4 (16.7)	ND-0.154 (0.053)
∑DDT	24	15 (62.5)	3 (12.5)	ND-0.120 (0.024)
Total samples	24			

ND:Not detected

showed that total HCH exceeding MRL of 0.1 mg kg-1 on whole milk basis. The average concentration of HCH residues was found as 0.135 mg kg-1. The detectable amount of DDT residues were found in 20 samples (83.3 %). DDT residues were present as p, p-DDE, p, p DDD and p, p-DDT. The predominant compound in most of the samples was p,p-DDE. DDT residues varied from ND-0.132 mg kg-1(0.122 mg kg-1). Five samples (20.8 %) contained residues above the prescribed MRL of DDT (0.05 mg kg-1). During 2013 (Table 3 and 4), out of 24 milk samples analyzed 16 samples (66.7 %) were found to be contaminated with HCH residues and total HCH residues varied from ND -0.154 mg kg-1(mean 0.053 mg kg-1). Four samples (62.5 %) and ranged from ND-0.120 mg kg-1(mean 0.024 mg kg-1). Three samples (12.5 %) had DDT residues above the prescribed MRL.

A perusal of the data (Tables 1) revealed that the residues of both HCH and DDT were detected in milk samples during 2012. HCH residues (ND-0.178 mg kg -1, mean value 0.135 mg kg-1) were detected in 75 per cent of samples, out of which 29.2 per cent samples contained residues above the prescribed MRL of 0.1 mg kg-1. In addition to HCH, 83.3 per cent milk samples were found to be contaminated with DDT (ND-0.132 mg kg-1, mean value (0.122 mg kg-1), out of which 20.8 per cent of the samples had total DDT exceeding MRL of 0.05 mg kg-1. The contamination level of both HCH and DDT residues in milk samples declined during 2013-14 (Table 4). Sixteen samples (66.7 %) contained HCH residues (ND-0.154 mg kg-1, mean value 0.053 mg kg-1) and only 16.7 per cent exceeded MRL. DDT residues (ND-0.120 mg kg-[1], mean value 0.024) were found in 62.5 per cent samples, out of which 12.5 per cent had total DDT residues above the MRL. The pesticide residue contamination levels observed in the present study are comparatively lower than those reported earlier form other states. It may be because of decreased HCH and DDT usage in the last one decade. The low level of pesticide residues in milk obtained in the present study may be explained as the use of pesticides per hectare of cropped area in Bihar is low (94g) in comparison to other states like Uttar Pradesh, Punjab, Haryana, Andhra Pradesh, Gujarat etc. in the country.

Agnihotri (1999) reported the residues of organochlorine pesticides in 487 milk samples and DDT residues were detected in 86.5 per cent of samples out of which

43.2 per cent samples contained residues above the prescribed MRL. In addition to DDT, 89.7 per cent of the samples were found contaminated with HCH (80.3 % with gamma-HCH) out of which 77.8 per cent had total HCH exceeding MRL. Vasanthi *et al.* (2003) conducted a study to determine level of contamination of milk with chlorinated hydrocarbon residues in Coimbatore, Tamil Nadu. Out of 69 samples analysed, 84 and 24 per cent of samples were contaminated with HCH and DDT, respectively. The Residues of p,p-DDE rangesd from 0.002 to 0.070 mg kg^{-1} in all milk sample collected from various farms. Residues of p,p-DDT were detected only in two samples, whereas, residues of p,p-DDD were detected in all the milk samples collected at different places and ranged between 0.001 and 0.007 mg kg^{-1}. In case of HCH isomers, α- HCH was not detected in any of the milk samples, β- HCH ranges from 0.003 to 0.025 mg kg^{-1}.The γ- HCH was detected in all the samples and ranged from 0.006 to 0.025 mg kg^{-1}. The residue of HCH and DDT-R detected in the milk samples were well below MRL values. Zhong *et al*. (2003) reported organochlorine pesticides and their metabolite residues in milk taken from super markets in Beijing, China. The average concentrations of total HCHand DDT were0.038 and 0.046 mg kg^{-1}on a fat body respectivety. Of 72 samples analysed, 3 from South China contained higher levels of DDT and HCH residues that exceeded the FAO/WHO accepted tolerance level. Aslam *et al.* (2013) reported pesticide Residue levels of P'P'- DDT of 0.038 and 0.033 ppm in buffalo milk samples of Delhi city, India .The gas chromatographic analysis of fodder samples of Zone 5 of Musi river showed the residues of dicofol at concentration of 0.07±0.0007 (0.071-0.077). Among organophosphorus compounds, dimetheoate was present in milk samples collected from Zone 6 at a level of 0.13±0.006 (0.111-0.167). DDT and HCH insecticides being lipophilic in nature can bind easy way into milk when the animals feed on the contaminated fodder. They persist in fat soluble form and usually do not get converted to the water soluble metabolites. Therefore, the pesticide contamination may occur by ingestion of contaminated feed and water by animals or by direct application of insecticides on the animals and their dwellings to control ectoparasites. Their deposits on walls gradually volatilize and the vapours are absorbed into the animal body through inhalation and direct absorption through skin.

Conclusion

From this study, it can be concluded that, during 2012, the residues of both HCH and DDT were detected in milk samples. Out of 24 samples, HCH residues (ND-0.178 mg kg^{-1}, mean value 0.135 mg $kg^{-1)}$ were detected in 18 samples (75 per cent) , out of which 7 samples (29.2 per cent) contained residues above the prescribed MRL of 0.1 mg kg^{-1}. In addition to HCH, 20 milk sample (83.3 per cent) were found to be contaminated with DDT (ND-0.132 mg kg^{-1}, mean value 0.122 mg kg^{-1}), in which 5 samples (20.8 per cent) have exceeded MRL of 0.05 mg kg^{-1}. The contamination level of both HCH (ND-0.154 mg kg^{-1}, mean value 0.053 mg kg^{-1}) and DDT residues (ND-0.120 mg kg^{-1}, mean value 0.024 mg kg^{-1}) in milk samples declined during 2013. Owing to effects on human, animal and environmental health of pesticide residues need for education and awareness among farmers about extensive use of pesticide was envisaged.

ACKNOWLEDGEMENTS

The authors are thankful to Department of Entomology of Rajendra Agricultural University, Pusa, Smastipur, Bihar for their kind cooperation, financial assistance and providing laboratory facilities.

REFERENCES

Agnihotri, N. P. Dewan, R. S. and Jain H. K. (1974). Residues of food commodities from Delhi II. High food content food materials. *Indian J. Entomol.,* 36: 203-208

Agnihotri, N. P. (1999). Pesticide safety evaluation and monitoring. All India Co-ordinated Research Project on Pesticide Residues, Indian Agricultural Research Institute, New Delhi, P. 173

Anonymous, (2001). Proceedings of IX Workshop of All India Co-ordinated Research Project on pesticide Residues held at KAU, Vellayani, P. 56

Aslam, M. Rais, S. and Aslam, M. (2013). Quantification of organochlorine pesticide residues in buffalo milk samples of Delhi city, India. *J. Environ. Prot.,* 4: 964-974

Dhaliwal, G. S. and Kalra, R. (1977). DDT residues in milk samples from Ludhiana and surrounding areas. *Indian J. Ecol.,* 4: 13-22

John, P. J. Bakore, N. and Bhatnagar, P. (2001). Assessment of organochlorine pesticide residues levels in dairy milk and buffalo milk from Jaipur city, Rajasthan India. *Environ Int.,* 26: 231-236

Kathpal, T. S. Kumari, Singh. S. and Singh, J. (2001). Multi residue analysis of bovine and Human milk in cotton growing belt in Haryana. Society of Pesticide Science, India. Extended abstract, International Conference on pesticide, Environment, Food Security, New Delhi: p. 236-238

Lemesm, V. R. R. kussumi, T. A. and Rocha, S. O. B. (2004). Monitoting of pesticide residues in milk consumed by the population of the state of Sao Paulo, Brazil, 2000 and 2002. *Revista do Instituto Adolfo Lutz,* 63 (1): 24-30

Mukherjee, I. and Gopal, M. (1996). HCH and DDT residues in animal feed and its composition in and around Delhi. *Indian J Plant Protec.,* 23: 61-63

Nicholas, M. J. (2008). Assessment of organochlorine pesticide residues in cow milk, sediments and water from Nairobi West region. *Earth Science Frontiers,* 15(5): 242–247

Srivastava, S. Narvi, S. S. and Prasad, S. C. (2008). Organochlorines and Organophosphates in Bovine Milk Sam-

ples in Allahabad Region .*Int. J. Environ. Res.,* 2(2): 165-168

Vasanthi, D. Kuttalam, S. and Jayakumar, R., Vijayalakashmi, K. and Chandrasekaran, S. (2003). Monitoring of bovine milk samples for chlorinated pesticides. *Pesticide Res. J.,* 15(2): 193-194

Zhong, W., Xu, D., Chai, Z. and Mao, X. (2003). 2001 survey of organochlorine pesticides from retail milk from Beijing,P.R.China. *Food Additives and Contaminants,* 20(3): 254-258