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DETECTION OF PESTICIDE RESIDUES IN MILK AND SOME DAIRY PRODUCTS

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ABSTRACT

The study were conducted to monitor and determine the presence organochlorine pesticide residues in raw buffalo's milk and some milk products. The studied samples were collected from different regions, Sharkia Governorate, Egypt. The effect of heat treatment on pesticide residue in raw buffalo's milk and butter was also studied. The results showed that the only detected organochlorine pesticide residues in all examined milk samples was p,p'-DDE. Resides of DDE detected in yoghurt were 2.7298±0.1126 ppb, with a mean value of 1.9161±0.0992 ppb., while no residues were detected in cottage cheese samples. The organochlorine pesticide residues detected in butter samples were lindane, heptachlor epoxide and p,p'-DDE with mean values of 7.428±1.667, 3.384±1.583 and 67.063±4.933 ppb on fat basis, respectively. Effect of common heat treatment on p,p'-DDE residues in raw buffalo's milk treated by pasteurization, boiling and sterilization, were 2.509, 0.932 and 0.2599 ppb on milk basis, respectively with degradation percent of 22.13, 71.16 and 92.8% , respectively. Resides detected in butter samples after heating processing to produce ghee were lindane, heptachlor epoxide and p,p'-DDE with a mean concentration 2.948, 1.935 and 15.609 ppb on fat basis respectively with degradation percent of 79.1, 87.50 and 76.76 %, respectively. We can conclude from the study that, the sterilization of raw milk is the most effective method for reduction of pesticide residues. The butter processing by heat treatment produced ghee (Samna) with marked decrease in organochlorine residue content.

Keywords: Detection, Pesticide, Residue, Milk, Dairy Product

INTRODUCTION

Milk is an essential nutritional food for infant's as well as all other age stages.

Contaminations of milk and dairy products with residues of pesticides poses serious risks to consumers.. (Abu-Zahw *et a*l., 1993).

Pesticides were identified as one of the major environmental pollutants. The environmental pollution with organochlorines has decreased due to restriction and /or banning of these compounds .However, several recent reports (Costabeber *et a*l., 2001; Pandit *et al*., 2002; Pardio *et al*., 2003; Waliszewski *et al*., 2003_b; Zhong *et al*., 2003 and Battu *et al*., 2004) indicated that pollution with organochlorines pesticides is still reported in many parts of the world.

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Animals are exposed to pesticide pollution from environment and ingestion of contaminated feed and water. The milk and its products are one of the important media for accumulation of organochlorine pesticides (Mukherjee and Gobal, 1993). Because of their environmental stability and lipophilic property, organochlorine pesticides and their metabolites tend to accumulate in the milk fat (Aman and Bluthgen, 1997). The rate of excretion into milk depends on the stage of lactation, the quantity of milk fat, the amount consumed daily and the duration of exposure (Vreman *et al*., 1977).

The contamination of milk and its products with organochlorine pesticide and their metabolites have been recorded in different countries. In Egypt, although the use of organochlorine pesticide has been banned in 1980 (El-Marsafy *et al*., 1999), high amount of these chemicals were found in milk and milk products collected from different localities (Dogheim *et al*.,1988; Dogheim *et al*., 1990; Abu-Zahw *et al*., 1993; Abd El-Kader *et al*., 1994; Zidan *et al.,1994; Ragab et al., 1996; Aman and Bluthgen, 1*997; Neamat-Allah, 1998; Dabiza *et al*., 1999 and Ayoub, 2000)

Organochlorine pesticides are a source of several chronic diseases, including cancer, liver and kidney disfunction, growth depression and neuritis (Oestreicher, *et al*., 1971, IARC 1974 and Sandhu 1992). Therefore, contamination of milk and milk products with pesticides need further emphasis over our country to probe and control such pollution.

The specific objectives of this study are:

1- Determination of organochlorine pesticide residues in raw buffalo's milk and milk products (yoghurt, kareish cheese and butter) collected from different districts at Sharkia Governorate.
2- Study the effect of heat treatment on the level of organochlorine pesticide residues in raw milk.
3- Study the effect of butter processing by heat treatment (ghee manufacture) on organochlorine pesticide residues.

MATERIALS AND METHODS

A- Collection of samples

A total of 138 random samples (72 samples of raw buffalo's milk, 24 yoghurt, 24 Kareish cheese and 18 butter) were collected from different districts, January 2002 to April 2003.

Raw buffalo’s milk samples (500 ml each) were collected from street peddlers and dairy shops in clean glass containers. While, yoghurt samples were collected from dairy shops and supermarkets in its original containers. The cottage cheese samples (250 gm each) and butter samples (one kg each) were collected from public markets in villages and kept in clean and dry sampling jars.

Each collected samples was labeled to identify the source, site and date of sampling, and transferred to the laboratory without delay. Delayed samples were stored in ice bag.

B. Preparation of Samples

1- **Milk and yoghurt:** Each sample of raw milk or yoghurt was thoroughly mixed and a sub sample of 10 ml was measured in small glass containers with screw Teflon capped stoppers and kept deeply frozen at -20 ^{o}C for pesticide residues analysis.

2- **Cottage cheese:** Twenty five gm of each cheese sample was thoroughly meshed with 50 ml of distilled water in a clean dry blender, then 10 ml from each prepared sample was measured into small glass containers with screw Teflon stoppers and kept deeply frozen at -20 ^{o}C for analysis.

3- **Butter:** Each sample was divided into 2 portions and each portion was kept in clean dry glass container. The first portion (250 gm) was warmed to about 50 ^{o}C until fat separate and decanted through dry filter paper No. 10, and then 3 gm were taken for pesticide analysis. The second portion (750 gm) was kept in deep freezer at –20 ^{o}C for processing by heat treatment (ghee manufacture)**.**

I- Determination of organochlorine pesticide residues in milk and other milk products

A-Extraction procedure

1- Milk, Yoghurt and Kareish cheese

The extraction of milk, yoghurt and kareish cheese samples was carried out according to Suzuki *et al*. (1979) and Riva and Anadon (1991)

2- Butter

The extraction of butter samples was conducted according to Corneliussen *et al*. (1984)

B-Clean up:

All extracts of milk, yoghurt, cheese and butter were subjected to clean up procedure according to Suzuki *et al*., (1979**)**

C-Quantitative determination of organochlorine pesticides

1- Preparation of stock standards

Pesticides stock standard were prepared in µg/ml concentration by dissolving 0.1 gram of the standard in 100 ml pesticide quality hexane using volumetric flask. The stock solution was transferred to ground glass Stoppard reagent bottles and stored in refrigerator.

2- Preparation of chromatographic working standards

Standard solutions were prepared from the stock solution using microsyrings. A typical concentration (1ng/1µl) was prepared by diluting 0.1 ml of stock solution to 100 ml using hexane. The standard solution were transferred to ground glass Stoppard bottle and stored in refrigerator.

3- Preparation of extracted samples

The extracted sample was diluted with 0.5 ml of nanograde hexane. Five µl of diluted extract were injected into Electron capture gas liquid chromatography using standard microsyring.

4- Analysis on Gas liquid chromatography

Gas liquid chromatography Hewlette packared 5890 Series II equipped with double Electron Capture Detectors (ECD's Ni^{63}) and double columns system was used for identification, quantification and confirmation of organochlorine pesticide residues under the conditions illustrated in Table (1). Confirmation of the results was carried out by injection two different columns

of gas liquid chromatography. Retention times for organochlorine pesticide were calculated relative to aldrin as show in Table (2) to avoid daily deviation in retention times.

Table (1): Conditions of gas chromatographic determination

Columns			Temperature Program				Conditions	
	Column 1	Column 2	Level	Rate oC/min.	Temp (oC)	Time (min)		
Name	PAS- 5%phenyl Methyl Siloxane	PAS-1701-14 Cyanopropyl phenyl Methyl	1 2 3	---- 20 6	90 150 270	2 0 25	Inject.Temp. Detec.Temp. Carrier gas	225 °C 300 °C N(2ml/m
Film thickness	0.52 um	0.25 um						
Length	25 m	25 m						
Column I.D.	0.32mm	0.32 mm						
Phase ratio	150	320						

Table (2): Retention times of organochlorine pesticide relative to aldrin

Organochlorine pesticides	Column(1) (PAS-5)	Column(2) (PAS-1701)
α-HCH	0.73	0.81
HCB	0.74	0.73
β-HCH	0.78	1.06
γ-HCH (lindane)	0.79	0.96
Heptachlor	0.93	0.94
Aldrin	1	1
Heptachlor epoxide	1.08	1.19
p,p'-DDE	1.20	1.27
Dieldrin	1.21	1.29
Endrin	1.25	1.34
p,p-DDD	1.29	1.37
o,p'-DDT	1.30	1.45
p,p'-DDT	1.35	1.48

5-Rate of recovery:

The reliability of analytical method was examined by fortifying the tested samples with known quantities of tested pesticides followed by the same procedure of extraction, clean up and analysis. The percentage rates of recoveries of organochlorine pesticides were recorded in Table (3).

Table (3): Recovery percent of organochlorine pesticides as a result of fortification of milk, cheese, butter and yogurt

Organochlorine pesticides	Milk	Cheese	Butter	Yoghurt
Alpha-HCH	89	79	78	80
Gamma-HCH	84	100	80	96
Beta-HCH	90	78	78	79
DDT-o,p	96	90	93	87
DDT-p,p	82	76	76	83
DDE-p,p	101	96	95	90
DDD-p,p	90	83	90	96
Heptachlor	96	85	87	75
Heptachlor epoxide	99	94	79	91
Dieldrin	80	90	96	85
Endrin	91	87	90	83

The results for the individual pesticide were calculated from the area displayed by the integrator attached to the instrument and corrected according to the rates of recovery obtained in Table (3).

II - Study the effect of common heat treatment on the level of pesticide residues in raw buffalo's milk:

A-Collection of samples:

Three raw buffalo's milk samples (4 liter each) were collected from dairy shops in Zagazig city, Sharkia Governorate. The samples were kept frozen till the experimental procedure was carried out.

B- Experimental procedure:

The raw buffalo's milk sample (4 liter) was elevated at room temperature and thoroughly mixed to ensure equally distribution of fat globules. Ten ml were taken and prepared by extraction and cleaned up for pesticide residue analysis as mentioned before in experiment (1). The raw milk sample was divided into 3 equal portions (one liter each) and subjected to heat treatment as the following:

1- Laboratory pasteurization: the 1st portion was heated at 62.8 °C for 30 minutes in thermostatically water bath and then immediately cooled.

2- Boiling: the 2nd portion was heated with stirring to boiling point for 5 minutes and then cooled.

3- Sterilization: the 3rd portion was heated at 121°C for 15 minutes in an autoclave and then cooled.

After heat treatment (pasteurization, boiling and sterilization), 10 ml were taken from each portion and prepared by extraction and cleaned up for pesticide residue analysis as previously mentioned in experiment (1).

III - Study the effect of butter processing by heat treatment (ghee manufacture) on pesticide residues.

1-Collection of samples:

From previously collected butter samples, 3 samples which had the highest concentration of pesticide residues were selected for this experiment.

2-Experimental procedure:

The butter samples (750 gm each) were elevated to room temperature. Each sample was melted gradually by heating in stain steel pen. Temperature was raised gradually till having characteristic turbid foams on

the surface. Finally, the temperature was raised again till formation clear supernatant layer, bring yellow color of solid not fat at the bottom (morta) and appearance the characteristic odor of ghee. The clear supernatant layer was drained carefully into special container and kept in refrigerator till analysis was carried out.

Three gm of each ghee samples were extracted and prepared using the same procedure employed for fresh butter as mentioned in experiment

RESULTS AND DISCUSSION

1- Organochlorine pesticide residues in raw buffalo's milk

Results, Table (4) showed that p,p'-DDE was the only detected organochlorine pesticide residues in all examined raw buffalo's milk, with a concentration varies between 0.899 to 4.714, with a mean value of 2.7298±0.1126 ppb on milk basis. These results are rather similar with those recorded by Acqua *et al*. (1982) and Losada *et al.* (1996). Higher residue levels of p,p'-DDE were reported by Dogheim *et al*. (1988).; Ali *et al*. (1993).; Abou-Zeid (1994).; Ejobi *et al*. (1996).; Abou-Arab (1997).; El Afi *et al*. (1997).; Barkatina *et al.* (1999).; El-Marsafy *et al*. (1999) and Pardio *et al*. (2003).

Table (4): Organochlorine pesticide residues (ppb on milk basis) in all examined raw buffalo's milk samples collected from various districts at Sharkia Governorate.

Organochlorine pesticide residues	No. of examined samples	Maximum	Minimum	Mean±S.E.
p,p-'DDE	72	4.714	0.899	2.729±0.1126

it was also noticed that p,p'-DDE was the only residue detected in all examined raw buffalo's milk samples, with no other organochlorine residues detected in the same samples. Similar results were obtained by also reported on bufflo's milk El-Marsafy *et al*. (1999).

Ahmed (1991) reported that for more than thirty years, DDT was extensively used for pest control and public health. More than 13000 metric tones of the active ingredient of DDT have been sprayed all over Egypt, causing significant contamination of many environmental segments

DDT was officially banned in Egypt in the early 1980s, however Kelthane, one of DDT derivatives that contains residues of DDT and DDE was still in use for some years, causing some notable contamination (Camoni *et al*.,1983).

Animals are exposed to pesticide pollution from the environment and ingestion of contaminated feed and water (Abu-Zahw *et al*., 1993). DDT in feed and water of the cow is converted into DDD by the action of microflora in the rumen. In animal tissues DDD is further metabolized to DDE which is more lipid soluble than DDT or DDD (Roos and Tuinstra 1991).

Levels of organochlorine compounds detected in buffalo's milk are shown in table (5

Table (5): Organochlorine pesticide residues (ppb on milk basis) in examined raw buffalo's milk samples in relation to different districts at Sharkia Governorate

Districts	No. of examined samples	p,p'-DDE		
		Maximum	Minimum	Mean ± S.E.
Zagazig	24	4.714	0.899	1.215±0.101
Abo-Hammad	24	4.021	0.990	2.501±0.367
Fakous	24	4.557	0.998	1.634±0.203

The results indicates that all examined raw buffalo's milk samples having p,p'-DDE residues, but the values in all examined samples were within the permissible limit (20 p.p.m. of total DDT and its derivatives) recommended by Codex Alimentarius Commission (2004). These results agreed with those obtained by Ahmed (1991), Abou-Zeid (1994).; Abou-Arab (1997) .; Mallatou *et al*. (1997).; Barkatina *et al.* (1999).; El-Marsafy *et al*. (1999) .; and Pandit *et al*. (2002) who reported that the p,p'-DDE residues were detected in raw milk samples with a concentration within the permissible limit. On the other hand, Puchwein *et al.* (1990) .; Awasthi and Ahuja (1995).; Wong and Lee (1997) and Dabiza *et al*. (1999) reported p,p-DDE residues above the permissible limit.

Although the p,p'-DDE in examined raw buffalo's milk did not exceed the permissible limit, the presence of such compound in buffalo's milk may act as a source of hazard for human health.

2- **Organochlorine pesticide residues in yoghurt**

Resides of organochlorine detected in yoghurt samples are shown in Table (6) .Results showed that p,p'-DDE was the only detected compound, with concentrations ranging between 0.887 to 2.860 with a mean value of 1.9161±0.0992 ppb. These concentrations are lower than those obtained by Kandil *et al*. (1987).; Ali *et al*. (1993).; Abou-Zeid (1994) and El-Marsafy *et al*. (1999). On the other hand, El-Hoshy (1997) reported that the processing of milk into yoghurt has a significant role in reducing of the insecticide residues in milk.

Table (7) would alsos show the various concentrations of organochlorines detected in various counties included in the study.

Table (6): Organochlorine pesticide residues in all examined yoghurt and Kareish Cheese samples (ppb) collected from different districts at Sharkia Governorate

Milk products	No. of examined samples	p,p-'DDE		
		Maximum	Minimum	Mean ± S.E.
Yoghurt	24	2.860	0.887	1.9161±0.0992
Kareish cheese	24	N.D.	N.D.	N.D.

N.D: Not detected

Table (7): Organochlorine pesticide residues in examined yoghurt samples (ppb) in relation to various districts at Sharkia Governorate

Districts	No. of examined samples	p,p'-DDE		
		Max.	Min.	Mean ± S.E.
Zagazig	8	2.201	0.887	1.594±0.112
Abo- Hammad	8	2.860	1.897	2.373±0.102
Fakous	8	2.313	1.373	1.987±0.118

Results revealed that all examined yoghurt samples were contaminated with p,p'-DDE, but the value were within the permissible limit (20 p.p.m. of total DDT and its derivatives) established by Codex Alimentarious Commission (2004). El-Marsafy *et al*. (1999) recorded that the only residues that can detected in yoghurt samples collected from Agricultural Research station of Kafer El Sheik, was p,p'-DDE with a concentration not exceeded the codex Alimentarious residue limits.

3-Organochlorine pesticide residues in kareish cheese

No residues of organochlorine were detected in cottage cheeseTable (6). This may be attributed to either absence or low fat percent in cottage cheese. Roose and Tuinstra (1991), recorded that most organochlorine insecticides are readily excreted in milk fat, because of their lipophilic nature and relatively stability.

On the other hand, El-Marsafy *et al.* (1999) reported that the average of p,p'-DDE residue amounts detected in cottage cheese were0.0109 mg/kg on fat content basis which less than that detected in raw, ripened and pasteurized milk samples and storage process caused a significant decreasing after 15 days of storage. Moreover Pietrino (1991) recorded that the microflora used in cheese making may degrade pesticide residues during cheese ripening.

4- Organochlorine pesticide residues in butter

Table (8) shows that the organochlorine pesticide residues detected in butter samples collected from various districts at Sharkia Governorate were lindane, heptachlor epoxide and p,p'-DDE.

Regarding lindane (gamma isomer of HCH) residues in butter samples, it clear that the residues were detected in butter samples collected from Abo-Hammad; Hehiaa; Abo-Kaber; Al-Husenia and El-Korine with a concentration of 10.91-10.10; 12.09-12.0; 13.02-12.88; 18.12-17.99 and 13.61-12.96 ppb on fat basis, respectively. On the other hand, lindane could not be detected in the examined butter samples collected from Zagazig, Fakous, Kafr-Sakr and Menia El-Kamh.

Table (8): Organochlorine pesticide residues(ppb on fat basis) in examined Butter samples collected from various districts at Sharkia Governorate

*Districts	Lindane	Heptachlor epoxide	p,p'-DDE
Zagazig	N.D. N.D.	N.D. N.D.	51.415 48.971
Abo-Hammad	10.91 10.10	17.05 16.90	65.08 64.899
Fakous	N.D. N.D.	N.D. N.D.	59.242 60.605
Hehiaa	12.09 12.00	N.D. N.D.	75.64 62.91
Abo-Kaber	13.02 12.875	N.D. N.D.	69.54 62.91
Kafr-Sakr	N.D. N.D.	N.D. N.D.	102.5 101.57
Al-Husenia	18.12 17.99	13.60 13.37	38.89 30.00
Al-Korin	13.61 12.96	N.D. N.D.	97.19 93.38
Menia El-Kamh	N.D. N.D.	N.D. N.D.	65.16 57.227
Maximum Minimum Mean ± Error	18.12 10.101 7.428±1.667	17.05 13.37 3.384±1.583	102.50 30.00 67.063±4.933

N.D. : Not detected
***2 samples were collected from each district**

In all examined butter samples, the maximum concentration of lindane was 18.12, while the minimum was 10.101 with a mean value of 7.428±1.667 ppb on fat basis. These results were nearly similar to those recorded by Noren *et al.* (1982). Higher lindane value were recorded by Abou-Zeid (1994).; Neamat-Allah (1998), Dabiza *et al*. (1999).; and Ayoub (2000). While, Pandit *et al.* (2002) reported lower concentration of lindane in examined butter samples.

Statistical analysis of different organochlorine pesticide residues in butter samples (Table 9) revealed that 10 (55.6%) out of 18 examined butter samples having lindane above the maximum residue limit (10 µg/kg on fat basis) as established by Codex Alimentarious Commission (2004). On the other aspect, the lindane were not detected in 44.4% of the examined butter samples. While butter samples having total HCH above the recommended tolerance level (0.1mg/kg) of FAO/WHO were reported by Neamat-Allah (1998).

Concerning the residues of heptachlor epoxide in butter samples, it is noticed that the residues were detected only in butter samples collected from Abo-Hammad and Al-Husenia with values of 17.05-16.90 and 13.60-13.37 ppb on fat basis, respectively. In other investigated districts the heptachlor epoxide could not be detected in butter samples.

It is found that the residues of heptachlor epoxide in all examined butter samples lie between 17.05 and 13.37 with an average of 3.384±1.583 ppb on fat basis. Abou-Zeid (1994).; and Waliszewski *et al*. (1997) detected

heptachlor epoxide in butter samples with higher values rather than that in our investigation.

Table (9): Frequency distribution of organochlorine pesticide residues in a total of 18 examined butter samples collected from various districts at Sharkia Governorate

Organochlorine Pesticide residues	M.R.L.* (ppb)	Not detected		Within P.L.		Over P.L.**	
		No.	%	No.	%	No.	%
Lindane	10	8	44.4	-	-	10	55.6
Heptachlor epoxide	6	14	77.8	-	-	4	22.2
p,p'-DDE	20	-	-	-	-	18	100

***M.R.L.: Maximum Residue Limit according to Codex Alimentarius Commission (2004)**
****P.L.: Permissible Limit**

Table (9) indicated that 4 (22.2%) out of 18 examined butter samples having heptachlor epoxide above the maximum residue limit (6 ppb on fat basis) as recommended by Codex Alimentarious Commission (2004). On the other hand, 14 (77.8%) of examined butter samples having no residues of heptachlor epoxide.

Regarding the residues of p,p'-DDE in butter samples, it is clear that all examined samples collected from different districts contained p,p'-DDE residues.

Estimation of p,p'-DDE in butter samples from different districts, revealed that the values were ranged from 30.0 ppb on fat basis (Al-Husenia district) to 102.50 ppb on fat basis (Kafr-Sakr district), with a mean of 67.063±4.933 ppb on fat basis. Nearly similar p,p'-DDE levels in butter samples were recorded by Takroo *et al*. (1985), Abou-Zeid (1994) and Ayoub (2000). On the other hand, higher p,p'-DDE levels were reported by Abu-Zahw *et al.* (1993), Neamat-Allah (1998) and Dabiza *et al*. (1999). Lower p,p'-DDE values were obtained by Noren *et al*. (1982).; Waliszewski *et al*. (1997).; and Pandit *et al.* (2002) .

It is shown from Table (9) that all examined butter samples (100%) having p,p'-DDE above the maximum residue limit (20 ppb on fat basis) as recommended by Codex Alimentarious Commission (2004). These results substantiate what has been reported by Abu-Zahw *et al.* (1993) who recorded that p,p'-DDE in all local butter samples were exceeded maximum residue limit (0.05 ppm) that established by Codex Alimentarious Commission (1992).

From a forementioned results, it noticed that butter samples having lindane, heptachlor epoxide and p,p'-DDE above the permissible limits.

From public health point of view, the chronic toxicity and sub lethal exposure of lindane and other HCH isomers produce liver tumors (IARC, 1974). Some authors (Unger and Olsen, 1980; Lopez *et al*., 1996.; and Jaga and Duvvi, 2001) evaluated the possibility of cancer occurring in humans from DDT exposure. On the other hand, Hayes (1994) recorded that heptachlor are significantly more toxic than DDT.

So, animal studies recorded by Jaga and Brosius (1999) showed a significant association between DDT administration and lymphoma, respiratory cancer, liver cancer and estrogenic effects on mammary tissue.

Therefore, with the persistence of DDT and DDE in the environment, the potential risk to the health of man, animals and the environment remains.

5-Effect of heat treatment on organochlorine pesticide residues

A-Effect of common heat treatment on organochlorine pesticide residues in raw buffalo's milk:

This experiment was conducted to determine the effect of common heat treatment (pasteurization, boiling and sterilization) on organochlorine pesticide residues in raw buffalo's milk.

Table (10) revealed that the p,p'-DDE was the only detected organochlorine pesticide residues in tested milk with a mean concentration of 3.21 ppb on milk basis. The milk was treated by pasteurization, boiling and sterilization, and the mean concentration of p,p'-DDE residues became2.509, 0.932 and 0.2599 ppb on milk basis, respectively with degradation percent of 22.13, 71.16% and 92.8, respectively.

Table (10): Effect of common heat treatment on organochlorine pesticide residues content of raw buffalo's milk

Sample number	p,p'-DDE						
	Concentration in raw milk	Pasteurization		Boiling		Sterilization	
		Concentration (ppb on milk basis)	Degradation %	Concentration (ppb on milk basis)	Degradation %	Concentration (ppb on milk basis)	Degradation %
1	2.980	2.332	21.75	0.864	71	0.238	92
2	3.88	3.109	19.88	1.1854	69.45	0.367	90
3	2.771	2.085	24.75	0.747	73.03	0.175	93.7
Mean	3.210	2.509	22.13	0.932	71.16	0.2599	92.8

From the previously mentioned results, it was noticed that the sterilization of milk had an extensive effect on degradation of p,p'-DDE in raw buffalo's milk followed by boiling and pasteurization. These results are agreement with those findings obtained by Hassan (1987).; Abd-Rabo *et al*. (1989_a).; and Abou-Zied (1994) who concluded that sterilization of milk is the most effective method for reduction of p,p'-DDE residues in milk followed by boiling and finally pasteurization. Moreover, El-Hoshy (1997) recorded that the reduction of p,p'-DDE residues in milk as the result of pasteurization was 21.11, while it was 70.35 due to boiling.

It was concluded that sterilization of raw milk is the most effective method for reduction of pesticide residues and consequently render the milk safe for human consumption.

B- Effect of butter processing by heat treatment (ghee manufacture) on organochlorine pesticide residues.

Table (11) showed the effect of ghee making on organochlorine pesticide residues content of butter. The analysis of butter samples before processing revealed that the detected residues were lindane, heptachlor epoxide and p,p'-DDE with a mean concentration of 14.213, 15.325 and

65.72 ppb on fat basis. After heat treatment for making ghee, the mean concentration of lindane, heptachlor epoxide and p,p'-DDE in produced ghee were 2.948, 1.935 and 15.609 with degradation percent of 79.1, 87.50 and 76.73, respectively.

Table (11): Effect of butter processing by heat treatment (ghee manufacture) on organochlorine pesticide residues

Sample number	Organochlorine pesticide residues								
	Lindane			Heptachlor epoxide			p,p'-DDE		
	Butter (ppb on fatbasis)	Ghee (ppb on fat basis)	Degradation %	Butter (ppb on fatbasis)	Ghee (ppb on fatbasis)	Degradation %	Butter (ppb on fatbasis)	Ghee(ppb on fat basis)	Degradation %
1	18.12	3.823	78.9	13.60	1.537	88.7	34.89	6.769	80.6
2	10.91	2.586	76.3	17.05	2.33	36.3	65.08	18.093	72.2
3	13.61	2.436	82.1	N.D.*	N.D.	N.D.	97.19	21.965	77.4
Mean	14.213	2.948	79.1	15.325	1.935	87.50	65.72	15.609	76.73

***N.D.: Not detected**

From the obtained results, it is found that butter making processes for production of ghee led to eliminate large quantities of organochlorine pesticide residues. Nearly similar results were obtained by Ayoub (2000) who recorded that heat processing of butter led to reduction of lindane and heptachlor and its epoxide with percentage of 75 and 88, respectively. Also, Zidan *et al*.(1994) reported that, in final product (samna) after boiling off method, the reduction percentage of lindane was 92.8. Moreover, Neamat-Allah (1998) recorded that more than 50% of added lindane to butter could be degraded or removed during processing into samna due to the effect of heat treatment.

Meanwhile, low degradation percentage was obtained by Abu-Zahw *et al*. (1993), the authors subjected butter samples to heat treatment to produce ghee and found that the loss percent of p,p'-DDE in cow and buffalo ghee were varied from 9.21 to 46.97. On the other hand, the reduction of organochlorine pesticide residues by butter boiling-off method can be attributed to heat degradation of pesticide residues and also partial transfer of pesticide residues into morta (Zidan *et al*., 1994 and Neamat-Allah, 1998).

In conclusion the butter processing by heat treatment (boiling-off method) yielded ghee (samna) with marked decrease in organochlorine pesticide residue content. Therefore, the produced ghee contained organochlorine residues within the permissible limit and render ghee (samna) safe for human consumption.

REFERENCES

Abd El-Kader, M.A.; Abu-Zahw, M.M.; Tork, I.Y. and Ayoub, M. A. (1994): Organochlorine pesticide residues in milk and some milk products. Egypt. J. Appl. Sci. 9 (9): 267-273.

Abd-Rabo, F.H.; Abou-Dawood, A.E.; Ahmed, N.S. and Hassan, F. A.M. (1989): Heat treatments change amounts of pesticide residues in buffalo's milk. Egyptian J. of Dairy Sci. 17 (2): 349-357.

Abou-Arab, A.A.K. (1999): Effect of processing and storage of dairy products on lindane residues and metabolites. Food Chemistry. 64 (4): 467-473.

Abou-Arab, A.A.K. (1997): Effect of Ras cheese manufacturing on the stability of DDT and its metabolites. Food Chemistry. 59 (1): 115-119.

Abou-Zeid, A.M.A. (1994): Studies on pesticide residues in milk and some dairy products. Ph.D. Thesis, Faculty of Vet. Med. , Cairo University, Egypt.

Abu-Zahw, M.M.; Abd El-Kader, M.A.; Hegazy, M.E.A. and Mansour, M.A. (1993): Pesticide residues in butter and ghee. Bull. of Suez Canal University Appl., Sci., 11 (1): 40-48.

Acqua, A.Del; Ribeiro, M.L.; Trevisan, L.M.V. and Cerqueira, M. (1982): Rapid and economic determination of organochlorine pesticides in milk. Ecletica Quimica, 7: 49- 54.

Ahmed, M.T. (1991): High performance liquid chromatography determination of DDT residues in some commercial brands of milk in Ismailia, Egypt. Z. gesamte Hyg. 37. Heft 3.

Ali, A.A.; Metwally, M.M.; Ahmed, N.S. and Anb, A.K. (1993): Pesticide residues in milk and their distribution among dairy products. Egyptian J. of Dairy Science. 21 (2): 291-299.

Aman, I.M. and Bluthgen, A. (1997): Occurrence of residues of organochlorine pesticides and polychlorinated biphenyls in milk and dairy products from Egypt. Milchwissenschaft. 52 (7): 394-399.

Awasthi, M.D. and Ahuja, A.K. (1995): Organochlorine insecticide residues in bovine milk and commercial baby milk powder. J. of Food Science and Technology. 32 (5): 420-422.

Ayoub, M.M.M. (2000): Monitoring of some organochlorine in some fatty foods. Ph. D. thesis, Department of agricultural Sci. Institute of Environmental Studies and Research. Ain-Shams University, Egypt.

Barkatina, E.N.; Pertsovsky, A.L.; Murokh, V.I.; Kolomits, N.D.; Shulyakovskaya, O.V.; Venger, O.N. and Makarevich, V.I. (1999): Organochlorine pesticide residues in basic food products and diets in the republic of Belarus. Bull. Environ. Contam. Toxicology. 63: 235-242.

Camoni, I.; Di Muccio, A.; Pontecorvo, D. and Citti, P. (1983): Study of p,p'-DDE impurities in commercial formulations based on dicofol (Kelthane). Nuvi Ann. Ig Microbial 33: 241-247.

Codex Alimentarius Commission (1992): Guide to codex Maximum limits for Pesticide Residues. Part (2) FAO, Rome, Italy.

Codex Alimentarius Commission (2004): Codex committee on pesticide residues. Thirty-Sixth Session. New Delhi, India.

Corneliussen, P.E.; McCully, K. A. ; McMahon, B. and Newsome, W.H. (1984): Pesticide and industrial chemical residues. A.O.A.C. official methods of analysis: 533-562.

Costabeber, I.; Trindade, R. and Fries, L.M. (2001): Organochlorine pesticides levels in cow milk. Alimentaria. 38 (319): 127-129.

Dabiza, N.M.A.; El-Senaity, M.H. and El-Dein, M.M.Z. (1999): Pesticide residues in some market dairy products and distribution of malathion in butter, ghee and Domiati cheese during manufacture. Egypt, J. of Dairy Sci. 27 (2): 345-357.

Dogheim, S.M.; Almaz, M.M.; Kostandi, S.N. and Hegazy, M.E. (1988): Pesticide residues in milk and fish samples collected from Upper Egypt, J. Association Official Analytical chemists. 71 (5): 872-874.

Dogheim, S.M.; Nasr, E.N.; Almaz, M.M. and El-Tohamy, M.M. (1990): Pesticide residues in milk and fish samples collected from two Egyptian Governorates. J. Association of Official Analytical Chemists. 73 (1): 19-21.

El-Afi, A.; Rahmani, M. and Abdennebi, E. H. (1997): Contamination of commercial milk by organochlorine pesticides in Libya. Assiut Veterinary Medical J. 36 (72): 306-312.

El-Hoshy, S.M. (1997): Insecticide residues in milk and influence of heat treatment and bacterial fermentation as safeguard against these pollutants. Assiut Vet. Med. J. 73 (37): 141-155.

El-Marsafy, A.M.; Alla, S.A.G.; Zidan, M.A.; Fahmy, S.M. and Dogheim, S.M. (1999): Monitoring of organochlorine pesticide residues in milk products. Egyptian J. of Agricultural Research. 77 (1): 205-216.

Ejobi, F.; Kanja, L.W.; Kyule, M.N.; Muller, P.; Kruger, J.; Nyeko, J.H.P. and Latigo, A.A.R. (1996): Organochlorine pesticide residues in cow's milk in Uganda. Bull. Environ. Contam. Toxicol. 56: 551-557.

Hassan, F.A.M. (1987): Effect of different processing on pesticide residues in milk. M.V.Sc. Thesis, National Research Centre, Dokki, Cairo, Egypt.

Hayes, A.W. (1994): Principles and methods of Toxicology. 3rd Ed. Raven Press, Ltd., New York ©.

IARC (1974): Monograph on the evaluation of carcinogenic risk of chemicals to man. 5, some organochlorine pesticides. International Agency for Research on Cancer (IARC), lyon, France.

Jaga, K. and Brosius, D. (1999): Pesticide exposure: human cancers on the horizon. Rev. Environ. Health. Jan.-Mar.; 14 (1): 39-50.

Jaga, K. and Duvvi, H. (2001): Risk reduction for DDT toxicity and carcinogenesis through dietary modification. J. R. Soc. Health Jun.; 121 (2): 107-13.

Juntunen, L.A.; Bidleman, T.F.; Harner, T. and Parkhurst, W.J. (2000): Toxaphene and other organochlorine pesticides in Alabama air. Environmental Sci. and Technology, 34: 5097-5105.

Kandil, M.A.; Hegazi, M.E. and EL-Sayed, M.A. (1987): Monitoring of pesticide in yoghurt. Egypt. J. Appl. Sci. Third Issue December, 41-46.

Lopez, Carrillo, L.; Torres, Arreola, L.; Torres, Sanchez, L.; Espinosa, Torres, F.; Jimenez, C.; Cebrian, M.; Walliszewski, S. and Saldate, O. (1996): Is DDT use a public health problem in Mexico? Environmental Health Perspectives.104 (6): 584-588.

Losada, A.; Fernandez, N.; Diez, M. J.; Teran, M. T.; Garcia, J. J. and Sierra, M. (1996): Organochlorine pesticide residues in bovine milk from Leon (Spain). Sci. of the Total Environ. 181 (2): 133-135.

Mallatou, H.; Pappas, C. P.; Kondyli, E. and Albanis, T. A. (1997): Pesticide residues in milk and cheeses from Greece. Sci. of the Total Environ. 196 (2): 111-117.

Mukherjee, I. and Gopal, M. (1993): Organochlorine pesticide residues in dairy milk in and around Delhi. J. of the Association of Analytical Chemistry International. 72: 283-286.

Neamat-Allah, A.A. (1998): Pesticides residues in butter from El-Beharah districts in Egypt and their stability during samna (ghee) processing. Annals of Agric. Moshtohor, 36 (3): 1597-1604.

Noren, K.; Linder, C.E. and Vaz, R. (1982): Levels of organochlorine pesticides and PCB's in Swedish butter, 1971-1976.

Oestreicher, M.L.; Shuman, D.H. and Wurster, C.F.(1971): DDE residues medullary bone formation on birds. Nature (London) 229:571.

Pandit, G.G.; Sharma, S.; Srivastava, P. K. and Sahu, S. K. (2002): Persistent organochlorine pesticide residues in milk and dairy products in India. Food Addit. Contam. Feb.; 19 (2): 153-157.

Pardio, V.T.; Waliszewski, K.N.; Landin, L.A. and Bautista, R.G. (2003): Organochlorine pesticide residues in cow's milk from a tropical region of Mexico. Food Addit. Contam. Mar; 20 (3): 259-269.

Pietrino, D. (1991): Pesticide residues and milk processing. Industria del Latte. 27 (2): 3-32.

Puchwein, G.; Eibeihuber, A.; Brodacz, W.; Muller, J.; Golles, J. and Unger, M. (1990): Pesticide residues in raw milk. Results of a study covering all of Austria, 1985-1988. Bodenkultur 41 (2): 153-175.

Ragab, A.A.; Abd El-Latif, M.S.; Ibrahim, A.M.A. and Smith, C.J. (1996): Indirect competitive ELISA for the quantitative and qualitative analysis of aldrin/dieldrin in Egyptian milk samples from different farm animal species. Food and Agricultural Immunology. 8 (4): 273-278.

Riva, C. Dela and Anadon, A. (1991): Organochlorine pesticides in cow's milk from agricultural region in northwestern Spain. Bull. Environ. Contam. Toxicology. 46: 527-533.

Roos, A.H. and Tuinstra, L.G.M. Th. (1991): Chlorinated insecticides. Monograph on residues and contaminants in milk and milk products. Chapter 5, p. 84. International Dairy Federation, Belgium.

Sandhu, T. S. (1992): Pesticides in food. 3rd World Congress. Food Born Infection and Toxications, 16-19 June Berlin.

Suzuki, T.; Ishikawa, K.; Sato, N. and Sakai K.I. (1979): Determination of chlorinated pesticide residues in food. 1- Rapid screening method for chlorinated pesticides in milk. J. Assoc. Off. Anal. Chem. 62: 681-684.

Takroo, R.; Kaphalia, B.S. and Seth, T.D. (1985): Chlorinated pesticide residues in different brands of butter. J. of Food Science and Technology, India. 22 (1): 57-59.

Unger, M. and Olsen, J. (1980): Organochlorine compounds in the adipose tissue of deceased people with and without cancer. Environmental Research 23: 257-263.

Vreman, K.; Tuinstra, L.G.M.T.; Van Den Hoek, J.; Be Visser, H. and Westerhuis, J. H. (1977): Neth. J. Agric. Sci. 25: 303. (Cited after Aman and Bluthgen, 1997)

Waliszewski, S.M.; Pardio, V.T.; Chantiri, J.N.; Rivera, J. and Aguirre, A.A. (1997): Organochlorine pesticide residues in cow's milk and butter in Mexico. Science of the total Environment 208: 127-132.

Waliszewski, S.M.; Villalobos-Pietrini, R.; Gomez-Arroyo, S. and Infanzon, R.M. (2003): Persistent organochlorine pesticide levels in cow's milk samples from tropical regions of Mexico. Food Addit. Contam. Mar.; 20 (3): 270-275.

Wong Siu-kay and Lee Wai-on (1997): Survey of organochlorine pesticides in milk in Hong Kong (1993-1995). J. of AOAC International 80 (6) 1332-1335.

Zapata Moran, A.L.; Santamaria Rios, M.M.; Alvarez Iris, M.; Salazar Vanegas, S. and Muller, U. (1996): Organochlorine pesticide residues in cow milk in Nicaragua. Boletin de la Oficina Sanitaria Panamericana. 120 (6): 483-490.

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

Zidan, Z.H.; Fayed, A.E.; Abou-Arab, A.A. and El-Nockrashy, S.A. (1994): Monitoring some contaminants during the manufacture of some conventional dairy products. Egypt. J. of dairy Sci. 22 (1): 19-29.

اكتشاف متبقيات مبيدات الافات فى اللبن و بعض منتجات الالبان

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أجريت الدراسة بهدف رصد وتقدير بقايا المبيدات الكلورونية العضوية في عينات من لبن الجاموس الخام وبعض منتجات الألبان و التي تم جمعها من مناطق مختلفة من محافظة الشرقية، مصر. وقد شملت الدراسة أيضا تأثير المعالجة الحرارية على مستويات هذه المبيدات في لبن الجاموس و الزبدة. وقد أظهرت النتائج أن مشابه الـ pp'-DDE هو الوحيد الذي تم التعرف عليه في جميع عينات اللبن التي تم فحصها. كما تم الكشف عن المشابه ال pp'-DDE في عينات الزبادي 0,1126 ± 2,7298 جزء من البليون، مع قيمة متوسط 0,0992±1,9161 جزء من البليون، في حين لم يتم رصد بقايا المبيدات الكلورونية في عينات الجبن. و في عينات الزبده كانت بقايا المبيدات الكلورية العضوية التي رصدت هي اللينـدين، وسباعي كلوروإيبوكسيد و pp'-DDE وكان متوسط قيم التركيزات المقدره هي 1,667±7,428 و 1,583±3,3841 و4,933±67,063 جزء من البليون على التوالي على أساس الدهون. وقد أظهرت نتائج دراسة تأثير المعالجة الحرارية بواسطة البسترة والغليان، والتعقيم على مستوى مشابه الـ pp'-DDE و التي تم رصدها في لبن الجاموس حيث كانت 2,509، 0,932 و 0,2599 جزء من البليون على التوالي على أساس حليب. بنسبة تدهور 71,16، 22,13 و 92,8٪ على التوالي. في حين كان تاثير المعاملة الحرارية لعينات الزبدة لإنتاج السمن على مستويات كل من اللينـدين، وسباعي الكلور إيبوكسيد وpp'-DDE هو الكشف عن مستويات لهذه المركبات 2,948، 1,935 و 15,609 على أساس جزء من البليون من الدهون على التوالي بنسبة تدهور79,1 و 87,50 و 76,76٪ على التوالي. و قد استخلصت الدراسة أن تعقيم الحليب الخام هو الأسلوب الأكثر فاعلية للحد من بقايا المبيدات الكلورونية العضوية كما أن المعالجة الحرارية للزبد لانتاج السمنة أدت إلى انخفاض ملحوظ في محتواها من بقايا المبيدات الكلورونية العضوية.

قام بتحكيم البحث

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