j.Egypt.net.med.Assoc 77, no 2. 277 – 296 (2017)

ESTIMATION OF MICROBIAL HAZARD AND HARMFUL RESIDUES IN TABLE EGGS

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ABSTRACT

A total of fifty egg samples (25 of each of baladi and farm eggs) were collected from different markets in Cairo and Giza Governorates to assess their microbiological status and detection of antibiotic, pesticide and estradiol residues. The mean values of Aerobic Plate Count in baladi and farm eggs were 3.1×10^2 $\pm5.8\times10$ and 2×10^2 $\pm6.5\times10$ cfu/g respectively, which were within the limit recommended by EOS, 3169/2007 (2.5×10^4 cfu/g). The mean value of *E. coli* count in baladi eggs were 3×10 $\pm0.79\times10$ cfu/g which was higher than the limit recommended by EOS, 3169/2007 (10 cfu/g), while it has not been inferred in farm eggs. *Salmonella* species, *Shigella* and *Clostridum perfrengens* could not be detected in all examined samples. The mean values of yeast and mould count in baladi and farm eggs were 1.5×10 $\pm0.45\times10$ and 3.5×10 $\pm0.78\times10$ cfu/g respectively, which were compatible with the limit recommended by EOS, 3169/2007 (5×10 cfu/g). The presence of antibiotic residues in egg samples were detected qualitatively by using modified four plate test (FPT). Pesticide residues were determined with Agilent gas chromatograph (GC). Estradiol hormone levels were determined wit Rida screen ELISA kit for tissue. The incidence of antibiotic residues in baladi and farm eggs were 2 (8%) and 10 (40%), respectively. The mean concentration of heptachlor epoxide in baladi and farm eggs was 0.085±0.02 ppm, and was present in a percent of 8 (32%), while it could not be detected in examined farm eggs, above the MRL cited by Codex (2016) (0.05 mg/kg). Endosulfan couldn't detected in farm eggs and was present in baladi eggs in a concentration of 0.105±0.003 ppm and incidence of 3(12%), above the MRL cited by Codex (2016) (0.03 mg/kg). Endrine couldn't be detected in baladi eggs and its mean value was 0.28±0.009 ppm in farm eggs and was present in a percent of 6(24%) above MRL cited by J F C R F (2016) (0.005 mg/kg). Dialdren not detected in baladi eggs but found in farm eggs in a percent of 4(16%) with mean concentration of 0.25±0.03 ppm which above MRL recommended by Codex (2016) (0.1 mg/kg). Methoxychlor was present in both baladi and farm eggs in a concentration of 0.037±0.001 and 0.016±0.002 ppm with percent of 11(44%)

and 6(24%) respectively, above MRL cited by J F C R F (2016) (0.01 mg/kg). Alfa-BHC, Gama BHC, Delta BHC, heptachlor, Aldrin, endosulfan 11, endosulfan sulfate, p,p-DDT, p,p-DDD, p,p -DDE and endrin aldehyde as well as organophosphorus pesticides couldn't be detected in both baladi and farm eggs. The mean value of estradiol in baladi and farm egg samples which were 0.030 ±0.001, 0.400±0.120 µg/kg respectively, within the permissible limit (1ppb) stated by Gracey (1986). All baladi egg samples are within acceptable daily intake (ADI) and 72% of farm eggs are within the acceptable daily intake (ADI) value for estradiol of 0.05 µg/kg body weight /day as assessed by **JECFA (1999).** The public health significance of the isolated organisms and harmful residues as well as recommended hygienic measures were discussed to ensure quality of eggs to safeguard the consumers.

Key words:

Eggs, microbial contamination, organochlorine pesticides and hormonal residues.

INTRODUCTION

From ancient times, eggs constitute an important part of human diets worldwide **(Musgrove *et al*., 2005)**. Egg is considered as a nutritionally complete food and an excellent source of protein, most eggs (90%) have been found to be sterile when laid, but they have the potential to become occasionally contaminated **(Ruxton *et al*., 2010)**. Freshly laid eggs following exposure to environmental conditions such as soil, dust and dirty nesting materials become contaminated with different types of microorganisms either by penetration through pores of the shells or through the transovarian route **(Smith *et al*., 2000 and Theron *et al*., 2003).** Eggs can be contaminated with micro-organisms such as bacteria and fungus. These microorganisms can evade the defense mechanism of eggs and penetrate inside the egg, thus increasing the risk of food borne illnesses or product spoilage **(Tan *et al*., 2012).** Food poisoning and food borne infection following consumption of eggs or dishes containing eggs are usually caused by *Salmonella*, as well as *Staphylococcus aureus*, *Escherichia coli* and other coli bacilli **(Przybylska, 2003).** Antimicrobials are used by the poultry industry to enhance growth and feed efficiency and to reduce bacterial diseases **(Donoghue, 2003).** In laying hens, antimicrobials are used to treat and to prevent bacterial infections. Antimicrobial classes used to treat poultry are similar to those used in human medicine which included aminoglycosides, tetracyclines, beta-lactams, quinolones, macrolides, polypeptides, amphenicols and sulphonamides**(Stolker and Brinkman,2005).**Organophosphorus pesticides (OPPs) are less persistent than organochlorine (OCPs), and frequently considered the

preferred choice for treatment because they provide efficacious, safe and cost effective control of a wide range of pests. The awareness that OPPs may also concentrate along the food chain has led to the establishment of low maximum residue limits (MRLs) in food, as set by European Union (UE). Consequently, this makes necessary for the control of this type of compounds in fatty matrices **(Gonzalez and Plaza 2006).** These compounds (OCPs and OPPs) are known of inducing or aggravating certain health problems in humans such as cancer, immune systems suppression and the disruption of hormonal functions. **(Vincenzo *et al*., 2006).** The endogenous estradiol in mature eggs probably results from diffusion of the steroid from follicular cells (steroid producing cells) to the ooplasm and yolk of preovulatory oocytes during oogenesis. The concentration of endogenous estradiol in ovulated eggs is lower than in the fluid of the ovarian cavity and in the intrafollicular oocytes. The estradiol content of fertilized eggs is also affected by concentrations of exogenous estradiol in the surrounding medium. In steelhead trout, it has been also suggested that endogenous steroid metabolism of maternally contributed sex steroid is active during the early stages of embryo development. The low estradiol concentration of ovulated eggs in ovarian fluid and the decline in E2 of eggs during incubation may be brought about not only by diffusion into the medium but also by metabolic degradation of the steroid by the egg itself **(Yoeh *et al*., 1996).** Therefore, the aim of this study was planned to assess the microbial hazards,organochlorine and organophosphorus pesticide and estradiol residues in both baladi and farm eggs.

MATERIAL AND METHODS

Collection of samples:

Fifty random samples of both baladi and farm chicken eggs (25 of each) were collected from different localities in Cairo and Giza Governorates. Samples were collected separately in sterile plastic bags, labeled and put in ice backed and transferred to the laboratory and subjected to the next examinations.

1-Microbiological examination:

1.1 Preparation of sample: (ISO 6887- 4: 2011).

The surface of each egg sample was cleaned using sterile cotton soaked in 70% (v/v) alcohol. A small opening was made at the tip of the egg using a sterile forceps. The content was then drained out carefully through the pore and transferred into a sterile beaker. Twenty five grams from each sample were aseptically placed in a sterile blender with 225 ml of peptone water

(1%) and homogenized for two minutes then serial dilution were prepared in sterile peptone water (1%) for decimal dilution up to 10^{-6} .

1.2 Aerobic Plate Count (APC): According to **APHA, (2001)** by using standard plate count agar medium.

1.3 Determination of *Escherichia coli* count: According to **(ISO 16649-2:2012)** by using Tryptose-Bile-glugorinide agar (TBX).

1.4 Yeasts and moulds count: According to (**ISO 21527-1:2008)** by using Dichloran Rose Bengal Chloramphenicol agar.

1.5 Isolation and identification of Salmonella: using standard method **(ISO 6579: 2012).**

1.6 Isolation and identification of Shigella: by using Xylose lysine Deoxycholate agar (XLD) **(ISO 21567: 2015).**

1.6 Isolation and identification of *Clostridium perfringens*: using Tryptose Sulfite Cycloserine medium (TSC) **(ISO 7937:2015).**

2- Detection of antibiotic residues: (Ehsani and Hashemi, 2015):

2.1 Preparation of samples: (AOAC, 2000).

The surface of each egg sample was cleaned using sterile cotton soaked in 70% (v/v) alcohol. A small opening was made at the tip of the egg using a sterile forceps. The content was then drained out carefully through the pore and transferred into a sterile beaker. Ten ml of phosphate buffer (pH 7) was added to 2 ml of homogenized egg. Using a sterile forceps, paper discs were dipped into homogenate, allowed to soak and drained from the discs before placing into the Petri dish containing organisms.

2.2 Detection of antibiotic residues:

The presence of antibiotic residues in egg samples were detected qualitatively by using modified four plate test (FPT). The bacteria used in FPT were *Bacillus subtils* (pH 6.0 and 8.0) and *Micrococcus luteus* that have been prepared from Department of Microbiology, Animal Health Research Institute. An overnight culture of the tested organisms in 10 ml of nutrient broth were used to inoculate plates in a concentration of 3×10^8 cfu/ml. Blank filter paper discs 0.6 cm in diameter were sterilized and used for inoculating samples and controls onto plates. After application of the test and control discs, plates were incubated at 37 ºC for 18-20 h and then investigated for the presence of inhibition zones (at least 2 mm in width) of test organism around the test and control discs.

3- Determination of pesticide residues according to Le Doux (2011):

3.1 Extraction of pesticides residues from eggs:

Three grams of egg content were homogenized with 5 g anhydrous sodium sulfate till fine homogenate was obtained. The mixtures were Soxhlet extracted for 12 h with 100 ml of acetone and dichloromethane (2:8 v/v). Extracts were processed in a rotary evaporator to remove acetone and dichloromethane before addition of 10 ml n-hexane and a second evaporation to approximately 3 ml. Extracts were transferred to a 250 ml separating funnel and extracted twice with 30 ml n-hexane saturated acetonitrile each time. Combined extracts were transferred to a 500 ml separating funnel. After the addition of 300 ml 5% sodium sulfate, solutions were extracted twice with 30 ml n-hexane each time. The combined n-hexane extracts were evaporated in the rotary evaporator to approximately 1 ml. Cleaned up extracts by transferring to a column packed with 10 g florisil (60 - 100 mesh) topped with one g anhydrous sodium sulfate and eluted with 25 ml n-hexane (discarded) and a 50 ml mixture of dichloromethane and n-hexane (2:3 v/v) in sequence at a rate of 1–2 ml/min.

3.2 Determination of pesticide residues:

The Agilent GC (6890), equipped with Ni^{63}-electron capture detector (ECD) was use for the chromatographic separation and was achieved by using DB-17 (J and W Scientific) capillary column (30m length x 0.32 mm internal diameter × 0.25 µm film thickness), carrier gas: N_2 at a constant flow rate of 4 ml/min. The injector and detector temperature were programmed at 300˚C and 320˚C, respectively. The initial column temperature was 160˚C for 2 min, raised at 5˚C/min, and then held at 260˚C for 10 minutes. The retention time, peak area and peak height of the sample was compared with those of the standards for quantization. The quantification limits obtained by GC with ECD have been reported to be mostly around 0.1-20 ng/g.

4- Determination of estradiol residue by ELISA method according to Mahgoub *et al.* (2006):

4.1 Extraction of estradiol residues:

Ten g of egg content was homogenized with 10 ml of 67mM PBS buffer then, vortex for 5 min. 2 g of homogenized sample was mixed with 5 ml of tertbutyl methyl ether in a centrifugal screw caped vial and shaken vigorously for 30-60 min. The contents were centrifuged at 3000 rpm for 10 min. The supernatant was extracted with 5 ml tertbutyl methyl ether.

The combined supernatant was evaporated then the dried extract was dissolved in 1ml of 80% methanol. The methanolic solution was diluted with 2 ml of 20 mM PBS buffer and applied to a RIDA C18 column (solid phase extraction column with C18 end-capped sorbent of an average particle size of 50µm) in the following manner: Column was rinsed by flowing of 3 ml methanol (100%). Column was equilibrated by injection of 2 ml PBS Buffer (20mn). Three ml of sample was loaded on column. Column was rinsed by injection of 2 ml methanol (40%). The column was dried for 3 min by pressing air through it. Sample was eluted slowly by injection of 1ml methanol (80 %) (15 drops / min).

4.2 ELISA method for determination of estradiol residue:

The test procedures were done according to the chart enclosed in the kits of RIDA® and RIDASCREEN® was registered trademarks of R-Biopharm AG. Manufacturer: R-Biopharm AG, Darmstadt, Germany. R-Biopharm AG is ISO 9001 certified. The detection limit of the test was 20 ppt. In order to obtain the estradiol residue concentration in ppb in the samples, the concentrations were read from the calibration standard curve which was established by using standard solutions at levels of 0, 0.050 ppt, 0.200, 0.800, 3.200, 12.800 ppb of 17ß-estradiol in aqueous solution Fig.(1). For the construction of the calibration curve, the mean of the absorbance values obtained for the standards were divided by the absorbance value of the zero standard and multiplied by 100 (percentage maximum absorbance). The absorbance is inversely proportional to the estradiol concentration.

Calculation: % absorbance = $\frac{\text{OD of (standard or sample)}}{\text{OD of (Zero standard)}} \times 100$

The values (% maximal absorbance) calculated for the standards were plotted (on the Y-axis) versus the estradiol equivalent concentration (ppb) on a logarithmic X-axis.

Statistical analysis:

A descriptive statistical analysis was performed to estimate the mean, minimum, maximum and the mean± standard error by MEAN Analysis Procedure, **IBM SPSS.20 (2011).**

RESULTS AND DISCUSSION

Table (1): Microbiological quality of examined baladi and farm egg samples (n=25 each).

	Baladi egg			Farm egg		
	Min	Max	Mean ±SE	Min	Max	Mean ±SE
Aerobic Plate Count (cfu/g)	$<10^2$	4×10^2	$3.1\times10^2 \pm 5.8\times10$	$<10^2$	2.4×10^2	$2\times10^2 \pm 6.5\times10$
E. coli count (cfu/g)	<10	1×10^2	$3\times10 \pm 0.79\times10$	<10	<10	<10
Yeast and mould count (cfu/g)	<10	2×10	$1.5\times10 \pm 0.45\times10$	<10	5×10	$3.5\times10 \pm 0.78\times10$
Salmonella	ND			ND		
Shigella	ND			ND		
Clostridumz perfrengens	ND			ND		

ND= not detected

Table (2): Incidence of antibiotic residues in examined samples.

Baladi eggs n=25				Farm eggs n=25			
positive samples		Negative samples		positive samples		Negative samples	
No.	%	No.	%	No.	%	No.	%
2	8	23	92	10	40	15	60

Table (3): Concentration of pesticide residues (ppm) in examined chicken egg samples.

pesticides	Concentration of pesticides		MRL mg/kg
	Mean ±SE		
	Baladi eggs	Farm eggs	
Alfa-BHC	ND	ND	0.01**
Gama-BHC	ND	ND	0.01**
Delta-BHC	ND	ND	0.01**
Heptachlor	ND	ND	0.2**
Heptachlor epoxide	0.085±0.02	ND	0.05**
Aldrin	ND	ND	0.1**
Endosulfan	0.105±0.02	ND	0.03**
Endosulfan11	ND	ND	-
Endosulfan sulfate	ND	ND	-
PP-DDT	ND	ND	0.1**
PP-DDD	ND	ND	-
PP-DDE	ND	ND	-
Endrin	ND	0.28±0.09	0.005*
Endrin aldehyde	ND	ND	-
Dieldrine	ND	0.25±0.03	0.1**
Methoxychlor	0.037±0.001	0.016±0.0002	0.01*

*** JFCRF, 2016(Japan Food Chemical Research Foundation).**

****MRL: Maximum Residual Limit (Codex, 2016).**

Table (4): Incidence of pesticide residues in examined baladi and farm chicken egg samples above the MRL (n=25 of each).

pesticides	Baladi eggs		Farm eggs	
	No. of positive samples	%	No. of positive samples	%
Heptachlor epoxide	8	32	zero	zero
Endosulfan	3	12	zero	zero
Endrin	zero	zero	6	24
Dialdrine	zero	zero	4	16
Methoxychlor	11	44	6	24

Table (5): Mean values of estradiol residues in examined chicken egg samples (ppb).

	Min	Max	Mean ±SE	Incidence				
				Within permissible limit <1 ppb*		Within physiological Level<0.05ppb		Acceptable daily intake (ADI)**
				No.	%	No.	%	
Baladi eggs n=25	ND	0.20	0.03±0.001	25	100	25	100	0.05ppb /kgbw/day
Farm eggs n=25	ND	0.65	0.40±0.120	25	100	18	72	

*Gracey, (1986).

**JECFA (1999).

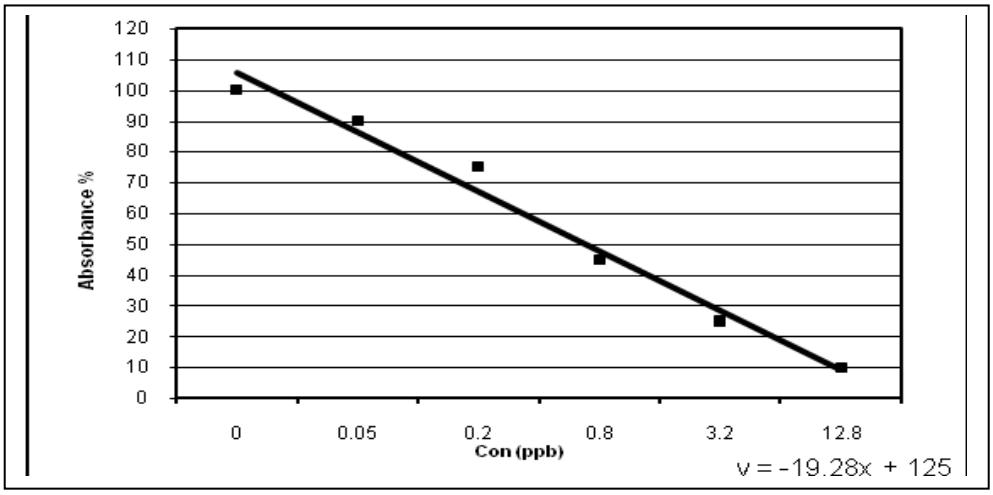

Fig.(1) oestradiol standard curve.

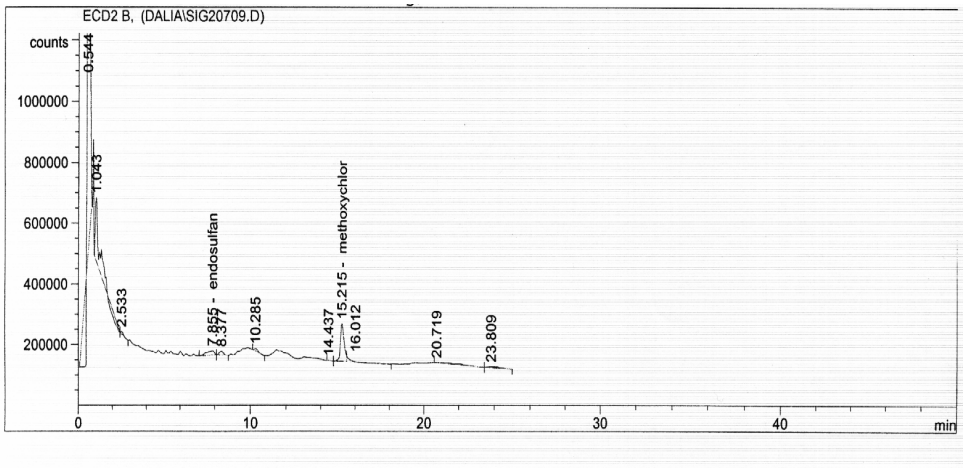

Fig (2): Detected pesticides residues in baladi eggs.

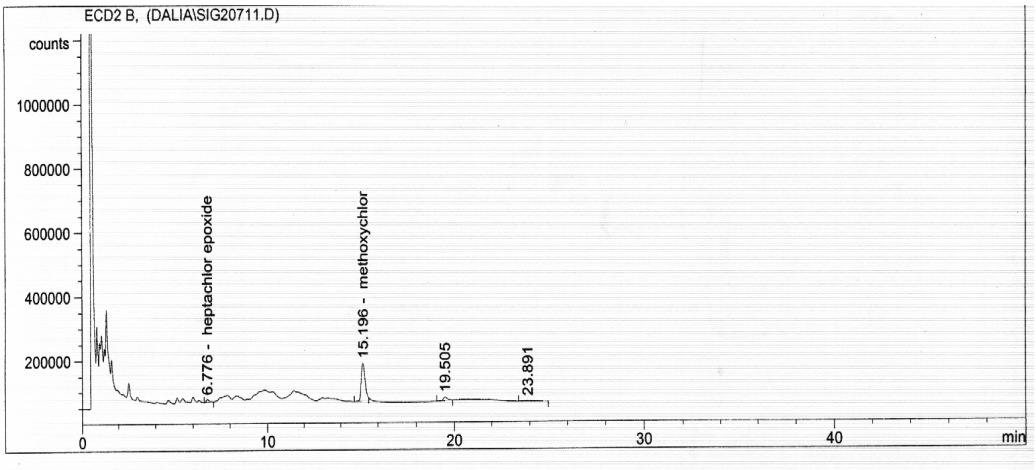

Fig (3): Detected pesticides residues in baladi eggs.

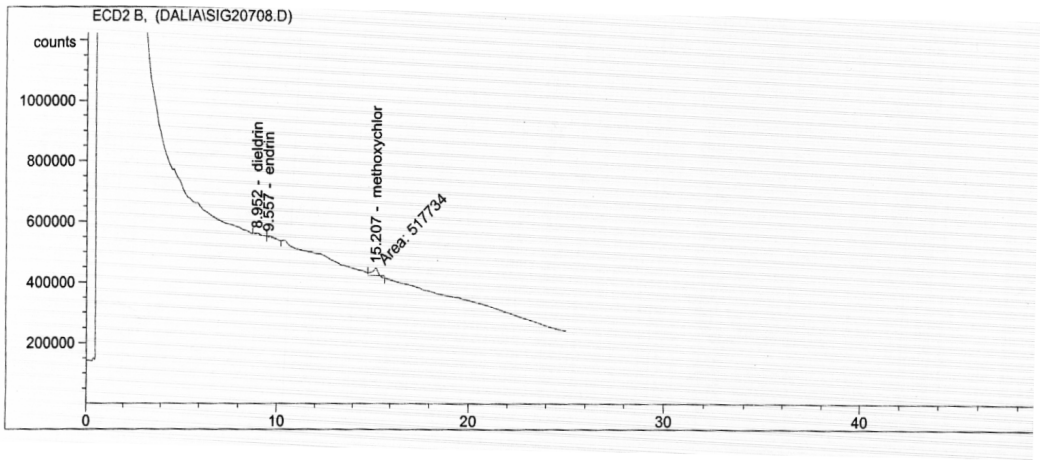

Fig (4): Detected pesticides residues in farm eggs.

Microbial quality:

Microbial contamination of table eggs in the process of production, handling and marketing should be concerned of a major public health importance. Until recently, little is known regarding microbial quality of table eggs and most of studies are concerned with the quality of hatching eggs **(Knape *et al*., 2002).** Eggs can be contaminated with microorganisms such as bacteria and fungi. These microorganisms can evade the defense mechanism of eggs and penetrate inside the egg, thus increasing the risk of food borne illnesses or product spoilage. **(Tan *et al*., 2012).** (Table 1) illustrated the microbiological quality of baladi and farm eggs. The mean values total aerobic plate count in baladi and farm eggs were $3.1\times10^2 \pm 5.8\times10$ and $2\times10^2 \pm 6.5\times10$ cfu/g with minimum and maximum values of $<10^2$,4×10^2 and $<10^2$, 2.4×10^2 cfu/g respectively, which were within the permissible limit recommended by **EOS, 3169/2007**

(2.5×10^4 cfu/g)**.** These results nearly similar to those obtained by **Abdul Aziz *et al*. (2012) and Olayemi and Charles (2013).**Higher results were obtained by **El-Kholy *et al*. (2014); Eman *et al*. (2015) and Ewonetu, *et al*. (2015).** The mean values of *E. coli* count in baladi eggs were $3\times10 \pm0.79\times10$ cfu/g with minimum and maximum <10 and 1×10^2 cfu/g which was higher than the permissible limit recommended by **EOS, 3169/2007** (10 cfu/g) with an incidence of 8 (32%); while it was <10 cfu/g in farm eggs. Lower result was obtained by **El-Kholy *et al*. (2014). Abdul Aziz, *et al*. (2012); Olayemi and Charles (2013) and Salihu *et al*. (2015)** could not detect *E. coli* in egg content. **Ghasemian Safaei *et al*. (2011); Eman *et al*. (2015) and Amal, *et al*. (2015)** could isolate *E. coli* from eggs in a percent of 19, 28.58 and 37.5%, respectively. Since *E. coli* serves as an indicator of sanitary quality as well as an index organism of pathogens, their numbers represent a measure of the efficacy of sanitation and disinfection procedures in the site of production and the degree of contamination and cross-contamination during processing **(Kornacki and Johnson, 2011).** *Salmonella* species, *Shigella* and *Clostridum perfrengens* could not be detected in the present study, this agreed with **Abdul Aziz *et al*. (2012); El-Kholy *et al*. (2014) and Sharmeen *et al*. (2014).** On the other hand, **Mohammad *et al*. (2015) and Salihu *et al*. (2015)** could isolate *Salmonella* from eggs in a percentage of 3, 13.5% respectively. The absence of *Salmonella* in the current study could be attributed to the fact that application of good poultry farmers practice, strict medication and care. The mean values of yeast and mould count in baladi and farm eggs were $1.5\times10 \pm0.45\times10$ and $3.5\times10 \pm0.78\times10$ cfu/gm with minimum and maximum values of <10, 2×10 and <10, 5×10 respectively which were compatible with the permissible limits recommended by EOS, 3169/2007 (5×10 cfu/g)**.** Higher results were obtained by **Abdul Aziz, *et al*. (2012)** in baladi egg samples, but couldn't detect yeast and mould in farm eggs. **Also Sharmeen, *et al*. (2014)** couldn't detect yeast and mould in egg contents but **El-Kholy *et al*. (2014)** found 2.6×10^2 cfu/gm in table eggs. The presence of yeast and mould may be due to bad storage of eggs in rooms with high temperature specially in summer months and under humid conditions, and could lead to several respiratory diseases through their spores like coccidioidomycosis, blastomycosis and histoblastomycosis when the fungal spores are inhaled by the humans and the birds **(Obi and Igbokwe, 2007).** From the public health point of view, certain strains of moulds were implicated in food poisoning outbreaks due to production of aflatoxins, as well as some moulds, are capable of forming toxins

that cause mycotoxicosis leukemia in man **(Ray, 2004).**

Antibiotic residues:

(Table 2) showed the incidence of antibiotic residues in baladi and farm eggs which were 2 (8%) and 10 (40%) respectively. Lower results were obtained by **Kabir *et al.* (2004); Fagbamila *et al.* (2010) and Donkor *et al.* (2011).** Higher results were obtained by **Al-Ghamdi *et al.* (2000) and Nonga *et al.* (2009).** On the other hand, **Alomirah, *et al.* (2007)** couldn't detect antibiotic residues in examined eggs. Antimicrobials are used by the poultry industry to enhance growth and feed efficiency and to reduce bacterial disease **(Donoghue, 2003).** In laying hens, antimicrobials are used only to treat and to prevent bacterial infections. Antimicrobials used to treat poultry are similar to those used in human medicine and included aminoglycosides, tetracyclines, beta-lactams, quinolones, macrolides, polypeptides, amphenicols and sulphonamides **(Stolker and Brinkman, 2005).**

Pesticide residues:

The mean concentrations of the following compounds have been determined in baladi and farm eggs and recorded in (Table 3). Alfa-BHC, Gama BHC, Delta BHC, heptachlor, heptachlor epoxide, Aldrin, endosulfan, endosulfan 11, endosulfan sulfate, p,p-DDT, p,p-DDD, p,p-DDE, endrin, endrin aldehyde, dieldrin and methoxychlor. The mean concentration of heptachlor epoxide in baladi and farm eggs recorded 0.085±0.02 ppm with an incidence of 8 (32%) which considered more than the maximum residue limit (MRL) cited by **Codex (2016)** which recommended maximum residue limit (MRL) of 0.05 mg/kg for heptachlor epoxide indicating contamination of the baladi eggs as shown in (Tables 3 and 4), while it couldn't be detected in farm eggs. Endosulfan couldn't detected in farm eggs and was present in baladi eggs in a concentration of 0.105±0.003 ppm and incidence of 3 (12%), above the MRL cited by **Codex (2016)** (0.03 mg/kg). Endrine couldn't be detected in baladi eggs and its mean value was 0.28±0.009 in farm eggs and was present in a percent of 6 (24%) above MRL cited by **J F C R F (2016)** (0.005 mg/kg). Dialdren not detected in baladi eggs but found in farm eggs in a percent of 4 (16%) with mean concentration of 0.25±0.03 ppm which above MRL recommended by **Codex (2016)** (0.1 mg/kg). Methoxychlor was present in both baladi and farm eggs in a concentration of 0.037±0.001 and 0.016±0.002 ppm with percent of 11(44%) and 6 (24%) respectively, above MRL cited by **J F C R F (2016)** (0.01 mg/kg). Alfa-BHC, Gama BHC, Delta BHC, heptachlor, aldrin, endosulfan 11 , endosulfan sulfate, p,p-DDT, p,p-DDD, p,p-DDE and endrin aldehyde as well as organophosphrous pesticides couldn't be

detected in both baladi and farm eggs. **Fontcuberta (2008)** couldn't detect DDT in examined samples. **Tao *et al.* (2009)** detected p,p-DDT, p,p-DDD, p,p-DDE in eggs in a concentration of 0.049, 0.015 and 0.089 ppm respectively. **Windala *et al.* (2009)** found DDT in a concentration of 0.06 ppm in 95% of examined samples. **Nida *et al.* (2010)** detected p,p-DDE, Heptachlor, Heptachlor epoxide and endrine in a concentration of 0.031 (15%), 0.058 (3,7%), 0.04 (10%) and 0.01 (12%) mg/kg, but couldn't detect endosulfan in examined egg samples. **Polder *et al.*, (2016)** detected dialderine and 14% of examined samples were exceeding the maximum residual limit. Although concentration of organochlorine pesticides in most of the samples were within the permissible limits, it must be emphasized that organochlorine pesticides are inherently unmanageable and they bio-accumulate in living species. Therefore, the acceptable standard for any organochlorine in any sample should ideally be zero **(Li *et al.*, 2006).**

Estradiol residue:

(Table 5) illustrates the mean value of estradiol in baladi and farm egg samples which were 0.030 ±0.001, 0.400±0.120 µg/kg with minimum and maximum value of ND, 0.2 and ND, 0.65 µg/kg respectively. All egg samples of both baladi and farm are within the permissible limit (1ppb) stated by **Gracey (1986).** All baladi egg samples are within acceptable daily intake (ADI) and 72% of farm eggs are within the acceptable daily intake (ADI) value for estradiol of 0.05 µg/kg body weight /day as assessed by **JECFA (1999).** These findings were higher than the value for estradiol obtained by **Aslam, *et al.*, (2013) and Xiaoxia, *et al.* (2014).** Nearly similar finding was obtained by **Sahar Abd Wafia (2015).** Administration of hormones to broiler chickens for performance-enhancing purposes may lead to deposit of residuals in their carcasses and eggs. The health concerns associated with hormonal compounds used as growth promotions are their carcinogenic and endocrine-disrupting potentials **(Henderson and Feigelson, 2000).**

CONCLUSION AND RECOMMENDATION

The pathogenic moulds found their way to penetrate and contaminate eggs and may produce their toxins under favorable conditions. Therefore, special attention should be directed to safeguard the eggs against their contamination through application of correct farm hygiene programs, good handling, processing and storage methods, as well as, the periodical examination of eggs and poultry feed **(Neamatallah *et al.*, 2009).** Finally, the most important

conclusions and recommendations stemming from the present study that people should be aware that eggs, substantially to the intake of organochlorine pesticides through food consumption.Although, the Egyptian Government has imposed a ban or restricted the use of various pesticides, there is need to continue the monitoring study of the organochlorine pesticides and other pesticide residues in foodstuffs from the view point of human food safety.

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تقدير المخاطر الميكروبية والمتبقيات الضارة فى بيض المائدة

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الملخص العربى

تم جمع عدد خمسين بيضة (25 من كل من البيض البلدي والمزارع) من مختلف الأسواق في محافظتى القاهره والجيزه لتقييم الوضع الميكروبيولوجي والكشف عن بقاياها المضادات الحيوية والمبيدات الحشرية وهرمون الاستراديول. وكان متوسط العد البكتيرى الهوائي في عينات البيض البلدي وبيض المزارع 3,1 × 10^2 ± 5,8 × 10 و 2 × 10^2 ± 6,5 × 10 خليه/جرام على التوالى والتي كانت ضمن الحد المسموح به الذي أوصت به المواصفات القياسيه المصرية 2007/3169 (2,5× خليه/جرام 10^4) وكان متوسط عد بكتيريا الإشيريشيا كولاى في البيض البلدي 3×10±7,9×10 وهو أعلى من الحد المسموح به الذي أوصت بـه المواصفات القياسيه المصرية 2007/3169 (10 خليه/جرام) ، في حين أنـه لـم يـتم الإسـتدلال عليهـا فـي بـيض المـزارع وكـذلك لـم يـتم عـزل بكتيريـا السالمونيلا، الشيجيلا و الكلوستريديوم بيرفرينجينز من جميع عينـات البـيض (بلـدى ومـزارع). وكـان متوسـط العـد الكلـى للفطريـات والخمائر في عينات البيض البلدي والمزارع 1,5 × 10 ± 0,45 × 10 و 3,5 × 10 ± 0,78 × 10 خليه/جرام علـى التـوالي والتـي كانت متوافقة مع الحد المسموح به الذي أوصت به المواصفات القياسيه المصرية 2007/3169 (5×10 خليه/جرام) .تـم الكشـف عـن وجود بقايا المضادات الحيوية في عينات البيض نوعيا باستخدام اختبـار (FPT) , تـم قيـاس بقايـا المبيـدات الحشـرية باسـتخدام جهـاز الكروماتوجرافي الغـازى (GC) و قيـاس بقايـا هرمـون الاسـتراديول باسـتخدام طريقـة ELISA للأنسـجه.كانت نسـبة تواجـد بقايـا المضادات الحيوية في عينات البيض البلدي والمزارع 2 (8٪) و 10 (40٪) على التوالي . وكان متوسط تركيز الهيبتـا كلورايبوكسـيد في عينات البيض البلدي 0,085 ± 0,02 جزء في المليون فوق الحدود القصوى التـي أوصـى بهـا الكـودكس (2016) (0.05 ملجـم / كجم) وكانت نسبه تواجده 8 (32٪) ولم يتم الإستدلال عليه في بيض المزارع . لم يتم الإستدلال على الإندوسلفان فـي بـيض المـزارع وكان موجودا في البيض البلدي بتركيز 0,105 ± 0,003 جزء مـن المليـون وكـان عـدد 3 (12٪) اعلـى مـن الحـدود القصـوى التـي ذكرها الكودكس (2016) (0,03 ملجم / كجم). لم يتم الإستدلال على الإندرين في البيض البلـدي بينمـا كـان متوسـط تركيـزه 0,28 ± 0,009 جزء من المليون في بـيض المـزارع أعلـى مـن الحـدود القصـوى التـي ذكرتهـا المؤسسـة اليابانيـه لأبحـاث الطعـام الكيميائيـة (2016) (0.005 ملجم / كجم) وكان نسبة تواجده 6 (24٪). لم يتم الإستدلال على الداي ألديرين في البـيض البلـدي ولكـن وجـد فـي بيض المزارع بنسبة 4 (16٪) بمتوسط تركيز 0,25 ± 0,03 جزء في المليون أعلى من الحدود القصوى التـي أوصـى بهـا الكـودكس (2016) (0,1 ملجم / كجم). كان متوسط تركيز الميثوكسيكلور في كل مـن البـيض البلـدي والمـزارع 0,037 ± 0,001 و 0,016 ± 0,002 جزء من المليون بنسبة تواجد 11 (44٪) و 6 (24٪) على التوالي أعلى من الحدود القصوى التـي ذكرتهـا المؤسسـة اليابانيـه لأبحاث الطعام الكيميائية (2016) (0,01 ملجم / كجم). لم يتم الإستدلال على كـل مـن ألفـا, جامـا و دلتـا البـى اتـش سـى، الهبتـاكلور، ألدرين، الإندوسلفان 11، كبريتات الإندوسـلفان، دي دي تـي، دي دي دي ، دي دي إي والأنـدرين ألدهايـد وكـذلك المبيـدات العضـويه الفسفوريه في كل من البيض البلدي والمزارع. كان متوسط بقايا الاستراديول فـي عينـات البلـدي والمـزارع 0,03 ± 0,001، 0,4 ± 0,120 جزء في البليون على التوالي ضمن الحد المسموح به (1 جزء فـي البليـون) (1986,Gracey). جميـع عينـات البـيض البلـدي ضمن الحد المسموح للاستهلاك اليومي (ADI) و 72٪ من بيض المزارع تقع ضمن الحد المسموح للاستهلاك اليـومي (0,05 جـزء في البليون من وزن الجسم / يوم (JECFA 1999) . وتمت مناقشة الأهمية الصحية للميكروبات المعزولة والبقايا الضارة المتواجده وكذلك التدابير الصحية لضمان جودة البيض لحماية المستهلك.