

# Investigation of Patulin in samples blood of persons in Karbala province

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## Abstract

**Objectives:** The study aim to investigation of patulin in samples blood collected from persons in Karbala .

**Methods:** A study of Investigation of Patulin in samples blood of persons in Karbala province included the collection of thirty six of blood samples persons , age of the persons to range 10 -70> years then extraction of patulin toxin from these samples by use chloroform and detection of patulin toxin in serum of persons by use thin layer chromatography technique ( TLC ).

**Results:** The results showed 22 out 36 samples (61.1%) of blood collected from persons were found contain patulin . the highest percentage blood samples contamination with patulin was ( 36%) that collected from person at age group 70> years , the percentage of blood samples that contamination with patulin that collected from female was( 54.5 %).

**conclusion:** persons in Karbala province high exposure to patulin toxin , and foods ( apples , pear and peach ) in local markets are contaminated with patulin .

**Key word :** Patulin , mycotoxin , *Penicillium* , *Aspergillus* , Biochemical parameters , TLC. Thin Layer Chromatography .

## Introduction

Patulin is mycotoxin that is produced by certain species of *Eupenicillium* , *Penicillium* , *Paecilomyces* and *Aspergillus* that may grow on a variety of founds including Apples , grapes , pears , apple juice , cheese and grains . <sup>1,2</sup>

Patulin effects in many biochemical parameter such hexokinase, aldolase and AT-pase glycogen phosphorylase , also cause decrease of glycogen concentration in liver , kidney and intestine tissues <sup>3, 4</sup> . Sugar level raised in blood to 60% when treated of animals lab with patulin , patulin toxin inhibited protein synthesis and causes it to decrease the proportion of glycogen in the kidney , liver and intestines because of high concentrations of the toxin and that the decrease of glycogen, which may be the result of the separation of insulin and reflected this result in the case of low concentration of insulin-dependent enzymes <sup>5</sup>. Many studies have shown that patulin toxin was toxic to the different cell types Such as protozoa, granular , cell

line <sup>6</sup>. When exposed to a ranged of about 0.1 - 50 $\mu$ M of patulin toxin most of the effects will appear in the process of imbalance in the concentrations and transport of ions over the plasma membrane influencing  $Ca^{+2}$  ,  $Na^{+}$  and  $K^{+}$  <sup>3</sup>.

Several genetic toxicological studies have been conducted both in vitro and in vivo And many laboratory tests with fungus, bacteria and mammalian cells , the results showed chromosomal aberrations , DNA breakage , DNA synthesis and reverse mutation <sup>7</sup>. This genetic toxicity of patulin toxin may be caused by its ability to interact with the sulfhydryl group, which in turn inhibits the participation of enzymes in the replication of genetic material <sup>8</sup>.

## **Materials and Method**

### **1- Collected of samples**

Thirty six blood samples (5ml ) collected from 18 male and 18 female . Age of the persons to range 10-70> year , each sample putting in gel tube and transported to clinical laboratory .

### **2- Extraction of Patulin**

**A.** added 3ml of chloroform to each tube and mixing well by vortex .

**B.** save the sample in closed tube in refrigeration .

**C.** After that isolating clear layer in clean glass tube and keep tube open and put in over at 40C° for day .

### **3- Detection of Patulin**

**A.** Thin layer chromatography (TLC ) technique used in detection of patulin in serum of persons .

**B.** Thin layer chromatography plates put in over (120C°) for one hour to activate it .

**C.** Make straight line on TLC plate in distance of about 1.5 cm from the base plate

**D.** Patulin stander (15 $\mu$ l ) put as spot on TLC plate by capillary tube and put 15  $\mu$ l on plate from each extracted samples with a distance 2cm between sample and another then let the spots to dry in laboratory condition .

**F.** Then put the plate in separation tank containing 100 ml from mixture chloroform : Aceton (8:2) .

**H.** plate exited from the tank and leaves it to dry under the laboratory condition .

**K.** Then plate examined under uv light (360 nm ) and compare the color and Relative flow (RF) of extracted samples with the standard toxin .

## Results

### Investigation of patulin in blood of samples persons.

The result showed 22 (61.1%) out 36 samples of blood collected from healthy person infected with patulin toxin with non significant difference of number of blood samples , not contamination with patulin (14 samples) , Table1.

Also this study clarified that highest percentage blood samples contamination with patulin that collected from persons at age 70> year ( 36% ) while group ( 10-20 year ) had the least percentage ( 9% ) . Table2 .

The result as show in ( table 3 ) the female and male infected with patulin were 54.5% and 45.5% respectively .

**Table 1. Number and percentage of samples blood contamination and non contamination with patulin toxin**

Case	No. of samples	Percentage (%)
Number of persons borne patulin toxin	22	<b>61.1</b>
Number of persons non borne patulin toxin	14	<b>38.9</b>

$$X^2 \text{ cal.} = 1.7$$

$$X^2 \text{ tab.} = 3.8$$

**Table 2. Effect of age on contamination blood with patulin toxin.**

Range of age	No. of persons borne toxin	Percentage (%)
10 – 20	2	<b>9</b>
21 – 40	7	<b>31.8</b>
41 – 60	5	<b>22.7</b>
70 <	8	<b>36</b>

**Table 3. Effect of Gender on contamination blood with patulin toxin.**

Gender	No. of borne toxin	Percentage (%)
Female	12	54.5
Male	10	45.5

$X^2$  cal. = 0.16

$X^2$  tab. =3.8

### Discussion

The common maximum level of patulin in apple juice is regulated within the range 25–50  $\mu\text{g}$  patulin/kg juice <sup>9</sup>. Fortunately, the flavor level identical with a level of patulin toxin near 50  $\mu\text{g}$  /kg juice , Data based on a United States Department of Agriculture (USDA) survey .In one study show a high consuming of apples and their derivatives in the beginning of life (6.4 g/kg per day), in proportion to the low age children 1– 6 years ; 2.4 g /kg per day children 7– 12 years; 1.0 g/kg per day to reduction 0.4 g/kg per day for adults <sup>9</sup>. Combining the appearance of patulin toxin in apple juices and children foods with intake of apples and apple products leads to an induction of an approximate amount of patulin by children and adults <sup>11</sup>. The highest amount is handled by children was 6.39  $\mu\text{g}$  patulin/kg juice , this is consistent with 40.9 ng/kg per day , For adults the approximate intake which is 0.24  $\mu\text{g}$  patulin / kg per day provided that the producers respect established limits <sup>12</sup>. In one study found zearalenone in blood plasma <sup>13</sup>. And in another search showed the concentration of Ochratoxin ranged from 90- 940 ng /L in blood <sup>14</sup>. the result were close to another study ,where they found 23.70% from the blood of patients specimen had Ochratoxin A , so healthy persons were had Ochratoxin A in their blood (10%) and showed the males highly in infected (87.5 % ) with Ochratoxin A while in female ( 70 % ) and ( 71-80 ) year age had highly infected ( 92.85 ) with Ochratoxin A <sup>15</sup>. In another study showed the evaluated intake of patulin toxin in body weight ranged from 0.1 – 1.5 ng /kg for the population , and from 0.3 – 5.1 ng /kg .w for the consumers only <sup>16</sup>.

**Conclusion:** persons in Karbala province high exposure to patulin toxin , and foods (apples , pear and peach ) in local markets are contaminated with patulin.

### Reference

1- Abrunhosa L, Paterson R R M, Kozakiewicz Z, Lima N and Venancio

- A ,Mycotoxin production from fungi isolated from grapes, *Letters in Applied Microbiology*. (2001), **32** (3) : 240–2.
- 2-** Moreau C. , Co-occurrence of patulin and citrinin in Portuguese apples with rotten spots, *Food Addit. Contam.*, (2002), **19**(1): 568–74.
- 3-** Wouters M F A and Speijers G J A., Toxicological evaluation of certain food additives and contaminants in food, Patulin, *WHO Food Additives Series*. (1996), **35**(4): 377–402.
- 4-** Moss, M. O. and Long, M. T. Fate of patulin in the presence of the yeast *Saccharomyces cerevisiae*. *Food Aditives and Contaminants*, (2002). vol. 19, no. 4, pp. 387–399 .
- 5-** Cunha, S.C. ,Faria M.A., Pereira V.L. , Oliveira T.M. and Pinto E., Patulin assessment and fungi identification in organic and conventional fruits and derived products. *Food Control*.(2014), vol. 44(3): pp. 185–190.
- 6-** Selmangolu , G. , Evaluation of the reproductive toxicity of patulin in rowing male rats. *Food and Chemical Toxicology*. (2006) ,. vol. 44, no. 12, p. 2019–2024
- 7-** Liu B-H, Yu F-Y, Wu T-S, Li S-Y, Su M-C, Wang M-C and Shih S-M . Evaluation of genotoxic risk and oxidative DNA damage in mammalian cells exposed to mycotoxins, patulin and citrinin, *Toxicol. Appl. Pharmacol.* (2003), **191**, 255–63.
- 8-** Puel, O., Galtier , P. and Oswald , I.P. ,Biosynthesis and toxicological effects of Patulin. *Toxins*.(2010), vol. 2, no. 4, pp. 613– 631.
- 9-** Rosner H., Mycotoxin Regulations: An update, *Revue Méd. Vét.* (1998), **149**(1) : 679–80.
- 10-** Plunkett L M, Turnbull D and Rodricks J V ., Differences between adults and children affecting exposure assessment, in Guzelian P S, Henry C J and Olin S S, *Similarities and Differences Between Children and Adults*, Washington DC, ILSI Press . (1992), 79–94.
- 11-** Thuvander A, Möller T, Enghardt-Barbieri H, Jansson A, Salomonsson A A C and Olsen M., Dietary intake of some important mycotoxins by the Swedish population, *Food Addit. Contam.* (2001), **18**(4) : 696–706.
- 12-** Rivka B.G. and Nachman P. , Mycotoxins in fruits and vegetables . Elsevier Inc. (2008), 225-247 .
- 13-** Magan N. and Olsen, M. , Mycotoxin in food, Detection and control . wood head publishing in food science and technology , limited .(2004 ) , pp, 471.
- 14-** Breitholz-Emanue lsson, A. , Olsen M. , Oskar soon A. , Palmiger hallen I. and Hult K. , Ochratoxin A in cow 's milk and human milk with corresponding human blood samples . *J. AOAC Int.* ( 1993 ) . , 76: 842-846 .
- 15-** AL- Musoui H.R.. , Study the relationship between some fungi and its toxins with kidney failure unknown case . Master Thesis , AL-Qadisiya university . (2015) .