EFFECTS OF ARTIFICIAL SWEETENERS ON RABBITS HEMATOLOGCIAL CHANGES +

Shakir M. Saied* **Asma A. Basheer**** **Ehsan G. Zakaria*****

Abstract:

The possible deleterious effects and hematological changes accompanying an extensively used artificial sweeteners(mixture of saccharin, aspartame and sodium citrate) have been studied on adult apparently healthy 9 males locally rabbits after feeding it for two months by juice containing these sweeteners at rate of 0.0 , 0.5, 1.0 and 1.5 g/kg body weight/day, for (control), first, second and third dose respectively.. The little changes which were observed by the first dose was increased at second dose. Here the anemia(which caused the bone marrow failure) was due to decreasing of total red blood cell(RBC), hemoglobin (Hb g/dl) and packed cell volume PCV (%),Finally , feeding by the third dose caused dead due to high drop in hematological values.

Key Words: Hematologcial changes, artificial sweeteners, Saccharin, Aspartame and Sodium citrate.

المستخلص

درست التأثيرات المحتملة والمصاحبة لاستخدام المحليات الاصطناعية (خليط السكرين والاسبارتام وسترات الصوديوم) على التغييرات الدموية لتسعة أرانب محلية بصحة جيدة،بعد تغذيتها لمدة شهرين بعصير يحوى هذه المحليات وبمعدل جرع ٠,٠ و٠,٥ و١,٠ و١,٥ غم\كغم من وزن الحيوان في اليوم لجرع (السيطرة) والأولى والثانية والثالثة على التوالي. التغييرات الطفيفة باستخدام الجرعة الأولى زادت في الجرعة الثانية. إن فقر الدم هنا(والذي أدى إلى فشل فئ نخاع العظم) هو بسبب انخفاض تركيز الهيموكلوبين وكريات الدم الحمراء والنسبة المئوية لحجم حزمة الخلايا . واخيرا فان الجرعة الثالثة سببت الوفاة الناشئ عن الهبوط الحاد بقيم التغييرات الدموية.

Introduction:

Over the years ,a great ambivalence was occurred between numerous studies which indicate that saccharin was a weak carcinogen in laboratory animals without this risk in humans(at moderate use),and another which indicated that saccharin caused canser[1].The scientists were told the National Toxicology Program, a division of the National Institute of Environmental Health Sciences, that declaring saccharin safe would "result in greater exposure to this probable carcinogen in tens of millions of people, including children (indeed, fetuses). If saccharin was even a weak carcinogen, this unnecessary additive would pose an intolerable risk to the public." [2].

+ **Received on 2007/3/5 , Accepted on 2008/12/10**

* Asst.Prof - Dept. of Pharmacy, Institute of Technical Mosul
** Lecturer - Dept. of Pharmacy, Institute of Technical Mosul
*** Ass. Lecturer- Dept. of Pharmacy, Institute of Technical Mosul

On another hand the possible risks of aspartame were it containing the naturally-occurring amino acid phenylalanine, which was a health hazard to the few people born with phenylketonuria, a genetic inability to process phenylalanine. A few studies had also recommended further investigation into possible connections between aspartame and diseases such as brain tumors, brain lesions, and lymphoma [3].
Cyclamate, the third was used in the United States in the early 1950s, were banned in 1969 (in Canada, only for general use) after a study of them in rats implicated them as possible carcinogens. Since the mid-1980s the Food and Drug Administration (FDA) has been considering a petition to reinstate them, Consult latest Government regulations on use in food [4] .

As part of continuous program directed toward the studying of the biological effects of some organic and inorganic compounds[5-8], it was become of interest to study effects of artificial sweeteners on hematologcial changes in laboratory rabbits.

Materials and methods

Nine adult males rabbits were used for this study, average weight, 1.18–1.73 kg were kept in standard rabbit cages (which were regularly cleaned and disinfected using standard procedures). The rabbits were allowed unrestricted access to normal rabbit chow and water, and allowed to acclimatize to their new environment for 7 days (at this period all animals are used as controls). The rabbits were divided into 3 equal groups (groups A– C), and then they were fed manually by the samples (extensively used artificial sweeteners).

Hematologcial Analysis

Blood samples were taken by vein puncture of the marginal vein of the ear, every 15 days at the same time for 60 days. Hemoglobin (Hb) concentration was measured by the cyanmethaemoglobin method [9]. Red blood cells (RBC) were counted with haemocytometer; the RBC counting methods were based on the dilution of obtained blood with diluting fluids in RBC counting pipettes [9]. Packed cell volume (PCV) was determined using the microhaematocrit method [9].

Statistical Analysis

The data were expressed as mean_ standard deviation (SD), the mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC) were calculated[10].The percentages of hematological changes were determined as : % = mean of individual value / mean value of control *100

Results and discussions:

The hematological changes in rabbits during the period of the study and those of the control group(7 days before the experiment) were presented in Tables 1-3,while the mean of these changes were presented in Tables(4-6).The percentages of these changes accompanying the doses of 0.5, 1.0,and 1.5 g/kg weight/day administration were:

PCV:
(89.07%,81.75%,64.02%,69.96%),(87.01%,54.10%,47.88%,34.7153%)and(96.72%,2.53%, dead) for periods of 15,30,45 and 60 days respectively.

Hb:(80.65%,81.99%,63,64%),(89.59%,54.34%,41.08%,54.80%)and(94.62%,51.62% and dead) for the same periods respectively.

RBC:(92.01%,81.85%,65.62%,57.38%),(89.59%,54.34%,41.08%,54.80%)and(95.13%,51.30%,and dead)for the same periods respectively.

These results showed the hematological values of the treated groups during the course of the experiment(60 days)were decreased as dose increased and the rabbits were died before the end of the experiment(60^{th} day).

Conclusion:

A major problem facing the experimental animals was related to increased the doses of artificial sweeteners. The total red blood cell(RBC), hemoglobin (Hb)and packed cell volume(PCV) decreased with this increasing. This reduction was due to cytotoxic effects of these compounds which activated in the bonemarrow ,mediated through disturbance in DNA function and produced inadequate production of red cell and other formed elements,led to boone marrow failure[11-12]. From the results of this study, it was hereby suggested that extensively used artificial sweeteners seious consequences on hematological parameters in rabbits[2].

Table (1) : Hematologcial changes accompanying artificial sweeteners 0.5 g/kg body weight/day administration in laboratory rabbits over a period of 60 days

Sample (n = 3)		Control	Period (Days)			
			15	30	45	60
1	PCV (%)	36.3 ±1.47	34 ±1.95	30.2 ±1.29	25.4 ± 2.73	25.1 ± 2.86
	Hb(g/dl)	12.24±0.50	11.56±0.67	10.2±0.44	8.47±0.91	8.58±0.97
	RBC($\times10^6$/µl)	3.852±0.16	3.71±0.21	3.217±0.14	2.675±0.29	2.70±0.30
2	PCV (%)	38.1 ± 0.92	38.2 ±1.08	33.1 ±1.44	25.1 ± 2.12	23.3 ± 2.38
	Hb(g/dl)	12.92±0.31	12.92±0.37	11.22±0.54	8.5±0.72	7.82±0.81
	RBC($\times10^6$/µl)	4.066±0.09	3.93±0.15	3.531±0.16	2.92±0.24	2.461±0.25
3	PCV (%)	38.3 ± 0.63	30.2 ± 0.82	32.1 ±1.35	24.2 ± 1.36	24.1 ±1.35
	Hb(g/dl)	12.89±0.24	10.20±0.28	10.84±0.47	8.16±0.46	8.16±0.46
	RBC($\times10^6$/µl)	4.039±0.07	3.477±0.20	3.42±0.14	2.568±0.14	2.568±0.15

Table (2) : Hematologcial changes accompanying artificial sweeteners 1.0 g/kg body weight/day administration in laboratory rabbits over a period of 60 days

Sample (n = 3)		Control	Period (Days)			
			15	30	45	60
1	PCV (%)	39.1±0.71	30.0±1.78	22.2±1.03	20±1.47	18±2.34
	Hb(g/dl)	13.26±0.24	10.2±0.61	7.48±0.32	6.8±0.50	6.12±0.79
	RBC ($\times 10^6$/µl)	4.173±0.08	3.477±0.18	2.354±0.12	2.147±0.16	1.926±0.25
2	PCV (%)	38.2±1.08	33.1±1.25	20.1±1.58	19.3±1.48	18±1.35
	Hb(g/dl)	12.92±0.36	11.22±0.37	6.80±0.53	6.46±0.50	6.12±0.46
	RBC ($\times 10^6$/µl)	4.066±0.12	3.531±0.18	2.140±0.16	2.163±0.24	1.926±0.17
3	PCV (%)	39.4±1.15	34.0±2.34	21.3±1.95	19.1±0.71	16±±1.91
	Hb(g/dl)	13.26±0.39	10.20±0.66	7.14±0.66	6.46±0.24	5.44±0.65
	RBC ($\times 10^6$/µl)	4.173±0.13	3.210±0.19	2.247±0.21	2.107±0.11	1.712±0.20

Table (3) : Hematologcial changes accompanying artificial sweeteners 1.5 g/kg body weight/dayadministration in laboratory rabbits over a period of 60 days

Sample (n = 3)		Control	Period (Days)			
			15	30	45	60
1	PCV (%)	39.1±0.91	38.0±2.71	21.2±0.85	17.0±1.08	*
	Hb(g/dl)	13.26±0.31	12.58±0.46	7.22±029	5.78±0.36	*
	RBC ($\times 10^6$/µl)	4.173±0.09	3.959±0.14	2.273±0.09	1.684±0.23	*
2	PCV (%)	38.2.±1.19	37.0±2.16	20.0±1.47	*	*
	Hb(g*dl)	13.09±0.40	12.58±0.36	6.8±0.50	*	*
	RBC ($\times 10^6$/µl)	4.044±0.55	3.959±0.12	2.113±0.13	*	*
3	PCV (%)	38.0±1.47	36.3±1.15	18.0±1.78	*	*
	Hb(g/dl)	12.85±0.62	12.16±0.38	6.12±0.61	*	*
	RBC ($\times 10^6$/µl)	4.102±0.26	3.825±0.12	1.926±0.19	*	*

*Dead

Table (4):Mean Hematologcial changes accompanying artificial sweeteners 0.5g/kg Body weight administration in laboratory rabbits over a period of 60 days

Haematolgical changes	Control	Period(days)			
		15	30	45	60
PCV(%)	38.3±.98	34.1±1.28	31.8±1.36	24.9±2.07	24.1±2.17
Hb(g/dl)	13.1±0.33	11.5±0.47	10.74±0.48	8.37±0.91	8.18±0.74
RBC(X10 ^{6}µl)	4.13±0.11	3.70±0.18	3.38±0.15	2.71±0.22	2.57±0.23

Table (4):Mean Hematologcial changes accompanying artificial sweeteners 1.0g/kg Body weight administration in laboratory rabbits over a period of 60 days

Haematolgical changes	Control	Period(days)			
		15	30	45	60
PCV(%)	38.9±0.98	34±1.79	21.2±1.52	19.4±1.34	17±1.86
Hb(g/dl)	13.1±0.33	10.5±0.54	7.14±0.50	6.57±0.41	5.89±0.63
RBC(X10 ^{6}µl)	4.13±0.11	3.70±0.18	2.24±0.16	2.13±0.17	1.85±0.30

Table (4):Mean Hematologcial changes accompanying artificial sweeteners 1.5g/kg Body weight administration in laboratory rabbits over a period of 60 days

Haematolgical changes	Control	Period(days)			
		15	30	45	60
PCV(%)	38.4±1.19	37.1±1.4	1.97±1.36	*	*
Hb(g/dl)	13.0±0.44	12.3±0.40	6.71±0.46	*	*
RBC(X10 ^{6}µl)	4.10±0.30	3.9±0.13	2.10±0.132	*	*

*Dead

References:

1-Redmond, W.A." Artificial Sweetener" *Microsoft® Encarta®* 2006 [DVD]. Microsoft Corporation,(2005).

2-Jacobson, M.F., " Saccharin Still Poses Cancer Risk, Scientists Tell Federal Agency " *Health-advocacy organization based in Washington, D.C., Comments on Saccharin*; (1997).

3-Dobersen, Thomas," Calculation of aspartame intake in children". *J Am Diet Assoc*. :pp831-833(1989).

4-Buckley, G.R. and Greenberger, C.E." Aspartame is no more likely than placebo to cause urticaria/angioedema: results of a multicenter, randomized, double-blind, placebo-controlled, crossover study". *J Allergy Clin Immunol*; 92:pp513-520(1993).

5-Basher, A.A."*Isolation and identification of some indoor dust fungi and their effect on the respiratory system"* MSc Thesis, College of Sciences; Univ. of Mosul;Iraq(2005)

6-Ayoub,M.T. and Saied ,SH.M. "Studies with Polyfunctionally Substituted Heterocyclics: Biological Evaluation of Coumarin SulphonamidesAs New Hypoglycaemic Agents [Part-II]", *Mu'tah Lil-Bohooth Wa Al-Dirasat*, Vol.15, No.4, 155, (2001).

7-Al-Taee, A.F. and Zakaria ,E.G.;"Effecte of *Eimeria tenella* Attenuted Vaccine With Levamisol On Immunity OF Broilers"; *Iraqi Journal of Veterinary Sciences*;vol.19,No *2(2005)*

8-Saied ,Shakir M. "Organic Microbiological Studies: Effects of Organic Dyes (Gentian Violet or Methylene Blue) On Laser Lethal photosensitization of Fungus Aspergillus flavus Treated with Boric Acid; accepted ,*J. Education and Science* ,accepted(2006).

9-Mitruka, B.M. and Rawnsley, H.M," Clinical, biochemical and hematological reference values in normal experimental animals". Masson Publishing, NewYork, USA (1977).

10-Schalm, O. W., JAIN, N. C. and Carroll, *J. Veterinary Haematology*; 3rd ed. Lea and Febiger. Philadelphia (1975).

11-Erah1, P.O." Canice C Asonye and Augustine O Okhamafe;" Response of Trypanosoma brucei brucei–induced anemia to a commercial herbal"; Full Length Research Paper ; *African Journal of Biotechnology* Vol. 2 (9), pp. 307-311, September (2003).

12-Jenkins,G.C.;McCrori,P.;Forsberg,C.M. and Brown ,J.L."Studies on the anaemi in rabbits infected with Trypanosoma brucei. 1-Evidence for haemolysi.J.Comp.Pathol.90:107-121,(1980)