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Effect of Coca Cola Consumption on Urinary Crystals Formation and Some Blood Biochemical Parameters

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<u>Abstract</u>

Coca cola consumption is widely distributed in recent years, therefore the study aimed to detect the side effects of coca cola intake on urinary crystals formation and some blood biochemical parameters including serum total calcium, ionized calcium, phosphorus ,uric acid and albumin. Blood and urine samples were collected from healthy volunteers who are divided into three groups :group 1 who drink 1 can (200ml) coca cola daily. Group 2 who drink 2 can or more (400ml) coca cola daily. These results compared with control(group 3) who didn't drink soft beverages . All parameters were determined in blood samples using spectrophotometer while urine samples examined under microscope. The results revealed significant (p<0.01) increase in serum phosphorus ,albumin and uric acid in group 1 and 2 compared to control group and significant decrease in serum total, corrected and ionized calcium in group 2 compared to control group. When compare between group1 and 2 the results revealed significant differences in serum total, corrected, ionized calcium and uric acid .Urine examination results appeared 39.1% of uric acid crystal and 30.4 % of amorphous urate crystals in group 1 while in group 2 drinking coca cola contribute to increased combined crystal formation which recorded uric acid with amorphus urate crystals 30% followed 15% of each Calcium oxalate with triple phosphate and Uric acid with triple phosphate.



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Conclusion: drinking coca cola in large quantities for long period effect ionized calcium and may lead to osteoporosis, kidney stone formation and decline in kidney function. **Keywords**:coca cola, calcium, phosphorus, uric acid, urinary crystals.

تأثير استهلاك الكوكا كولا على تكوين بلورات في الادرار وعلى بعض المعايير الكيوحيوية في الدم اسراء أنور محمد جمعة كلية التقنية-كركوك /قسم التحليلات المرضية/كركوك

الخلاصة

نظرا لاستهلاك الكوكا كولا بصورة واسعة في السنوات الاخيرة . فقد هدفت هذه الدراسة الى التحري عن التأثيرات الجانبية لاستهلاك كوكا كولا على تكوين بلورات الادرار وعلى مستوى الكالسيوم الكلي والمتأين ،الفوسفور،حامض اليوريك،والالبومين الدم. تم جمع عينات الدم والادرار من اشخاص اصحاء وتم تقسيمهم الى ثلاث مجاميع :المجموعة الاولى الذين يشربون علبة واحدة (200مل)يوميا ،المجموعة الثانية الذين يشربون علبتين او اكثر يومياوتمت مقارنتها مع مجموعة السيطرة. وتم تقدير تركيز الكالسيوم الكلي والمتأين ،الفوسفور،حامض المربون علبتين او اكثر يومياوتمت معاريقة طيفية بأستخدام المطياف الضوئي اما عينات الادرار فقد تم فحصها تحت المايكروسكوب .

أظهرت النتائج زيادة معنوية (p<0.01) في تركيز الفوسفور اللاعضوي ، حامض اليوريك والالبومين في المجموعتين الاولى والثانية مقارنة بمجموعة السيطرة ونقص معنوي في تركيز الكالسيوم الكلي والمصحح والمتأين في المجموعة الثانية مقارنة مع مجموعة السيطرة. خلال فحص عينات الادرار اظهرت النتائج تكوين 39.1% من بلورات حامض اليوريك و 30.4% من بلورات السيطرة. خلال فحص عينات الادرار اظهرت النتائج تكوين 39.1% من بلورات حامض اليوريك و 30.4% من بلورات المتعادية في المجموعة الاولى بينما في المجموعة الثانية أدت الى تكوين البلورات المركبة والتي سجلت بلورات حامض اليوريك مع يورات امورفس نسبة 30% تلتها 15% كلا من بلورات اوكز الات كالسيوم مع فوسفات الثلاثي وبلورات حامض اليوريك مع فوسفات الثلاثي .

الاستنتاج:شرب الكوكا كولا بكميات كبيرة لمدة طويلة يمكن ان يؤدي الى هشاشة العظام ،وتكوين حصى في الكلى ونقص في وظيفة الكلية.

الكلمات المفتاحية: كوكا كولا ،كالسيوم ، الفوسفور ، حامض البوليك ، بلور ات الادر ار .



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Introduction

Soft drinks and beverage has an increasing percentage of use and Cola is one of the common commercial drink that is worldwide distributed (1). Owing to increasing percentage of soft drinks and beverages energy intake, there are public health rising due to increased beverages and soft drinks consumption (which are sugar-sweetened) and relation to obesity, and other metabolic disturbance (2, 3). Cola is a worldwide popular soft drink which contains sugar, caffeine, phosphoric acid, and citric acid (4,5). According to the '2008 Global Soft Drinks' report, America alone represents 25% of the total beverage market energy intake consumption of more than 50 billion liters of soft drinks per year (6). Developing countries like those in Asia, Latin America and Africa represent the largest growth markets for soft drink producers. Consumers in 7 developing countries are being targeted by the soft drink industry with aggressive marketing often viewed by children (6). According to studies cola side effects may be due to excessive sugar intake which lead to obesity in addition to side effects of caffeine and phosphoric acid which contribute to subsequent chronic conditions including urinary stones(7).

The consumption of soft drinks is increasing constantly, especially among younger age group and become part of their eating habits . In general these soft drinks are rich in caffeine and artificially flavored and sweetened in addition to preservatives and high calories. Coca-Cola is the name of a multinational company in Atlanta and it is non-alcoholic soft drink (8). Nowadays daily consumption of coca cola estimated more than 1.5 billion drinks in comparison to early years where 9 drinks were sold per day (8). In general the main characteristics of most soft drinks are high sugar level, carbon dioxide, acidity and artificial sweeteners and Coca-Cola is one of them (9). The bubbles and froth which are characteristic of carbonated beverages arising from dissolved carbon dioxide in water under pressure(9). A decrease in renal function is expected among individuals who consume a lot of artificially sweetened drinks , especially when there is a consumption of more than two beverages a day. The metabolism of sweeten soft drinks may lead to hyperuricemia leading to formation of



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uric acid crystals in the kidney and Cola drinks lead to increased demineralization of calcium from bones and consequently the formation of insoluble calcium stones in the kidney. The risk of developing kidney stones increase by 15 percent in persons who drinking three cans of soft drink per week (10).

There is more than 10 teaspoons of sugar in each 8-ounce of *Pepsi*. It is hard to realize that it is so harmful to health in spite of so good tastes and so common . Intake of phosphoric acid containing soft drinks (like colas) causes an elevated level of phosphorus in the blood. Furthermore, increased intake of sugar from highly caloric soft drinks lead to impairment of kidneys reabsorption causing increased loss of calcium, chromium, copper, zinc, magnesium, and sodium . The parathyroid hormone (PTH) will be activated due to low calcium level in the blood causing release of calcium from the bones leading to osteoporosis, arthritis, bursitis, and gout.(11).

Ionized calcium has significant biological roles in bone homeostasis and it is physiologically active fraction of serum calcium that facilitates the nerve impulse transmission, interferes in the blood clotting ,couples the excitation with the muscle contraction, reduces the membrane permeability and control of hormone secretions such as vitamin D3 and parathyroid hormone, activates metabolic and digestive enzymes , influences the utilization of iron (12). Only 10% of the protein-bound calcium is linked to a variety of globulins while 90% is bound to albumin, therefore the interpretation of total calcium depends on serum albumin and total protein (13). The purpose of the study is to estimate the side effects of coca cola consuming in large quantities on urinary crystals formation and their effect on some serum biochemical parameters including serum total calcium ,phosphorus, uric acid, albumin and ionized calcium.

Materials and Methods

During the period from the beginning of August to the end of October 2015, out of 55(34male and 21 female) volunteers in Kirkuk city with no clinical history of any disease and no history of renal stone and their ages 15-40 years were enrolled in the present study. The participant population were classified into three fundamental groups,



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A-Group 1 (n=20) who drink 200ml(1 can) of coca cola daily.

B-Group 2 (n=20)who drink 400ml(2 can) of coca cola daily.

C- Group 3Control (n=15) who didn't drink soft beverage.

Urine samples and three milliliters of venous blood samples were collected from each group without application of tourniquet.

Microscopic urine examination

It involves the collection of 10 ml of first voided morning specimen of urine in a sterile wide mouthed container after taking necessary precautions. The sample was analyzed in the laboratory after centrifugation at 2000 rpm for 5 minutes, after draining the supernatant, a drop of the sediment was placed on a clean glass slide covered it with the cover slip. Then was studied under normal microscope first under low power &then under high power lens. It can be analyzed under contrast phase microscopy equipped with polarized filters for clear demonstration of crystals. (14)

Blood Examination: Blood Samples were transported into new plane tube, left for 15 minutes at room temperature for clotting, then centrifugated at 2000 rpm for 10 minutes, the separated serum was transferred into Eppendrof tube, the tubes were stored at -20c° until analysis.

The concentration of serum albumin was estimated using modified bromocresol green colorimetric method who described by Doumas et al. (15). The assay for total calcium was carried out colorimetrically using a kit supplied by biomerieux company/ France .(16), serum phosphorus was determined by Gamst (17) and serum uric acid by uricase(18) are preformed using a kit supplied by (biomerieux company/ France) by spectrophotometer in Technical college biochemistry lab.

As the ionized calcium that is physiological active and with clinically important ,therefore total calcium was used to calculate the ionized calcium according to the formula have been based upon plasma albumin(19):

Free or ionized Ca+² (mg/dl)= Ca measured in (mg/dl) –[albumin g/dl+4]/2



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In hypo or hyperalbuminemia corrected total calcium value was calculate ,the same data was used according to the formula(18):

Corrected total Ca(mg/dl)=total Ca(mg/dl)+0.8(4-albumin g/dl)

Statistical Analysis: The results were described as percentage, mean \pm standard deviation, Chi square and t-test was used for the comparison between the participants and control groups in the measured parameters were calculated using Microsoft Excell 2007 program. The results considered statistically significant when 'p' value < 0.05.

Results

The results showed drinking coca cola daily lead to formation of uric acid crystals with 39.1% followed by amorphous urate ,triple phosphate and calcium oxalate with percentage 30.4%,17.3 and 13% respectively (table1).

Table 1- Types and numbers of urinary crystals appeared in 20 individuals in group 1.

Type of urine crystals	No. of crystals	%	
Uric acid	9	39.1	
Amorphous urate	7	30.4	
Triple phosphate	4	17.4	
Calcium oxalate	ASITI COLLEGE	13.1	
total	23	100	

Types of crystals according to age in group 1 revealed significant results with high incidence of uric acid and triple phosphate crystals at age 20-25 years old , while at age 26-30 years appeared amorphous urate and uric acid crystals (Table 2).



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Table 2- Types of urinary crystals found in group 1 according to age

Age	Uric acid	Amorphous urate	Triple phosphate	Calcium	
				oxalate	
20-25	5	1	3	1	
26-30	2	4		1	
31-35	2	2	1	1	

(p< 0.05) Chi square=21.07

The results in Table 3 demonstrated that drinking coca cola two can or more contribute to increased total number of crystals into 38 crystals in group 2 compared to 23 crystals in group 1 and increased combined crystal formation in group 2 which recorded uric acid+ amorphus urate crystals with 30% followed 15% of each calcium oxalate+ triple phosphate and uric acid +triple phosphate

Table 3-Types of urinary crystals appeared in 20 individuals in group 2.

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Type of crystals	No. of cases	%
Uric acid+ Amorphous urate	6	30
Calcium oxalate+ triple phosphate	3	15
Uric acid +triple phosphate	3	15
Calcium oxalate + Uric acid	2	10
Calcium oxalate + Amorphous urate	2	10
Amorphous urate+ triple phosphate	2	10
Uric acid	1	5
Amorphous phosphate	1	5



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The results in Table 4 revealed uric acid crystals appeared at age 20-30 and 36-40years old, Amorphous urate crystals at age 20-30years old ,triple phosphate at ages 20-25 and 36-40 years old ,while calcium oxalate crystals approximately at ages 20-40 years old .

Table 4- Types of urinary crystals present in group 2 according to age.Chi square=30.7 (p<0.05)</td>

Age	Uric acid	Amorphous	Amorphous	Triple	Calcium
years		urate	phosphate	phosphate	oxalate
20-25	6	4	A A	4	2
26-30	3	5	1	5	1
31-35	6	1		1	2
36-40	3	C		3 2	2

The present study focused on effect of coca cola drinking on some biochemical parameters which revealed significant(p<0.01)increase in serum uric acid, albumin and phosphorus in group 1 and 2 compared with control group. Effect of coca cola on serum total, corrected ,ionized calcium revealed no significant results in group 1 while group 2 appeared significant decrease on corrected total and ionized calcium. When compare between group1 and 2 the results revealed significant differences in serum total ,corrected ,ionized calcium and uric acid ,this mean quantity of coca cola drinking affect serum calcium and uric acid .



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Table -5 – Some biochemical parameters in people consumed coca cola in 1 and 2groups comparison with control group .

parameters	Group 1	Group 2	Control	p- value	p-value	p-value
	No.=20	No.=20	No.=15	group1 & control	group2 &control	group1 & 2
Total serum Ca (mg/dl)	8.39±2	6.1±1.7*	7.7±2.07	0.1	0.03	0.001
Corrected total calcium(mg/dl)	7.3±1.8	4.49±1.6*	7.3 ±1.9	0.4	0.001	0.000
Serum ionized Ca (mg/dl)	3.75±1.2	2.3±0.8*	3.8±1.3	0.4	0.007	0.000
Serum albumin (g / dl)	5.4± 0.8 **	5.2±0.5**	4.3±0.53	0.000	0.000	0.14
Serum uric acid (µmol/l)	259.3±34.4*	284.7±41.2*	218±36.1	0.04	0.005	0.03
Serum phosphorus (mmol/l)	3.12±1.61**	3.2±1.8**	1.7±1	0.000	0.001	0.4

Values are given as mean \pm SD; No.=number * (p<0.05) **(p<0.001)

Discussion

In calcium metabolic disturbance, clinical studies showed that free calcium is a better indicator than total calcium regarding diseases such as multiple myeloma, acute acid-base disturbance, dysproteinemia, hyperparathyroidism, renal stones and renal insufficiency(20) therefore ionized calcium also was measured in this study.

The present study revealed coca cola intake daily lead to significant decrease in serum total calcium, corrected ,ionized in group 2 and significant increase in serum phosphorus in groups 1 and 2 compared to control group ,this could be explained that during the formation of new



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bones and remodeling of old bones, the body maintains a steady phosphorus and calcium ratio in the bloodstream. Intake of phosphoric acid containing soft drinks (like colas) causes an elevated level of phosphorus in the blood (11). Furthermore, increased sugar intake from the highly caloric soft drinks affect impair renal reabsorption system leading to increased urinary loss of calcium, magnesium, chromium, copper, zinc, and sodium. So parathyroid hormone (PTH) is activated by low level of calcium in the blood and this leads to the release of calcium from the bones to maintain the balance. Over the time end results of this present is osteoporosis (weakened bone structure) so the bone is fragile and more liable to fracture due to deficiency of calcium or hormonal change or vitamin D deficiency (22). The elimination of excess calcium in the blood is by increased urine excretion of calcium leading to kidney stones formation and calcified plaque and deposition of calcium in joints leads to osteoarthritis, bursitis and gout. Therefore with less calcium available, the bones become more porous and prone to fracture (11). The tolerable upper limit of phosphorus in children is 3-4 grams. A 12-ounce can of carbonated soft drink contains phosphoric acids averaging about 30 milligrams. Even this low amount can be damaging if consumed in excess especially during the peak bone-building years of childhood and adolescence. Phosphoric acid in the drink can leach toxic amounts of aluminum (from the can) into the soft drink. Exposure to Aluminum causing a decrease in new bone formation and increases in bone breakdown with excessive loss of calcium in the urine resulting in osteoporosis associated with severe bone pain (10,21).

The results reported significant increase in serum uric acid and albumin in groups 1 and 2 compared to control group. Persons who drink a lot of artificially sweetened drinks are more liable to suffer from a decrease in kidney function which lead to increase in serum albumin and uric acid; especially when there is a consumption of more than two beverages a day. Cola drinks can lead to increased demineralization of calcium from bones leading to formation of insoluble calcium stones in the kidney. The metabolism of high-fructose corn syrup used to sweeten soft drinks may lead to hyperuricemia leading to formation of uric acid crystals in the kidney. the risk of developing kidney stones increase by 15 percent in persons who drinking three cans of soft drink per week (10,11,27).The study appeared 39.1% of uric acid crystal and 30.4 % of amorphous urate crystals in group 1 while in group 2 contribute to increased combined



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crystal formation which recorded uric acid with amorphus urate crystals with 30% followed 15% of each calcium oxalate with triple phosphate and uric acid with triple phosphate . Clinical studies regarding correlation between long-term ingestion of cola beverages and the risk of stone formation are currently ongoing and no definite results can be obtained (23, 24, 25) . For example, (Ferraro et al.) showed that intake sugar sweetened cola had a 23% higher risk to develop renal stones, whereas consumption of artificially sweetened cola was associated with a trend to reduce the risk (25). Rodgers group in one such study analyzed the biochemistry of volunteers urine who ingested 2 liters of cola regularly per 24 hr. and their findings were unfavorable changes to risk factors associated with calcium oxalate (CaOx) stone formation (24). One of the major contents of cola is caffeine and it has reported previously for responsibility of CaOx stones formation [26].

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