Determination of Some heavy metals in Different vinegar Samples Applied in folk medicine by flame atomic absorption Spectrophotometry

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Abstract:

A study was carried out to analysis of some heavy metals in nine different types of vinegar, belong to Grape, Apple, Synthetic White, Date, Hawthorn, Garlic, Cactus, Pomegranate and Ginger vinegar, which are locally available in Iraqi folk medicine markets. The concentrations of heavy metals in the studied samples including, Cr, Mg, Mn, Zn, Fe, Cd, Ni, Pb and Ag, were determining by using flame atomic absorption spectrophotometry. All data were subjected to statistical analysis by calculating accuracy, precision and correlation coefficient for each concentrations level. The results indicate that Ni was recorded the highest concentration in all studied samples except, Ginger and cactus vinegar, each one receded the highest concentration value of Mn(8.520µg.mL^{-1}) and Fe (3.500µg.mL^{-1}) respectively. Also it can be noticed that Ni shows the highest total concentration value (26.250µg.mL^{-1}), while Cr recorded the lowest total concentration value (0.347µg.mL^{-1}). The concentrations of each metal studied were in the limits of health intake and not considered a health risk.

Key word: heavy metals, Vinegar, Folk medicine, Flame atomic absorption.

Introduction:

Interest in vinegar minerals has recently increased, because they can be used as an index of processes that affect the properties and healthy aspects of vinegars, such as the production process, environmental pollution, sanitary conditions and the quality of the raw material [1].

Vinegar is a stringent product [2], important fermented condiment [3], becoming more and more popular throughout the world [4]. It is produced by alcoholic fermentation or from chemical synthesis from natural gas and petroleum derivatives, resulting in a highly concentrated acetic acid solution [5]. It has been used for thousands of years as both a condiment and a preservative [2]. In folk medicine traditions, it has been used to promote calcium absorption

[6], antihypertensive (blood pressure regulator) [7], blood glucose regulator [8], stimulate the appetite [9], and regulate aid digestion [10].

Minerals play an important role in human nutrition, because they are not synthesized in the body [11], the human body requires both metallic and nonmetallic elements within certain permissible limits for growth and good health. Therefore, determination of element compositions is essential for understanding their nutritive importance [12].

In recent years, there has been increasing interest in evaluating the macro (major) elements and micro (minor/trace and-ultra-trace) elements in a variety of food samples [13]. Heavy metals which are also called trace element due to their presence in trace or in ultra trace quantities in the environmental matrices [14], are individual metals and metal compounds that can impact human health [15], contaminated of heavy metals in vinegar might be due to used raw material or the procedures during processing and production, on the other hand it may be coursed from endogenous or anthropogenic origin [16]. The most toxic effect of heavy metals results from potentially toxic elements such as lead, silver and cadmium. The potentially toxic metals can be health threading either at low or high concentration when ingested by humans [3]. Therefore metals or heavy metals can play a harmful at high concentrations [11]

Several attempts have been used to determine metals in vinegar [1, 3, 17], soil [18], blood [19], water [20], plants and foods [13, 16] Atomic absorption spectropotometry are popular for their sensitivity in the assay of elements and hence, considerable attentions for the quantitative determination of many elements.

In this present work, nine vinegar samples belong to Grape, Apple, Synthetic White, Date, Hawthorn, Garlic, Cactus, Pomegranate and Ginger vinegar were collected from Iraqi folk medicine markets and analyze the concentration of its contents of heavy metals including, Cr, Mg, Mn, Zn, Fe, Cd, Ni, Pb and Ag by using flame atomic absorption spectrophotometer.

Apparatus:

Mineral concentrations, Cr, Mg, Mn, Zn, Fe, Cd, Ni, Pb and Ag were determined by using Phoenx-986, flame atomic absorption spectrophotometer (air/acetylene flame), K-PLC Series Centrifuge, Sartorius BL 210S balance, and a Pentium 4 computer (DELL 1545) were used for data processing.

Experimental:

Material and Reagents:

All chemicals used analytical reagent and grade, Analytical grade nitric acid (65% Merck) was used for the mineralization of the samples [1]. Analyte stock solutions containing 1000 μg.mL^{-1} of the individual elements. Distilled and deionized water was used for dilution.

Samples:

Nine vinegar samples, representing the common types of vinegars used in folk medicine and readily available to consumers included, Grape, Apple, Synthetic White, Date, Hawthorn, Garlic, Cactus, Pomegranate and Ginger vinegar, were obtained from Iraqi folk medicine markets, (herbs markets). The majority of the vinegars were in glass bottles; however, several were in plastic bottles. Sample containers were (100-500 mL) and an identification code was assigned for each sample. It Were stored in refrigerator at 4±1°C prior to analysis. Each sample was re obtained from the same herbs markets after one month; this procedure was utilized to include different batches of each product.

Procedure for analysis of minerals in vinegar samples:

The vinegar samples were diluted 1:1with 0.2% (v/v) HNO_3 and centrifuged for 20 mints at 2000 rpm. All sample cups, and glass-ware were cleaned by soaking in 10% (v/v) HNO_3 and dilute with de-ionized water prior to use [1]. The appropriate standards for each element were made within the concentration range of the elements in the samples. The results were obtained from triplicate measurements.

Evaluation of matrix effects:

Signal suppression or enhancement because of matrix effects can severely compromise quantitative analysis of the metals. Consequently, the matrix effects must be evaluated and discussed in the context of method development before method development. The matrix effect was evaluated using the following two sets solutions. Set A, standard solutions in calibration. And set B, solutions of real vinegar samples. The absence or presence of matrix effects was evaluated using the ratio of the signal for the two sets of samples. This ratio ranged from 97.3-101.2%, which indicates that there are no matrix effects [17].

Statistical analyses:

All obtained data were subjected to statistical analysis that was performed by using statistical software (STATISTICA 6.0), Stat-Soft Co. USA and Excel program office.

Results and discussions:

Information on the metal content in vinegar is important to evaluate the potential human health risks of its consumption, and the quantification of these elements can serve as a tool to characterize vinegar quality and authenticity. Therefore, in this respect the concentrations of heavy metals such as Cr, Mg, Mn, Zn, Fe, Cd, Ni, Pb and Ag, were determining by using flame atomic absorption spectrophotometer, in nine different kinds of vinegar samples which are used in Iraqi folk medicine. These minerals play an important role in the treatment of diseases, but it is when founds in execs concentrations becomes toxics and harmful for human body also deficiency of each one will coursed many diseases and problems in humans growth. Table 1 shows the concentrations of each metal in each vinegar sample.

Chromium is considered an essential nutrient and a health hazard. Trivalent chromium is an epigenetic carcinogen factor since it can form stable compounds with macromolecules, Such as DNA and glutathione [21]. According to Food and Drug Administration (FDA), Cr concentration should not exceed 1 milligram per liter (1ppm) in bottled water [15]. Figure1 shows that the concentrations of Cr were between (0.009-0.050) $\mu g.mL^{-1}$ in all studied samples, the highest concentration value in Grape vinegar, while the lower concentration value in Synthetic White vinegar.

Magnesium is one of the most essential mineral in human body [22], has an important function in many biochemical and physiological process, which directly effects human Lung function, joint pain, smoothening of muscle function, neuromuscular excitability, immune function, inflammation, cardiovascular disease, blood sugar and blood pressure regulator[23, 24]. Figure 2 shows that the concentrations of Mg were between (0.060-0.630) $\mu g.mL^{-1}$.

Manganese is a common element in the earth's crust, water, and particulate matter in the Atmosphere [25]. It considered as essential element for normal life processes. Mg safe intake requirements are 2.5 to 5.0 mg/day [26]. Toxicity of manganese called "manganese-induced Parkinsonism," because some of the symptoms are similar to those seen in cases of Parkinson's disease [27]. Figure 3 shows that highest concentration of Mn in Ginger vinegar (8.520$\mu g.mL^{-1}$). The concentrations of Mn were between (0.062-8.520) $\mu g.mL^{-1}$ in all samples studied.

Zinc is necessary for the growth and multiplication of cells [28]; it is involved in the activity of about 100 enzymes of human body. There is 2–3 g of Zn present in the human body and about 1 mg/L in plasma [29]. Zn deficiency causes loss weight [28], it is common in underdeveloped countries (mainly associated with malnutrition), affecting the immune system, wound healing, the senses of taste and smell. [30]. Figure 4 shows that Ginger vinegar recorded the highest concentration of Zn (4.330$\mu g.mL^{-1}$), while Synthetic White recorded the lowest value of Zn (0.140$\mu g.mL^{-1}$).

Iron plays an important role in the metabolic process. The role of iron in the body is clearly associated with hemoglobin and the transfer of oxygen from lungs to the tissue cells [31]. Serum Fe is about 1.3 mg/L, Fe deficiency causes anemia. Chronic Fe intoxication occurs frequently associated to genetic and metabolic diseases, repeated blood transfusions, or excessive intake [32]. Figure 5 show that Cactus vinegar contains the highest value of Fe (3.500$\mu g.mL^{-1}$), while Hawthorn vinegar contains the lowest value of Fe (0.210$\mu g.mL^{-1}$).

Cadmium is a very toxic metal and carcinogens [15]. This metal causes adverse health effects in human and their presence in the human environment comes from anthropogenic activities [33]. Smokers get exposed to significantly higher Cd levels than non-smokers. Severe damage to the lungs may occur through breathing high levels of Cd, kidney disease and fragile bones. According to FDA health intake of drinking water should not exceed (5.000μg.mL^{-1}) of Cd [14, 15]. Figure 6 shows that the concentrations of Cd were between 0.031μg.mL^{-1}(Synthetic White) and 0.060μg.mL^{-1}(Hawthorn vinegar) in all studied samples.

Nickel is an essential element for animal nutrition. The Nickel requirement of humans has been estimated to be (25-35) μg/day [34]. Ni is a toxic mineral when present in execs amount because it induces carcinogenesis through several processes including DNA hypermethylation [35]. Figure 7 shows that Hawthorn vinegar recorded the highest value of Ni (4.600μg.mL^{-1}), while Grape and Cactus vinegar contains the lowest value of Ni (2.400μg.mL^{-1}).

Lead is the most common industrial metal that has become widespread in air, water, soil and food[14], so it is very toxic for the nervous system, kidneys and others tissues of the body [36]. The recommended intake (safe intake) of lead 15-280μg/day for adults and10-275μg/day for children [14]. Figure 8 shows that the concentrations of Pb were between (0.200-1.250) μg.mL^{-1} in all studied samples.

Silver, is not classifiable as a human carcinogen, but when presence in high level may result in a condition called arygria, a blue-gray discoloration of the skin and other body tissues. Exposure to high levels of Ag in the air has resulted in breathing problems, stomach pains, lung and throat irritation. Most food contains traces of silver in the range of 10-100μg/kg [37]. Figure 9 shows that the concentrations of Ag were between 0.033 μg.mL^{-1}(Ginger vinegar) to 0.044 μg.mL^{-1} (Synthetic White and Pomegranate vinegar).

The variation in elements concentrations of vinegar samples due to natural composition of the raw materials, the constituents formed during fermentation, contact with production and storage equipment, concentration of the production environment and sometimes, the substances formed during the aging process[2].

In this study we can observed that the lowest concentrations values for Mn and Ag will appears in Garlic and Ginger vinegar respectively, as well as grape and cactus vinegar recorded the lowest concentrations values for Ni (2.400μg.mL^{-1}), so as Cr, Zn, Cd, Pb and Mg, Fe recorded the lowest concentrations values in Synthetic White and Hawthorn vinegar respectively. While the highest concentrations values for Cr, Fe and Pb will appears in grape, cactus and Date vinegar respectively, in addition to, hawthorn and ginger Vinegar will recorded the highest concentrations of Cd, Ni and Mn, zn respectively, also it is notice that the highest concentrations values of Ag (0.044 $\mu g.mL^{-1}$) will appears in Synthetic White and Pomegranate vinegar.

Generally, Manganese recorded the highest concentration value (8.520 $\mu g.mL^{-1}$), while Chromium recorded the lowest concentration value (0.009$\mu g.mL^{-1}$).Totally, Nickel shows the highest total concentration value (26.250$\mu g.mL^{-1}$), while Chromium recorded the lowest total concentration value (0.347$\mu g.mL^{-1}$) (Figure 10).

Accuracy and precision:

The accuracies of the proposed methods were confirmed by analyzing three replicate analyses for each sample by calculating the relative error percentage (RE %), which were found to be in the range of 0.958-2.609%. The precision was determined in each case by calculating the percentage relative standard deviation (RSD %) for three determinations at each of the studied vinegar samples and were found to be in the range of 0.865-3.121%. The results indicated good accuracies and precisions of the proposed method. (Table 2).

The method limit of detection and method limit of quantitation:

The method limit of detection (MLOD), which set as $3 \times RSD_{MB} \times P_{BEC}/100$ and method limit of quantitation (MLOQ) which set as $10 \times RSD_{MB} \times P_{BEC}/100$, in addition to coloration coefficient(R), were also calculated (table 3), were, RSD_{MB}, is the relative standard deviation for five measurements of the blank, P_{BEC} is the background equivalent concentration [38,39]. The results indicated of the high sensitivity of the proposed method, and all metals could be quantified, because their concentrations were highest than the MLOQ [17].

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Table1: Concentrations (μg.mL^{-1}) of each metal in each vinegar samples

No	Vinegar Sample	Metal								
		Cr	Mg	Mn	Zn	Fe	Cd	Ni	Pb	Ag
1	Grape	0.050	0.410	0.390	0.430	1.500	0.034	2.400	0.240	0.040
2	Apple	0.043	0.280	0.610	0.190	0.230	0.033	2.620	0.230	0.040
3	Synthetic White	0.009	0.110	0.110	0.140	0.230	0.031	2.760	0.200	0.044
4	Date	0.033	0.410	0.330	0.750	0.610	0.035	2.800	1.250	0.041
5	Hawthorn	0.040	0.060	0.100	0.500	0.210	0.060	4.600	0.240	0.035
6	Garlic	0.048	0.110	0.062	0.920	0.330	0.051	2.430	0.620	0.037
7	Cactus	0.046	0.630	0.920	0.760	3.500	0.034	2.400	0.620	0.036
8	Pomegranate	0.045	0.310	0.940	0.250	0.230	0.032	3.100	0.730	0.044
9	Ginger	0.033	0.240	8.520	4.330	1.800	0.033	3.140	0.220	0.033

Table 2: Evaluation of accuracies and precisions of each metal in all vinegar samples

R.S.D.* %	Relative Error %	Metal concentrations(μg.mL^{-1})			Metal
		range	mean	Total	
0.865-1.321	1.111-2.543	0.009-0.050	0.039	0.347	**Cr**
1.436-1.806	1.414-1.735	0.060-0.630	0.284	2.560	**Mg**
1.812-2.223	2.300-2.609	0.062-8.520	1.331	11.982	**Mn**
2.592-2.891	1.500-1.831	0.140-4.330	0.919	8.270	**Zn**
2.993-2.628	0.984-1.626	0.210-3.500	0.60	8.640	**Fe**
2.471-2.395	1.935-2.405	0.031-0.060	0.042	0.343	**Cd**
2.513-3.121	0.981-1.776	2.400-4.600	2.917	26.250	**Ni**
1.635-1.885	1.414-1.749	0.200-1.250	0.483	4.350	**Pb**
0.994-1.829	0.958-1.581	0.033-0.044	0.039	0.350	**Ag**

*Average of three determinations.

Table 3: Regression coefficient, method limit of detection and method limit of quantitation for each metal in all vinegar samples

MLOQ* ($\mu g.mL^{-1}$)	MLOD* ($\mu g.mL^{-1}$)	Regression Coefficient (R)	Metal
$1.320x10^{-4}$	$3.960x10^{-6}$	0.9996	**Cr**
$3.960x10^{-4}$	$1.188x^{10-5}$	0.9964	**Mg**
$3.840x10^{-4}$	$1.152x10^{-4}$	0.9999	**Mn**
$3.360x10^{-4}$	$1.008x^{10-4}$	0.9993	**Zn**
$1.620x10^{-3}$	$4.860x10^{-4}$	0.9999	**Fe**
$3.240x10^{-4}$	$9.720x10^{-5}$	0.9991	**Cd**
$2.661x10^{-2}$	$7.985x^{10-3}$	0.9994	**Ni**
$3.612x10^{-3}$	$1.084x10^{-3}$	0.9987	**Pb**
$3.120x10^{-4}$	$9.360x10^{-5}$	0.9999	**Ag**

*Average of five determinations.

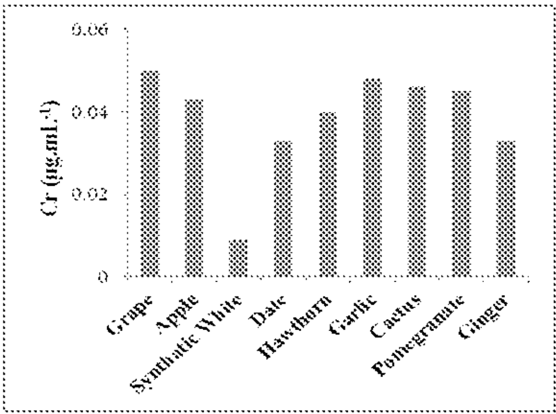

Figure1: Concentration of Cr in Vinegar samples studied.

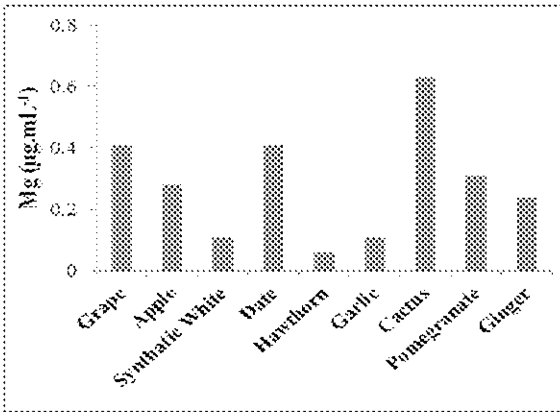

Figure2: Concentration of Mg in Vinegar samples studied.

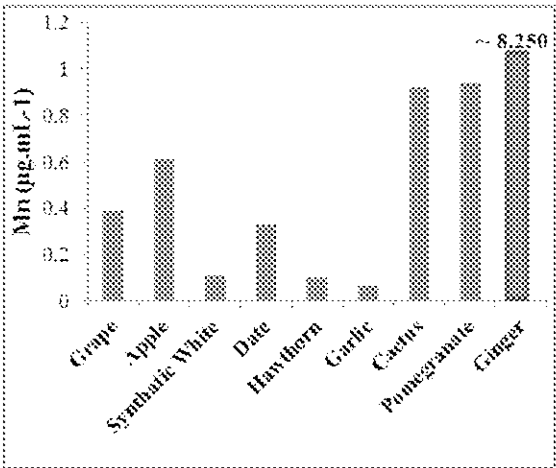

Figure3: Concentration of Mn in Vinegar samples studied.

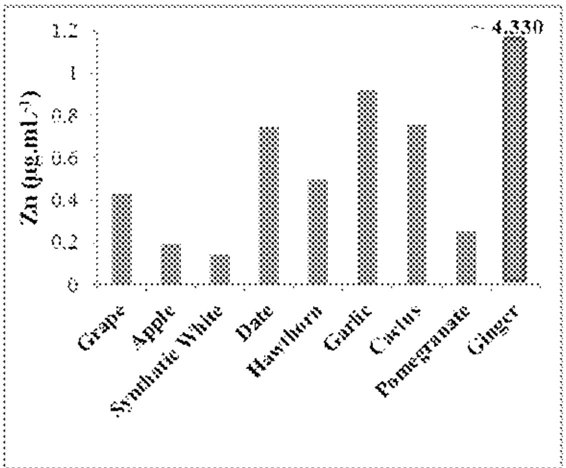

Figure4: Concentration of Zn in Vinegar samples studied.

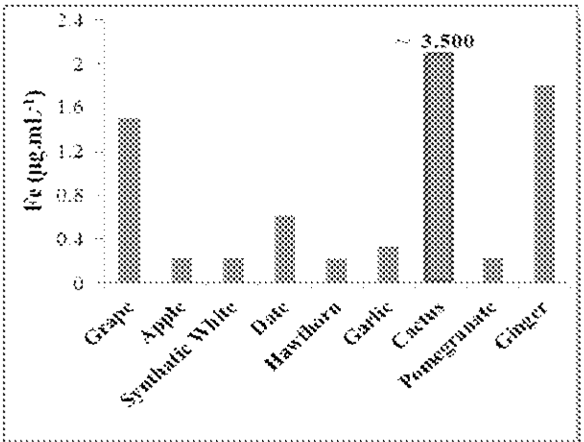

Figure5: Concentration of Fe in Vinegar samples studied.

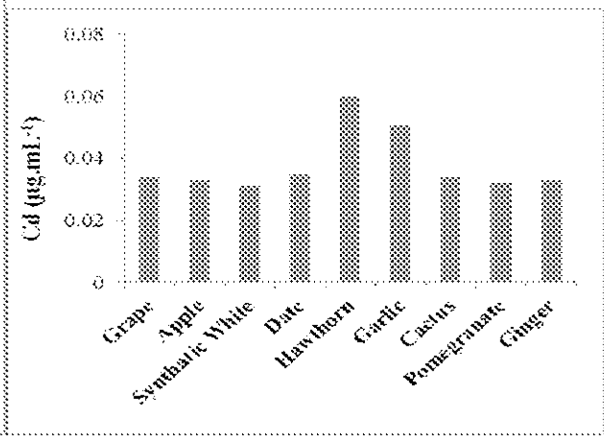

Figure6: Concentration of Cd in Vinegar samples studied.

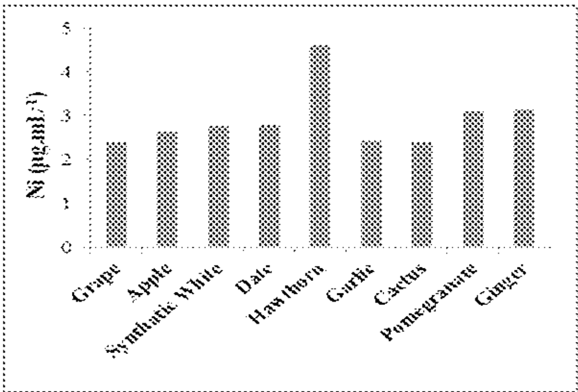

Figure7: Concentration of Ni in Vinegar samples studied.

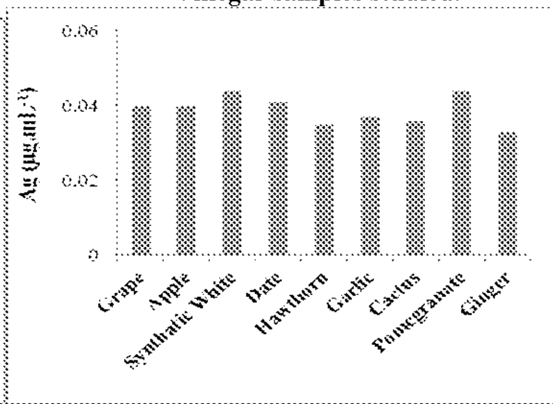

Figure8: Concentration of Pb in Vinegar samples studied.

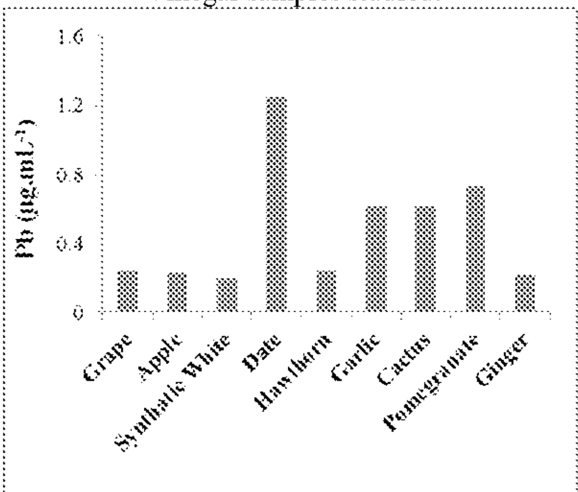

Figure9: Concentration of Ag in Vinegar samples studied.

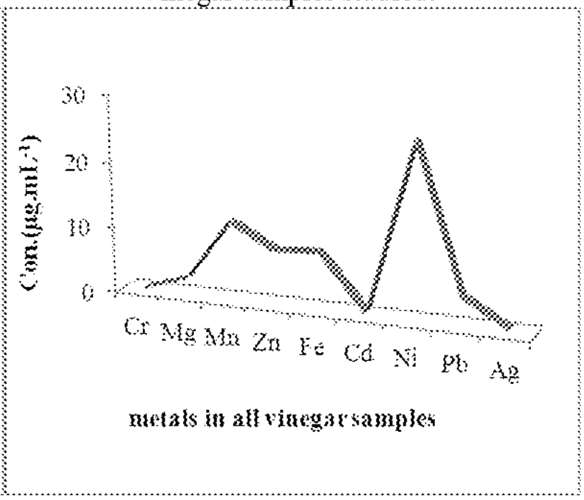

Figure10: Total concentration of each metal ($\mu g.mL^{-1}$) in all Vinegar samples studied

تقدير بعض العناصر الثقيلة في نماذج مختلفة من الخل المستعمل في الطب الشعبي بأستخدام مطيافية الامتصاص الذري اللهبي

علي خليل محمود
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الكلمات المفتاحية: العناصر الثقيلة، الخل، الطب الشعبي، الامتصاص الذري اللهبي.

الخلاصة:

تناولت هذه الدراسة تقديرتراكيز بعض العناصر الثقيلة في تسع انواع مختلفة من الخل المستخدم للاغراض الطبية (الطب الشعبي) في العراق ، والتي تضمنت خل العنب، التفاح، الخل الصناعي الابيض، التمر، الزعرور، الثوم، الصبار، الرمان و خل الزنجبيل. حللت العناصرالمدروسة (الكروم، المغنيسيوم، المنغنيز، الزنك، الحديد، الكادميوم، النيكل، الرصاص والفضة) بأستخدام مطيافية الامتصاص الذري اللهبي، وعولجت النتائج احصائيآ، حيث تم حساب قيم الدقة والضبط ومعامل الارتباط. تشير النتائج الى ان النيكل سجل اعلى التراكيز في النماذج المدروسة، ماعدا خل الصباروالزنجبيل، حيث اظهر كل منهما اعلى التراكيز لعنصري المنغنيز(8.520مايكروغرام/مل) والحديد (3.500مايكروغرام/مل). تم حساب مجموع التراكيز الكلية للعناصر في جميع النماذج المدروسة، والتي اظهر فيها النيكل اعلى قيمة (26.250 مايكروغرام/مل) في حين سجل الكروم اوطأ قيمة (0.347 مايكروغرام/مل)، بينت النتائج ان تراكيز العناصر المدروسة كانت ضمن الحدود المسموح بها.