Estimation of Cadmium Concentration in Blood Serum of Mothers and Umbilical Cord

Nadiaa Ahmed Salih[**] **Hussam Dawood Abdullah** **Wasan Nazhan Hussien**

Tikrit University- College of Education- Department of Chemistry

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Abstract

Determining the level of concentration of cadmium in the blood of the umbilical cord of neonatal and their mothers may serve as an indicator of the extent of the pollution of the environment with this element. It may also indicate the extent of the exposure to this element especially that this element has the capability of passing through placenta. This study aims at determining the concentration of (Cadmium, Zinc and Copper) and the concentration of Glutathione in the blood of the umbilical cord of neonatal and their mothers in Tikrit city and Arbeda town which is an agricultural town. For this study, 160 samples of mothers blood and of the umbilical cords of their neonatal were collected and 50 blood samples were collected as a control group. Metal elements were discovered in the blood of the mothers and their neonatal by using the atomic absorption spectrometer. The results of the current study showed that the rate of mothers and neonatal who have the cadmium in their blood was higher in Tikrit city than those in Arbeda town. The results also showed a significant decrease $P<0.05$ in the concentration of zinc and glutathione in the serum of pregnant women and the umbilical cord compared to the control group. An increase in copper concentration rate in the blood of pregnant women and umbilical cord compared to the control group.

Keyword: Cadmium , Zinc , Glutathione, Copper, Umbilical Cord, Neonatal .

Estimation of Cadmium Concentration in Blood Serum of Mothers And Umbilical Cord

Nadiaa Ahmed Salih Hussam Dawood Abdullah Wasan Nazhan Hussien

تقدير تركيز الكادميوم في مصل دم الأمهات والحبل السري

نادية احمد صالح** **حسام داود عبدالله** **وسن نزهان حسين**
جامعة تكريت كلية التربية قسم الكيمياء

الملخص

تم دراسة مستوى تركيز كل من العناصر المعدنية (الكادميوم، الخارصين ،النحاس) بالإضافة الى تركيز الكلوتاثايون في عينات مأخوذة من دم الحبل السري للأطفال حديثي الولادة وأمهاتهم ،اذ يعتبر مستوى تركيز الكادميوم في هذه العينات مؤشرا لمدى تلوث تلك البيئات بهذه المعادن ومدى تعرضهم لها خاصة وان عنصر الكادميوم له خاصية المرور عبر المشيمة. تم جمع 160 عينة دم للام والحبل السري لأطفالهم و50 عينة دم من النساء غير الحوامل كمجموعة سيطرة وقد تم جمع هذه العينات من مدينتي تكريت واربيضة لكون الثانية من المدن الزراعية .استخدم جهاز امتصاص الطيف الذري للكشف عن العناصر المعدنية في العينات المأخوذة . وأوضحت النتائج بان عنصر الكادميوم يتواجد بنسب اعلى في عينات مدينة اربيضة مقارنة بمدينة تكريت ، ولوحظ وجود انخفاض معنوي عند مستوى $P< 0.05$ لتركيز الخارصين والكلوتاثيون في مصل الأمهات الحوامل والحبل السري مقارنة مع مجموعة السيطرة ، وكما وجد أيضا ارتفاع بمستويات تركيز النحاس بدم النساء الحوامل والحبل السري مقارنة بمجموعه السيطرة .

الكلمات المفتاحية: الكادميوم، الزنك، الكلوتاثيون، النحاس، الحبل السري، الاطفال حديثي الولادة.

Introduction

Environmental pollution became one of the most dangerous problems not only on the third-world level but also on the whole world level. By its spread to the whole components of the ecosystem, environmental pollution became a crucial issue that human being coexist with it obligatorily. It is said that human being is now a refugee in his environment; its water, air, food, and clothing became polluted and sometimes poisonous. This happens in the time that the whole world suffers from a shortage in the sources of food and water, until human started to die not because of thirst and hunger, but due to suffocation by polluted air[(1)]. Until now, human being haven't encountered a danger of such enormity and spread, which is resulted from many factors each of which became enough to cause insolvable problems

which all together means that human pains will increase to a shocking extent in the near future. In the time at which humanity aims at securing its needs, although we don't doubt the effectiveness of the solutions for that, if we put aside our selfish personal interests, the environment in which we live now is deteriorating in an unprecedented fast way. This is obvious in some parts of the world. What affects a part of a particular position may affect other positions in the world. An examples on that are the pollution of the air and the atomic and nuclear pollution, transmission of the poisonous materials like heavy metals such as lead and cadmium in addition to the agricultural pesticides which are of the direct or indirect negative effects jointly or solely food chain of human being, the remnants of which were discovered in the remnants of the birds and animals that live in places away from areas in which these compounds are used[1,2] .In addition to the fast industrial and social development during the last four years, population and city expansion contributed to increasing the rate of pollution which affects the public health and the safety of the environment. So, caring for the environment and its variables is one of the collective duties that contribute to avoiding disasters and menaces which unintentionally harms the health of human being. In this way, the concerns of humanity are all about the environment pollution especially the pollution caused by heavy metals that result from either natural factors such as erosion factors and microscopic organisms, or of artificial factors as a result of industrial, agricultural, and civil wastes. This problem obtained a great attention of many researchers[2,3]. Heavy elements are found in the tissues of the organisms with very low rates. Some of these elements are necessary for the continuity of the life of organisms despite the fact that the concentrations of the needed elements is too little that it hardly amounts parts of a million. Furthermore, the increasing of the concentrations of the elements to certain rates inside the tissues of the organisms may be harmful or poisonous to these organisms[4]. Many studies conducted on the rate of heavy elements in the blood of the pregnant mothers and the extent of its ability to pass through the placenta, and the effects of some environmental factors such as smoking, residence, period of pregnancy and number of pregnancies on the pregnant mothers and their neonatal to show the extent of these factors on the existence of the rates of cadmium in the blood of the pregnant mothers and their neonatal . It was proved by such studies that smoking affects the rate of cadmium in the blood of the pregnant mothers and the umbilical cords and that the decrease of the weight of the infant is attributable to the smoking

of the pregnant mothers[5].Cadmium is one of the highly toxic and dangerous heavy metal that it has the ability to accumulate in the tissues of the plants and due to the similarity between Cadmium and Zinc, the plant may absorb the cadmium instead of the zinc necessary for the plant. In this way cadmium transmits into human through vegetables, fruits, and cereals[2,5]. Scientists consider this metal as the most dangerous metals in the food chain. It accumulates in the body of the organism especially the aquatic ones such as fishes and crustaceans then it moves from such organisms to humans through the food chain. Cadmium is found in the kidneys and livers of cows and sheep with high rates, so that people eating them have high rates of this element in their bodies[2,5]. Smoking increases the concentration of cadmium in the liquids of the body, its rate in the blood and sperms of the smoker is higher than that in the blood and sperms of non-smokers. Cigarette is often the source of cadmium in the air. The cigarette contains 1-2 microgram of cadmium and 10% of the cadmium found in the cigarette is inhaled. Cadmium is one of the modern poisonous metals and one of the important metals for many uses. It is used as a base for galvanizing surfaces and electro coat for its anti-scratch characteristics. It also enters into the formation paints and plastic in addition to its use for the anode of the Nickel and Cadmium batteries. Moreover, cadmium is considered as a byproduct from mines and from smelting Zinc and Lead. The permissible rate of cadmium in the blood of non-smokers ranges from 0.04 to 0.1 microgram/deciliter. Some studies revealed the toxicity of cadmium on the genital tissues that cadmium can pass through the placenta to affect the fetus[6]. Zinc is an important element in the meal in small amounts, a plenty of which causes harmful effects such as stomach spasm then nausea and vomiting[7]. Some studies show that people with arteriosclerosis have a high rate of zinc and copper in their blood serum which increases the accompanying lipids. There are many health problems resulted from shortage of zinc such as ovary weakness, delay of wound healing, and constitutional growth delay[8]. Shortage in zinc leads to general weakness, osteonecrosis, anemia, diarrhea, and inefficiency of respiratory system, zinc cause many health disorders, especially durning the primary stages of growth, causes growth delay, wound healing difficulties, and sexual weakness, since the production and movement of the sperms are affected by the low concentration of zinc[9]. Many studies and researches carried out to estimate the concentration of zinc in the blood and its relation to many physiological cases. One of the modern studies showed that the concentration

of zinc decreases with the progress in age in healthy persons. It was noticed that the concentration of zinc decreases remarkably in the aged people compared to middle-aged people. This decrease was seen in both males and females equally. Copper is one of the rare elements necessary for the body. It enters into the vital reactions such as physical operations and iron balance in the body. It also can be linked to many enzymes and proteins that play a key role in the cells. The free copper participates in oxidation reactions that release the free radicals which negatively reflects the performance and function of the cell. It may affect the balance of copper inside the cell then it affects the copper of the oxidation-preventing characteristic as is the case with Haper-Weis Reaction[11]. Glutathione is a peptide consists of three amino acids which are Glutamate, Cysteine, and Glycine[12]. It is an active antioxidant found in living the animal and plant tissues[13]. Glutathione has a role in removing toxicity of many metabolic operations linked to the free radicals (Peroxidase Flotation) that organizes the function of the immunity system. Glutathione consists of two steps L-Glutamate and L-Cysteine and Glycine with breaking ATP molecule and converting it into ADP and phosphate for each formed peptide bond. The most effective group in the Glutathione is the amino acid cysteine that contains the thiol group (-SH)[14]. GSH enters into multiple metabolic reactions such as protein (in terms of analyzing and creating) and creating deoxidized ribose. It also serves as an organizer (oxidation and reduction) that recalls the role of the tokophrole[15]. It also contributes to transferring the amino acids through the cell membrane and a helping factor for some enzymatic factors and aims at recalling the disulfide bond. In addition it works as an important reductive in keeping the balance of the membrane of red blood corpuscles[16]. The effect of glutathione interacts with the other antioxidants such Vitamin C they will emit and promote each other. The function of GSH is to organize the oxidation and reduction. It contributes to the reduction of Vitamin C that reacts with the free radicals and converts into Dehydroascorbate. When glutathione reduces the antioxidant compounds, it will be oxidized into GSSG and converted into the oxidized form GSH by the Glutathione Reductase (GSH-RD)[17]. It is found in most of the organs of the body in red blood corpuscles, kidney, liver, eye, and brain. It has an importance in preserving the membrane of the cells against oxidation and oxidatory damage. It performs like the folic acid for pregnant women to prevent natal defects and works as an antioxidant that it protect the mother and the fetus against the harms of the effects of the free radicals and oxidatory

distress[18]. The aim of the research is to measure the metal elements and knowing the extent of its poisonous effect on both mother and infant in addition to knowing the extent of the effect of smoking on the concentration of such biochemical variables.

Materials and Methods

The samples were collected from Tikrit Teaching Hospital within the period from 11/08/2011 to 24/01/2012. Eighty samples are collected from expectant mothers, 40 of which are from Tikrit city and 40 from Arbeda Town. The same division is applied to the samples of the umbilical cord. The samples of the umbilical cord are collected straight on the time of birth before cutting the umbilical cord. In addition, 80 samples of umbilical cord are collected and 50 samples from non-pregnant women (as a control group). The control group is divided into two parts; 25 samples for Tikrit city and 25 samples for Arbeda town, the ages selected ranges from 15-40 years. All the samples are divided on the criterion of smoking.

Detection of Cadmium Element

In the blood serum, the cadmium content in blood is realized through the following steps: diluting the blood samples; by pulling200 microliter of the blood sample and 200 microliter of TritonX solution the diluted (TritonX-100 0.1%) by using an electronic pipette in the small pipe of the centrifuge. Then mixing every pipe using a vibrating device. After that it will be put in the centrifuge. The next step is taking the serum by pipette and then putting the serum in a small cup. Then the sample is analyzed using the atomic absorption spectrometer at a 228.8 nanometer wavelength. The standard solutions samples for cadmium were prepared at different concentrations (0, 0.3, 0.6, 1.2, 2.5, 5) microgram/deciliter. The samples from 0.3 to 5 were put in the device with the blood samples of the unknown concentrations. The device will draw the standard curve of the Cadmium with its concentration and the unknown concentration of the sample will be read through the standard curve[19].

Detection of Zinc Element in the Blood Serum

The content of zinc in blood was estimated after diluting (0.5ml) of the serum sample and diluting it with to eight volumes from (HNO3, V/V %). Then the signal of the atomic absorption of the produced solution is recorded under the operation conditions of the inflammable atomic

absorption device. The standard zinc solution was prepared at the concentration of (1000µg/ml) by solving (1.2 gm) of zinc in a quantity of deionized water and adding HCL (5M) until completing the solving process then the volume reaches (1000ml) in the deionized water. A chain of the standard zinc diluted solutions was prepared with (2-0.5µg/ml) concentrations by diluting from the standard storing solution in the deionized water.

Detection of Copper Element in Blood Serum

It was prepared with concentration 1000µg/ml by solving 1gm of copper in 50ml of HNO3 with 5M then completing the volume to 1 liter in distilled deionized water. Standard solutions were prepared from the standard solution with concentrations of (0.2, 0.4, 0.8, 1.2, 1.6,2µg/ml) . the absorbance was measured at 324.7 nm wavelength.

Detection of Glutathione In Blood Serum

The glutathione in serum was estimated by using the modified method used by researchers. This method depends on using Ellman'sreagent [5,5- dithiobis (2-Nitrobenzoic acid)] (DTNB Ellman's Reagent) which reacts rapidly with Glutathione and reduced by Sulfhydryl Group (SH Group) of Glutathione resulting a colored product the absorption of which is read at 412nm. The concentration of such product depends on the concentration of Glutathione of the serum.

Statistical analysis

The results were statistically analyzed using SPSS Program (Statistical Package for Social Sciences) (Version10), the significant levels of variables are calculated using T-test to compare two variables and find differences between the values resulted through the Probability Values (P Values).

Results and Discussion

The current study revealed that the rate of the mothers who have cadmium in their blood is higher in those in Arbeda town than those in Tikrit city. The concentration of cadmium element in Tikrit city 0.07µg/dl while in Arbeda the concentration of cadmium amounts 0.192 µg/dl. This may be attributable to the environmental factors surrounding the expectant mother as shown in Table No. (1). For the control group, there is a significant decrease at probability level $P<0.05$. The decrease in cadmium concentration for the two areas (Tikrit&Arbeda) is

approximate as shown in Table No. (1). This decrease in cadmium concentration of the control group emphasizes that pregnant women have the capability of double absorption of cadmium during pregnancy, and that these results are consistent with the conducted study[21], which showed that the concentration of cadmium in the blood of mothers was 0.053µg/dl and these results are also consistent with what was found[22], which proved that the average concentration of the element in the blood of the mothers was 0.073µg/dl and the concentration ranges from 0.2 to 0.04 µg/dl, the average concentration of Cadmium in the blood of the umbilical cord was 0.66 µg/dl and ranges from 0.015 to 0.02 µg/dl. The scientist[23]found that the concentration of Cadmium ranges from 0.08 to 2.52 µg/dl in the blood of the mothers and ranges from 0.002 to 0.548 µg/dl in the blood of the umbilical cord. The results of the research also showed that the concentration of cadmium in the blood of the mothers and their infants was higher in Arbeda town compared to those in Tikrit city. This may be attributable to difference in kinds of food and the spread of smoking habit amongst most of the women in Arbeda town which increase the Cadmium rate, so that we find that the rate of Cadmium is higher in the blood of the smoker women than that in the blood of non-smoker women as shown in Table No. (2). This result is consistent with what was found by the scientists[23,24]. Smoking during pregnancy increases the concentration of Cadmium in the placenta and leads to the occurrence of some morphological changes in the placenta. The exposure of the mothers to the cadmium through smoking results in delivering low-weight babies, this may be due to the damage of placenta or due to disorders in the transference of food which leads eventually to lessening the weight of the fetus and the formation of the bones[26]. The results also showed that the concentration of cadmium was higher in some infants (the umbilical cord) than in their mothers, this is consistent with with[27] , in that the concentration of cadmium in the blood of the umbilical cord is higher than that in the blood of their mothers. The accumulation of cadmium in high quantities in the placenta especially in the smokers women encourages the synthesis of Metalothionein in the placenta with high quantities as a reaction to the accumulation of cadmium which decreases the transference of cadmium to the fetus. In addition, [28] found that the increasing of copper and iron in the blood of mothers reduces the ratio of the passage of cadmium through the placenta and this also consistent with the results reached through the research and shown in the Tables No. (1,2,3,4). There was an increase in the copper concentration in the blood of the mothers, so

that the food containing copper stops the effect of the cadmium that represses the phosphatic acids and basis in the liver and kidney. When pregnant mothers are exposed to low levels of Cadmium during pregnancy, it will overlap with the functions of the placenta then the transference of zinc and calcium to the fetus will decrease and results an abnormal growth[25]. There are cellular defense means for the placenta that both the molecules of Glutathione and Metallthionein (MT) have tendency to be linked to cadmium, then the linked Cadmium becomes non-toxic for the placenta until a particular level but when it increases above the critical limit. Metallthionein can be linked to copper and zinc and helps to transmit them from the mother to fetus and this helps in the growth and development of the fetus. Metallthionein works on storing the necessary metal for the body. The metabolism of zinc and calcium is linked to Metallthionein, both the calcium and zinc induce the synthesis of Metallthionein. Metallthionein-Cadmium protects the cells against the toxicity of Cadmium and that this compound is highly toxic for the renal tubes when it comes in the glomerular filtrate[29]. The results in Table No.(1) indicate a significant decrease in the probability value $P<0.05$ in the concentration of zinc in the blood of both mothers and the umbilical cord compared to the control group. This may be due to the usage of zinc in large quantities as a co-enzyme for Superoxide Dismutase (SOD) which shows its activity in the presence of the metal elements (zinc, copper, and manganese) and consequently uses these metals in removing roots of negative superoxide and getting rid of them by converting it into hydrogen peroxide which is also one of the oxidizing compounds which is not containing free radicals[30]. An insignificant decrease was also found in the concentration of Glutathione in the serum of the blood of the mothers compared to the control group and this is due to using it as a co-enzyme for Glutathione S-transferase. It also contribute to removing oxidation directly using the thiol group which contains it in its composition. It works to protect the cellular components against free radicals as an electron-donor through the thiol group. The reaction may occur automatically or in the presence of enzymes such as Glutathione peroxidase (GPx)[31]. The concentration of Glutathione in the umbilical cord was low compared to its concentration in the blood of the mothers, but an increase in its concentration was found in the umbilical cord with the aging process, this was only in Arbeda town and this may be due to the aforementioned reason that the placenta is the maker of it or because of smoking by mothers as shown in Table No. (4).

This result is consistent with the results by the scientist[28]. The Tables (3) and (4) show that the age has an effect on the concentrations of the metal elements. Due to this research it is assumed that by the process of aging, the absorption of cadmium will increase which consequently increases the toxicity of the cadmium. It is also assumed that by aging, the concentrations of the zinc and copper increase, and it is assumed that the concentration of Glutathione is high by the aging regarding the control group, pregnant women, and the umbilical cord and these results are consistent with[32].

Table No. (1): the concentration of cadmium, zinc, copper, and glutathione in the serum of the blood of the mothers (pregnant women), non-smokers and non-pregnant and the umbilical cord (infant) in the cities of Tikrit and Arbeda.

Type Of Sample	Variables	Cdmiumμg/Dl Mean ± SD		Zinc μg/Dl Mean ± SD		Copper (Cu) μg/Dl Mean ± SD		Glutathione μ M/L Mean ± SD	
City		Tikrit	Arbeda	Tikrit	Arbeda	Tikrit	Arbeda	Tikrit	Arbeda
Control Group		0.01 ± 0.002	0.019 ± 0.0013	1.079±0.025	1.34 ± 0.027	0.087± 0.02	0.11±0.05	8.41± 2.11	6.793±1.22
Pregnant Women (Mothers)		0.07± 0.005 (P)0.001**	0.192 ± 0.045 (P)0.03*	0.845 ± 0.01 (P)0.07	0.987 ± 0.011 (P)0.01*	0.187± 0.07 (P)0.002**	0.200±0.069 (P)0.001**	6.39±1.54 (P)0.18	4.76±1.05 (P)0.22
UmbilicalCord (Infants)		0.042 ± 0.008 (p)0.001*	0.131 ± 0.026 (P)0.04*	0.432 ± 0.008 (P)0.05	0. 533 ± 0.009 (P)0.01**	0. 105±0.083 (P)0.0001	0.188±0.072 (P)0.01	5.52±1.81 (P)0.13*	3.09±0.96 (P)0.11*

- A significant difference seen at P<0.05*
- A significant difference seen at P<0.01**

Table No. (2): the concentration of cadmium, zinc, copper, and glutathione in the serum of the blood of the mothers (pregnant women), smokers and non-pregnant and the umbilical cord (infant) in the cities of Tikrit and Arbeda.

Type Of Sample	Variables	Cdmiumμg/Dl Mean ± SD		Zinc μg/Dl Mean ± SD		Copper (Cu) μg/Dl Mean ± SD		Glutathione μ M/L Mean ± SD	
City		Tikrit	Arbeda	Tikrit	Arbeda	Tikrit	Arbeda	Tikrit	Arbeda
Control Group		0.03 ± 0.005	0.049 ± 0.0013	1.1 79±0.025	1.22 ± 0.021	0.087± 0.02	0.11±0.05	6.793± 1.208	12.41±3.11
Pregnant Women (Mothers)		0.09± 0.008 (P)0.04*	0.212 ± 0.075 (P)0.13	0.945 ± 0.021 (P)0.067*	1.13 ± 0.016 (P)0.001*	0.187± 0.07 (P)0.062*	0.200±0.069 (P)0.018*	5.76±1.11 (P)0.23	11.39±1.77 (P)0.02*
UmbilicalCord (Infants)		0.102 ± 0.003	0.191 ± 0.046	0.532 ± 0.009	0. 729 ± 0.011	0. 145±0.08	0.168±0.078	4.033± 2.76	10.52±5.01

Table No. (3): the concentration of cadmium, zinc, copper, and glutathione in the serum of the blood of the mothers (pregnant women), smokers and non-pregnant and the umbilical cord (infant) in the cities of Tikrit and Arbeda of the ages between 15-20 **years.**

Type Of Sample	Variables	Cdmiumμg/Dl Mean ± SD		Zinc μg/Dl Mean ± SD		Copper (Cu) μg/Dl Mean ± SD		Glutathione μ M/L Mean ± SD	
City		Tikrit	Arbeda	Tikrit	Arbeda	Tikrit	Arbeda	Tikrit	Arbeda
Control Group		0.03 ± 0.005	0.049 ± 0.0013	1.1 79±0.025	1.22 ± 0.021	0.087±0.02	0.11±0.05	10.41± 3.11	6.793±1.2 08
Pregnant Women (Mothers)		0.06± 0.003	0.112 ± 0.075	0.101 ± 0.020	0.711 ±0.009	0.099 ±0.05	0.16 ±0.021	8.19±1.09	6.76±1.02
UmbilicalCord (Infants)		0.042 ± 0.003	0.122 ± 0.040	0.500 ± 0.007	0. 432± 0.003	0. 095±0.013	0.188±0.072	7.12±3.01	5.933±1.71

Table No. (4): the concentration of cadmium, zinc, copper, and glutathione in the serum of the blood of the mothers (pregnant women), smokers and non-pregnant and the umbilical cord (infant) in the cities of Tikrit and Arbeda of the ages between 25-40 years.

Type Of Sample	Variables	Cdmiumµg/Dl Mean ± SD		Zinc µg/Dl Mean ± SD		Copper (Cu) µg/Dl Mean ± SD		Glutathione µ M/L Mean ± SD	
City		Tikrit	Arbeda	Tikrit	Arbeda	Tikrit	Arbeda	Tikrit	Arbeda
Control Group		0.03 ± 0.005	0.064 ± 0.007	1.198 ±0.035	1.733 ± 0.051	0.097±0.011	0.16±0.053	12.41±4.11	11.711±3.708
Pregnant Women (Mothers)		0.08± 0.006	0.199 ± 0.091	0.171 ± 0.029	0.829 ± 0.017	0.11 ±0.076	0.11 ±0.041	9.19±1.89	9.86±2.92
UmbilicalCord (Infants)		0.083 ± 0.009	0.192 ± 0.081	0.677 ± 0.021	0. 52± 0.0043	0. 122±0.003	0.168±0.062	7.42±2.01	10.91±4.71

Conclusion

The study showed that the rate of mothers and neonatal who have the cadmium in their blood was higher in Tikrit city than those in Arbeda town. The results also showed decrease in the concentration of zinc and glutathione in the serum of pregnant women and the umbilical cord compared to the control group. An increase in copper concentration rate in the blood of pregnant women and umbilical cord compared to the control group.

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