

# **Evaluation of Traceability of Dietary Urine Biochemistry Changes with Commercial Urine Strips**

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## ABSTRACT

**Objective:** This research aims to (i) examine the effects of nutrition on urine biochemistry and (ii) compare the two different measurement methods (laboratory and commercial strip). This means it is desired to bring a new direction to the literature.

**Methods:** The study involved 42 women aged 20-30 from Turkey and examined the urinary excretion of calcium, vitamin C (smoker and non-smoker), sodium, and magnesium based on nutritional status. The collected urine samples were applied onto commercial urine strips, and the resulting color changes were recorded by smartphone; at the same time, it was sent to the laboratory for comparative analysis. The obtained data were used in regression and correlation statistical analysis. All statistical analyzes were performed using IBM SPSS 28.0.

**Results:** While evaluating the regression analysis results in which the excretion due to nutrition was examined, each nutritional level was compared to the restricted intake. ANOVA sig values <.001, t values 1.96 <, in all metabolites (calcium, vitamin C, sodium, magnesium) evaluations. The following rates (R<sup>2</sup> values) were obtained restricted/optimal nutrition in calcium, vitamin C, sodium, and magnesium: .636, .575, .386, and .209 respectively; restricted/high nutrition in calcium, vitamin C (non-smoker), vitamin C (smoker), sodium, magnesium: .442, .308, .482,.413 and .337 respectively; restricted/supplement in calcium, vitamin C, magnesium: .273, .698 and .799. Calcium and magnesium strips correlated strongly with lab results, correlation coefficients are .679 and .59 respectively. Sodium and creatinine strips correlated very strongly with lab results, correlation coefficients are .876 and .884 respectively.

**Conclusion:** The study revealed that nutrition significantly affected urine excretion levels for calcium, vitamin C, sodium, and magnesium. Additionally, the results showed that urine strips had a correlation with laboratory results indicating their usefulness for pre-diagnosis purposes.

Keywords: Nutrition, urine biochemistry, urinary excretion, commercial urine strip, statistical analyzes.

## **1. INTRODUCTION**

Metabolic phenotypes, including signals from diet and endogenous metabolism, are the product of environmental and genetic interactions. These products can be used to establish meaningful relationships between health outcomes and nutrients. Longitudinal twin studies quantify the heritability of metabolic phenotypes; it also shows that nutrition has various effects on a person's metabolic phenotype (1,2). Metabolomics can be used to identify dietary biomarkers in nutritional research (2). Urine samples contain a higher amount of foodderived metabolites than blood samples (3). Controlled trials on metabolites found in urine show that urine samples can provide an objective measure of an individual's nutrition status (3,4). Studies have shown the relationship between 46 metabolites in urine and nutrition. Some metabolites have been associated with glucose, fructose, citrus, and vitamin C intake, while other metabolites have been associated with alcohol intake, as well as the consumption of meat (3). In addition, some metabolites such as sodium have also been found to be associated with various health conditions like high blood pressure and obesity (3-6).

Urine analysis, commonly referred to as urinalysis, is a diagnostic test that analyzes the urine in order to assess and monitor different aspects of health. This straightforward procedure provides information about the functioning of the urinary system and can aid in diagnosing various medical conditions. Urine analysis is one of the most crucial aspects of the healthcare, being a multipurpose and also informative diagnostic tool (7). There are many advantages to using urine as a diagnostic specimen over other biofluids. Urine sampling is non-invasive, allowing for repeated collection

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Content of this journal is licensed under a Creative Commons Attribution-NonCommercial 4.0 International License. in the desired quantity (8). Among the major goals of urine analysis is to diagnose and detect a wide range of medical conditions. Abnormalities in urine composition may indicate various conditions such as kidney disease, diabetes, UTI, and liver disorders (9,10). Also, the measurement of urine composition is important for body's general wellness.

Creatinine in urine was also examined to aid in evaluating the measurement of calcium and magnesium in urine samples. Calcium (Ca) is the fifth most abundant element in the human body and constitutes 2% of an adult's body weight. The body absorbs about 30-40% of the calcium found in the foods consumed through one's diet, leaving the rest to be excreted through urine and feces (11,12). Vitamin C plays a role in immune defense by supporting the cellular functions of the innate and adaptive immune systems (13,14). Vitamin C protects against environmental oxidative stress by keeping the skin's oxidant cleansing activity and epithelial barrier function (15,16). Sodium is vital for human health and the main cation of the extracellular fluid and is found naturally in foods. The required amount of sodium for the body is determined by the kidneys (17). Magnesium (Mg) is the fourth most abundant cation in the body (after Na+, K+, and Ca2+) and the second most abundant cation in the intracellular compartment after K+ (18,19). The body of a healthy adult contains approximately

24 g of magnesium (19,20). About 65% of this magnesium is found in the mineral phase of the bone, 34% in the intracellular space, and 1% in the extracellular space (19).

With the 'new normal' introduced by the pandemic, this research aims to explain new approaches in telemedicine that enable individuals to use test kits from home to control and diagnose their health. This research investigates (i) examine the effects of nutrition on urine biochemistry and (ii) compares the two different measurement methods (laboratory and commercial strip).

# 2. METHODS

## 2.1. Source Population

Data from a diet-induced urinary metabolite excretion study conducted between July 2022 and October 2022 were used.

The subjects were 42 individuals who participated in living in Turkey. Inclusion criteria were that participants had to be 20-30 years old, permanent residents of Turkey, not have any health problems, and not doing sports professionally (Table 1). Groups were assigned based on the number of people who voluntarily wanted to participate in the study.

Table 1. Information of participants					
(	Group	Calcium	Vitamin C	Sodium	Magnesium
	Mean ± SD	24.8 ± 2.18	24.86 ± 2.13	25.07 ± 2.14	25 ± 1.99
Age (year)	Range	21-30	21-30	21-30	22-30
Hoight (cm)	Mean ± SD	162.94 ± 6.14	163.07 ± 5.63	163 ± 6.23	162.86 ± 5.94
Height (Chi)	Range	152-177	152-174	152-177	152-177
Woight (kg)	Mean ± SD	57.86 ± 8.86	58.38 ± 9.02	58.96 ± 9.56	57.4 ± 9
weight (kg)	Range	41-78	41-78	45-78	41-78
Gender Woman Woman Woman Woman					
Ethnicity		Turkish	Turkish	Turkish	Turkish
Number of Participants		35 (7 in each group)	42 (7 in each group) 28 (7 in each group)		35 (7 in each group)
* Participants do not have any health problems.					
** Darticinants are not professional athletes					

Participants are not professional athletes

The study of Evaluation of Traceability of Dietary Urine Biochemistry Changes with Commercial Urine Strips was accepted by Bezmialem University Non-Interventional Ethics Committee with file number 2022/70. Participants were informed and gave written consent.

## 2.2. Dietary Record

The applied nutrition programs were prepared by a dietitian who is an expert in her field. The nutrient content of local foods was taken from standard nutritional tables, while the content of commercial foods (e.g. pizza and ready-to-eat foods) was derived from labeled ingredients and nutrients.

The main food groups used are 1) cereals and cereal products (breakfast cereals; bread, rice, pasta, and other cereal-based products); 2) meat products (turkey, chicken, fish, red meat); 3) sdairy products (cheeses, milk, and yogurts); 4) vegetable

products; 5) soups; 6) fruits; 7) other foods (pulse, bean, canned fruit, and pickle). The contribution of these 7 food groups to calcium, vitamin C, sodium, and magnesium intake was calculated. Diet lists were based on the amounts of metabolites found in these food groups by our co-author Esra Kozan, who is an expert dietitian.

The diet program was applied for one week for each parameter. For one week, the participants applied different nutrition programs for calcium, vitamin C, sodium, and magnesium. Participants were then given a one week break between each diet program. The experiment was made of calcium, vitamin C, sodium, and magnesium, respectively. Five study groups were prepared for calcium, six study groups for vitamin C, four study groups for sodium, and five study groups for magnesium. The daily intake amounts of each study group are different, these amounts are determined according to the standards (21) (Table 2).

Table 2. Study groups

Group	Restricted Diet (mg/d)	Optimal Diet (mg/d)	High Diet (mg/d)	Supplement (mg/d)	Control
Calcium	0-30	1000	1500	Solgar Calcium 600 (Oyster Shell) calcium carbonate (600 mg) + Vitamin D3 (75) mg	were not subject to any diet.
Vitamin C	0-10	90	200 (non-smoker) 250 (smoker)*	Solgar Chewable Vitamin C 500- Vitamin C (500 mg) + Acerola fruit extract (10 mg) + Rosehip fruit powder (4 mg)	were not subject to any diet.
Sodium	500-1000	1500	2000	**	were not subject to any diet.
Magnesium	0-30	310	350	Solgar Magnesium Citrate – trimagnesium citrate (200 mg)	were not subject to any diet.
* This group has been added because vitamin C absorption is less in smokers					
** No sodium supplement					

#### 2.3. Data Collection

The data collection started with the calcium study. Participants were asked to comply with the nutrition programs and pay attention to their supplement intake. On the first, third, and seventh days of the study, urine was collected from the participants three times in the morning, noon, and evening. Urine containers were provided to the participants to collect their urine. Collected urine was transferred to urine tubes (10 ml) and sent to an accredited laboratory for analysis. Analysis with commercial urine strips was also performed. The study was continued for 1 week. After the end of the study, a one-week break was taken and the vitamin C study was continued similarly for sodium and magnesium respectively.

Calcium and magnesium were analyzed by photometric method, creatinine by enzymatic photometric method, and sodium by ion selective electrode method in Abbott/Architect C8000 device at the laboratory. As there is no laboratory standard for vitamin C in urine, it has been analyzed only with commercial urine strips. In commercial urine strips, for each strip, one drop of urine was dropped on the parameter being studied, and the color change was photographed by smartphone. The results were determined using a reference color chart. The obtained data (Supplementary A Table 1-60) were used in statistical analysis.

#### 2.4. Statistical Analysis

All statistical analyzes were performed using IBM SPSS 28.0. *P* value of <.05 was accepted to indicate statistical significance. Bivariate Correlation analysis was used to compare laboratory and strip results. Pearson's correlation coefficient was used to determine and test the strength of the relationship. Linear regression analysis was used to examine the association between dependent and independent variables.

There was no dependent-independent variable in the correlation, but there was a reciprocal relationship. In regression, there is an 'effect'. The method of analysis provides information on the interactions between the dependent and independent variables. Impact includes prediction and forecasting. There is a cause-effect relationship. While there

is one independent dependent variable in simple regression analysis, there is more than one independent variable and one dependent variable in multiple regression analysis. When performing multiple regression analysis, the first ANOVA table is checked. If the model is found to be significant, then an interpretation is made. If sig(2-tailed) <.05, the established model is significant. Secondly, when the 'model summary' table is examined, the (R Square)x100= gives the power of the independent variable to explain the dependent variable. Third, the "coefficient" table is checked. This table shows the importance of the independent variables on the dependent variable. The higher the 't' value, the higher the degree of importance. If 1.96 < t, it is "meaningless".

The correlation analysis method gives the relationship between two metrics. It is not possible to determine causality where there is a relationship. In general, a correlation between .1-.3 is considered "weak," .3-.5 is considered "moderate," .5-.8 is considered "strong," and .8-1 is considered "very strong." The correlation/correlation coefficient takes values between "-1" and "+1". The correlation weakens as it approaches "0"; the negative correlation increases as it approaches "-1"; and the positive correlation increases as it comes to "+1". ( – or + has no mathematical meaning, and only indicates the direction.). There is no distinction between dependent and independent variables, as there is a reciprocal relationship. Pearson Correlation examines the relationship between two variables. If sig(2-tailed) <.05, there is a significant relationship.

#### **3. RESULTS**

The study involved 42 participants from Turkey and examined the urinary excretion of calcium, vitamin C (nonsmoker), vitamin C (smoker), sodium, and magnesium based on nutritional status. This research aims to (i) examine the effects of nutrition on urine biochemistry by regression analysis and (ii) correlation of the results obtained from two different methods by correlation analysis.

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#### 3.1. Regression Analysis

According to the data obtained (Supplementary A Table 1-60), the effect of nutrition on excretion was interpreted using regression analysis (Table 3).

Table 3.	Rearession	analysis	results	between	nutrition	and	excretion
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Group	Restricted/Optimal	Restricted/High	Restricted/ Supplement			
Calcium	.636	.442	.273			
Vitamin C	.575	.308 (non-smoker) .482 (smoker)*	.698			
Sodium	odium .386 .413 **					
Magnesium .209 .337 .799						
* This group has been added because vitamin C absorption is less in smokers.						
no soaium supplement						

While evaluating the regression analysis results in which the excretion due to nutrition was examined, each nutritional level was compared to the restricted intake. ANOVA sig values <.001, t values 1.96 <, in all calcium evaluations (Supplementary B Table 7-9). In this case, the models are meaningful and can be interpreted. The following effect rates were obtained in calcium: restricted/optimal nutrition, 63.6% (Supplementary B Table 7); restricted/high nutrition 44.2% (Supplementary B Table 8); and restricted/supplement 27.3% (Supplementary B Table 9).

ANOVA sig values <.001, t values 1.96 <, in all vitamin C evaluations (Supplementary Table 10,11,12,13). In this case, the models are meaningful and can be interpreted. The following rates were obtained in vitamin C: restricted/optimal nutrition, 57.5% (Supplementary B Table 10); restricted/high (non-smoker) nutrition 30.8% (Supplementary B Table 11); restricted/high (smoker) nutrition 48.2% (Supplementary Table 12) and restricted/supplement 69.8% (Supplementary B Table 13).

ANOVA sig values <.001, t values 1.96 <, in all sodium evaluations (Supplementary Table 14,15). In this case, the models are meaningful and can be interpreted. The following rates were obtained in sodium: restricted/optimal nutrition 38.6% (Supplementary B Table 14), and restricted/high nutrition 41.3% (Supplementary B Table 15).

ANOVA sig values <.001, t values 1.96 <, in all magnesium evaluations (Supplementary Table 16-18). In this case, the models are meaningful and can be interpreted. The following rates were obtained in magnesium: restricted/optimal nutrition 20.9% (Supplementary B Table 16), restricted/high nutrition 33.7% (Supplementary B Table 17), and restricted/ supplementary 79.9% (Supplementary B Table 18).

## 3.2. Correlation Analysis

According to the data obtained as a result of the study (Supplementary A Table 1-15; Supplementary A Table 34-60), comparative analyzes of the two measurement methods used were made and the correlation coefficients were calculated (Table 4). As there is no laboratory standard for vitamin C in urine, it has been analyzed only with commercial urine strips. Therefore, it was not included in the correlation analysis.

<b>TUDIE 4.</b> CONTENTION OF TUDOFULORY UND SEID FESUL	Table 4.	Correlation	of	<sup>f</sup> laboratorv	and	strip	results
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Group	Lab-Strip Correlation Coefficient*		
Calcium	.679		
Calcium/Creatinine	.604		
Magnesium	.59		
Magnesium/Creatinine	.511		
Creatinine	.884		
Sodium	.876		
* Correlation of laboratory and strip results with each other			

As a result of the correlation analysis in which the strip and laboratory results were evaluated, the calcium strip was .679 (Supplementary B Table 1), and the calcium/creatinine ratio was .604 (Supplementary B Table 2), the sodium strip was .876 (Supplementary B Table 3), magnesium strip was .59 (Supplementary B Table 4), and the magnesium/creatinine ratio was .511 (Supplementary B Table 5), the creatinine strip was .884 (Supplementary B Table 6).

Valuable results were obtained in this study on calcium, magnesium, sodium and vitamin C, which are necessary for our body. Based on above results, it is possible to confirm that nutrition affects calcium, vitamin C, sodium and ,magnesium excretion in urine.

When smokers and non-smokers were compared during the vitamin C study, it was found that vitamin C excretion was higher among smokers. It was thus observed again that smoking affects vitamin C absorption. In addition, it is also possible to confirm that the use of supplements seriously affects vitamin C excretion in urine.

This is due to decreased absorption with increased intake of vitamin C, making the bioavailability of vitamin C in supplements less than that of food. When daily doses of 30-60 mg of vitamin C is consumed, almost no vitamin C is excreted in the urine in a 24-hour period. In cases where there is moderate vitamin C intake, approximately 70-90% of the amount taken in is absorbed, while in cases where there is vitamin C intake above 1,000 mg, vitamin C absorption falls below 50%, as unmetabolized ascorbic acid is excreted through the urine (21).

When evaluating sodium results, environmental factors, water and salt consumption should be considered that affect sodium excretion rates.

Although spot magnesium ratios are low in restricted diets, magnesium/creatinine ratios are at optimal levels because of low creatinine excretion, which has been associated with high water consumption. Because of this, the effective rates were lower than expected, but it is still possible to say that nutrition affects the excretion of magnesium in urine.

In addition, it is also possible to confirm that the use of supplements seriously affects magnesium excretion in urine.

## 4. DISCUSSION

The results of this study significantly contribute to understanding the interrelationships between nutrition and the urinary excretion of crucial elements, including calcium, vitamin C, sodium, and magnesium. Our analysis shows that nutritional status affects the excretion of these elements directly and quantitatively. The regression analysis shows that the levels of nutritional intake significantly influence calcium excretion. The finding that limited diets result in increased calcium excretion relative to optimal or high nutrition implies a counterbalancing mechanism in the body to keep calcium homeostasis. This is important in understanding bone health and the prevention of osteoporosis, especially in diverse populations with different eating patterns (22).

Our data suggest that vitamin C excretion is higher in smokers than in non-smokers. This is consistent with the assumption that smoking increases oxidative stress, leading to increased demand for vitamin C in the body (23). The decreased bioavailability of supplemental vitamin C, as observed through increased excretion with higher supplement consumption, supports the notion that vitamin C levels are tightly regulated, mainly by renal excretion (24). This suggests that a balanced diet should be prioritized over supplementation as the primary source of vitamin C intake. Our results on sodium excretion support the role of dietary intake and environmental effects. These findings emphasize the difficulty of understanding sodium regulation in the body and the need to take into account external factors when evaluating sodium excretion. Our findings of elevated magnesium excretion from supplementation and restricted diets are also interesting. This indicates that, just like vitamin C, the body controls magnesium concentration via urinary excretion, which is based on dietary intake.

Another goal of this study was to assess the accuracy and feasibility of commercial urine strips compared to traditional laboratory analysis methods. It is a very crucial comparison, because it directly addresses the possible implementation in the clinical practice where speed and accuracy are key. Commercial urine strips provide a fast, very inexpensive, and also convenient method for initial urine testing (25,26). These strips are very useful in the situations where quick results are needed, such as emergency medicine or for regular screening in outpatient settings. Their ease of use makes them accessible to non-specialists, expanding the scenarios in which they can be employed in a healthcare setting. But although these strips give quick outcomes, their precision and specificity may be much less than those of the laboratory analyses. The main source of this variance is the semi-quantitative nature of the strip readings and also the possibility of user error in the interpretation. Conversely, laboratory analysis which is carried out using 24 hour urine, despite being lengthy and also costly, provides a greater level of precision and sensitivity. The 24-hour

urine test is considered the gold standard method for measuring biochemical parameters, but it is time-consuming and difficult to implement (27). Instead, the equivalent is obtained by determining concentrations of creatinine in spot urine and putting them into proportion with the component. Typically, this method estimates the 24-hour excretion rates of protein, potassium, sodium, magnesium, calcium, urea, and uric acid (28). The comprehensive analysis conducted in a stable setting reduces the many chances of wrong results, making it alot more effective in complicated cases or where the accurate quantification of analytes is essentially essential. However, the time and cost of the laboratory analysis are a constraint in the emergencies or resource-poor settings.

When comparing these two methods, it can be seen that each has its own strengths and also weaknesses. The decision between the use of commercial urine strips and the laboratory analysis is dependent upon the setting of the testing. For example, in a primary care environment where fast screening is critical, commercial urine strips can be a very good first test. But for precise diagnostic purposes, especially when more subtle information is needed, the laboratory analysis remains the better option.

Commercial urine strips and laboratory analysis both play their own role in the medical diagnostics. The choice of the method should be based on factors such as precision, available resources, and the urgency of the situation. This research highlights the necessity of a delicate balance, combining the many strengths of each method to achieve the best possible care for the patients.

# **5. CONCLUSION**

This study provides a new perspective on scientific literature by exploring the impact of nutrition on urine biochemistry, a relatively understudied field.We compare two different urine measurement methods - laboratory analysis and commercial strips of urine. This comparison is useful because it assesses the feasibility and trustworthiness of urine strips relative to the conventional laboratory approach. The conclusions drawn from this study are based on solid statistical analyses, including regression and correlation. The statistical techniques are also appropriate and accurate for the nature of collected data. This research provides evidence for the relationship between nutrition and urine excretion of various metabolites, contributing to a better understanding of this field. Showing the association between urine strips and laboratory findings not only validates the use of these strips but also indicates their ability to pre-diagnosis. This might have great practical implications in the clinical settings or for personal health monitoring.

The focus on a specific age group and location in this study may limit the external validity of the results to other populations. However, the sample size we used is sufficient for conducting the study. On the other hand, a larger sample from different country could give more generalizable results.

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# Finally, our research provides a useful and unique method of evaluating the effects of nutrition on urine biochemistry. However, the results are constrained by the small sample size and regional scope. Further studies could strengthen these results, by including a larger and more representative sample, a larger number of nutritional and lifestyle factors, and exploring the possible applications in different clinical or health settings.

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