

# **Content Analysis of Locally Marketed Energy Drinks: Turkish Market**

# Yerel Pazarda Sunulan Enerji İçeceklerinin İçerik Analizi: Türkiye Pazarı

## Zafer Peker<sup>©</sup> and Dursun Ali Köse<sup>\*®</sup>

Hitit University, Graduate Education Institute, Department of Forensic Sciences, Çorum-Türkiye, 19100.

### ABSTRACT

n the Turkish Food Codex Communique on Energy Drinks, energy drinks are defined as "flavored non-alcoholic beverage containing caffeine, containing taurine, glucuronolactone, inositol, carbohydrates, amino acids, vitamins, minerals and other food and components". The amount of caffeine in the composition of the energy drink cannot be more than 150 mg/L, inositol 100 mg/L, glucuronolactone 20 mg/L, and taurine 800 mg/L. It has been reported that consumption of beverages containing caffeine, taurine and glucuronolactone in healthy young adults increases arterial blood pressure and platelet aggregation and decreases endothelial function. For healthy adults, consuming more than 400 mg of caffeine daily and consuming more than 500 ml of energy drinks in energy drinks, as well as consuming energy drink with or together with alcohol, is risky and not recommended. Energy drinks are risky for children, people under the age of 18, the elderly, diabetics, those with high blood pressure, pregnant and lactating women, those with metabolic diseases, kidney failure and people who are sensitive to caffeine. The use of energy drinks has been increasing rapidly in the world and in our country in recent years. Usually energy drinks are used by young people, athletes and people who live actively. Energy drinks are also preferred as a pleasure or to increase mental, physical and cognitive performance. The aim of our study is to analyze the components of the content and the effectiveness of the components by making a qualitative analysis with the GC-MS (Gas Chromatography-Mass Analysis) device of many energy drinks used in the market. In addition, it is the evaluation of the energy drinks available in the market by examining the effects of the determined substances and their amounts on human health.

#### **Key Words**

Energy drinks, GC-MS analysis, caffeine, trehalose, forensic science.

ÖΖ

Türk Gıda Kodeksi Enerji İçecekleri Tebliği'nde enerji içecekleri, "kafein, taurin, glukuronolakton, inositol, karbonhidratlar, amino asitler, vitaminler, mineraller ve diğer besin ve bileşenleri içeren aromalı alkolsüz içecek" olarak tanımlanmaktadır. Enerji içeceğinin bileşimindeki kafein miktarı 150 mg/L, inositol 100 mg/L, glukuronolakton 20 mg/L ve taurin 800 mg/L'den fazla olamaz. Sağlıklı genç erişkinlerde kafein, taurin ve glukuronolakton içeren içeceklerin tüketiminin arteriyel kan basıncını ve trombosit agregasyonunu arttırdığı ve endotel fonksiyonunu azalttığı bildirilmiştir. Sağlıklı yetişkinler için günde 400 mg'dan fazla kafein tüketmek ve enerji içeceklerinde 500 ml'den fazla enerji içeceği tüketmek, ayrıca enerji içeceklerini alkolle birlikte tüketmek risklidir ve önerilmez. Enerji içecekleri çocuklar, 18 yaş altı kişiler, yaşlılar, şeker hastaları, yüksek tansiyonu olanlar, hamileler ve emzirenler, metabolik hastalığı olanlar, böbrek yetmezliği olanlar ve kafeine duyarlı kişiler için risklidir. Enerji içeceklerinin kullanımı son yıllarda dünyada ve ülkemizde hızla artmaktadır. Enerji içecekleri genellikle gençler, sporcular ve aktif olarak yaşayan kişiler tarafından kullanılır. Ayrıca keyif verici olarak veya zihinsel, fiziksel ve bilişsel performansı artırmak için tercih edilir. Çalışmamızın amacı piyasada kullanılan birçok enerji içeceğinin GC-MS (Gaz Kromatografi-Kütle Analizi) cihazı ile kalitatif analiz yaparak içeriğindeki bileşenleri ve bileşenlerin etkinliğini analiz etmektir. Ayrıca piyasada bulunan enerji içeceklerinin belirlenen maddelerin ve miktarlarının insan sağlığı üzerindeki etkilerinin incelenerek değerlendirilmesidir.

#### Anahtar Kelimeler

Enerji içecekleri, GC-MS analizi, kafein, trehaloz, adli bilimler.

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Correspondence to: D.A. Kose, Hitit University, Graduate Education Institute, Department of Forensic Sciences, Çorum-Türkiye, 19100. E-Mail: dalikose@hitit.edu.tr;

# INTRODUCTION

nergy drinks were introduced to the world for the first time in 1949 by the American scientist Dr. Enuf. It has been produced as an alternative to sugary sodas, supported by vitamins and sugar. Subsequently, it started to be used in Asia and Europe in 1960 [1,2]. Its widespread use in the world started in the 1980s, but in our country, the year it was released for sale was in the 1990s. It is known that approximately 25 different brands of energy drinks are sold in our country, many more brands are on the market shelves in small batches, and the total number of brands has reached 42 [2]. Energy drinks can be defined as calorie-rich drinks prepared by combining energy-boosting substances such as caffeine, taurine, herbal extracts and B vitamins [1]. Caffeine in energy drinks is an important psychoactive substance and its amount in energy drinks can vary. In addition, many substances can be found in energy drinks, including guarana, ginseng, ginkgo Biloba, L-carnitine, milk thistle (Silybum marianum, thistle), B vitamins, taurine, an amino acid, and other substances to increase energy and mental performance. For example, guarana, a plant containing the highest amount of caffeine in the world, can be added to energy drinks in addition to increasing the amount of caffeine contained in energy drinks [3-7]. They fall into the category of functional drinks, which includes energy drinks, sports drinks, and nutraceutical drinks [8]. Sports drinks are caffeine-free drinks that are consumed before or during exercise, designed to prevent water loss and provide carbohydrate and electrolyte support [9-12]. Nutraceutical beverages, on the other hand, are beverages designed to support and improve health, generally containing biologically active ingredients such as tea, concentrated extracts from fruits, vegetables or herbs. Some nutraceutical beverages can be supplemented with vitamins, minerals, and antioxidants, especially polyphenols. Various energy drinks can be included in the scope of nutraceutical drinks depending on the composition of the substances they contain. In the world, energy drinks cover 47.3% of the general market share of functional drinks. Between 2004 and 2009, the market share of energy drinks has grown considerably in other countries, especially in the USA [8]. The positive effect of energy drinks on cognitive and psychomotor functions, the effect of reducing fatigue by increasing long-term attention, the effect of increasing attention in driving ability, and increasing physical performance and endurance. While energy drinks have these effects, they cause excessive

release of dopamine, serotonin, noradrenaline and adrenaline and their prolonged effects, just like addictive drugs. Substances such as L-tyrosine, which is the precursor of dopamine production, cause more neurotransmitter release, leading to the discharge of neurotransmitter stores in the body and the formation of a negative balance in the body. Intense fatigue, especially after 5 hours of consumption of the energy drink, should be perceived as a sign of a negative situation [13,14]. Turkish Food Codex; it is the institution that sets the standards for food products to be sold in Türkiye. In the Turkish Food Codex (TFC) Energy Drinks Communiqué, energy drinks are defined as "flavored non-alcoholic beverage containing caffeine, containing taurine, glucoronolactone, inositol, carbohydrates, amino acids, vitamins, minerals and other food components". In that notification, the amount of caffeine to be included in the energy drink cannot be more than 150 mg/L, inositol 100 mg/L, glucuronolactone 20 mg/L, and taurine 800 mg/L. It is also stated that ethyl alcohol will not be added as a component to energy drinks. However, the amount of ethyl alcohol that can be used in processes such as dissolving and transporting the components that make up the product, that can be found naturally in the components, that can be formed during the production phase and that can be transported to the final product, is at most 3.0 g/L. It has also been stated that flavored soft drinks, which may contain one or more of taurine, inositol and glucuronolactone together with caffeine in their composition, cannot be supplied to the market with any food other than energy drinks. The rate of fruit in energy drinks called fruit should be at least 4% by weight in carbonated ones and at least 10% by weight in non-carbonated ones. There is a requirement that the energy value provided from carbohydrates in energy drinks should be at least 45 kcal/100 ml [15-17]. The aim of our study is to make gualitative analyzes of many energy drinks used in the market with the GC-MS device, to determine which components the content consists of and to examine the effects of the determined substances on human health.

### **MATERIALS and METHODS**

Nine different energy drinks offered to the market in local markets in Turkey were collected and 10 ml liquid samples were taken from each of them for the gas chromatography mass analysis method. Content screening of the energy drink samples taken was done by gas chromatography-mass analysis method using TR5MS column (60m x 0.32mm) in Thermo DSQ II GC-MS device.

### **Results and Discussions**

GC-MS chromatogram graphs of energy drink samples recorded between 0-14 minutes are given in Figure 1.

The mass analysis diagrams obtained from the strong peaks selected by examining all of the GC-MS chromatogram curves are shown in Figure 2. Possible ingredient formulas found by scanning the relevant peak diagrams in the device library are also shown above the diagrams. When the mass analysis diagrams obtained from the peaks selected from the chromatograms were examined, it was determined that nine different energy drinks were very similar to each other in terms of content. The possible ingredient contents of each energy drink are summarized in Table 1 in detail.

According to the data obtained, all of the products whose content analyzes were made, except for the ED (energy dirink) no. 9, contain a high rate of "Pholoroglucinol" compound in their content, which starts to come with a high peak at approximately 6.75-6.99 minutes. The "Trehalose" molecule, which is thought to be used for sweetening purposes as a common component in all structures, has been identified. The peaks belonging to the Trehalose molecule (it is a natural disaccharide sugar consisting of glucose and glucoside units), which appear in more than one time region in the chromatogram spectrum, show the content of this molecule in the structure of energy drinks.

Glycolic acid, which was determined to come in the first minutes of the chromatogram spectra of the analyzed energy drinks, is preferred as a basic raw material or additive agent in the production of many products (from adhesives to skin creams). In the food industry, it is often used as an additive for flavoring and preservative purposes. It was observed that the intensity of the peak of the molecule, which was detected in the structure of all products, except for two products (ED no 4 and 8), which was observed to arrive at 1.52-1.69 minutes, was low [18-20].

It indicates the peak dextroamphetamine compound that comes in almost the same minute (weak strength) in seven of the nine different energy drinks studied. The peak of the compound indicated in the energy drinks numbered 4 and 8 was not detected. Carbohydrazide, which is observed to peak in the range of approximately 3.82-3.85 minutes, is a chemical compound with the formula  $OC(N_2H_3)_2$ . White crystalline solid molecule with good solubility in water, a number of carbazide derivatives are known in which one or more N-H groups have been replaced by other substituents [21-26].

The peak of the chemical levulinic acid, which gave a peak in 5.35 minutes, was detected only in the ED no 5. Levulinic acid or 4-oxopentanoic acid is an organic compound with the formula  $CH_3C(O)CH_2CH_2CO_2H$  and is also classified as a keto acid. This white crystalline solid is soluble in water and polar organic solvents [27-29].

Succinic acid, which was detected only at the 7.05<sup>th</sup> minute in ED 9, is a dicarboxylic acid with the chemical formula  $(CH_2)_2(CO_2H)_2$ . In living organisms, succinic acid takes the form of succinate, an anion with multiple biological roles as a metabolic intermediate that is converted to fumarate by the enzyme succinate dehydrogenase in the electron transport chain involved in making ATP. It is marketed primarily as an acid-regulating food additive in the food and beverage industry with the code E363 [30].

Like succinic acid, citraconic acid, which is detected only in the ED 9 with a peak at the 7.94<sup>th</sup> minute, is one of the isomeric dicarboxylic acids produced by the distillation of citric acid or as metabolites by microorganisms [31-33].

Caffeine is the only molecule commonly detected at 11.00-11.03 minutes in all energy drink samples for which gas chromatography mass analysis was performed, and this molecule is a methylxanthine class central nervous system (CNS) stimulant. It is the world's most widely consumed psychoactive substance, a bitter, white crystalline purine, chemically related to the adenine and guanine bases of deoxyribonucleic acid (DNA), ribonucleic acid (RNA). Unlike many other psychoactive substances, it is legal and unregulated in nearly all parts of the world. Caffeine blocks adenosine receptors, preventing the onset of adenosine-induced sleepiness. In addition, caffeine is the most preferred stimulant in energy drinks today due to its ability to increase sprint performance, athletic performance in cycling and running, in aerobic (especially endurance sports) and anaerobic conditions, in moderate doses (approximately 5 mg / kg) [34-41].

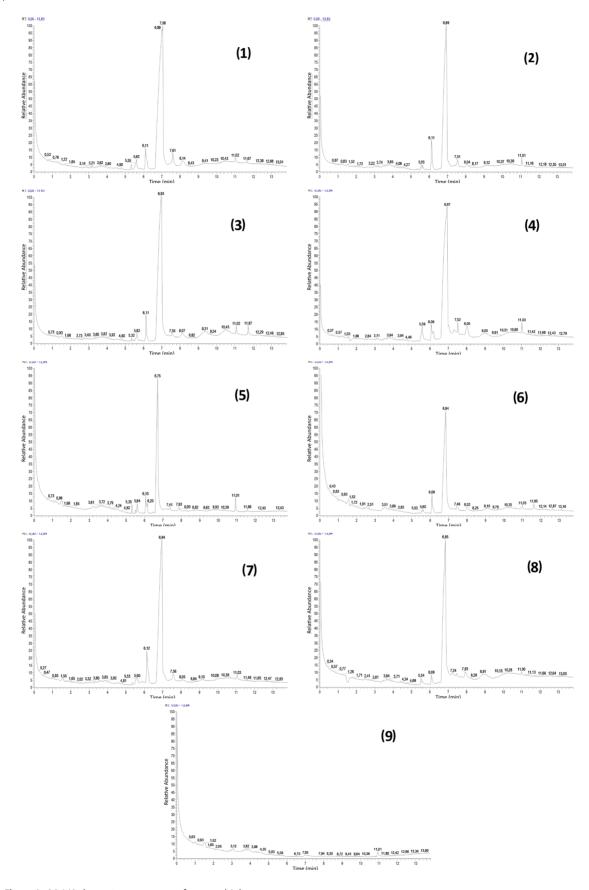


Figure 1. GC-MS chromatogram curves of energy drinks

Glycolic acid (hydroacetic acid or hydroxyacetic acid) with a weak peak in approximately 1.52-1.69 minutes; chemical formula  $C_2H_4O_3$  (also spelled HOCH<sub>2</sub>CO<sub>2</sub>H) is the smallest  $\alpha$ -hydroxy acid (AHA). It was not detected in only two of the nine different energy drink samples studied. Glycolic acid is a colorless, odorless and hygroscopic crystalline solid, highly soluble in water. Ethyl 2-furoate, which was not observed only in the energy drink sample No. 9, appears with a weak peak at approximately 5.54-5.62 minutes. The substance, which has a colorless to pale yellow color, is liquid and has a berry, fruity, wine-like, heavy-smelling aroma taste [42].

# Conclusions

9 different energy drink samples that are currently on sale in the Turkish market have been examined, and when the recorded content content analyzes are taken into account, it has been determined that there is no extra ingredient that will actually provide energy support. When energy drinks were first introduced to the market in our country, it was determined that the chemical "taurine", which could be considered as an energizing additive, was also removed from the ingredients. It has been determined that they contain "caffeine" as a component that increases attention, concentration and sports endurance, instead of the energizing content that is mostly detected. The amount of caffeine to be taken from the energy drink, the daily consumed coffee, etc. It is thought that it can be easily obtained from beverages. It has been revealed that the energy drinks, which are determined to contain several types of sugar derivatives that can be metabolized in the body, other than caffeine, do not have very serious energy-increasing ingredients, which are actually advertised.

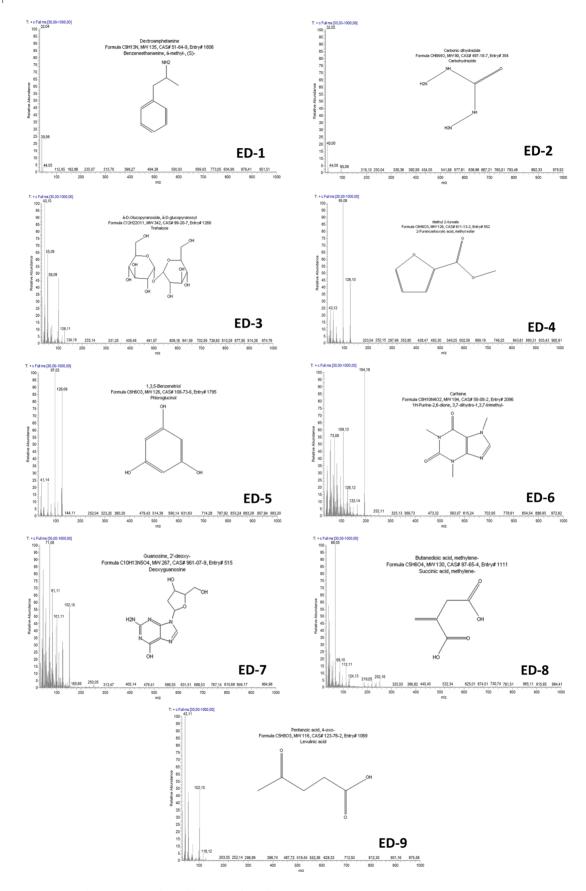


Figure 2. Mass analysis patterns and possible content formulas

ED 1		ED 2		ED 3		ED 4		ED 5	
Glycolic acid	1.65	Glycolic acid	1.52	Glycolic acid	1.68			Glycolic acid	1.69
Dextroamphetamine	3.21	Dextroamphetamine	3.22	Dextroamphetamine	3.40			Dextroamphetamine	3.72
Carbohydrazide	3.62	Carbohydrazide	3.85	Carbohydrazide	3.82	Carbohydrazide	3.82	Carbohydrazide	3.85
Trehalose	4.80			Trehalose	4.80			Levulinic acid	5.35
Trehalose	5.55	Trehalose	5.55	Trehalose	5.32	Ethyl 2-fruoate	5.59	Ethyl 2-fruoate	5.62
Ethyl 2-fruoate	5.62	Ethyl 2-fruoate	5.62	Ethyl 2-fruoate	5.62	Trehalose	6.08	Trehalose	6.10
Trehalose	6.11	Trehalose	6.11	Trehalose	6.11	Benzoic acid	6.22	Benzoic acid	6.20
Pholoroglucinol	6.99	Pholoroglucinol	6.89	Pholoroglucinol	6.93	Pholoroglucinol	6.94	Pholoroglucinol	6.75
Pholoroglucinol	7.06								
Trehalose	7.61	Trehalose	7.51	Trehalose	7.51	Trehalose	7.52	Trehalose	7.41
Trehalose	8.14	Trehalose	8.04	Trehalose	8.07	Trehalose	8.05	Deoxyguanosine	7.93
Trehalose	9.41	Trehalose	9.12	Trehalose	9.31	Trehalose	9.00	Trehalose	8.82
Trehalose	10.43	Trehalose	10.39	Trehalose	10.43	Trehalose	10.31	Trehalose	10.39
Caffeine	11.02	Caffeine	11.01	Caffeine	11.02	Caffeine	11.03	Caffeine	11.01
Trehalose	11.67	-	•	Trehalose	11.67	Trehalose	11.68	Trehalose	11.68
ED 6		ED 7		ED 8		ED 9			
Glycolic acid	1.69	Glycolic acid	1.65			Glycolic acid	1.52		
Carbohydrazide	3.82	Dextroamphetamine	3.32			Dextroamphetamine			
ı	ı	Carbohydrazide	3.85	Carbohydrazide	3.71	Dextroamphetamine			
ı		Trehalose	4.81						
Ethyl 2-fruoate	5.60	Trehalose	5.53	Ethyl 2-fruoate	5.54				
Trehalose	6.09	Ethyl 2-fruoate	5.60	Trehalose	60.9				
Ţ		Trehalose	6.12						
Pholoroglucinol	6.84	Pholoroglucinol	6.94	Pholoroglucinol	6.85				
·	ı					Succinic acid	7.05		
Trehalose	7.46	Trehalose	7.56	Trehalose	7.41	Citranoic acid	7.94		
Trehalose	8.02	Trehalose	8.05	Trehalose	7.93				
Trehalose	9.10	Trehalose	9.10	Trehalose	8.91				

11.01

-Caffeine

10.28 11.00

10.38 11.03 11.65

Trehalose Caffeine Trehalose

10.25

11.03 11.65

Trehalose Caffeine Trehalose

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11.66

Trehalose Caffeine Trehalose

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