ETHANOL LEVELS OF THE NON-ALCOHOLIC BEVERAGES SOLD IN MARKETS IN TÜRKİYE

TÜRKİYE' DE MARKETLERDE SATILAN ALKOLSÜZ İÇECEKLERİN ETANOL SEVİYELERİ

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ABSTRACT

Although ethanol is one of the basic chemicals used in many different fields, when included in the human diet, it has been one of the major subjects of discussion in terms of medical, social and religious issues due to its intoxicating and detrimental effects. The control of presence, amounts and effects of ethanol in beverages and foods is one of the important research fields. Regulations regarding non-alcoholic beverages vary among countries. In Türkiye, the highest allowable level of ethanol content is 3.0 g / L according to Non-alcoholic Beverages Notification of Turkish Food Codex (2007/26). Non-alcoholic beverages, according to the codex, are defined as the drinks that contain ethanol less than 0.3% by volume. In this study, colas, orange sodas, 100% fruit juices, fruit nectars, sodas with fruit aromas, energy drinks, ayran and kefir of the well-known brands were used as the non-alcoholic beverage samples. The test method TS 1594 of Turkish Standards Institution, Titrimetric Method of the Determination of Ethanol Content in Fruit and Vegetable Products, was used to determine the ethanol levels with a modification. The ethanol from the samples was distilled by hydrodistillation with Clevenger type distillation apparatus, which made the method more practical. The findings showed that the highest per thousand concentration of ethanol in the samples were as follows: colas 0.14, orange sodas 0.53, other fruit flavored sodas 0.88, energy products 0.47, fruit juices 1.46, and milk products 0.25. Ethanol levels of all the samples were found below the allowed limit according to the codex.

Keywords: Non-Alcoholic Beverages, Ethanol, Hydrodistillation, Clevenger Distillation Apparatus.

ÖZET

Etanol, birçok farklı alanda kullanılan temel kimyasallardan biri olmasına rağmen, diyetle alındığında sarhoş edici ve zararlı etkileri yüzünden tıbbi, sosyal ve dini konular bakımından önemli tartışma konularından biri olmuştur. Gıdalardaki ve içeceklerdeki etanolün varlığı, miktarı ve etkilerinin kontrolü önemli araştırma alanlarındandır. Alkolsüz içeceklerle ilgili düzenlemeler ülkeden ülkeye farklılık gösterir. Türkiye'de Türk Gıda Kodeksi Alkolsüz İçecekler Tebliği'ne (2007/26) göre izin verilen en yüksek etanol miktarı 3,0 g/L'dir. Kodekse göre, hacimce % 0,3' den daha az etanol içeren içecekler, alkolsüz içecekler olarak tanımlanır. Bu çalışmada; kolalar, portakallı sodalar, %100 meyve suları, meyve nektarları, meyve aromalı sodalar, enerji içecekleri, ayran ve kefirin bilinen markaları alkolsüz içecek test numunesi olarak kullanıldı. Bu amaçla, Türk Standartları Enstitüsü'nün TS 1594 Meyve ve Sebze Ürünleri Etanol Muhtevası Tayininin Titrimetrik Yöntemi bir değişiklik yapılarak kullanıldı. Numunelerdeki etanol Clevenger tipi destilasyon aparatıyla hidrodestilasyonla destillendi, ki bu uygulama yöntemi daha uygulanabilir hale getirmiştir. Numunelerde bulunan en yüksek binde etanol konsantrasyonları sırayla kolalarda 0,14, portakallı kolalarda 0,53, meyve aromalı sodalarda 0,88, enerji içeceklerinde 0,47, meyve sularında 1,46 ve süt ürünlerinde 0,25 olarak bulundu. Bütün numunelerin etanol seviyesi kodekste izin verilen sınırın altında bulundu.

Anahtar kelimeler: Alkolsüz İçecekler, Etanol, Hidrodestilasyon, Clevenger Destilasyon Sistemi.

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INTRODUCTION AND AIM

Alcohol consumption, as a risk factor in cancer, has continually been investigated. Some types of cancer may be associated with alcohol. According to recent data, it may be responsible for 5% of all cancer death in some societies.[1] If long term alcohol consumption increases, its effects on the risk of cancer become more important.

Alcohol effects esophagus and stomach crucially, and recently, in these two organs, a significant increase has been observed in mortality. Alcohol irritates stomach inner surface covering layer, causing vomiting and small tears in the lower part of the esophagus. The most significant result is probably the serious increase in the development of esophageal cancer.[2]

In addition, excessive use of alcohol has adverse effects on the brain and nervous system. Alcohol can also cause temporary memory loss. Besides, excessive use of alcohol by pregnant women can cause significant damage to the developing nervous system of the fetus.[3] Continuous and excessive alcohol use ruins myocardium and results in conditions from arrhythmias to heart failure, which may lead to various serious problems.[4]

Food demands are met from the products of unnatural industrial sources in order to increase efficiency, reduce costs, and extend the shelf life. So, a large number of additives are generally used in processed foods. Many religions are worried about certain foods and drinks as they may contain the ingredients that are forbidden according to religious laws. The views of different religions, e.g. Islam, Judaism, and Christianity, on alcohol vary.[5]

In Islam, according to the Qur'an, the direct and final revelation of God to humankind, alcohol intake is forbidden and it is described as "najis" (unclean).[5] Prophet Muhammad (SAW) says that Allah (SWT) has cursed not only drinking but also indirect link to alcohol generating, manufacturing, delivering, buying, selling and profiting from it.[6]

In Judaism, the followers serve wine in important religious rituals. Purim Jews support excessive drinking, even though Jewish tradition usually backs up drinking in moderation and not to get drunk, which is linked to non-Jews.[7]

In Catholics, wine which was part of the diet and religious rituals for the Old Testament Jews was said to be allowed and blessed at the Last Supper for the New Testament by Jesus.[8,9]

The Church of Jesus Christ of Latter Day Saints (known as the Mormons) allowed wine in moderation in the 19th century; however, today the Mormons unequivocally ban alcohol for better health.[10]

Until this time, determination of ethanol in alcoholic beverages have been fulfilled in many ways: oxidation of distillate[11,12,13], dichromate oxidation spectrophotometry[11,12,14], refractive index method[11,12], picnometry and densimetric analysis[13], boiling point depression of the ethanol solution relative to water[11], enzymatic method[15,16], biosensor[17], potantiometry[18], gas chromatography (GC)[19,20], capillary gas chromatography[20], capillary electrophoresis[21], high performance liquid chromatography (HPLC)[22,23], modular Raman spectrometry[24], near-infrared (NIR) spectroscopy[25], and flow injection analysis.[26,27]

Densimetric analysis and picnometry are not practical for small amount of samples. More than 5 mL of sample volume is required in oxidation of the distillate and dichromate oxidation spectrophotometry. For enzymatic method, biosensor and potantiometry, stability, reproducibility and accuracy are low.[28]

Capillary electrophoresis and Raman spectrometry are not prevalent because of demanding expensive instruments. HPLC provides a comparatively low sensitivity. In

establishing calibration curves, NIR spectroscopy is time consuming and has low accuracy as there can be interferences from other alcohols. GC is the most suitable and fast method for determining ethanol content in alcoholic beverages. The capillary gas chromatography technique is one of the most significant modern techniques which has high resolution and sensitivity.[20]

According to Turkish Food Codex, non-alcoholic beverages are fruit drinks, aromatic beverages, fruit syrups, aromatic syrups, fruit drink powder, aromatic drink powder, fruit drinks with natural mineral water, aromatic drinks with natural mineral water, artificial mineral waters, colas, tonics and mineral waters. Up to now, the only investigation about the alcohol content in non-alcohol beverages has been conducted on sodas by the Consumer Association in Türkiye, in 2008. The reason of conducting this study was that there was no information about the alcohol content in the soda products label and also that there was a statement reporting nonexistence of alcohol in the products. For this study, the most prevalent soda brands were preferred, and in their alcohol content has been tested in TUBITAK Marmara Research Center by using IFFJ Modified Rebel Method, 1983.[29]

In this study, the answer to the question "Is there alcohol in non-alcoholic beverages?" is sought for a variety of sample types. The use of hydro-distillation with Clevenger type apparatus was described for the first time here, to determine ethanol from non-alcohol beverages. For the measurement of ethanol, the test method TS 1594 of Turkish Standards Institution, Titrimetric Method of the Determination of Ethanol Content in Fruit and Vegetable Products, was used.[30] In this study, colas, orange sodas, 100% fruit juices, fruit nectars, sodas with fruit aromas, energy drinks, ayran and kefir of the well-known brands were used as the non-alcoholic beverage test samples. Several brands of each of non-alcoholic beverage type were evaluated among themselves comparatively. Also, the effect of storage time on alcohol content was investigated. The concentration of ethanol in six different kinds of non-alcoholic drinks kept at room temperature in a dark cabinet was determined at 30 day intervals for a 60 day period, by opening a new bottle for each analysis time.

MATERIALS AND METHODS

Materials and Chemicals

In the selection of samples; various well-known brands of fruit juices and nectars, gaseous, colas, fruit sodas, orange sodas and milk products, which are defined as non-alcoholic beverages, were purchased from markets in Türkiye.

Iron(II)-1,10-phenanthroline was purchased from Merck. Potassium dichromate was purchased from Abbott. Ammonium Iron(II) Sulphate and ethanol were purchased from Riedel-de Haen. Sulphuric acid was obtained from Carlo Erba and calcium oxide was obtained from Horasan Kimya. All of the reagents used in the experiments were of analytical grade. Deionized distilled water was used to prepare all the reagent solutions.

Sample Preparation

Non-alcoholic beverages were shaked well, and 100 mL from each beverage was used to determine ethanol content with the test method TS 1594 of Turkish Standards Institution named "Titrimetric Method of the Determination of Ethanol Content in Fruit and Vegetable Products".

Distillation

100 mL beverage sample was diluted with 150 mL deionized distilled water and transferred to a flask. Although normal distillation glassware is used in the standard method for ethanol distillation, a Clevenger

type distillation apparatus was adopted for the first time for better practicallity (Figure 1). Before distillation was started, the sample was made basic with calcium hydroxide suspension (pH: 7.8-8.2). To control the speed of boiling, boiling stones were placed in the distillation flask. Ten mL pure water was added to 100 mL empty volumetric flask, and closed end of the distillation apparatus was placed into water, and the ethanol from the sample was distilled by hydro-distillation for 1 h, cooling fluid circulator set at -10 °C.

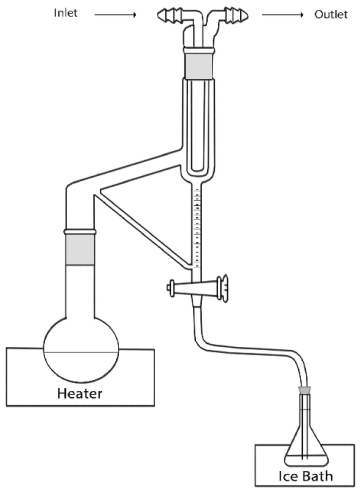

Figure 1. Clevenger type distillation system

About 80-85 mL of distillate was collected and diluted to 100 mL.

Oxidation

Five mL of potassium dichromate solution and 15 mL deionized distilled water were added into 250 mL volumetric flask, and then 20 mL sulphuric acid solution was added and stirred. Ten mL of distillate was added into this flask and incubated for 30 min.

Titration

The excess of dichromate was back-titrated by ammonium iron (II) sulphate. When the color of the solution turned blue-green, four drops of ferrous(II)-1,10-phenanthroline solution was added. Then ammonium iron (II) sulphate solution was added continuously until the color of the solution turned from blue-green to brown.

Calculation

The ethanol content was calculated as mass per volume percentage according to the formula available in the TS 1594 standard.[30]

RESULTS AND DISCUSSION

Various flavors are used in the non-alcoholic beverages. These essential ingredients are mostly non-polar in character and insoluble in water. These essences are dissolved in a suitable carrier solvent system like ethanol. Besides, ethanol can be found in very small amounts in the fruits used in the production of drinks. In addition, ethanol can occur as fermentation product as well.

Because of changing world conditions and growing population, foods which are obtained naturally are becoming insufficient; therefore a large number of additives are generally used in unnatural processed foods to meet food demands. So, Muslims are worried about certain food and drinks as they may contain the ingredients that are forbidden according to Islamic law, i.e. haram.[31]

In the production of non-alcoholic beverages small amounts of ethyl alcohol may be used as solvents of additives, which cause a controversy in Islamic countries. The main problem here is that if that small amount of alcohol which does not naturally exist in the components of the drink is considered unclean. The question is whether that amount is intoxicating or istihlak has occurred in the soft drink where small amount of alcohol is diluted dramatically.[32]

When non-alcoholic beverages are evaluated in terms of the concept of Halal Food, the small amount of ethyl alcohol

used as solvent remain unchanged and is absorbed through digestion. So, these beverages are considered to be illegal (haram) or suspicious.[32]

Regulations regarding non-alcoholic beverages vary according to countries. In Türkiye, production and control of non-alcoholic beverages are done according to Non-alcoholic Beverages Notification of Turkish Food Codex (2007/26). In non-alcohol beverages, the highest allowable level of ethyl alcohol content is 3.0 g / L.[33]

In this investigation, the ethanol contents of non-alcoholic beverages were determined. The test samples selected were the non-alcoholic beverages 100% fruit juices, fruit nectars, sodas with fruit aromas, colas, orange sodas and milk products of the well-known brands in Türkiye. For the measurement of ethanol, the test method TS 1594 of Turkish Standards Institution, Titrimetric Method of the Determination of Ethanol Content in Fruit and Vegetable Products, was used. The method was based on the distillation of ethanol followed by oxidation of ethanol in distillate with potassium dichromate in sulfuric acid solution and then titration of the remaining dichromate with ammonium iron(II) sulfate using iron(II)-1,10-phenanthroline as indicator. A variation was made in the method by using Clevenger type apparatus for distillation of ethanol from the sample for simplicity and better reproducibility.

Firstly, standard calibration graphs were prepared with the standard ethanol with and without distillation in duplicate measurements. Because the curves of the two experiments showed near full overlap (Figure 2) proving high recovery for the distillation method with the use of Clevenger system, calibration graphs used in subsequent tests were obtained with ethanol solutions directly without distillation.

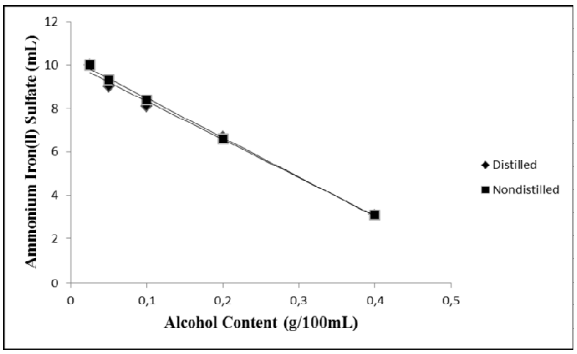

Figure 2. Standard Calibration Graph for Ethanolic Standards with Distillation and without Distillation

The findings showed that the highest concentrations (mg/L) of ethanol in the sample series were as follows: colas 0.14, orange sodas 0.53, other fruit flavored sodas 0.88, energy drinks 0.47, fruit juices 1.46, and milk products 0.25 (Figures 3-6).

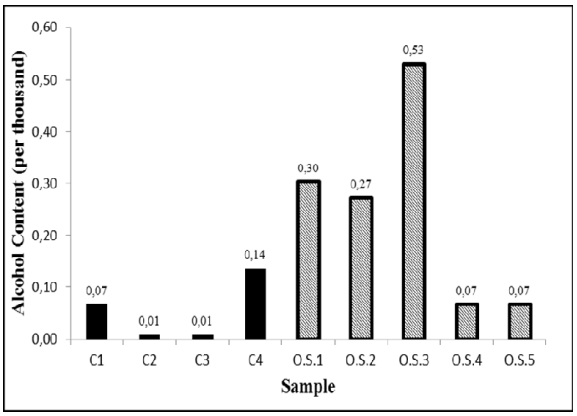

Figure 3. Quantities of Alcohol per Thousand in Several Brands of Colas and Orange Sodas; C: Cola; O.S.: Orange soda

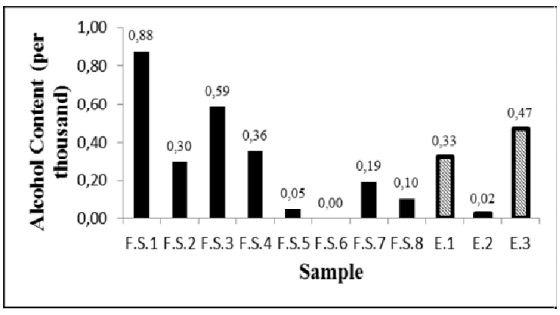

Figure 4. Quantities of Alcohol Per Thousand in Several Brands of Fruit Sodas and Energy Drinks F.S.: Fruit Soda, E.: Energy Drinks

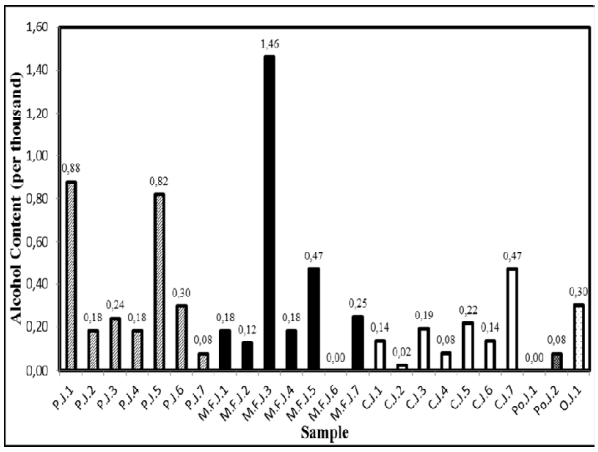

Figure 5. Quantities of Alcohol Per Thousand in Several Brands of Fruit Juices, P.J.: Peach Juice, M.F.J.: Mix Fruit Juice, C.J.: Cheery Juice, Po.J.: Pomegranate Juice, O.J.: Orange Juice

Moreover, the concentration of ethanol in six different kinds of non-alcoholic drinks was determined for a 60-day period to show the effect of storage time on alcohol content in 2010 (Figure 7). While no regular increase or decrease was observed in cola 1, cola 2 and fruit soda 9, there was an increase in fruit soda 1 and mix fruit juice 4, and there was a decrease in fruit soda 6 during the storage.

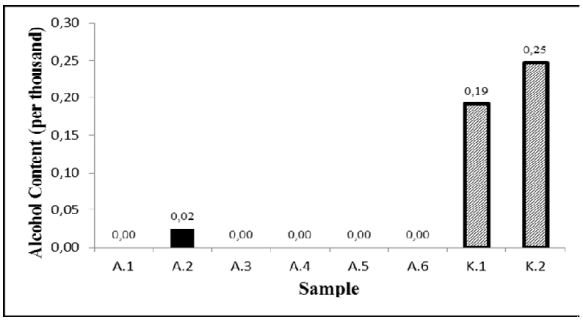

Figure 6. Quantities of Alcohol Per Thousand in Several Brands of Ayran and Kefir, A.: Ayran, K.: Kefir. All the ayran samples except A.2 did not show any detectable ethanol levels.

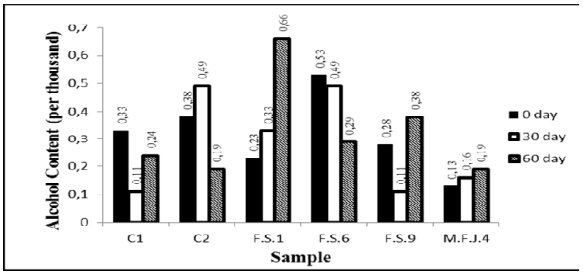

Figure 7. Alcohol Amount According to The Shelf Life of Non-Alcohol Beverages, C: Cola, F.S.: Fruit Soda, M.F.J.: Mix Fruit Juice

CONCLUSION

Hydro-distillation of ethanol with a Clevenger-type distillation apparatus applied for the first time here is simple, showed good compatibility to the reference method TS 1594 of Turkish Standards Institution, Titrimetric Method of the Determination of Ethanol Content in Fruit and Vegetable Products. Recovery was high as the curves of the standard ethanol with and without distillation showed near full overlap (Figure 1), and also system design has been simplified.

The highest per thousand concentration of ethanol in the samples were as follows: colas 0.14, orange sodas 0.53, other fruit flavored sodas 0.88, energy products 0.47, fruit juices 1.46, and milk products 0.25. These values fall well below the highest allowable level of 3.0 g/L according to Non-alcoholic Beverages Notification of Turkish Food Codex.

The ethanol determined in the samples is not thought to be the result of ethanol addition during preparation or processing. A well determined control and testing system is needed to determine intentional alcohol addition, as for example solvent for aroma compounds, and new methods should be developed.

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REFERENCES

1. Danaei, G. Hoom, S. V. Lopez, A. D. Murray, C. J. Ezzati, M. (2005). "Causes of cancer in the world: comparative risk assessment of nine behavioural and environmental risk factors". Lancet, 366(9499): 1784-93.
2. Teyssen, S. Singer, M. V. (2003). "Alcohol-related diseases of the oesophagus and stomach". Best Practice & Research Clinical Gastroenterology, 17(4): 557–573.
3. Eckardt, M. J. File, S. E. Gessa, G. L. Grant, K. A. Guerri, C. Hoffman, P. L. Kalant, H. Koob, G. F. Li, T. K. Tabak, B. (1998). "Effects of Moderate Alcohol Consumption on the Central Nervous System". Alcohol Clin Exp Res., 22(5): 998-1040.
4. Gemes, K. Janszky, I. Ahnve, S. Laszlo, K. D. Laugsand, L. E. Vatten, L. J. Mukamal, K. J. (2016). "Light-to-moderate drinking and incident heart failure — the Norwegian HUNT study". International Journal of Cardiology, 203: 553–560.
5. Wild, S. (2015). "Muslim Translators and Translations of the Qur'an into English". Journal of Qur'anic Studies, 17(3): 158-182.
6. Michalak, L. Trocki, K. (2002). "Alcohol and Islam". Kettil Bruun Society Annual Meeting, Paris. 523.
7. Glassner, B. Berg, B. (1985). "Jewish-Americans and alcohol: processes of avoidance and definition". The American Experience with Alcohol, 93–108.
8. Stivers, R. (1985). "Historical meanings of Irish-American drinking". The American Experience with Alcohol, 109–130.
9. Ablon, J. (1985). "Irish-American Catholics in a west coast metropolitan area". The American Experience with Alcohol, 395–410.
10. Hilton, M. E. (1986). "References to alcohol in the Mormon scriptures". Drinking Drug Pract. Surveyor, 21: 13–16.
11. Caputi, A. Ueda, M. Brown, T. (1968). "Spectrophotometric Determination of Ethanol in Wine". Am. J. Enol. Vitic, 19: 160-165.
12. Calull, M. Marce, R. M. Borrull, F. (1992). "Determination of carboxylic acids, sugars, glycerol and ethanol in wine and grape must by ion-exchange high-performance liquid chromatography with refractive index detection". Journal of Chromatography A 590(2): 215-222.
13. Caputi, A. J. Wright, D. (1969). "Collaborative study of the determination of ethanol in wines by chemical oxidation". J. Assoc. Off. Anal. Chem., 52: 85-88.
14. Singh, N. M. Berkes, I. (1980). "Spectrophotometric determination of ethanol by an enzymatic method with 2, 2'-azino-di(3-ethylbenzthiazo- line-6-sulfonate". Analytica Chimica Acta, 115: 401-405.
15. Jones, A. W. (1995). "Measuring ethanol in saliva with the QED enzymatic test device: comparison of results with blood and breath alcohol concentration". J. Anal. Toxicol., 19: 169-174.
16. Mason, M. (1983). "Report of the technical subcommittee on ethanol determination via immobilized enzyme". J. Am. Soc. Brew. Chem., 41: 89-90.
17. Mason, M. (1983). "Ethanol determination in wine with an immobilized enzyme electrode". Am. J. Enol. Vitic., 34: 173-175.
18. Kokovkin, V. V. Smolyakov, B. S. (1995). "Ethanol determination in aqueous solutions and wine stocks by potentiometry with ion selective electrodes". J. Anal. Chem., 50: 569-573.
19. Antonelli, A. (1994). "Ethanol determination by packed GLC: a quick method with small sample amount and high sensitivity". Vitic. Enol. Sci., 49: 165-167.
20. Naviglio, D. Romano, R. Attanasio, G. (2001). "Rapid determination of ethanol content in spirits and in beer by high resolution gas chromatography". Italian Food Bever. Technol., 24: 19-21.
21. Collins, T. S. Miller, C. A. Altria, K. D. Waterhouse, A. L. (1997). "Development of a rapid method for the analysis of ethanol in wines using capillary electrophoresis". Am. J. Enol. Vitic., 48: 280-284.
22. Kupina, S. A. (1984). "A simultaneous quantitation of glycerol, acetic acid and ethanol in grape juice by high performance liquid chromatography". Am. J. Enol. Vitic., 35: 59-62.
23. Yarita, T. Nakajima, R. Otsuka, S. Ihara, T. A. Takatsu, A. Shibukawa, M. (2002).

"Determination of ethanol in alcoholic beverages by high-performance liquid chromatography-flame ionization detection using pure water as mobile phase". J Chromatogr A., 976(1-2): 387-391.

24. Sanford, C. L. Mantooth, B. A. (2001). "Determination of ethanol in alcohol samples using a modular Raman spectrometer". J. Chem. Edu., 78: 1221.
25. Van den Berg, F. W. J. Osenbruggen, W. A. Smilde, A. K. (1997). "Process analytical chemistry in the distillation industry using near-infrared spectroscopy". Proc. Contr. Qual., 9: 51-57.
26. Mattos, I. L. Sartini, R. P. Zagatto, E. A. G. Reis, B. F. Gine, M. F. (1998). "Spectrophotometric flow injection determination of ethanol in distilled spirits and wines involving permeation through a silicon tubular membrane". Anal. Sci., 14: 1005-1008.
27. Wagner, K. Bilitewski, U. Schmid, R. D. (1992). "Flow injection analysis of wine - accomplishments and needs". Microchem. J., 45: 114-120.
28. Mason, M. (1983). "Report of the technical subcommittee on ethanol determination via immobilized enzyme". J. Am. Soc. Brew. Chem., 41: 89-90.
29. (https://bilinmeyenler.wordpress.com/2007/06/03/gazoz-cola-fanta-asitli-iceceklerin-hepsinde-alkol-var/). Erişim: 03.06.2007.
30. Turkish Standard – TS 1594, ISO 2448 (2001). Fruit and vegetable products – determination of ethanol content, Türk Standartları Enstitüsü, Ankara.
31. Çelik, T. (2013). "Halal and Healthy Food in Hadith Relations". Int J Health Nutr., 4(3): 70.
32. Akkaya, K. M. İlim, E. Şimşek, M. (2013). "Istihlak and Istihalah in Islamic law". Int J Health Nutr., 4(3): 72.
33. Anonymous. Official Journal Date and Number: 15. 06. 2007-26553, Ministry of Agriculture and Rural Affairs, Non-alcoholic Beverages Notification of Turkish Food Codex, Notification Number: 2007/26.