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Study of High Performance Liquid Chromatography Grade Water Quality in Iran

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ABSTRACT

Background: The use of high performance liquid chromatography (HPLC) grade water is very important for laboratory studies, especially food quality control. These studies have a great impact on diagnosing food health. Food health improves the health of the community and these two parameters are closely related. HPLC grade water is ultra-pure water with low UV absorption ability. This water is often purified through a micron filter and all contaminants are removed. **Methods:** Thirty samples of HPLC grade water with Iranian and non-Iranian brands purchased from Tehran were tested for electrical conductivity and silica. **Results***:* The quality of all the tested samples was approved which was lower than the allowable limit. None of the Iranian and non-Iranian samples exceeded the standard limit. **Conclusion:** This study indicated the good quality of this product. It was concluded that all the samples had desirable quality.

Keywords: *Water; HPLC; Food control*

Introduction

High performance liquid chromatography (HPLC) is a valid and reliable technique that has become a practical tool in many laboratories. The HPLC Analytical Laboratory faces problems that can waste time and resources. One of the most common problems in HPLC is the quality of the solvent/mobile phase. Contaminants in the solvent can affect the performance of HPLC in a variety of ways (Safavizadeh *et al.*, 2020, Safavizadeh *et al.*, 2021, Snyder *et al.*, 2012).

In the HPLC analysis, water is essential in the preparation of mobile phases and samples. If impurities are present in the moving phase, they can amplify the background signals. In this way, the base noise and drift increase or cause other phenomena, such as negative peaks or unknown peaks during slope washing. Also, the presence of impurities in the sample solutions can cause unknown peaks. HPLC grade water is a type of water that loses its impurities due to the distillation process (Arabi *et al.*, 2020, Wang and Kasperski, 2018).

In fact, water is boiled and distilled using special devices, and finally HPLC grade water is obtained by separating gases and suspended mineral particles. Depending on the type and amount of distillation, this water can be up to 95 pure. As a result, there are almost no minerals in it. This water is used for various and important purposes (Saini *et al.*, 2017).

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Ordinary water, or treated city water, does not contain any pollutants or dangerous salts. However, it is not completely pure and contains salts, such as calcium and magnesium. These minerals, which are found in mineral water, do not pose a threat to the health of the body, and sometimes even their presence is necessary for the better functioning of the body organs. However, sediments left by these salts may in some cases pose a threat to human health (Senevirathne *et al.*, 2018).

HPLC grade water, in addition to being free of salts and minerals, has no calories or sugar. On the other hand, the most important difference between this water and municipal water is how it is processed. HPLC grade water is obtained by the process of evaporation at high pressure. For this reason, it can be said that HPLC grade water is the product of changes in the structure of urban drinking water (Zhang *et al.*, 2017).

HPLC grade water does not contain any solutes. For this reason, it does not have the property of contrasting with some chemicals. This feature has led to the use of HPLC Grade water in laboratories and scientific and research centers for activities, such as dissolving chemicals or even washing laboratory dishes. On the other hand, the highest consumption of this water is in the pharmaceutical industry and medical applications due to its high purity. You have seen the smallest example of using HPLC grade water in chemistry and food analysis (Ghafurian *et al.*, 2019).

One of the most important properties of HPLC grade water is that it does not leave any sediment due to evaporation or excessive use. For this reason, it is also used to keep car batteries moist, since ordinary water sediment can damage the battery and eventually the car engine after a while (Senevirathne *et al.*, 2018).

HPLC grade water is also used to make raw materials in various factories, especially food. The purity of this water has led many people to drink it instead of municipal water. Measuring pH, oxidizing materials, and residues after evaporation in this type of water is not practical due to measurement problems of this purity (Zhang *et al.*, 2017).

HPLC grade water free of any minerals is essential. Drinking this water, although in many cases recommended for some people with certain diseases, but also carries risks. The purity of this water reduces the error in the tests and increases the accuracy and has a very high impact on the results (Ouyang *et al.*, 2017). The present study aimed to evaluate the quality of HPLC grade water available in the market for the first time.

Materials and Methods

Thirty samples of HPLC grade water were purchased from the relevant stores in Tehran during the period of January 1st to February 1st 2020. The samples had 5 domestically produced samples and 25 foreign samples.

The electrical conductivity was measured using COND 7310 conductivity meter, WTW Co., Germany, and its standard limit according to Iranian standard 1728 is 0.01 MS/s.

In general, the instructions for measuring silica in water were generally divided into the five or six categories as follows:

Dye formation: For 50 ml of the sample, 10 ml of 1 + 1 hydrochloric acid and 2 ml of ammonium molybdate reagent were added to the sample in rapid sequence. They were mixed at least 6 times and placed on the mixer for 5 to 10 min. Two ml of dissolved oxalic acid was added to the sample and mixed thoroughly again. After 2 to 15 min, the color value was recognized, the desired time was calculated from adding oxalic acid to the sample. Given the yellow color of the sample may follow the beer rule, it was better to read the color value with a photometer.

In order to find some unreacted silica solids, the sample was digested with NaHCO3 before the dye formation step. Certainly this amount of digestion is not sufficient to convert all amounts of unreacted silica molybdate to reacted silica molybdate. Silicate compounds as well as silicate polymers have a high melting point with alkalinity at high temperatures. The digestion step can be omitted if all known silicates react with molybdate. Then,

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200 mg of NaHCO3 was added without digestion and placed in digestion reactors for 1 h. The analysis cannot be stopped in time, but all the steps must be continued until the end. Fifty ml of Nessler tube was carefully transferred to it and its value was marked (Regnault *et al.*, 2004, Toppner, 2014).

Preparation for colorimetry (or determination of turbidity): For each sample that needs to be prepared, there was a separate blank or control sample. For the measurement, two of the above cases should be made according to the function of the instructions. The necessary reagents should be added to one of them, and the other sample, which is the control and blank sample, should be oxalic acid and hydrochloric acid, without adding Molybdate. The control sample or blank without any molybdenum was put in the device and did not show the device below (Paul *et al.*, 2019).

Calculations and measurements: The device was turned on and the absorption rate was reduced to zero with HPLC grade water and all the standards were read, including control reagents, in front of the HPLC grade water blank. Plot micrograms of silica eventually appeared in the 55 ml solution for reading in the Fluorimeter. The blank sample was put in the machine and a standard was applied for each group of samples, so that the calibration curve drawn by the machine was not disturbed (Regnault *et al.*, 2004, Toppner, 2014).

$$\text{mg } SiO_2/L = \frac{\mu g\ SiO_2 \text{ (in 55 mL final volume)}}{\text{mL sample}}$$

Results

The samples were analyzed in three replications for electrical conductivity and silica. The results of electrical conductivity at 25 °C of HPLC grade water samples are shown in **Tables 1** and **2**.

Table 1. The results of electrical conductivity at 25 °C of HPLC grade water samples.

Samples	The number of samples higher than the standard level ms/s 0.01	Highest electrical conductivity ms/s	Lowest electrical conductivity ms/s	Average electrical conductivity ms/s
Imported (n=25)	0	0.01	0.006	0.009
Made in Iran (n=5)	0	0.01	0.005	0.01

Table 2. Measurement of silica in HPLC grade water samples.

Samples	The number of samples higher than the standard level mg/l 0.01	The highest amount of silica mg/l	The lowest amount of silica mg/l	Average amount of silica mg/l
Imported (n=25)	0	0.01	0.0001	0.001
Made in Iran (n=5)	0	0.01	0.003	0.006

Discussion

HPLC is a reliable and established technique used in laboratories to separate, identify, and quantify components in a mixture (Gika *et al.*, 2016). In HPLC analysis, water is used in mobile phases and sample preparation. Water is a universal solvent as it dissolves most substances, but ordinary tap water cannot be used for laboratory applications due to the presence of contamination (Treder *et al.*, 2020). Water contaminants include dissolved atmospheric gases, natural minerals, and organic substances, dissolved solids, and suspensions and bacteria or microorganisms if the necessary nutrients and environmental conditions are present to support them (Edition, 2011). Water for laboratory use requires different degrees of treatment for the required applications. Reagent grade water is defined as water suitable used in a specified procedure, so that it does not interfere with the specificity, accuracy, and the precision of the procedure (Furusawa, 2013). In HPLC analysis, water is essential in preparing mobile phases and samples. If impurities are present in a mobile phase, they can increase background signals that can increase baseline noise, drift, or cause other phenomena, such as negative peaks or unknown peaks during gradient elution. Similarly, if impurities are present in sample solutions, they can cause unknown peaks (Raval and Patel, 2020). If impurities include a target component being analyzed, they can even affect quantitation values. To prevent struggling with such problems and to increase data reliability, this study discussed some basics about water (Paul *et al.*, 2019). General laboratory applications include glassware washing and rinsing, reagent and buffer solution preparation, making blanks and standard solutions for calibration purposes (Parriott, 2012). Ultra-pure or HPLC grade water is used for HPLC analysis. HPLC grade water is ultra-pure water with low UV-absorbance wherein specific conductivity is maintained by water purification systems. They are often filtered through a 0.2 micron filter and sealed in solvent-rinsed containers under an inert atmosphere to avoid contaminations (Blum, 2014, Sailaja *et al.*, 2014). Reputable suppliers of chemicals and reagents provide water for HPLC in bottles; however, it is advisable and economically viable to install commercially available dispensing units in the long run which will cater to your water requirements for other sophisticated analytical techniques other than HPLC. Such systems are designed for large volume outputs and provide other useful features, such as handheld controlled dispensing, sensors indicating the levels inside the internal storage tank, and external spillage prevention. System components normally include pre- and post-filtration capsules for removing dissolved solids above 0.2 μ size (Lindsay and Barnes, 1992). Activated carbon filters provide freedom from organics and dissolved chlorine. Mixed bed deionizers control conductivity and UV irradiation source remove organic traces. Reverse osmosis system and a water storage tank complete such systems (Horváth, 2013, Xiao and Oefner, 2001). So far, no similar research has been conducted in this area and it was examined for the first time in the present study. This study showed that the HPLC grade waters used in Iran are of good quality and are suitable to be used in research studies.

Conclusion

The results showed that the samples available in the Iranian market have all the required quality in terms of standards and none of the samples in terms of electrical conductivity and silica have values higher than the national standard, which is a sign of high quality of these products. This study was conducted for the first time in this regard and its results will be shared with other researchers. It can be concluded that the quality of these products is generally acceptable and suitable for important tests, such as food quality control. It seems that due to the appropriate quality of domestic products, more planning should be done on more production and quality of this product in the country.

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Authors' contributions

The both authors involved in study design, analysis, and writing of the manuscript.

Conflict of interest

The authors declare that there is no conflict of interest in this study.

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