



# Evaluating the Effect of Ultraviolet Radiation on the Total Number of Microbes in Apple Juice, Grape Juice and Orange Juice at Different Times of Radiation

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## Abstract

**Background and aim:** Fruit juice is one of the best drinks that provide a major part of the body's needs for vitamins and is considered a suitable source to compensate for wasted water, but if health standards are not met, it is considered as a potential source of contamination. The purpose of this research is to evaluate the effect of ultraviolet radiation on the microbial load of apple, grape and orange juice during the radiation period.

**Materials and Methods:** 500 ml of fruit juice including natural apple, grape and orange juice were randomly provided from fruit juice supply centers in the Shahrekord city and then the fruit juice was passed through device to perform tests (manufactured by Pars UV company, model 1X). The microbial load measurement method was carried out in accordance with the standard-4207 of Iran's National Standard Organization and using serial dilution method, and 4 and 3 dilutions of the tubes were prepared. Then, they were radiated in zero, 30, 60 and 120 second treatments. The data was statistically analyzed by SPSS software and the one-way ANOVA method was used to evaluate the data.

**Results:** Based on the statistical analysis, it was found that the highest lethality was related to the time of 120 seconds in all three types of fruit juice and there was a significant relationship between the duration of UV radiation and the reduction of the microbial load in the fruit juice.

**Conclusion:** According to the analyzes obtained from the UV method on the reduction of the microbial load of fruit juices, it could be said that if this method is radiated effectively to liquids, then it can have a significant effect on reducing the bacterial contamination.

**Key words:** UV, Fruit juice, Microbial bar, Shahrekord, Microbiology

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## Introduction

Drinks that are sold on the streets are considered to be one of the traditional foods of any country. These drinks are important for knowing the local food habits around the world, in addition, they play an important role in preserving cultural and social heritage. Since they provide an income for sellers, they are also important in creating employment. These drinks are highly demanded by both sellers and consumers. Approximately 2.5 billion people around the world consume street foods and beverages every day. Making and selling drinks on the streets provides a steady income for millions of people who have poor health knowledge (Worku *et al.*, 2011).

Due to the tremendous growth of food vendors on the side of the roads and crowded parts of every region who do not have sufficient understanding of the basic issues of food safety, the prevalence of infections and poisonings has increased. The main sources of microbial contamination include the place of preparation, cooking and serving utensils, raw materials, abuse of time and temperature of cooked foods and personal hygiene of sellers (Lattanzi *et al.*, 2019). Various studies have identified the sources of food safety issues involved in street foods as microorganisms belonging to the genus *Bacillus*, *Staphylococcus*, *Clostridium*, *Vibrio*, *Campylobacter*, *Listeria*, *Salmonella* and *Escherichia coli* and almost all the Enterobacteriaceae family. In this regard, one of the foods that is consumed on the streets and also has many fans are fruit juices. Due to their acidity, fruit and vegetable juices were not recognized as the cause of food-borne diseases until recently, but recently, several outbreaks of *Escherichia coli* O157:H7, *Salmonella typhimurium* and *Listeria monocytogenes* have occurred in fruit and vegetable juices. In the United States, between 1995 and 2005, 21 human outbreaks associated with fruit juice were reported to the Centers for Disease Control and Prevention (CDC) (Ndlovu, 2020; Petruzzi *et al.*, 2020).

Juice is a liquid that is usually found in plants and is one of the most common drinks of people around the world or in other words, it is a non-fermented but fermentable product, in some cases transparent and blur in some items, which is obtained by mechanical or manual methods from healthy and ripe fruit and it is directly used for drinking and due to having all the

micronutrients, salts and vitamins, it provides a significant part of the body's needs and it is a suitable source for compensating the water lost by the body due to heat or sports activities. Fruit juices contain natural antioxidant compounds that prevent many diseases, including cardiovascular diseases and certain cancers (Conidi *et al.*, 2020; Ho *et al.*, 2020; Scheffers *et al.*, 2020).

Irradiation in food means exposing the food to a certain amount of radiation, in order to prevent the germination of some food products such as onions and potatoes and also, controlling storage pests, reducing microbial and fungal contamination of some products such as saffron and spices, and delaying the ripening of some fruits in order to increase their storage time and ultimately reduce the microbial load in juices and other fat-free liquid materials, and this method is also used in studies related to plant nutrition, like the way of absorbing fertilizers and elements, that by using the technique of nuclear radiation, the desired genetic changes can be used to improve the product in plant masses (He *et al.*, 2021). The radiation used in the food industry and food hygiene has its own characteristics. For example, they must have a short wavelength and in a general classification they are divided into two separate groups including: A. Radiation with a lower frequency and less energy, such as ultraviolet, which has enough energy to only excite molecules. This spectrum is used in food hygiene and food industry and is called ultraviolet radiation. B. Radiations with higher frequencies that have a large amount of energy and are able to break down molecules into ions, and hence they are called ionizing radiations which include: X-rays, gamma rays, cathode rays, beta rays, protons, neutrons and alpha rays (Gayán *et al.*, 2014). Food is usually irradiated using gamma waves and through a radioisotope source, electrons or X-rays produced through an electron accelerator. The radioisotope used in most radiations is natural cobalt (60) and cesium 137 (Van Dyk *et al.*, 2013). In this regard, it is very important to investigate the effect of radiation on the reduction of bacterial load and the purpose of the present study is to evaluate the effect of ultraviolet radiation on the microbial load of apple juice, grape juice and orange juice offered in the juice shops of Shahrekord city at different times of irradiation.

## Materials and Methods

500 cc natural apple juice, grape juice and orange juice offered in the juice shops of Shahrekord city were prepared in such a way that it is free of suspended substances. The fruit juices were passed through a suitable filter to remove suspended matters. Then, the fruit juice was purified from the UV device (manufactured by Pars UV Company, Model 1X). Using a turbidity meter, the turbidity of each fruit juice was measured and they were irradiated before and after irradiation in 4 treatments before irradiation (zero), 30, 60 and 120 seconds. The microbial load measurement method was carried out in accordance with standard 4207 of the National Standards Organization of Iran and using the serial dilution method. The dilution was done for all 3 types of juice in 3 and 4 tubes according to the standard and agar plate culture medium was used for the total number of bacteria at a temperature of 37 degrees and after 24 hours, the colonies of each sample were counted.

## Statistical analysis

For statistical analysis, SPSS software version 22 and one-way ANOVA statistical analysis were used; Also, The level of significance in this study was considered ( $P < 0.05$ ).

## Results

Based on the results obtained from the statistical analyses, the microbial load between the 3 samples of the tested fruit juices was decreased. The amount of colonies in the culture medium decreased with the increase in the time of fruit juice storage against radiation and could be seen in a stepwise manner (table 1). In this experiment, the higher the treatment time; microbial load decreased. The amount of material concentration can have a high impact on this case, the higher the turbidity of the material, the higher the microbial load, and the lower the turbidity, the lower the microbial load could become, because the amount of light absorbed by fruit juice increases and penetrates deeper into the liquid (table 2).

Juice Type	Apple Juice	Orange Juice	Grape Juice
<b>Irradiation Amount (Based on Time)</b>			
<b>Zero</b>	$35 \times 10^3$	$45 \times 10^3$	$40 \times 10^3$
<b>30</b>	$29 \times 10^3$	$35 \times 10^3$	$32 \times 10^3$
<b>60</b>	$17 \times 10^3$	$21 \times 10^3$	$19 \times 10^3$
<b>120</b>	$4 \times 10^3$	$7 \times 10^3$	$4 \times 10^3$

**Table 1.** The average of fruit juices colonies from 3-step dilution series.

Juice Type	Apple Juice	Orange Juice	Grape Juice
<b>Irradiation Amount (Based on Time)</b>			
<b>Zero</b>	$33 \times 10^4$	$38 \times 10^4$	$35 \times 10^4$
<b>30</b>	$27 \times 10^4$	$31 \times 10^4$	$29 \times 10^4$
<b>60</b>	$16 \times 10^4$	$19 \times 10^4$	$15 \times 10^4$
<b>120</b>	$2 \times 10^4$	$5 \times 10^4$	$3 \times 10^4$

**Table 2.** The average of fruit juices colonies from 4-step dilution series.

## Discussion

The studies of Baykus *et al.*, in 2021 on the reduction of microbial load in the effect of radiation showed that 120 seconds of ultraviolet radiation causes a reduction of 2.5 bar logarithm of pollution,

which is equal to the result of this research (Baykuş *et al.*, 2021). Andrea *et al.*'s study in 2018 on the effects of radiation on reducing the microbial load of fruit juice showed that the amount of 120 seconds causes a significant decrease in the amounts of bacteria, such

that in this research, the microbial load has decreased by 90% (Acevedo *et al.*, 2018). In a similar study conducted by Nyhan *et al.*, they reported that UV rays reduced the effects of up to 3 log on microorganisms (Nyhan *et al.*, 2021). Another similar study conducted by Turkmen *et al.* on reducing the amounts of microorganisms in fruit juices showed that irradiation for 5 minutes caused the destruction of all pathogenic microorganisms (Türkmen & Takci, 2018). In 2019, Cava *et al.*, showed that 16 seconds of UV radiation to fruit juice caused a 14% reduction in pathogenic microorganisms in fruit juice (La Cava & Sgroppo, 2019). In the study of Keyser *et al.*, in 2008, they showed that the destructive power of UV rays at 230 nm caused a 67% reduction in the amount of contamination in juices (Keyser *et al.*, 2008).

Quintero-Ramos *et al.*'s studies in 2004 to investigate the reduction of bacterial contamination showed that the effect of 10 seconds of UV rays caused a 5% reduction of bacterial contamination, which is somewhat consistent with the results of this study (Quintero-Ramos *et al.*, 2004). In a similar study, Gayán *et al.* found that the effect of UV rays on reducing the microbial load in orange juice was significant and has caused a significant decrease in *Escherichia coli* bacteria, which is consistent with the present study (Gayán *et al.*, 2013). In a similar study conducted on the reduction of the bacterial load of *Escherichia coli* O157H7 in carrot juice, the researchers found that 5 logarithmic cycles compared to the time before irradiation were reduced in a time equivalent to 23 seconds which is somehow equal to the results of the present research (Yin *et al.*, 2015). Keyser *et al.*'s studies in 2008 showed that UV rays had no significant effect on juices that have a high concentration and puree and the period of 10 seconds has only a logarithmic effect on the reduction of bacteria which is consistent with the present study in this part, because in juices with high turbidity, the effect of radiation did not have a great effect on reducing the amount of bacterial load (Keyser *et al.*, 2008).

### Conclusion

Since fresh fruits have microbial contamination on their surface, therefore, some of the high pollution load can be due to the use of low-quality fruits, improper washing of fruits, non-observance of hygiene principles and standards during the process

of peeling fruits, use of old equipment and lack of disinfection of fruit juice machines and improper washing of the containers for preparing fruit juice, which can increase microbial contamination and this in turn increases the risk of poisoning caused by fruit juice. In addition, another part of the microbial load may be due to the possible contamination of the water used to prepare fruit juice or the ice that is used to cool them, keeping and storing juice in unhygienic conditions, contamination caused by inappropriate containers and juice supply equipment and also, the poor personal hygiene of the workers preparing handmade fruit juices and the inappropriate location can cause the transfer of various bacterial agents, including *Escherichia coli*, to the products.

### Conflict of interest

The authors have no conflicts of interest to declare.

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## ارزیابی اثر اشعه ماورای بنفش بر میزان شمارش کلی میکروب های آب سیب، آب انگور و آب پرتقال در زمان های مختلف تابش

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### چکیده

**زمینه و هدف:** آبمیوه یکی از بهترین نوشیدنی هایی است که بخش عمده ای از نیازهای بدن به ویتامین ها را تامین نموده و منبعی مناسب برای جبران آب هدر رفته به حساب می آید، اما در صورت عدم رعایت استانداردهای بهداشتی به عنوان منابع بالقوه آلودگی به حساب می آید. هدف از این تحقیق، ارزیابی اثر اشعه ماورای بنفش بر بار میکروبی آب سیب، انگور و پرتقال در زمان های تابش است.

**مواد و روش ها:** ۵۰۰ میلی لیتر آبمیوه شامل آب سیب، انگور و پرتقال طبیعی به صورت تصادفی از مراکز عرضه آبمیوه در شهرستان شهرکرد تهیه شد و سپس آب میوه ها جهت انجام آزمایش از دستگاه UV (ساخت شرکت پارس یووی مدل ۱X) عبور داده شد. روش سنجش بار میکروبی مطابق با استاندارد ۴۲۰۷ سازمان ملی استاندارد ایران و با استفاده از روش سریال دابلوشن انجام گرفت و رقت های ۴ و ۳ لوله ای آماده شد. سپس در تیمارهای صفر، ۳۰، ۶۰ و ۱۲۰ ثانیه ای مورد تابش قرار گرفتند. داده ها توسط نرم افزار SPSS مورد آنالیز آماری قرار گرفت و از روش آنوای یکطرفه برای ارزیابی داده ها استفاده گردید.

**یافته ها:** بر اساس آنالیز آماری داده ها مشخص شد که بیشترین میزان کشتندگی مربوط به زمان ۱۲۰ ثانیه ای در هر سه نوع آبمیوه بوده و رابطه معنی داری بین مدت زمان مورد تابش UV و کاهش بار میکروبی در آبمیوه ها بوده است.

**نتیجه گیری:** با توجه آنالیزهای به دست آمده از تاثیر روش UV بر میزان کاهش بار میکروبی آبمیوه ها، می توان گفت که این روش چنانچه به صورت موثر به مایعات تاییده شود، می تواند تاثیر معنی داری بر کاهش آلودگی های باکتریایی داشته باشد.

**واژه های کلیدی:** UV، آب میوه، بار میکروبی، شهرکرد، میکروبیولوژی

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