

# [Quantitation of caffeine in sports gels using hplc](https://assignbuster.com/quantitation-of-caffeine-in-sports-gels-using-hplc/)

Quantitation of caffeine in sports gels using HPLC

Background

It is well known that caffeine is one of the most widely used drugs in the world. Caffeine is a central nervous system stimulant, which can increase blood pressure and heart rate. Otherwise, it can improve athletic performance in aerobic (especially endurance sports ) and anaerobic conditions by delaying the onset of muscle fatigue and central fatigue. Therefore some athletes and fitness persons often use sports gels contained caffeine to boost their brain and body. It is safe to ingest proper caffeine for humans. However, if ingested too much, caffeine would lead to some side effects include restlessness, anxiety, tremors, irregular heartbeat and difficulty of sleeping. More seriously, excessive long-term intake of caffeine may also promote headaches, migraines and high blood pressure in some people. In 2004, European Union began to require packaging beverages with a caffeine content of more than 150 mg/L should be labeled “ high caffeine content” and express caffeine content by mg/100 mL. 1 Considering the harmfulness of excessive caffeine for human body and the requirement of EU, it is necessary to discovery and use a simple and fast method to quantify caffeine.

Extraction method

Caffeine in the sports gels need to be extracted first before being quantified. However, there are not too much experimental paper conducted an extraction method about caffeine in sports gels. From the researches on the extraction of caffeine from tea, coffee, beverage plants and chocolate products, there are two methods work better and can use for reference.

The first one is Soxhlet extraction 1 . Although Soxhlet extraction is more complicated and time-consuming than other simple extraction methods, it is much better from a quantitative aspect. Soxhlet extraction can remove most doubts about efficiency of extraction by making heat, reflux and solvent combined to use. The sample should be crushed into a fine powder and weigh out 10 g into Soxhlet extraction thimble. Place it in the extraction apparatus. Use a 250ml round -bottom as a container and add 100ml ethanol and anti-bumping stones into it. Heat mantle and refluxing mixture for 1. 5-2 hour. After that, allow the extract solution to cool before adding it to a 10% (w/v) aqueous solution of magnesium oxide. The ethanol should be evaporated in a steam bath until a brown pulp is obtained. Add about 125 mL distilled water, and boil the mixture 30 minutes on the steam bath. Filter through a conical coarse filter paper. Add 10 mL of 0. 1 M sulfuric acid solution to the filtrate and boil until volume is reduced to half. Let it cool. Transfer to a dispensing bottle and add 12 mL dichloromethane. Remove the yellow layer after shaking and repeat two more times with fresh dichloromethane. 8 mL of 0. 1 M potassium hydroxide solution was added to the combined extract into the new extract solution. This should be enough to eliminate yellow. Remove the organic layer and wash the base solution layer twice with 5-ml volume of dichloromethane. Add these volumes to earlier extracts. The combined extract was evaporated to approximately 10 mL in a pre-weighed beaker in a steam bath. Finally, a white crystalline precipitate is obtained. Recrystallization with ethanol and gain the caffeine.

The second method is Ultrasound-Assisted extraction method (UAEM) 3 . It is more efficient and can gain better result when applied in different samples of chocolate. First, the fats of the sample need to be removed with petroleum. Poured one gram solid sample into a 250-mL beaker, and add 100 mL of distilled water at 80°C. The ultrasound extraction is carried out at 240 W for 180 s. Then, add 5 mL of Carrez I reagent and NaHCO 3 until precipitation stopped. The mixture is filtered off. Transfer the remaining filtrate into a 100-mL volumetric flask and dilute with distilled water. Next, add 5. 5 mL of 1 mol/L NaOH to the solution make the pH about 12. 5-12. 7 and add 10 mL of chloroform. Put ultrasound probe into the glass at 0. 5 cm from the top surface of the solution. Apply 160 W power for 30 s. The mixture is separated and centrifuged at 6, 000 RPM for 5 minutes, then transfer 1 mL of chloroform phase to a 10 mL volumetric flask and diluted with chloroform to a certain volume to measure caffeine.

Analytical methods

With time elapsing, there are many different kinds of analytical methods were developed to quantify caffeine. For example, in 2000, Wenrui Jin, Daiqing Yu and two people 2 quantified caffeine in human serum and a cola drink by Capillary zone electrophoresis method with end-column amperometric detection at a carbon microdisk array electrode. The result was satisfactory, and the main advantage of this method is excellent selectivity. In 2003, Magali Laasonen, Tuulikki Harmia-Pulkkinen and two people 3 developed and validated a near-infrared spectroscopic method to determine the caffeine concentration in intact single tablets. The result showed that near-infrared analysis of caffeine in tablet form was more flexible and much faster than that performed with HPLC method. At the meantime, High Performance Liquid Chromatography is widely used for the quantitation of caffeine.

## References

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