

# Optimization of colorimetric analysis conditions of flavonoids in pomelo

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## Introduction

The pomelo is one of the most frequently used medical herbs in traditional Chinese medicine. The active principle of this plant consists essentially of a combination of flavonoid compounds. Flavonoid compounds have a high level of physiological activity and many beneficial effects on human health. The increasing applications of pomelo in pharmaceutical and food industries make it necessary to develop an analysis method of flavonoids.

In the chemical laboratory, the quantitative determination of flavonoid compounds was performed using the HPLC method. Despite having good precision, the application of the HPLC method in industrial production is limited, for some reasons such as: high maintenance budget and complicated sample pretreatment. In this article, we look at the analysis of flavonoids by UV-Visible spectrophotometer, which is a relatively simple method and specially adapted to online real-time analysis.

This work aimed to choose a color reagent and optimizing reaction conditions including: color reagent amount and reaction time. The common color reagents used for flavonoids include  $\text{AlCl}_3\text{-NaOH}$ ,  $\text{NaNO}_2\text{-Al}(\text{NO}_3)_3\text{-NaOH}$  and  $\text{KOH}$ . The most suitable color reagent was chosen, by comparing the spectrums after the color reaction. The influence of, the color reagent amount and the reaction time, on the spectrophotometric behavior of flavonoids were also studied by different assays.

## Experimental Conditions

The naringin, which constitutes more than 70% of total flavonoids in pomelom was used as a test sample. The standard solution of naringin was treated with 3 different color reagents. All UV-VIS spectrums were registered in the range of 200 – 500 nm. After a comparison of the spectrums, the reaction conditions were studied with the chosen optimal color reagent. Color reagent amount and reaction time were tested in order to ascertain the relation between their variation and the absorbance of naringin standard solution at 420 nm.

## Results and conclusions

The color reagent was used to avoid interference absorption in the range of 250 – 350 nm. It can be seen from figure 1 that the farthest peak from the interference domain was obtained with  $\text{KOH}$ , which made it the optimal color reagent.

Figure 2 shows the variation of absorbance with the volume of  $\text{KOH}$  solution. The absorbance increased until it reached a maximum with the volume of  $\text{KOH}$  at around 0.5 mL. The decrease after maximum can be explained by naringins known oxidization under very basic conditions. A volume of 0.5 mL was chosen as the optimal color reagent amount.

Figure 3 shows that the absorbance increases with reaction time and stabilizes after 50 min. The optimal reaction time was fixed at 60 min, in order to obtain a compromise between absorbance stability and analysis time.

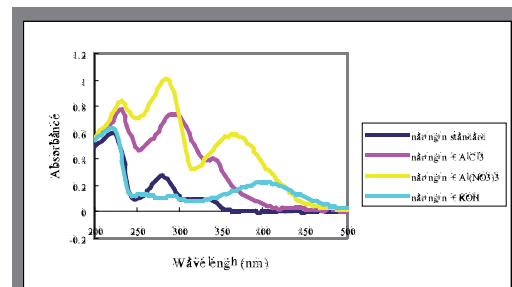


Figure 1: Spectrums of naringin before and after color reactions