## Astringency estimation by Localized Surface Plasmon Resonance

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## Abstract

The popularity of phenolic compounds has increased in the past years due to their antioxidant capacity and association for the prevention of heart diseases <sup>1</sup>, chronic inflammation <sup>2</sup> and cancer <sup>3</sup>. Human diet is the main source of polyphenols provided by products derivate from plants such

Human diet is the main source of polyphenols provided by products derivate from plants such as fruits, vegetables and beverages, and the long term consumption suggests a contribution for potential health benefits. In addition, polyphenols also contribute for the sensorial perception of food products being astringency one of the known sensory characteristics.

Astringency is a mechanism that is still not fully understood. Nevertheless, several studies support the concept that astringency is a tactile sensation rather than a taste. The astringency mechanism is thought to be caused by the polyphenols ability to bind salivary proteins, thus forming complexes that lead to precipitation by promoting the dryness roughening and pucker typical perceived. Furthermore astringency is also an important parameter used to determine the wine quality and it is usually estimated based on sensorial panels which are expensive, time consuming and conferring a certain subjectivity to the process <sup>4</sup>.

In order to overcome these disadvantages, a label free sensory system was performed based on localized surface plasmon resonance (LSPR) by detecting the interaction between a salivary protein and the polyphenols, mimicking the natural interaction which occurs in the mouth. The LSPR is a powerful tool because the metallic nanostructures can be excited by incident light, thereby promoting a collective oscillation of the conduction electrons at a specific wavelength. Structural changes in the local refractive index are induced, when the target molecules bind to the receptor on the surface of a nanoplasmonic, those being tracked by the monitorization of variations at the correspondent wavelength of maximum extinction.

The nanostructures used in this work were gold nanodisks attached to a glass surface fabricated by sparse colloidal lithography. The substrates were prepared according to the following steps : i) cleaning the glass coverslips; ii) spin coating of PMMA, followed by 2 min. in a hot plate; iii) deposition of a triple layer of polyelectrolytes for 30 min each followed by the deposition of polystyrene particles 100 nm size; iv) deposition of 20 nm Ti as a mask; v) removal of particles by tape stripping followed by etching for 10 min vi) deposition of 2 nm Ti and 20 nm Au followed by acetone rising. The resultant nanostructures were gold nanodisks as shown in figure 1. After the fabrication of gold disks,  $\alpha$ -amylase was immobilized on the gold surface according to the optimal conditions previously tested, and the subsequent interaction with pentagalloyl glucose (PGG) was measured by LSPR. The interacting between the polyphenol PGG and the immobilized alpha-amylase displayed a red shift in the spectra which kept shifting with the increasing of PGG concentration as can be seen in figure 2. The interaction between the polyphenol and the salivary protein provided a linear behavior for a concentration range from 0.5 to 155 µM of PGG, which can be seen in figure 3. Red wine samples which were previously analyzed by a sensory panel were also measured and classified in terms of astringency. The wine astringency presented the same order of astringency levels when compared with the sensory panel, indicating the close agreement between both methodologies.

Therefore the proposed sensor offers a simple approach for the estimation astringency based on protein-polyphenol interaction, being the application to real samples successful. Additionally the LSPR nanostructure enhanced the sensing ability and included a simple fabrication due

to the recent advances in nanotechnology.

## References

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Figures

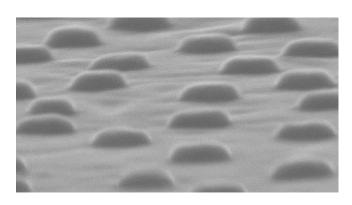
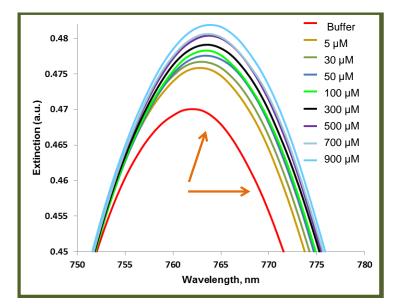
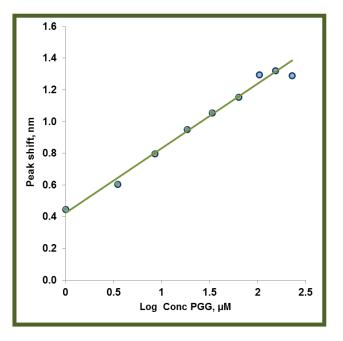


Figure 1 - Side view of the fabricated gold nanodiscs.



**Figure 2 -** LSPR peak shift according resulting from the interaction of the salivary protein with the increasing concentrations of PGG (polyphenols)



**Figure 3** - Linear correlation between immobilized alpha-amylase and PGG (polyphenol) on the surface of gold nanodiks.