CHAPTER 7

Conclusions and Future Direction of Research

Contents

7.1 Conclusions

From previous works on detection and reduction of limonin from citrus fruits, the drawbacks are found to be time-consuming, expensive and not designed for on-site testing. The sensors developed till date such as amperometric biosensor, OECT based sensor are also expensive, time consuming in terms of fabrication. Hence, there is a need for finding out inexpensive device to overcome the present limitations. This work is motivated to design and develop a capacitive sensor based on IDE structure for on-site detection of limonin content as well as reduction in citrus juice with high selectivity and sensitivity.

Firstly, previous works for detection of limonin using various analytical/electrochemical methods have been reviewed in terms of material synthesis, cost, complexities in processing/fabrication, response time, selectivity, sensitivity, etc. It is seen that, in comparison to many existing methodologies, the capacitive sensor based on IDE structure approach for the quantification as well as measure of its reduction is simpler, easier and inexpensive.

An Interdigitated electrodes capacitance sensor and its mathematical-model has been studied considering different electrical fields generated in the device. Simulation of the sensor was carried out in COMSOL multiphysics and MATLAB tool to achieve the effective design parameters of the device such as the matellization ratio, width and gap of the electrode fingers, number of electrode fingers etc. Using these design parameters, the sensor was fabricated with the sensing materials on the IDE patterned paper substrate with Ag as the electrode material. The variation in the sensing film's dielectric property upon exposure to analyte enhances the capacitive response of the sensor. The transverse and fringe capacitances of the capacitive sensor were included in the construction design specifics that are presented. An efficient design for the development of capacitive sensors in sensing applications was provided by device simulations based on the developed capacitance model. We used the aforementioned design settings for the design parameters to construct the inexpensive IDE sensor.

A novel CeO_2 NPs based interdigitated electrodes capacitive sensor has been designed and fabricated for measuring limonin concentration to monitor bitterness in citrus fruit juices. The aqueous extract of D. indica was used to synthesize CeO_2 NPs for the first time utilizing a green synthesis method. For samples of synthesized NPs annealed at various temperatures, UV spectra show peaks in the 400–600 cm^{-1} range, which are attributed to Ce–O stretching. The crystal size of the polycrystalline cerium nanoparticles with fcc structure was observed to rise with an increase in annealed temperature and was determined to be 7.05 nm and 26.15 nm respectively for as-synthesized and annealed (400°C) NPs. Here, green synthesized NPs produced by D. indica have smaller crystallite sizes than NPs produced at the same calcination temperature by pectin and Acalypha indica leaf extract. FESEM images show a size of NPs to be of 70 nm approximately with spherical to polyhedral shape morphology. The IDE capacitive sensor based on CeO_2 exhibits good specificity for measuring limonin in fruit juices. With a sensitivity of ~ 9.62 ±0.095 µF/ppm, a very quick response time of ~13 s, and reusability with less than 13.8% loss of initial capacitance value after one cycle. Two fruit juice samples namely- Citrus limetta and Citrus grandis were investigated for limonin concentration. Limonin concentration for Citrus limetta juice were found out to ~ 4.16 ppm, 9.68 ppm and 10.15 ppm for the samples at 0 h (as- prepared juice), at 6 h and at 10 h of storage time respectively. When tested with the other fruit sample (citrus grandis), limonin content of 17.98 ppm (at 0 h), 20.40 ppm (at 6 h) and 24.03 ppm (at 10h) were measured with the sensor. Utilizing a conventional process of quantification, such as HPLC analysis, the sensor's performance was validated, and for the quantification of Citrus limetta and Citrus grandis fruit juices, accuracy rates of 14.9% and 5.20% respectively, were attained. The inexpensive, flexible paper substrate-based capacitive sensor ensures quick, simple, and environmentally friendly on-site monitoring of limonin during the preservation of fruit juice.

Further, we have reported on the development of a novel IDE sensor system based on a composite of magnesium silicate and PVA for detection as well as measuring the reduction in limonin content through its selective adsorption using Magnesium silicate. The composite material made of magnesium silicate and PVA has been effectively prepared and characterized. The Si-O-Si stretching vibration of magnesium silicate is shown by the vibration band at 1108 cm^{-1} in the FTIR spectrum of the composite. The vibration bands at 1091 cm^{-1} indicating stretching of C-O and bending of OH (amorphous sequence of PVA), confirms the presence of PVA in magnesium silicate / PVA composite. In addition, the peaks that occurred at 468 cm^{-1} and 459 cm-1 reveal Si─O bending vibrations of the MgO. The band at 666 cm^{-1} is due to a Si–O bending motion for magnesium silicate and the same bending is observed at 676 cm^{-1} for $MgSiO_3.xH_2O$- PVA composite. Magnesium silicate's X-ray diffraction (XRD) pattern exhibits broad peaks that show the particles' non-crystalline structure. The prepared sample's average crystal size (D) was determined to be 2.12 nm. The sensor based on $MgSiO_3.xH_2O$- PVA composite was accomplished with a not remarkable shift in output in the presence of other elements like ascorbic acid, citric acid, sucrose, fructose, glucose, mamlic acid and naringin present in the juice that interfere with the detection process. With a quick response time of just under 6s, the device offers a sensitivity of nearly 2.3920.3 µF/ppm in identifying the rise in limonin in citrus juice. When the fabricated sensor is compared to HPLC analysis while testing

Citrus limetta juice, it exhibits an approximate variation of 0-12%. After 6 hours of juice processing, a 19% rise in limonin content was noted. After keeping juice for six hours, it was exposed for 60 s to a magnesium silicate surface that reduces the amount of limonin from 5.77 ppm to 4.29 ppm. The sensor can be gently washed with DI water, dried, and reused up to two times with a degradation of less than 10%. When magnesium silicate treated juices were analysed for sensory, it shows not remarkable changes in sweetness and tartness but gives noticeable changes in bitterness. The TFC, TPC and anti-oxidant properties of juice after its treatment with the material magnesium silicate shows negligible changes in those parameters signifying retention of its valuable properties TFC, TPC and anti-oxidant properties after treatment. Because the components magnesium silicate and PVA are not poisonous, fruit juice is not subject to acute toxicity.

Both the developed sensors were compared with the previous reported methods in terms of response time, LOD, selectivity, sensitivity, reusability, and flexibility, etc. The previously reported OECT shows a high LOD as compared to our developed sensors, but requires complex fabrication steps and hence in terms of complexity and fabrication cost the present devices are beneficial. The odorant-binding protein-modified screen-printed electrodes do not show specificity to limonin while the two sensors developed show specificity in the detection process. The surface molecularly imprinted polymers (SMIPs) exhibit only debittering of limonin, taking a time of ~5 mins. The electrochemical device introduced by Puri et al. takes a much longer time (~20 mins) for its response compared to the present sensors developed based on IDE. The response time is fairly less in the sensors developed in the present work. The OECT sensor/device can be reused multiple time by re-depositing its sensing layers that needs complex fabrication steps again whereas the IDE based sensor system in the present work need the replacement of inexpensive sensor for its repeated use. For the sensors, on-site monitoring is still a crucial concern. The previously mentioned devices/sensors require a complex measuring tool, like an impedance analyzer, for their output measurement, which is bulky and unsuitable for onsite monitoring. Our portable, Arduino-based measuring system that is integrated with the sensor overcomes this constraint. Here, a straightforward fabrication procedure helps reduce the need for highly qualified personnel. Moreover, the current sensors can be produced in large quantities at a low cost, which makes them favourable in contrast to other methods. As a result, our developed sensors demonstrate significant benefits over past efforts, making them appropriate for measuring limonin in citrus fruit juices. The magnesium silicate based capacitive device developed is used for both detection and measure of limonin reduction which seems to be first of its kind. Therefore we can conclude that the developed sensors are easy in use, less expensive, selective and having the facility of onsite monitoring with environment friendly nature.

7.2 Future Direction of Research

The freshness of fruit juices (especially juices with health benifits) is a vital issue not only in food industry but also the health. The research related to bitterness assessment (detection and debittering) opens the doors to many interesting future scope which are as follows:

- Detection of other biomolecules like nomilin etc using the array of sensor devices.
- Further miniaturization of the device dimension to increase in of production reducing the effective cost of the device.
- Increasing efficiency of the evaluation system.
- Exploring other methods of doping and composites materials in the sensor to improve stability and repeatability.
- Design and fabrication of device for the detection of of naringin and limonin using same sensor set-up.
- Debittering of fast and delayed bittering caused by naringin and limonin respectively using same device or same experimental setup.

Here, we have performed quality assessment of bitterness in Citrus limetta and Citrus grandis fruit juices. The results of the experiments are encouraging and motivate us for implementation of the same on other fruit juices.