

Carbon quantum dots prepared by using microwave irradiation-based total green synthesis from vegetables exhibit *in vitro* antibacterial activity.

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Repeated usage of synthetic antibiotics leads to the evolution of antibiotic-resistant strains of pathogenic bacteria that are not only harmful to humans but also for animals (poultry, livestock, fish and seafood). Alternative approaches are urgently needed to efficiently control the environmental contamination by synthetic antibiotics and to control the evolution of antibiotic-resistance. In this study we prepared carbon quantum dots (CQDs) from raw vegetables as an alternative source for synthetic antibiotics and evaluated if they have any antibacterial activity. The CQDs were prepared using a single step-microwave irradiation-based total green synthesis methodology from vegetables. The CQDs were tested for their fluorescence under ultraviolet rays and were confirmed to be fluorescent compared to the vegetable pulp before microwave irradiation. Our results show that the CQDs were successful in inhibiting the replication of the common laboratory strain of Gram negative *E. coli* (DH5-alpha). In conclusion, we believe that our one-step green synthesis methodology can provide natural and ecofriendly alternatives to synthetic antibiotics towards the one health concept.

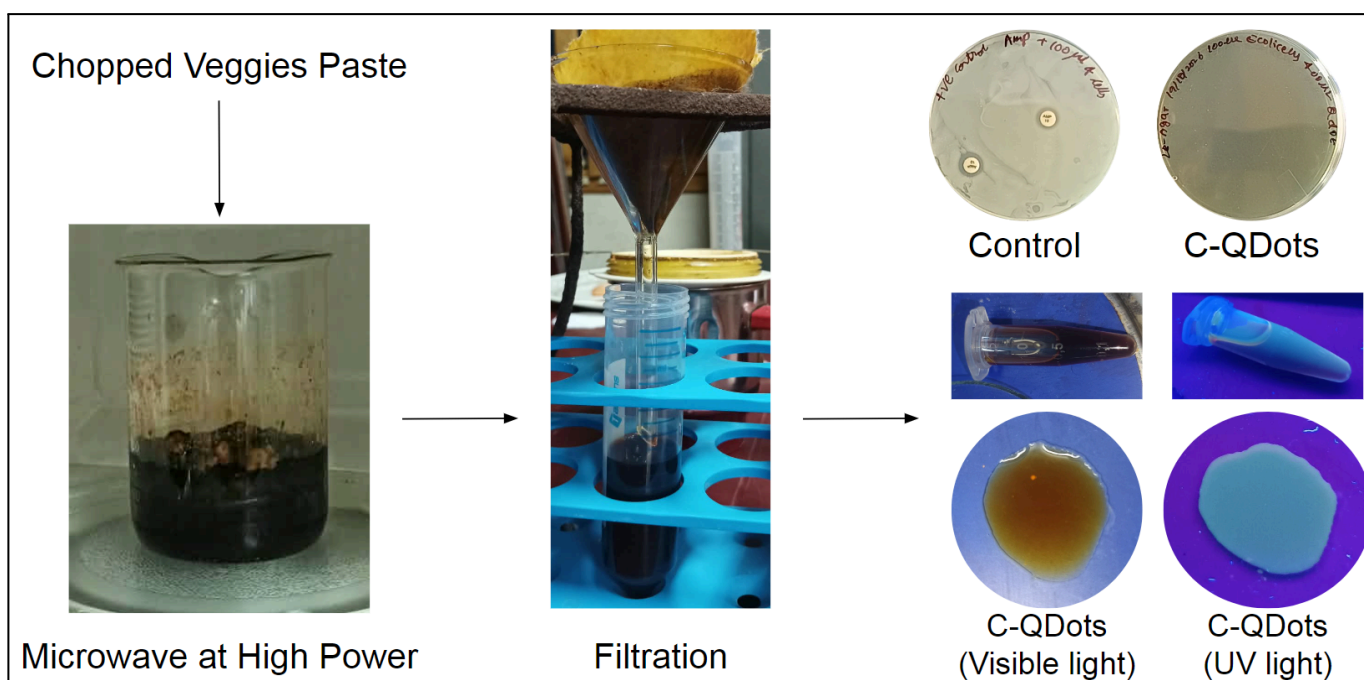


Figure 1. Overall process of carbon quantum dots preparation in the current study and their antibacterial activity.

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Antimicrobial-resistance (AMR) is one of the rising global medical challenges these days that is claiming millions of lives [1, 2]. According to the latest one health concept, overuse of synthetic antibiotics is not only fueling the evolution of antibiotic-resistant bacterial strains in humans and animals (poultry, livestock, fish and sea food) but also polluting the water bodies in the environment [3]. In order to control the above mentioned problems alternative approaches to the synthetic antibiotics are urgently needed. Accordingly, natural and eco-friendly sources for antibiotics preferably that can incorporate waste recycling methods would be highly sustainable approaches supporting the one health concept in the future.

Quantum dots are clusters of atoms prepared usually from metals and metal oxides in general. However, carbon quantum dots (CQDs) have gained popularity in recent years due to their low toxicity [4]. CQDs have been used for various medical purposes such as anticancer agents, bio-imaging, etc. In this study, we reasoned whether the CQDs possess any antibacterial activity with a goal to identify natural and eco-friendly alternatives to synthetic antibiotics (Figure 1). We used the pulp/paste made from fresh and semispoiled vegetables as the starting material which was subjected to high power microwave irradiation until all the liquid evaporated leaving behind a solid/semi-solid black residue in the glass beaker (Figure 1). The black residue was collected into a tube and deionized water was added to it until a thick liquid was obtained at room temperature. This liquid was filtered through filter paper using gravity without any pressure and the filtrate was directly used for testing fluorescence. As shown in Figure 2 the CQDs preparation (concentrated) and a 10-fold diluted version of the same was spotted on a glass slide along with the spots of the vegetable extract/juice before microwave irradiation. The spots were checked under visible and UV lights to confirm the fluorescence of the CQDs. Fluorescence was observed in both concentrated and diluted spots of the CQDs while a similar fluorescence was not visible for the vegetable extract.

The CQDs prepared as above were directly used for antibacterial assay without any further dilutions. The goal of this assay was to check whether the crude preparation of CQDs possessed any antibacterial activity. As shown in Figure 3, the CQDs exhibited potent antibacterial activity with 100% inhibition of bacterial growth when tested on the routine laboratory strain of *E. coli*, DH5-alpha. Evidently, the LB agar plates containing 500 µl and 400 µl of CQDs exhibited potent inhibition (100%) of bacterial growth while the LB agar plates containing 300 µl and 200 µl of CQDs exhibited partial inhibition (<50%) of the bacterial growth. The LB agar plates containing 100 µl, 50 µl and 10 µl of CQDs did not show any bacterial inhibition and bacterial lawns were seen.

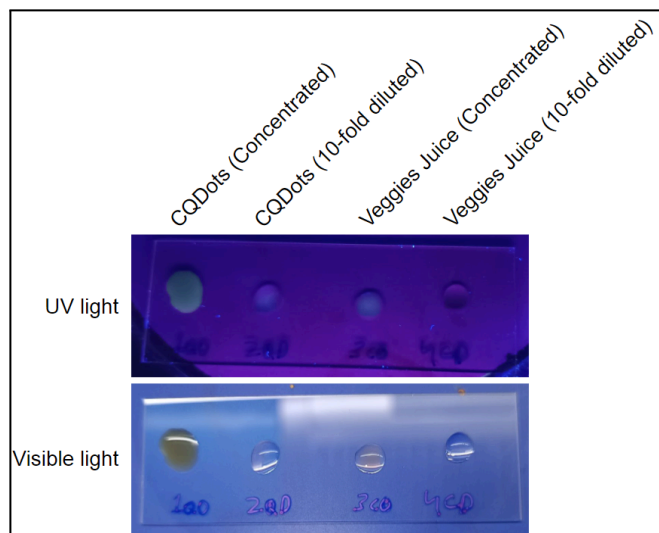


Figure 2. Fluorescence analysis of CQDs. Among the four spots on the glass slide, the CQDs containing spots exhibited fluorescence compared to the non-irradiated veggie extract/juice.

In conclusion the results from the present study suggest that the crude CQDs that were prepared from vegetables using the one step microwave-based green synthesis exhibit antibacterial activity. These CQDs are potentially a natural and eco-friendly alternative to synthetic antibiotics towards the one health concept.

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Conflict of interest: The authors declare no conflict of interest in this study.

Author contributions: A.M. and Ch.V.M.G. performed all the wet lab experiments. R.S.Y., principal investigator, provided laboratory space/facilities needed. S.B.B.V. supervised A.M. R.S.Y. wrote, edited, finalized the manuscript approved by all authors.

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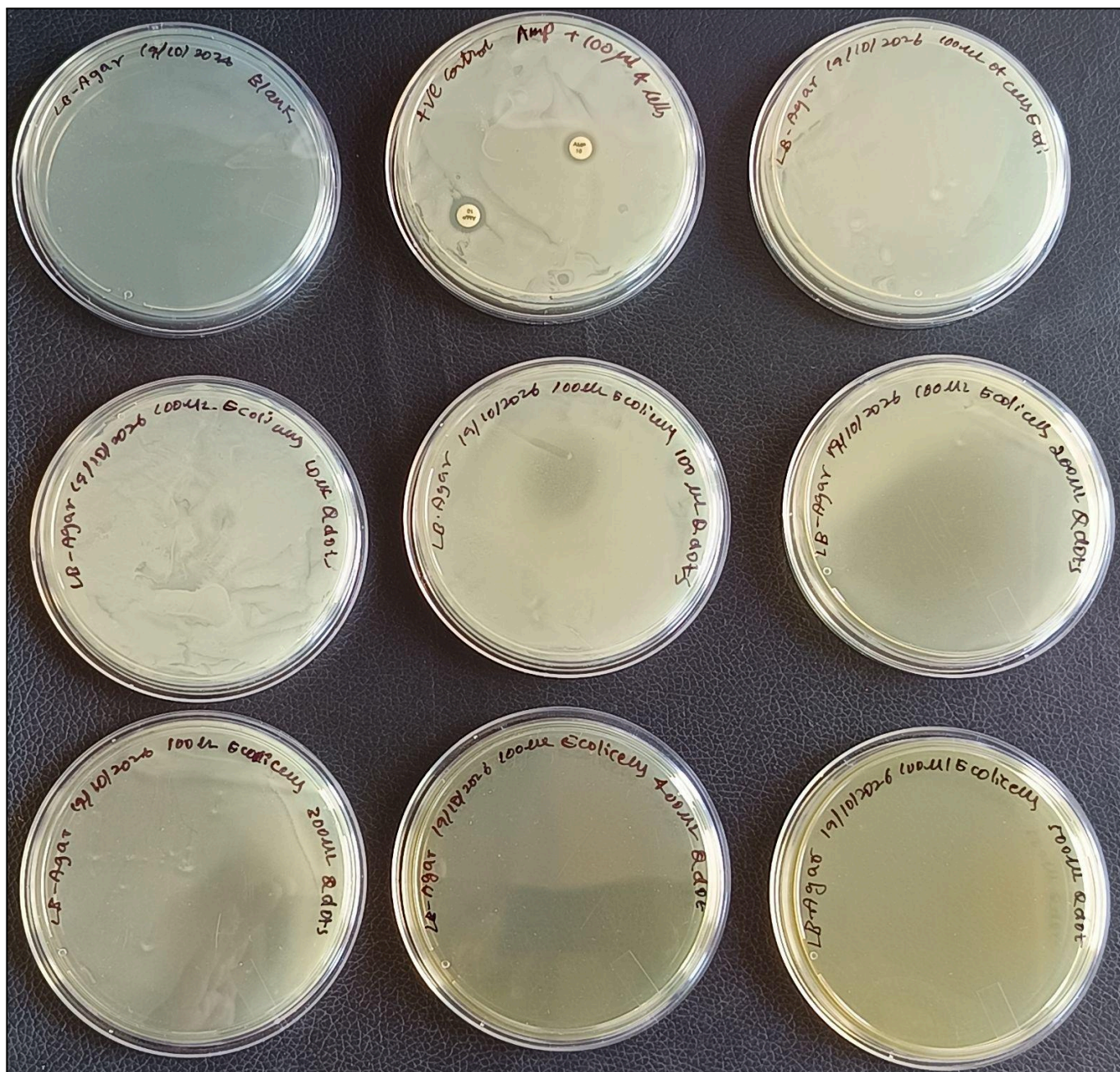


Figure 3. Results from the CQDots antibacterial activity assay. The crude preparation of CQDots was used without any further dilutions. Both LB agar plates containing 400 µl and 500 µl of CQDots exhibited 100% inhibition while the plates containing 200 µl and 300 µl exhibited partial (<50%) inhibition of the Gram negative *E. coli* (DH5-alpha) bacteria. The plates containing 100 µl, 50 µl and 10 µl of CQDots exhibited bacterial lawns comparable to the negative control. Ampicillin discs (10 mcg) were used as positive control and a blank LB agar plate is shown as a control for lack of any general contaminations in the assay.