Optical coherence tomography detection and quantitative evaluation method to examine the effect of polyethylene microplastics (PEMPs) on lentil seed germination

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1. Introduction

Microplastics (MPs) are accumulated in the environment at an alarming rate giving a considerable impact on seed germination because of overwhelming usage and mismanagement of plastic. Depending on the formation, MPs can be categorized into two groups, namely primary and secondary MPs. The adverse effect of MPs on seed germination and plant growth have been reported [1]. According to the existing literature, PEMPs could reduce the seed germination rate and seedlings growth. The effect was measured using conventional measurements such as germination test, root length, and leaf length [2]. In general, the effect of external stressors on seed germination and seedling growth are assessed by destructive post-harvesting conventional measurements such as germination test, cold test, tetrazolium test, etc., that require a relatively long period of time. Hence, there is an urgent requirement for novel methods that can monitor the effect of MPs on seed germination and seedling growth at an early stage. Here, we proposed the Biospeckle Optical Coherence Tomography (bOCT), to observe the effect of polyethylene microplastics (PEMPs) on lentil (*Lens culinaris*) seed germination and seedling growth. bOCT is an ultra-high accurate non-contact, non-destructive, in vivo monitoring technique to visualize the change of internal activity of a biological object. Earlier we demonstrated the potential of using bOCT to investigate the effect of polystyrene microplastics, and alumina NPs on seed germination [3].

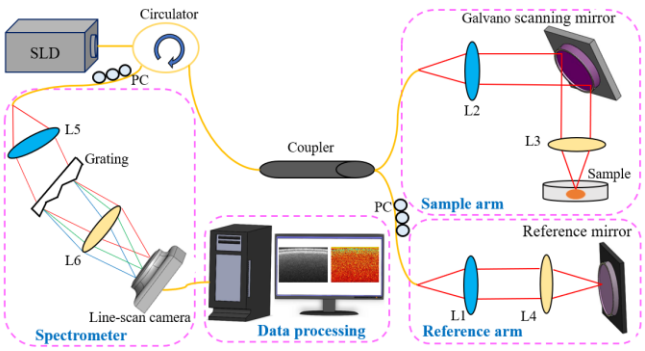

Fig.1. Schematic diagram of the OCT experimental system.

2. Experiments and Discussion

The OCT experimental system is illustrated in Fig.1. In bOCT, from the acquired OCT structural images, the biospeckle contrast, γ, defined as the ratio of standard deviation of the intensity at each pixel to the mean value of pixels across total period of the scan was calculated according to the following equation:

$$\gamma(x,y) = \frac{1}{<I_{OCT}(x,y)>}\left[\frac{1}{N}\sum_{j=1}^{N}\{I_{OCT}(x,y,t_j) - < I_{OCT}(x,y) >\}^2\right]^{\frac{1}{2}},$$

$$< I_{OCT}(x,y) >= \frac{1}{N}\sum_{j=1}^{N} I_{OCT}(x,y,t_j),$$

where x, y represents the pixel coordinates, j is the scan number, and N indicates the total number of scans. In the experiments, 0, 10, 50, and100 mg/L PEMPs concentration were used. For each sample, 6 seeds were placed on a filter paper in 9 cm Petri dishes. Thereafter, the Petri dishes were kept in a growth chamber at 27° C temperature, a relative humidity >70% and a constant illumination of 4000 lux. The bOCT observations were taken at 6h, 12h, and 24h. The average local contrast of the bOCT images was calculated using six ROIs as shown in Fig.3. A clear reduction of internal activity for all PEMPs treatments was observed compared to control emphasizing the adverse effect of PEMPs on seed germination, and the effect was significant just after 6h of exposure, due to inhibition of nutrient and water uptake through the physical blockage of pores of the seed coat [4].

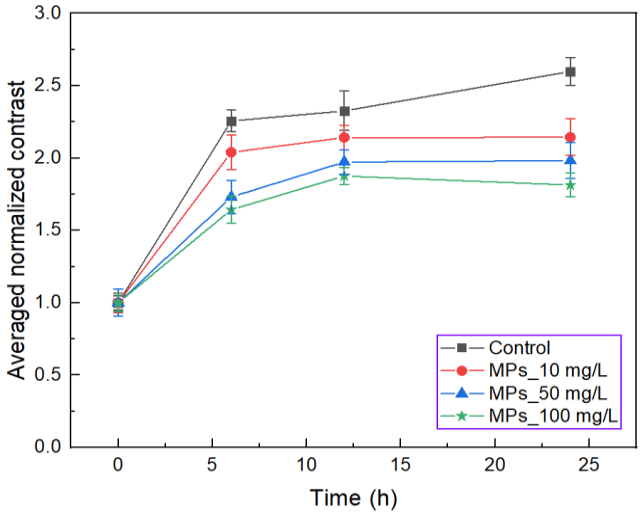

Fig.3. Averaged normalized contrast of bOCT image under PEMPs exposure.

3. Conclusion

This research proposes bOCT to monitor the adverse effect of PEMPs on lentil seed germination. Furthermore, conventional measurements such as seed germination rate, root length, shoot length, fresh and dry weight of seedlings were also measured for comparison. The bOCT results show that the tested PEMPs concentrations has negative influence on seed germination with a clear reduction of the internal activity of seed observed just after 6h of exposure. Hence, the proposed method is able to observe the effect of PEMPs at an early stage before the germination. Currently we are working with synergic effect of microplastics and Zn on seed germination.

References

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