Efficacy of cold plasma for strawberry cultivation on fruit ripening process

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Abstract: We periodically treated the strawberry seedlings during their cultivation with cold plasma by two methods such as direct plasma irradiation and indirect plasma-activated sodium lactate. Both treatments activated the fruits ripening on their process due to the increase of anthocyanin content in the fruits. The results suggested that the cold plasma treatment had the effectiveness for the strawberry cultivation because the high-quality fruits can be stably produced, which leads to the innovation as the new agricultural technology.
Keywords: Agricultural application, Cold plasma, Strawberry cultivation, Anthocyanin accumulation

1. Introduction

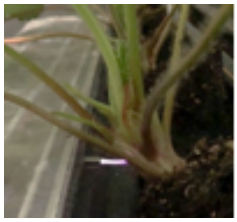
Recently, the applications of non-equilibrium atmospheric-pressure plasma (cold plasma) to agriculture has much attentions. Many reports have been shown that the plasma treatments improve the germination rate and the plant growth. However, most of them were limited to the seed or initial growth, and there have been few results to verify the actual yield so far.

Previously, we developed the ultrahigh electron-density plasma source,[1,2] and reported various biological applications, including medicine and agriculture, by using the plasma source. Cancer cells were selectively killed by the direct irradiation and the indirect treatment with plasma-activated Ringer's lactate solution (PAL).[3,4] In addition to the killing or inactivation, we quantitative demonstrated the growth promotion effects on the budding yeast cells with the oxygen radical treatment.[5] These results indicated that the plasma treatment can induce various living organisms to both of the activation and inactivation by regulating the treatment condition.

On the basis of the results, in order to verify the efficacies of plasma treatment on the cultivars in the actual field, we have treated rice in a paddy and strawberry in a greenhouse. Successfully, not only the yield was increased, but also the quality was improved.[6,7] It was effective that rice plants were treated in the vegetative growth stage. The yield was increased by 15%, and the grain ripening was activated due to the decrease of the ratio of immature grains to the whole grains. Furthermore, the strawberry seedlings in a greenhouse were periodically treated during the cultivation with the plasma, and the anthocyanin contents in harvested fruits were increased. Anthocyanin is one of important factors to determine the quality of strawberry fruits due to the high antioxidant activity.[8] The results demonstrated that the cold plasma can stably produce the high-quality crops and fruits by using in the actual cultivation field. In this study, based on the stage of fruit ripening, further investigations were carried out in order to analyze the effects on high accumulation of anthocyanin in the strawberry fruits.

2. Experimental

Strawberry seedlings (*Fragaria* × *ananassa,* cv. Beni-hoppe) were planted on the planter beds in a greenhouse in Nishio city, Aichi, Japan on October 13th 2020. The seedlings were cultivated according to the conventional methods. For plasma treatment on site, we used two cold plasma devices; the automatically direct plasma irradiation system and the automatic equipment for producing the plasma solution as shown in Figs. 1a and b, respectively. In the former, the plasma unit with the pen-type He plasma jet (60 Hz-9 kV) was mounted on transport belt in 3 m, and enclosed in a lab case with stainless-steel frames and acrylic covers. The plasma unit was stopped at the planted point of seedling by setting the photosensor, and the seedlings in a row were automatically treated with the plasma under program. The latter had the ultrahigh electron-density atmospheric pressure plasma source (60 Hz-9kV), pumps and tubes. Sodium lactate solution of 27.6 mM was continuously treated with the plasma for 5 min by program control, the plasma-activated sodium lactate solution (P-SL) was automatically prepared. Then, P-SL solution was 16-fold diluted by distilled water and watered onto soil. For a control, equal volume of distilled water (DW) and non-plasma-treated sodium lactate solution (SL) were used. These treatments were carried out three time a week from November 10th to March 26th together with the cultivation.




(a) Direct irradiation (b) Preparation of PAL

Fig. 1 Plasma treatment during strawberry cultivation

For the investigation of the effects of plasma treatment on fruit ripening, in order to uniform in fruit size, the young green fruits with vertical fruit length of 21±2 mm (approximately 7 days after flowering) were tagged in each treatment area on Mar 6th. The tagged fruits were harvested on 7, 16 and 21 days after, by the number from 4 to 18 fruits for each of treatment condition and ripening stage, and stored at -80°C. Then, anthocyanin content in each fruit was measured.

3. Results and discussion

Figure 2 shows the anthocyanin content in the fruits harvested at 7, 16 and 21 days after tagged in each treatment area, such as control (Ctrl), direct irradiation (Direct), DW, SL and P-SL. At 7 days after tagged, all fruits in all treatment areas were still green (data not shown). And, those anthocyanin contents in them were less than 2.4 mg/100 g fresh weight (Fw), although those in direct plasma irradiation area were significantly higher than those in control area.

Then, at 16 days after tagged, the fruits in control area with a variety of color, such as white, partial red, and full red, were mixed, which indicated that those in control area were still on the progress of ripening. Therefore, the average anthocyanin content in the control fruits was varied largely. On the other hand, all the fruits from direct irradiation area were red in their appearance, and their anthocyanin content was less variable and 2.4-fold higher than those in control. Similarly, since the fruits in the DW and SL area were still almost white, the anthocyanin contents in both areas were low less than 3.0 mg/100 gFw. On the other hand, all the fruits from P-SL area were already red and the content was significantly higher than those from DW and SL areas. These results indicated that the both plasma treatments by direct irradiation and P-SL solution promoted the fruits ripening.

Finally, at 21 days after tagged, all the fruits in control area were fully ripened. The average anthocyanin contents from them were 21.6 mg/100 gFw. On the other hands, those from direct plasma irradiation area were 22% higher than those in control area. Similarly, the anthocyanin contents in the fruits from P-SL area were 14% and 26% higher than those from DW and SL areas, respectively. The results were consistent with our previous results.

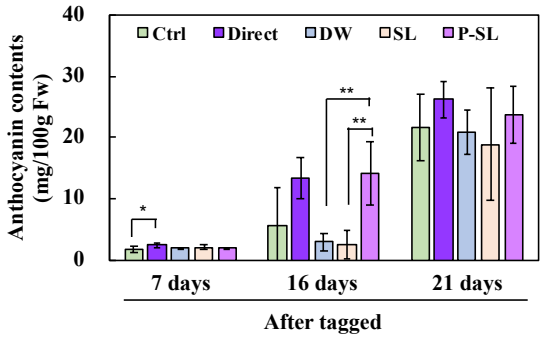


Fig. 2 Anthocyanin content in tagged strawberry fruits. Significant differences identified with Student's t-test at $p<0.05$ and 0.01 are indicated by *and**.

These results suggested that plasma treatment activated the anthocyanin biosynthesis from the early or medium stage of the fruit ripening process, and that the activation was maintained even to the late stage, resulting in the high accumulation of anthocyanin in the fruits. From the viewpoint of the application in agriculture, various efficacies are conceivable, such as shortening the period from flowering to harvest, and suppression of the individual variations, as well as the improvement of the quality of fruits. We will report and discuss the detailed results containing the physiological analysis at the meeting.

4. Conclusions

We periodically treated strawberry seedlings during the cultivation with cold plasma, such as direct plasma irradiation and indirect P-SL treatment. and verified the effects on the fruit ripening on the basis of anthocyanin contents together with the progress of the ripening stage. Accumulation of anthocyanin in the fruits earlier was observed in plasma treatment areas, and the contents in the mature fruits were higher than control. Since anthocyanin has an antioxidant activity, the results will be expected for the innovation to produce the high-quality fruits by cold plasma technology in terms of our health.

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