

Research on Emission Mechanisms of Biophotons from Cucumber

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Abstract: If plants such as cucumber are injured, the wound emits many biophotons. We have applied this phenomenon to measure effects of the non-contact healing known as laying-on-of-hands. Our previous studies suggested that natural biophotons from injured cucumber samples are generated by plural biochemical reactions. In the present study, we discuss possible mechanisms: 1) oxidation reaction of ascorbic acid; 2) biosynthesis of C9/C6 aldehyde and alcohol (so-called green order); and 3) other defense reactions. We supplied alpha-linolenic acid, linoleic acid, ascorbate oxidase (ASOD) and lipoxygenase (LOX) onto cut pieces of cucumber. We found these compounds increased the intensity of biophotons emitted from cucumber.

Keywords: biophotons, cucumber, mechanism, healing, green order, ascorbic acid

1. Introduction

It is well known that the intensity of biophotons emitted from a wound on a plant surface is much greater than the intensity emitted from a non-wounded surface. Using cucumber samples, we have applied this phenomenon to develop a standard measuring method for non-contact healing such as laying-on-of-hands¹⁻⁶.

Fig. 1 shows the time transition of intensities of biophotons from cucumber samples ($n = 102$) in our previous work on non-contact healing⁵. In the first 4 hours (Phase I), there was no difference in intensities between experimental and control samples. After 5 hours, however, the intensity of biophotons from experimental samples was more than that from the control samples (Phases II & III). This suggested that biochemical systems for emitting photons in Phases II & III were different from those in Phase I. Moreover, there seemed to be a possibility that non-contact healing could affect the biochemical systems of Phases II & III, but not Phase I.

In the present study, we discuss possible biochemical reactions which emit photons from injured pieces of cucumber⁷.

2. Possible Reactions

We used a biophoton measuring system C2400-47 (Hamamatsu Photonics) which has an image intensifier

(I.I.). The system can measure wave lengths from 280-650 nm (sensitivity, 60 mA/W at 400 nm). Although we can measure visual light, we cannot measure near infrared light. Moreover, we can detect only strong emissions because the samples are set 250 mm from the camera lens. Considering these points, we assumed the following chemical reactions as those which can generate biophotons in our experiments.

2-1 Oxidization of Vitamin C

Cucumber contains ascorbic acid (Vitamin C) at an average concentration of 14 mg/100g. Also cucumber contains ascorbate oxidase (ASOD). If a cucumber is cut, oxygen in the air makes contact with cytoplasm of cucumber and then ASOD starts to oxidize ascorbic acid immediately. This reaction is considered as a source of biophotons.

2-2 Biosynthesis of Green Order

There are many studies on the details of biosynthesis of the so-called green order in plants. In this section, we have discussed possible mechanisms, based on Hatanaka's book⁸.

When cytoplasm of cucumber is exposed to oxygen by cutting, lipase is activated immediately. And then lipase generates alpha-linolenic acid and linoleic acid from phospholipids and natural lipids. Immediately, alpha-linolenic acid and linoleic acid are oxidized to hydroperoxides by lipoxygenase (LOX). LOX of cucumber is distributed in cytoplasm of cells. Next, the fatty acid hydroperoxidelyase (HPOL) decomposes those acid hydroperoxides into aldehydes and alcohols of C9 or C6 (Fig. 2). The major component of the aroma of cucumber is C9 aldehyde, 2E,6Z-nonadienal (violet leaf aldehyde, 52.6%).

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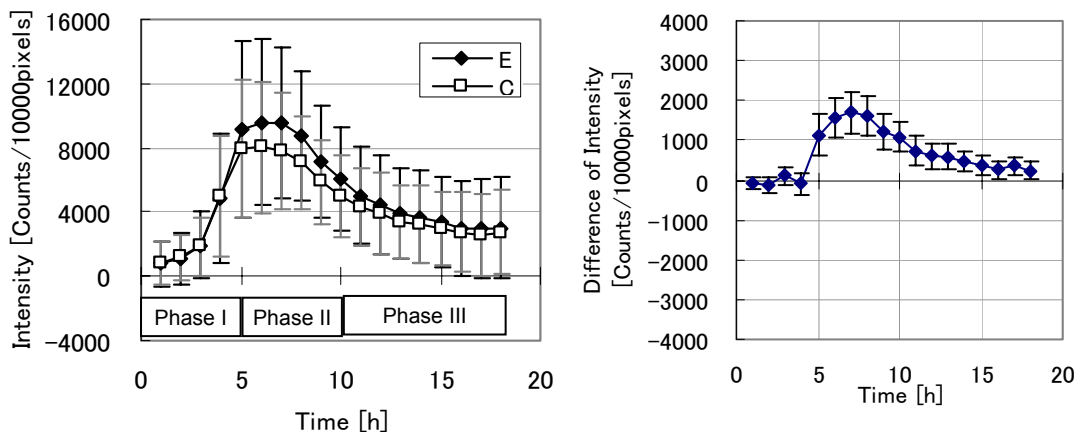


Fig. 1 Transition of Intensity of Photon Emission of Non-contact Healing (E) and Control (C) Samples (n = 102)
 Left: Intensity and standard deviation. Right: Difference between E & C and 95% confidential interval.
 (Kokubo & Yamamoto, Ref. 5)

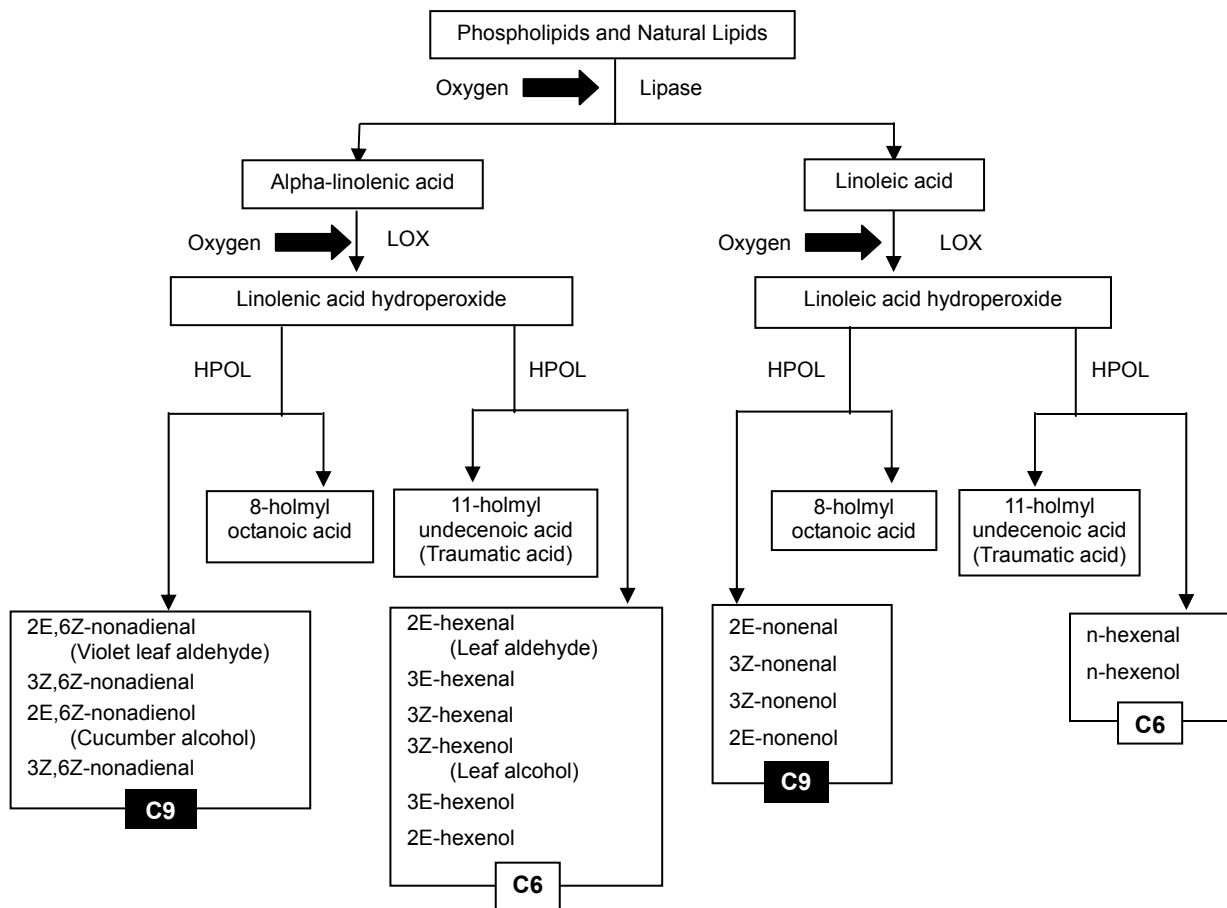


Fig. 2 Biosynthesis of C9 & C6 Aldehyde and Alcohol in Cucumber

Biosynthesis of green order is the first defense reaction of most plants, not only cucumber, when plants are damaged by insects or diseases. Violet leaf aldehyde and leaf aldehyde (2E-hexenal) are strong germicides, and also have a role in information transfers to other individuals.

2-3 Induced Biosynthesis

Some components which are generated in green order biosynthesis induce other defense reactions of plants. For example, in case of Dalmatian pyrethrum, leaf alcohol, leaf aldehyde, 3Z-hexenal and acetate of leaf alcohol induce biosynthesis of terpene, and pyrethrin is produced⁸⁾. We have supposed similar defense reactions are induced by green order in the case of cucumber, and some of the reactions emit photons. However, in the present paper, we omit discussion of this because we do not have enough knowledge about those reactions yet.

3. Heating Experiment

To examine whether biophotons are generated in the process of biosynthesis, we deactivated enzymes by heating cucumber samples. The sample preparation and arrangement of samples and procedures in the experiment were the same as our previous studies^{2,5)}. Whole cucumbers were heated in a microwave oven (DMW-95D2, Daewoo Electronics Corporation, South Korea) for 81 s, 60 s and 30 s, and then cut into 8 sample pieces.

Fig. 3 shows 18-hour transitions of intensities of photons from heated samples and non-heated (control) samples. For samples with 30 s and 60 s of heating, photon intensities became larger than those of control samples after 10 hours. The increase of photon emission occurred in the middle area of a sample cross section, not in the sarcocarp.

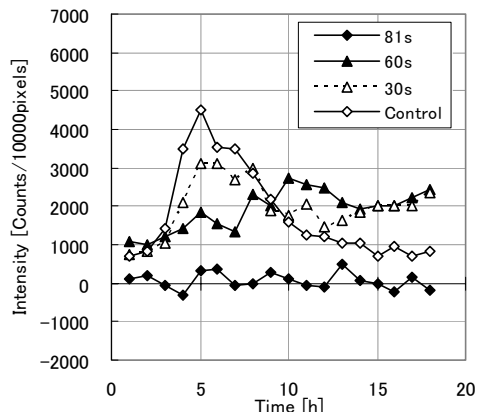
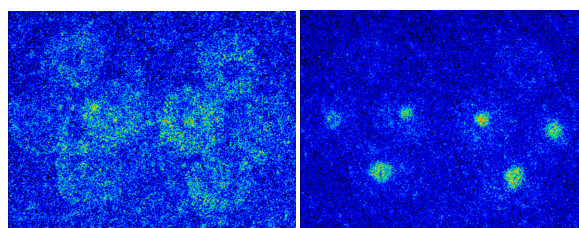


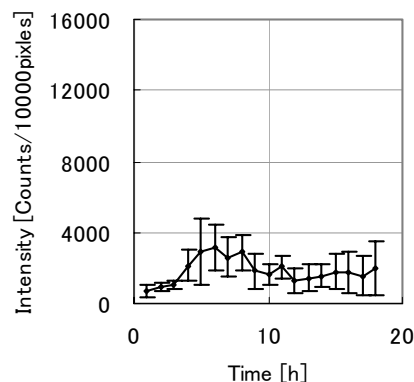
Fig. 3 Time Transition of Intensity in Heating Experiment

Photos reproduced in Figs. 4a & b show accumulations of photons of the first and the second 9 hours. Figs. 4c & 4d show time transitions of intensity at the sarcocarp and the middle area. Intensity of photon emission increased with time for the middle area. We considered that oxygen in the air permeates easily inside of samples reaching their middle area because the tissue along the axial center of cucumber was hurt more than other parts due to concentration of heating effect of the microwave oven.

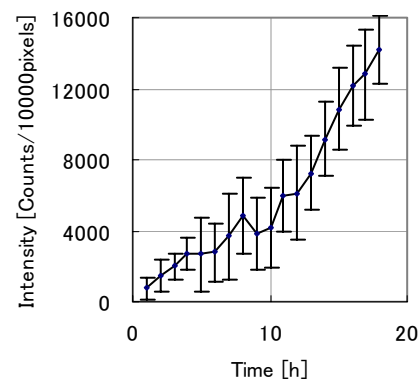
However, biophoton emission was not observed for the 81s heating.



a) First 9 hours b) Second 9 hours



c) Sarcocarp



d) Middle Area

Fig. 4 30s Heating Experiment

4 Additive Experiments

From the heating experiment, we saw that biophotons were generated by enzyme reactions. Next, we carried out additive experiments with alpha-linolenic acid, linoleic acid, ascorbate oxidase (ASOD) and lipoxygenase (LOX). The experiments were done from September to November 2007. The preparation and arrangement of samples were the same as our previous studies.

4-1 Alpha-linolenic acid and linoleic acid

Undiluted alpha-linolenic acid (chemical grade, Nacalai Tesque, Japan) was supplied dropwise onto each experimental sample by using a disposable polyethylene syringe (each amount: 0.05 ml). After that, alpha-linolenic acid was uniformly spread on the sample surface by using the bottom surface of a tea spoon. During the acid addition, 0.5 ml acid was placed in each of two small Petri dishes to check photon emission of the non-enzyme reaction which is a direct reaction of the acid with oxygen in air. Nothing was given to 4 control samples. Small dishes were set near the dishes containing the cucumber samples and biophoton emissions were measured simultaneously.

The same procedure was used to supply linoleic acid (chemical grade, Wako Pure Chemical Industries, Japan) to samples.

4-2 Ascorbate oxidase (ASOD)

Ascorbate oxidase (ASOD) (originated from cucumber, ASOD-03, 800 units/3.7mg, Oriental Yeast, Japan) was diluted with 1 ml distilled water, shaken well and then allowed to stand for several minutes. Next, 0.5 ml of the solution were supplied dropwise to 4 experimental samples by using a disposable polyethylene syringe and the solution was uniformly spread on the sample surface by using the bottom surface of a tea spoon (100 units/sample). For 4 control samples, 0.5 ml distilled water were dropped using a disposable syringe and spread on the sample surface by using the bottom surface of a tea spoon.

Additionally, another experiment was done at 50 units/sample of ASOD.

4-3 Lipoxygenase (LOX)

We could not get lipoxygenase (LOX) produced from cucumber. Therefore, we used LOX produced from soybean (15-Lipoxygenase, soybean P1, 26.9 Munits/ml, Cayman Chemical, USA) which has $\omega 6$ position specificity which is the same as cucumber. First, frozen LOX was thawed at room temperature and diluted with distilled water, and then the experiment was done using 6500 units/sample. However, the first experiment failed unfortunately because the intensity of photon emission of cucumber was often very small in winter. Therefore, after 50 hours, we thawed LOX which had been kept at -20 °C,

and repeated the experiment again. Preservation time of activity of LOX is 12 hours at -8 °C although LOX is stable at -40 °C. Because activity of LOX was considered as very low, we supplied 2 drops (0.1 ml) of undiluted LOX to each experimental sample and spread it on the sample surface by using the bottom surface of a tea spoon (2.4 Munits/sample). Biophotons were measured for 12 hours in this experiment on account of the laboratory schedule.

4-4 Results

Fig. 5 shows accumulation of photons for 18 hours. Both alpha-linolenic acid (α Lnn) and linoleic acid (Lin) increased the numbers of biophotons remarkably. This showed biosynthesis of green order produces photons. Intensities of photon emissions from both undiluted alpha-linolenic acid and linoleic acid were less than background noise, therefore we could not recognize the positions of undiluted acids. This meant that increased intensity of biophotons is caused by enzyme reactions when alpha-linolenic and linoleic acids are supplied to cucumber.

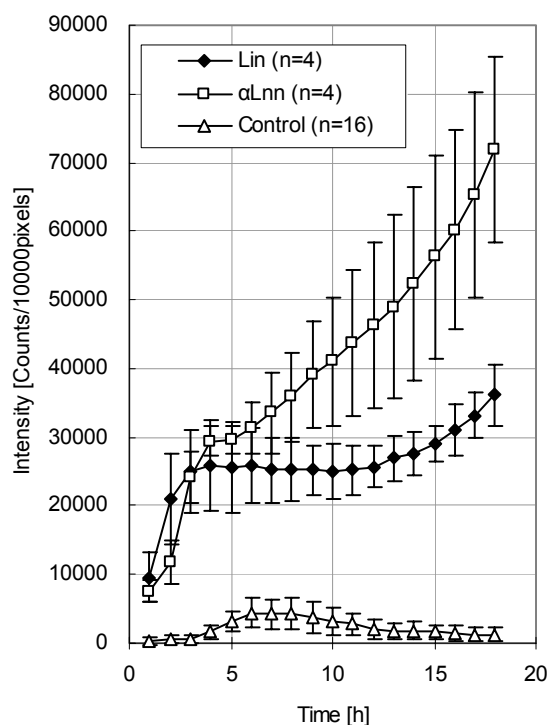


Fig. 5 Alpha-Linolenic Acid and Linoleic Acid

Intensities of photons increased linearly after 6 hours for alpha-linolenic acid and after 12 hours for linoleic acid. The reason was that intensity increased linearly for the placenta. It is considered that acids gradually permeated deeply into samples through sieve tubes or parenchyma of the placenta. Especially, after the linoleic acid experiment, all the samples had

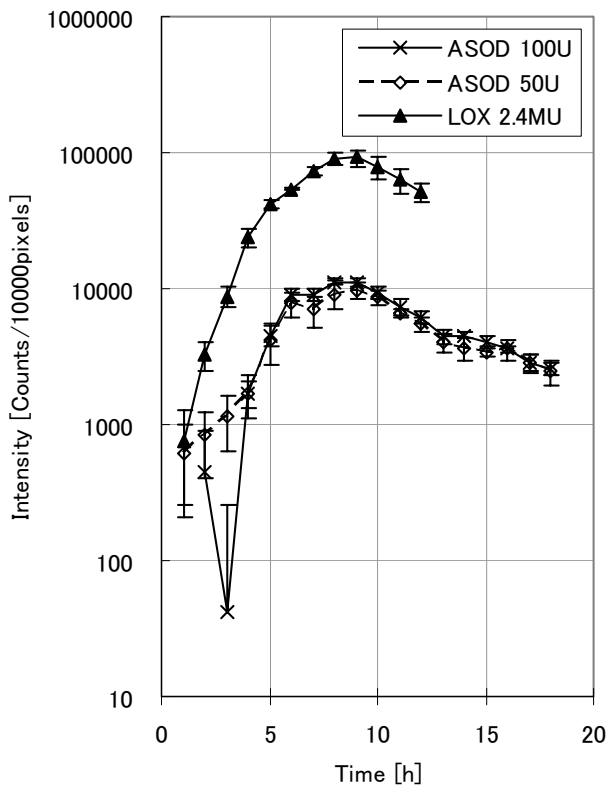


Fig. 6 ASOD and LOX

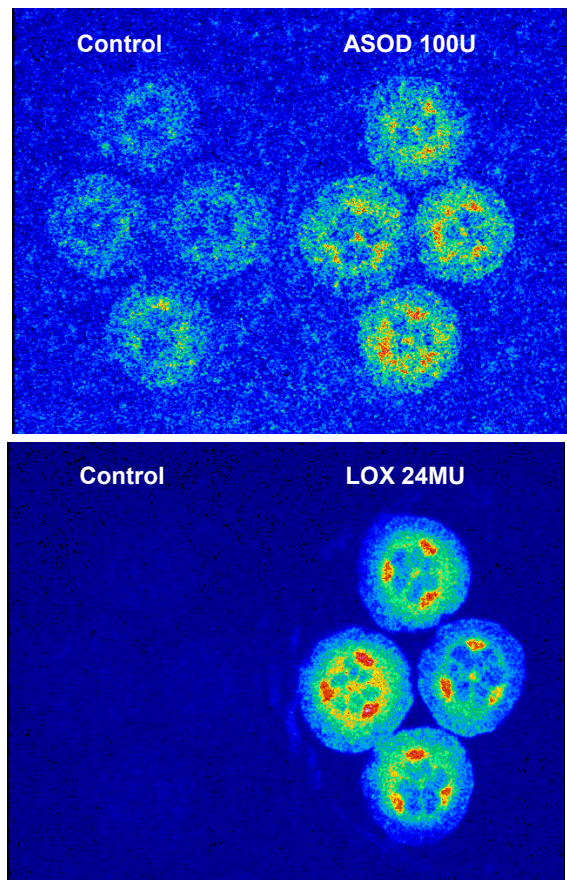


Fig. 7 Distribution of ASOD and LOX

apparently become pickles in the cooking oil and contracted a little, moreover there were small dents on the surface of each cross section.

Fig. 6 shows results for ASOD and LOX. Both gradients of intensity changed at 5-6 hours and reached maximum at 9 hours although amount of enzymes were very different and intensities were very different.

Fig. 7 shows accumulations of photons after 18 hours for ASOD and LOX, but intensity is shown as relative intensity. Each was similar to the intensity distribution of control samples. This suggested that oxidization of ascorbic acid and biosynthesis of green order are two origins of biophotons when cucumber is cut and exposed to the air without other treatments.

5 Discussion and Conclusions

We examined whether oxidization of ascorbic acid and biosynthesis of green order were sources of biophotons emitted from pieces of cucumber. However,

it is unknown yet whether different defense reactions emit photons or not because we could not test them in the present study. And, we did not succeed in identifying specific reactions corresponding to Phase I and Phase II. Further studies are needed.

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