Anatomical and Histochemical Studies of the Abscission Process in the Juncture Tissue between the Perianth and Receptacle Tissue on *Lilium* ‘Pollyanna’

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Keywords: anatomy, ethylene, flower longevity, histochemistry

Abstract
Abscission layer formation and development and abscission process of cut lily perianth were investigated anatomically and histochemically by applications of ethephon. At three days after application with ethephon, cell walls of the juncture tissue between the perianth and receptacle were disintegrated. Histochemical study showed that the pectic materials were dissoluted from the middle-lamella of the abscission region. The results indicate that the cell degrading enzymes such as pectinase may play a role in perianth abscission.

INTRODUCTION
It is now apparent that many, but not all, flowers, leaves, and fruits produce considerable amounts of ethylene in correlation with maturation and abscission. Also, aborting, injured, infected, or detached organs, such as leaves, petals, and fruits, produce ethylene (Burg, 1962). Abscission is one of the conspicuous responses to ethylene. In particular, leaf, petal and fruit abscission is commonly accelerated by ethylene. In *Lilium* plants, the flower bud absceses if light intensities are reduced in the greenhouse during winter season. In cut flowers of *Lilium*, when the flower matures, the perianth absceses at the juncture tissue between the perianth and receptacle tissue. Although ethephon (2-chloroethylphosphonic acid) accelerates abscission, little information about the abscission process has been reported. In this study, we observed the abscission process anatomically on perianth following an application of 100 ppm ethephon.

MATERIALS AND METHODS

Plant Materials
Cut lilies ‘Pollyanna’ were harvested from Mr. Moriyama’s commercial greenhouse in Niigata Prefecture, Japan when the largest flower bud on the stem was yellow color (Fig. 1). The stem was adjusted to 50 cm in length in the laboratory at Tamagawa University and sprayed with 100 ppm ethephon (Nissan Chemical Co., Ltd.). Each stem was maintained at 25°C under continuous cool white fluorescent lamps (20 µmol s⁻¹ m⁻²). Vase life was defined by morphological observation.

Anatomical and Histochemical Study
Flower samples at five mature stages were collected from the plants: 1) full flowering stage (Fig. 2), 2) one day after treatment, 3) two days after treatment 4), 7 days after treatment and 5) 10 days after the onset of the full ripe stage. Collected flower tissue were fixed in FAA solution (30% formaldehyde: 100% ethanol: 30% acetic acid = 8:1:1, v/v), dehydrated through a graded tertial butyl alcohol series, and embedded in paraffin. The embedded tissues were cut to longitudinal 15 µm thick sections with a slice microtome. The sections were stained with 0.1% toluidine blue-O as described in Tabuchi (1998) and Tabuchi et al. (1999, 2000, 2001). For pectic materials and cellulose detection, sections were stained with 0.01% ruthenium red and 1.0% hydrochloric acid (Arai et al., 1998; Tabuchi et al., 2001), respectively. The abscission zone of a juncture tissue between the perianth and receptacle tissue was divided into three histological regions: the central...
parenchymatous pith, vascular tissue (xylem and phloem) and the epidermis. The morphological and histochemical changes of the abscission zone in the juncture tissue between the perianth and receptacle tissue were observed more than 20 longitudinal sections at five development stages of flower.

RESULTS AND DISCUSSION

At three days after the onset of the anthesis stage of flower development (Fig. 3), cell walls of the juncture tissue between the perianth and receptacle were disintegrated (Fig. 4). These responses were found initially in the epidermis and cortex regions. Intercellular cavities were found in the epidermis and cortex region, and then spread to the central parenchymatous region at the juncture tissue between the perianth and receptacle. Perianth abscission then occurred (Fig. 3).

Staining with ruthenium red to detect pectin materials at the juncture tissue between the perianth and receptacle indicated that primary walls of these cells stained bright red color (positive) at the anthesis stage, whereas with the ethephon application, the disintegrated cell walls of the abscission zone absorbed less stain (negative) (Fig. 5). These results indicate that chemical changes in the cell walls in the juncture tissue between the perianth and receptacle, accompanies structural changes. For example, pectinase may play a significant role in perianth abscission. Thus, perianth separation occurred by dissolution and breakdown of the middle-lamella of the disintegrated cell walls. The same results have been obtained in seed-propagated geranium (Arai et al., 1998) and processing tomato (Tabuchi et al., 1998, 1999).

The use of zinc chloride to detect cellulose materials (Fig. 6) indicated that cellulose was located on the cell walls of the abscission zone during separation. In the proximal side of the abscission zone, walls of these cells thickened and stained darkly with toluidine blue-O, indicating they were heavily lignified. At this stage, the surface exposed cells on the proximal side of the peduncle stained darkly because their cell walls were thick and lignified.

In *Lilium* ‘Pollyanna’ perianth, lignified cell layers occur only in the proximal surface cells of the receptacle (Fig. 4), even after flower abscission occurs: subsequently, we concluded that lignified cells or periderm may act as a protective layer against the invasion of micro-organisms.

Literature Cited


Figures

Fig. 1. Large bud stage of flower development.  Fig. 2. At anthesis stage.

Fig. 3. At three days after application with ethephon. Abscission process proceeds.
Fig. 4. Longitudinal sections of the perianth abscission region in cut lily ‘Pollyanna’ stained with toluidine blue-O. Left: At anthesis. Right: At three days after applications with ethephon.

Fig. 5. Longitudinal sections of perianth abscission region staining with ruthenium red. Left: At anthesis. Right: At three days after application with ethephon.

Fig. 6. Longitudinal sections of perianth abscission region staining with zinc chloride. Left: At anthesis. Right: At three days after application with ethephon.