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Effects of Light on Disassembly of Chloroplast during Senescence of Detached Leaves in *Phaseolus vulgaris*

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Effects of light on leaf senescence of *Phaseolus vulgaris* were investigated by measuring the disassembly of chlorophyll-protein complexes in detached leaves which had been kept in the dark or under light. The loss of chlorophyll accompanied by degradation of chlorophyll-protein complexes. PSI (photosystem I) complex containing LHCI (light harvesting complex of PSI) apoproteins was rapidly decreased after the early stage of dark-induced senescence. RC(reaction center)-Core3 was slightly increased until 4 d and slowly decreased thereafter. As disassembly of LHCII trimer progressed after the late stage of senescence, there was a steady increase in the relative amount of SC(small complex)-2 containing LHCII monomer. On the other hand, white and red light adaptation caused the structural stability of chlorophyll-protein complexes during dark-induced senescence. Particularly, red light was more effective in the retardation of LHCII breakdown than white light, whereas white light was slightly effect in protecting the disassembly of PSI complex compared to red light. These results suggest, therefore, that light may be a regulatory factor for stability of chlorophyll-protein complexes in the senescent leaves.

Key words : senescence, chlorophyll-protein complexes, *Phaseolus vulgaris*, light

1. Introduction

Leaf senescence is an important sequence of biochemical and physiological events during which plant cells cause characteristic metabolic and structural changes, which finally induce cellular breakdown and death (Nooden, 1988). The initiation of leaf senescence can be caused by environmental factors such as shading, mineral deficiency, drought and pathogen infection (Thomas and Stoddart, 1980), as well as by developmental process such as an age-related manner in many species (Hensel, 1993; Jiang, 1993). One of the most conspicuous changes that occur during leaf senescence is yellowing, which is due to the preferential degradation of chlorophylls in comparison with carotenoids. And these syndromes of leaf senescence are accompanied by the disassembly of chloroplast (Gut, 1987; Thomson and Platt-Aloa, 1987). The loss of chlorophyll during leaf senescence can be accompanied by a decline in photosynthesis rate (Tobias, 1995). Also, there is a general increase in the degradation of proteins, nucleic acids and membranes, and the subsequent transport of the nutrients resulting from this degradation to other regions of the plant, although some new synthesis of those is necessary (Woolhouse, 1984; Smart, 1991). Recently it is proposed that the amount of mRNAs encoding proteins which play an important role for photosynthesis decrease during senescence, which those involved in the senescence program increase (Bate, 1991; Graham, 1992; Taylor, 1993; Lohman, 1994).

During leaf senescence it is possible that the rapid breakdown of chlorophyll, the most widely used biomarker for the senescence syndrome, is associated with the changes of chloroplast internal structure (Woolhouse, 1984; Thomson and Plat-Aloa, 1987). Of particular interest is the observation using bean that the synthesis of the majority of thylakoid-associated photosynthetic proteins, such as the 68 kD apoprotein of PSI, ATPase, light-harvesting chlorophyll binding protein, and cytochrome *f* and *b6,* decreases by two- and four-fold during senescence, whereas there is no change in the synthesis of the D1 protein of PSII (Robert, 1987; Droillard, 1992). Sopory *et al.* (1989) have proposed that the degradation of D1 protein is induced by activated oxygen, which causes it to undergo a changes in conformation. Also it is suggested that PSII proteins extrinsic to the thylakoid membranes, such as the 33 kD protein of oxygen-evolving complex, is rapidly degraded during leaf senescence of *Festuca pratensis* (Emyr-Davies, 1990).

However, the studies on the disassembly of chlorophyll-protein complexes during leaf senescence in detail are not fully understood comparing with greening in plant development, though a few researches have been dealt with the structural and physiological changes of chloroplast in senescing leaves (Siffel, 1991; Pancaldi, 1996). Fractionation of chloroplast proteins by native green gel electrophoresis, by which chlorophyll-protein complexes are solubilized and separated with minimum loss of noncovalently bound chlorophyll, is a very important method in the study of thylakoid membrane composition, organization and degradation (Thornber, 1986). Current native gel system with little release of free pigment resolves multiple PSI-LHCI complexes, multiple PSII-LHCII complexes, four oligomeric LHCII complexes, several reaction center complexes, and a number of small complexes from chlorophyll-protein complexes (Allen and Staehelin, 1991). On the other hand, there are many investigations using detached leaves *in vitro* on leaf senescence because of the relative ease of manipulating the experimental protocol compared to the complexity of using attached leaves. Also there is a remarkable variability in experimental results during senescence in attached leaves because of the uncertainty of the exact time onset of senescence (Thomas, 1990; Thomas, 1992). Therefore, experimental system in which detached leaves are used will offer an excellent system in which to investigate changes of chlorophyll-protein complexes during leaf senescence.

In the present study, the first objective was to identify changes in chlorophyll-protein complexes during leaf senescence of detached leaves in *Phaseolus vulgaris*. The second objective was to investigate effect of light on the changes of chlorophyll-protein complexes during leaf senescence. We have, therefore, analyzed bean leaves treated with dark condition in the physiological and biochemical levels, and compared them with light-illuminated leaves.

2. MATERIAL AND METHODS

2.1. Plant material

The bean seeds (*Phaseouls vulgaris* L.) were soaked in running tap water for 4 h and planted on the pot, and grown in growth chamber at 25 ℃/18℃ (light/dark) temperature cycle under 18 h of light / 6 h of dark regime condition with 70% humidity for 3 weeks. And the leaves of 21 day-old plants were excised by razor blades and the detached leaves, grown hydroponically in 3 mM MES (2-(N-morpholine) ethanesulfonic acid) buffer (pH 5.6) under dark condition or irradia-

tion of different light quality for 12 days, were used as the experimental materials.

Light source was a white fluorescent light enriched acryl filters for red light. Light intensities treated with bean leaves were 491 erg cm^{-2} sec^{-1} for white light and 47 erg cm^{-2} sec^{-1} for red light with radiometer (Metrologic, 60-535, USA) as described in Lee *et al.* (1995).

2.2. Thylakoid membrane isolation

For the isolation of thylakoid membranes, the detached leaves were homogenized with Waring blender in homogenization buffer consisted of 50 mM HEPES (pH 7.6), 0.3 M sorbitol, 10 mM NaCl and 5 mM $MgCl_2$. To remove cellular debris the homogenate was filtered through four layers of cheesecloth, and the filterate was centrifuged at 350 g for 10 min. The supernatant was pelleted at 5,000 g for 10 min. The membrane pellet was washed twice in washing buffer consisted of 50 mM HEPES (pH 7.6), 0.1 M sorbitol, 10 mM NaCl and 5 mM $MgCl_2$. The resulting pellet was resuspended in a small volume of the same buffer with 10% glycerol, aliquoted and frozen at -80℃. All procedures of thylakoid membrane isolation were performed at -4℃.

2.3. Thylakoid membranes solubilization

For native green gel electrophoresis thylakoid membranes was washed twice in 2 mM Tris-maleate (pH 7.0), and the washed pellet was resuspended in solubilization buffer containing 2 mM Tris-maleate (pH 7.0), 10% glycerol, 0.45% octyl glucoside, 0.45% decyl maltoside, and 0.1% LDS (lithium dodecyl sulfate). Solubilization buffer was added to give a ratio of total nonionic detergent to chlorophyll of 20 : 1 (w/w). Samples were incubated on ice for 30 min and centrifuged at 15,000 g for 10 min to remove insoluble materials.

2.4. Native green gel electrophoresis

Native green gel was consisted of a stacking gel containing 5% acrylamide, 25 mM Tris-HCl (pH 6.3), 50 mM glycine and 10% glycerol, and a resolving gel containing 8% acrylamide, 25 mM Tris-HCl (pH 8.3), 50 mM glycine and 10% glycerol. The acrylamide/bisacrylamide ratio was 100 : 1. The gel was polymerized by adding 0.05% TEMED (tetramethylethylenediamine). The electrode buffer contained 25 mM Tris (pH 8.3), 192 mM glycine and 0.1% SDS (sodium dodecyl sulfate) as described by Allen and Staehelin (1991). After the gel was prerun at 10 mA for 1 h in cold chambers, samples (about 13.5 μg chlorophyll) were loaded onto the gel. The amount of chlorophyll was determined with spectrophotometer (Shimadzu, UV 240, Japan) according to the method of Lichtenthaler (1987). The gel was electrophorized at 10 mM constant current, 4℃ for 3 h.

2.5. Denaturing SDS-PAGE

For two-dimensional gels, gel slices were excised from native green gel lanes, incubated for 15 min at 55℃ in solubilization solution containing 25 mM Tris-HCl (pH 6.3), 50 mM glycine, 2% SDS, 2% β-mercaptoethanol, and 10% glycerol. Gel slices treated with the solution were loaded directly onto the gel of 12% polyacrylamide. PAGE (polyacrylamide gel electrophoresis) in the presence of SDS was performed as described by Laemmli (1970).

2.6. Densitometry scanning

Gel lanes of a native green gel were scanned using a TLC scanner (Shimadzu, CS-930, Japan). The measuring wavelength of the densitometer was 675 nm. Peak areas for each green band in the lane was measured and represented as a percentage of the total chlorophyll in chlorophyll-protein complexes excluding free pigment. The values for each green band were the average of three independent experiments.

3. RESULTS

3.1. Changes in the contents of chlorophyll

A good representative of leaf senescence is yellowing, which is due to the loss of chlorophyll. The detached leaves of 21 day-old bean plants after the 4 d of artificially dark-induced senescence showed the first visible sign of senescence, leaf yellowing and the leaves became completely yellow after the 12 d of the senescence. Leaf yellowing started at the whole area of senescing leaves in this experiment, though yellowing of naturally senescing leaves started at the tip of the leaf (Lohman, 1994). Fig. 1 shows the changes of chlorophyll contents from the leaves which were kept in the dark during leaf senescence. The contents of total chlorophyll were gradually decreased along with senescence period, thus those of total chlorophyll were given rise to a decrease of about 66% during leaf senescence period investigated. To examine the effect of light quality for leaf senescence, chlorophyll contents of the leaves, which had been kept under illumination with white or red light for 12 d, were shown in Tables 1 and 2. When compared to the leaves which were kept in the dark during senescence, the illumination of white or red light was more effective in the suppressing of chlorophyll degradation during leaf senescence, and white light strongly retarded the breakdown of chlorophylls comparing to red light.

Chl a/b ratio is an important parameter to investigate the structural changes of thylakoids and light acclimation in the senescing leaves of bean plants. As the detached leaves of 21 day-old bean plants progressed through senescence, Chl a/b ratio ranged from 2.14 to 2.90 (Fig. 1, Tables 1 and 2). Chl a/b ratio was slightly increased during the 8 d of dark-induced senescence, but remarkably increased in the late stage of leaf senescence. That fact suggests that the rapid increase of Chl a/b ratios is due to the degradation of light-harvesting chlorophyll complexes containing both Chl a and Chl b. On the

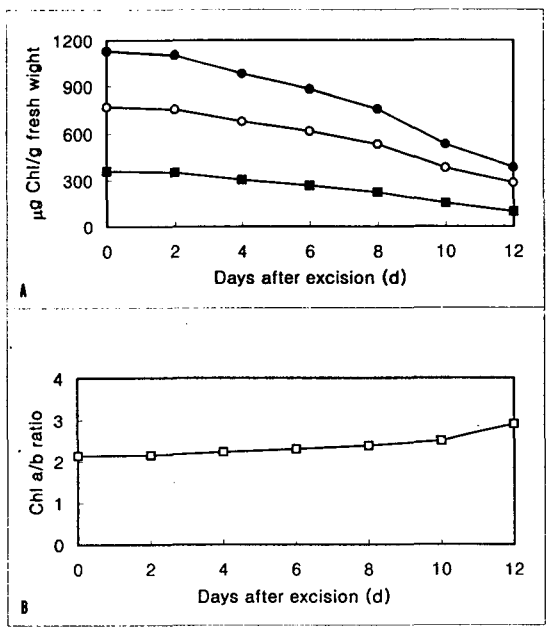

Fig. 1. Changes in chloropyll contents (A) and Chl a/b ratios (B) from bean leaves which were kept in the dark for 12 d. The values are the average of three independent experiments. (A) -●-; tatal Chl; -○-, Chl a; -■-, Chl b; (B) -□-, Chl a/b ratio.

Table 1. Changes in chloropyll contents and Chl a/b ratios from bean leaves which were kept under white light for 12 d.

μg Chl/g fresh weight

days after excision (d)	Chl a	Chll b	Chl a/b	total Chl
0	771.5[a)]	360.5	2.14	1132
2	776.9	363.1	2.14	1140
4	721.5	332.5	2.17	1054
6	640.3	289.7	2.21	930
8	589.8	262.2	2.25	852
10	538.6	224.4	2.40	763
12	399.9	158.1	2.53	558

[a)]The values are the average of three independent experiments.

Table 2. Changes in chloropyll contents and Chl a/b ratios from bean leaves which were kept under red light for 12 d.

μg Chl/g fresh weight

days after excision (d)	Chl a	Chl b	Chl a/b	total Chl
0	771.5[a)]	360.5	2.14	1132
2	768.8	359.2	2.14	1128
4	680.1	314.9	2.16	995
6	621.7	281.3	2.21	903
8	569.1	252.9	2.25	822
10	487.1	209.9	2.32	697
12	346.6	144.4	2.40	491

[a)]The values are the average of three independent experiments.

other hand, Chl a/b ratio of leaves illuminated with white or red light was not significantly altered compared to those which were kept in the dark. Particularly, red light was more effective in the stability of Chl a/b ratio during leaf senescence than white light.

3.2. Relative distribution of chlorophyll-protein complexes

The bean thylakoids were solubilized with nonionic detergent/chlorophyll weight of 20 : 1 and applied to native green gel system (Fig. 2). Ten main pigmented bands shown in the green gel were nomenclatured in order of increasing mobility as follows: two bands of RC(reaction center)-LHC, three bands of RC-Core (RC-Core1, RC-Core2 and RC-Core3), LHCII, three bands of SC (small complexes, two bands of SC-1 and SC-2) and FP (free pigment). The band patterns were similar to those of *Chlamidomonas* chlorophyll-protein complexes (Allen and Staehelin, 1991). For analysis of chlorophyll-protein complex polypeptide composition in detail, the vertical strips obtained in the green gel system were incubated in SDS-solubilization buffer, and were applied to a fully denaturing gel (Fig. 3). PSI complex, main band of RC-LHC, consisted of

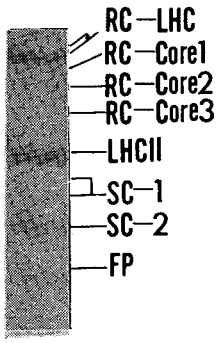

Fig. 2. Native green gel after electrophoresis of chlorophyll-protein complexes from bean leaves. Letters on the right-hand side indicate the designation of chlorophyll-protein complexes resolved. RC-LHC, reaction center-light harvesting complex; PSI, photosystem I complex; RC-core, reaction center-core complex; LHCII, trimeric form of the main light harvesting antenna PSII; SC, small complex; FP, free pigment.

P700 apoproteins, LHCI apoproteins and smaller PSI polypeptides. We resolved a number of core complexes associated with PSI and PSII, and RC-Core region associated with PSII particularly contained CP43/CP47 and D1/D2 apoproteins. LHCII was preserved as trimers of the main light harvesting antenna of PSII, and the band was highly fluorescent (data not shown). The fact indicates that the presence of antenna complexes not attached to reaction centers, and it agrees with that using *Chlamidomonas* chlorophyll-protein complexes (Allen and Staehelin, 1991). The region of SC contained a number of complexes, most of which appeared to be partially dissociated PSII components. Particularly, SC-2 contained a number of monomeric LHCII apoproteins.

3.3. Disassembly of chlorophyll-protein complexes during dark-induced senescence

The changes of relative distribution of chlorophyll-protein complexes in the detached leaves kept in the dark for 12 d were shown in Fig. 4. The relative amount of each complexes in the leaves of 21 day-old bean plants were 26.4% of PSI, 3.5% of RC-Core1, 3.8% of RC-Core2, 4.2%

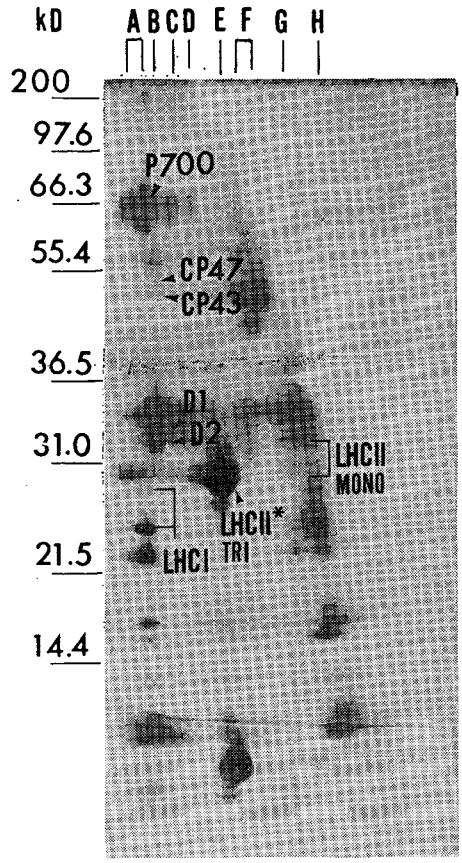

Fig. 3. Two dimensional gel in which bean chlorophyll-protein complexes have been separated on native green gel in the first dimension, and on a fully denaturing SDS-PAGE gel in the second dimension. Letters on the top indicate the designation of chlorophyll-protein complexes resolved: A, RC-LHC; B, RC-Core1; C, RC-Core2; D, RC-Core3; E, LHCII; F, SC-1; G, SC-2; H, FP. Silver-stained gel. P700, P700 apoproteins; CP47/CP43, internal chlorophyll a-binding of PSII; D1/D2, core apoproteins of PSII; LHCII*, apoproteins of LHCII.

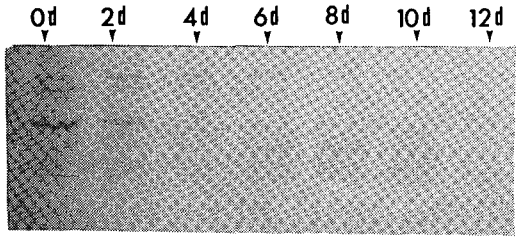

Fig. 4. Changes in the disassembly of chlorophyll-protein complexes from bean leaves which were kept in the dark for 12 d. Thylakoid membranes were solubilized with octyl glucoside, decyl maltoside and lithium dodecyl sulfate.

of RC-Core3, 35.7% of LHCII, 12.0% of SC-1 and 14.4% of SC-2 excluding free pigment. The disassembly of PSI complexes containing LHCI apoprotein, main band of RC-LHC, remarkably appeared to occur after 2 d of leaf senescence, and then the complexes was not detectable at 8 d of senescent leaves in the green gel. RC-Core1 and 2 was rapidly degraded at the early stage of senescence, whereas RC-Core3 was slightly increased until 4 d and slowly degraded thereafter. There was relatively stable in the region of LHCII throughout the senescence compared to those of the other complexes. As the gradual disassembly of LHCII which seemed to be trimeric form progressed, there was a steady increase in the amount of SC-2 during leaf senescence. As shown in Fig. 3, SC-2 contained a number of monomeric LHCII apoproteins. Therefore the fact suggests that trimeric LHCII apoproteins are degraded into monomeric LHCII apoproteins and the monomeric LHCII apoproteins seems to be accumulated in the thylakoid membranes as a stable form, in the senescing leaves. On the other hand, the rapid degradation of SC-1 appeared to occur after 4 d, and it agrees with the previous results using *Arabidoposis* (Oh and Lee, 1996).

3.4. Effect of light on the disassembly of chlorophyll-protein complexes

The above experiments for the changes of chlorophyll contents and Chl a/b ratios indicated that light was a regulator in the stability of chlorophylls during leaf senescence. The following experiments, therefore, conducted to examine how light regulates the disassembly of chlorophyll-protein complexes in the senescing leaves. The detached leaves illuminated with white light showed the structural stability of chlorophyll-protein complexes during senescence when compared to those kept in the dark (Fig. 4 and Table 3). The breakdown of PSI complexes was

Table 3. Changes in the disassembly of chlorophyll-protein complexes from bean leaves which were kept under white light for 12 d.

days after excision (d)	Relative amount of chlorophyll in chlorophyll-protein complexes						
	PSI	RC-Core1	RC-Core2	RC-Core3	LHCII	SC-1	SC-2
0	26.4	3.5	3.8	4.2	35.7	12.0	14.4
2	11.8	1.2	4.9	3.5	45.2	15.2	18.2
4	8.5	-	7.1	4.8	47.6	4.8	27.2
6	6.0	-	6.5	16.4	40.4	4.3	26.4
8	3.9	-	-	2.2	50.9	3.9	39.1
10	2.0	-	-	2.9	29.7	3.7	61.7
12	-	-	-	-	37.5	-	62.5

Table 4. Changes in the disassembly of chlorophyll-protein complexes from bean leaves which were kept under red light for 12 d.

days after excision (d)	Relative amount of chlorophyll in chlorophyll-protein complexes						
	PSI	RC-Core1	RC-Core2	RC-Core3	LHCII	SC-1	SC-2
0	26.4	3.5	3.8	4.2	35.7	12.0	14.4
2	19.6	2.4	7.1	3.8	39.3	13.2	14.6
4	7.3	1.2	6.3	4.8	45.2	10.2	25.0
6	5.2	1.0	7.5	6.9	40.4	8.2	30.8
8	2.2	-	0.8	2.0	43.0	5.0	47.0
10	1.2	-	-	-	34.7	3.3	60.8
12	-	-	-	-	43.8	-	56.2

obvious after 6 d, and PSI complexes were almost degraded after 10 d. In addition, the disassembly of RC-Core3 and SC-1 was suppressed by the illumination of white light compared to dark condition. Red light was, particularly, more effective in the suppression of LHCII degradation than white light during leaf senescence, but the leaves illuminated with red light had lower levels in the accumulation of SC-2 when compared to those illuminated with white light (Tables 3 and 4). Also, white light had only a weak protecting effect on the disassembly of PSI complexes when compared to red light.

4. DISCUSSION

The relative distribution of overall chlorophyll-protein complexes in the detached leaves of 21 day-old bean plants grown in light was observed in native green gel (Fig. 2). Ten main pigmented bands were separated in order of increasing mobility, and the bands were classified as follows: two bands of RC-LHC, three bands of RC-Core (RC-Core1, 2 and 3), LHCII, three bands of SC (two bands of SC-1 and SC-2) and FP. The band patterns appeared to be similar to those reported by others using *Arabidoposis* and *Chlamidomonas* as well as oat seedlings (Allen and Staehelin, 1991; Peter and Thornber, 1991; Lee, 1996). For analysis of chlorophyll-protein complex polypeptide composition in detail, the gel slices obtained in the native green gel system applied to a fully denaturing gel system (Fig. 3). PSI holocomplexes, main band in the region labeled RC-LHC, were easily seen in the second dimension gel by the presence of P700 apoprotein, a number of LHCI apoprotein and the smaller PSI subunits. In the RC-Core regions named to RC-Core1, RC-Core2 and RC-Core3, there were a number of core complexes associated with PSI and PSII. In this experiments, RC-Core complexes were seemed to be associated with PSII by the presence of a number of CP43/CP47 and D1/D2 apoproteins, but it is not clear whether or not RC-Core complexes named to RC-Core1, RC-Core2 and RC-Core3 were associated with PSII. Therefore, it is necessary to identify correct composition of RC-Core complexes associated with PSI or PSII. Although trimeric LHCII complexes had known to be composed by at least three types of LHCII using the native green gel system (Allen and Stahelin, 1991), those were composed by one type of LHCII in the present study using nondenaturing green gel. These

results are consistent with the observation using barley and oat seedlings (Dreyfuss and Thornber, 1994; Lee, 1996). SC complexes were composed by a number of complexes, most of which appeared to be partially dissociated PSII components. SC-1 complexes were seemed to contain smaller PSII complexes such as oxygen evolving complexes as shown in the previous reports (Dreyfuss and Thornber, 1994; Preiss and Thornber, 1995; Oh and Lee, 1996), whereas SC-2 complexes were appeared to include a number of LHCII monomer.

Fractionation of chlorophyll-protein complexes using native green gel system, by which the complexes were solubilized and separated with minimum loss of noncovalently bound chlorophylls led to excellent results in the study of disassembly of chlorophyll-protein complexes during leaf senescence. The status of disassembly of the various chlorophyll protein complexes was easily represented throughout leaf senescence (Fig. 4). PSI complexes declined steadily until the early stage of leaf senescence and decreased significantly thereafter. RC-Core1 and 2 were rapidly degraded at the early stage of leaf senescence, whereas RC-Core3 complexes were slightly increased until 4 d and slowly decreased thereafter. These results are similar to other observations used oat and barley seedlings that core complexes associated with PSII was increased for the early stage of greening and gradually decreased thereafter (Dreyfuss and Thornber, 1994; Lee, 1996). There was more stable in the region of LHCII throughout leaf senescence than those of the other chlorophyll-protein complexes. Mae *et al.* (1993) also reported that the decrease of LHCII protein was delayed until later in the leaf senescence of *Lolium tumelentum*. On the other hand, gradual decrease in LHCII induced the increase in the relative amount of SC-3. Therefore, the facts suggest that the disassembly of LHCII trimer is reorganized into the pigmented monomeric LHCII, as a stable form during dark-induced senescence.

The gradual loss of chlorophyll was a good representative of leaf senescence, and induced yellowing of leaf area. In this experiment, leaf yellowing started at the whole area of senescing leaves, but in the natural senescence, the yellowing started at the tip of the leaves (Lohman, 1994). Light affects chlorophyll contents, Chl a/b ratio and the disassembly of chlorophyll protein complexes in the detached leaves of bean plants. The illumination of white or red light significantly caused the suppression of chlorophyll loss during leaf senescence when compared to dark condition, and white light strongly retarded the breakdown of chlorophylls compared to red light (Fig. 1, and Tables 2 and 3). But Okada *et al.* (1992) has proposed that effect of very weak white light is equivalent to that of red light which retards chlorophyll loss. Therefore, it is not clear whether light quality or intensity controls the suppression of chlorophyll degradation. Nevertheless, it can be suggested that white light may be more effective in protecting chlorophyll loss than red light. The changes of Chl a/b ratio was almost not altered until the middle stage of dark-induced senescence, but Chl a/b ratio was remarkably increased in the late stage (Fig. 1). The increase of Chl a/b ratio may be due to the degradation of light-harvesting chlorophyll complexes containing both Chl a and Chl b. On the other hand, white and red light caused the stability of Chl a/b ratio when compared to dark condition, and red light was more effective in the stability of Chl a/b (Fig. 1, and Tables 2 and 3). These results suggest that no significant changes in the Chl a/b ratio may cause the degradation in Chl a-binding proteins of PSI and PSII, and LHCII containing both Chl a and Chl b in parallel, and the increase of Chl

a/b ratio may induce the rapid degradation of LHCII. The facts agree with the notion using the senescent leaves of rice and *Hibiscus* (Okada, 1992; Siffel, 1991). The changes of Chl a/b appear to be an adaptive response of leaves to changing environments.

From the results for Chl a/b ratio it could be suggested light induced differences in the distribution of chlorophyll between chlorophyll-protein complexes in the senescent leaves. White light caused the structural stability of chlorophyll-protein complexes during leaf senescence when compared to dark condition (Fig. 4 and Table 3). The disassembly of PSI holocomplex was significantly retarded by the illumination of white light compared to dark condition. It is consistent with the observation that PSI complex degradation in the rice leaves is strongly suppressed by illumination of white light (Okada, 1992). Red light was, particularly, more effect in the retardation of LHCII breakdown than white light during leaf senescence, whereas white light was slightly effect in protecting the disassembly of PSI holocomplexes (Tables 3 and 4). The results support the notion that light quality causes a reorganization of the components of chlorophyll-protein complexes such that plants grown in white light were enriched in PSI complexes, while those grown in red light were enriched in PSII complexes containing LHCII (Glick, 1986; Deng, 1989). However Lee *et al.* (1996) have proposed that thylakoids from oat seedlings adapted to red light have lower amount of LHCII than those adapted to white light. These results with the results for Chl a/b ratio suggest, therefore, that the effect of light quality on the composition of chlorophyll-protein complexes during leaf senescence is an important regulatory factor, but not the same for all plant species.

The results of the present study have identified changes in the disassembly of chlorophyll-protein complexes during dark-induced senescence and suggested the important role of light that controls the breakdown of chlorophyll-protein complexes of senescing leaves. Further studies are necessary to clarify molecular mechanism that allows the structural changes of chloroplast to adapt to light because the changes are attributed by the plastid mRNA encoding the proteins for two photosystem.

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