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Variation in Photosynthesis and Leaf Pigments of Susceptible *Pinus densiflora* and Resistant *Pinus rigida* Following Pine Gall Midge Attack[1]

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솔잎혹파리 侵害에 따른 感受性樹種인 소나무와 抵抗性樹種인 리기다소나무에 있어서 光合成 및 葉의 色素變異에 관한 研究[1]

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ABSTRACT

Susceptible trees of *Pinus densiflora* and resistant trees of *Pinus rigida* following pine gall midge (*Thecodiplosis japonensis* Uchida et Inouye) attack were seasonally compared to examine the variation in needle growth and photosynthetic ability, respiration rate, chlorophyll contents, carotenoid and anthocyanin contents. Also, carotenoid and anthocyanin contents of larvae both from soil and from galled tissue were compared during March and September, respectively. The plantation damaged severely by this insect consisted mostly of 10-to 15-year old *P. rigida* and *P. densiflora*. The results obtained in this study were as follows: 1) The length of the infested needles of *P. densiflora* decreased by 48.1 percent compared with the normal needles, while that of *P. rigida* did 37.4 percent. 2) All of *P. densiflora* and *P. rigida* showed higher photosynthetic ability in normal needles than in infested needles. The maximum photosynthetic ability of *P. densiflora* was shown in mid-August, while that of *P. rigida* in mid-October. In contrast to that, respiration rate of infested needles was higher than that of normal needles in both species. The respiration rate of *P. rigida* was higher than that of *P. densiflora*. 3) *P. rigida* had higher total chlorophyll contents than *P. densiflora*. The total carotenoid contents tents in infested needles were higher than those in normal needles of both species. 4) Total carotenoid contents were generally higher in *P. rigida* than in *P. densiflora* during the growing season. The total carotenoid content (0.094mg/g) in larvae from soil was similar to that (0.092mg/g) in larvae from galled tissues. 5) Infested needles of both species showed higher anthocyanin contents than normal needles. Higher anthocyanin contents in galled needles were due primarily to its active formation stimulated by larval attack. Thus, reddish-brown coloration occurred only in galled needles of *P. densiflora*.

Key words: Pinus densiflora; Pinus rigida; *pine gall midge; photosynthesis; anthocyanin; carotenoid.*

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要 約

솔잎혹파리(*Thecodiplosis japonensis* Uchida et Inouye) 侵害에 따른 生長 減少와 色素變異를 調査比較하기 위해 지난해에 솔잎혹파리 被害가 甚한 地域－京畿道 水原市 巴場洞－에서 感受性 樹種인 소나무(*Pinus densiflora* S. et Z.)와 抵抗性 樹種인 리기다소나무(*Pinus rigida* Mill.)를 대상으로 솔잎혹파리 侵入前, 後의 針葉生長, 光合成能力 및 呼吸率, 葉綠素 그리고 carotenoid 및 anthocyanin 色素含量의 變化를 1983年 6月부터 10月까지 月別로 調査하였으며, 또한 3, 4月에는 土壤속에서 越冬中인 幼虫과 9月에 솔잎속에서 生長하는 幼虫의 carotenoid와 anthocyanin色素 含量을 調査한 結果는 다음과 같다. 1) 針葉生長에 있어서 솔잎혹파리에 侵害를 받은 被害葉이 健全葉에 比하여 소나무에 있어서는 48.1%, 리기다소나무에 있어서는 37.4%정도의 生長 減少를 보였다. 2) 光合成 能力에 있어서는 두 樹種 모두 健全葉이 被害葉보다 높았으며, 소나무에서는 8月에 最大値를 보였으나 리기다소나무에서는 10月에 最大値를 보였다. 한편, 呼吸率 있어서는 리기다소나무가 소나무보다 높았으며, 두 樹種 모두 被害葉이 健全葉보다 높았다. 3) 葉綠素 含量에 있어서는 리기다소나무가 소나무보다 높았으며, 두樹種 모두 被害葉이 健全葉보다 높았다. 4) 一般的으로, 솔잎혹파리에 抵抗性인 리기다소나무가 感受性인 소나무보다 carotenoid 含量이 많았다. 또한 土壤中 幼虫에서는 carotenoid 含量이 0.094㎎/g, 虫癭속 幼虫에서는 0.092㎎/g을 보여 서로 비슷하였으며, 이는 針葉에서 나타난 含量보다 적었다. 5) Anthocyanin 含量에 있어서는 두 樹種 모두 被害葉이 健全葉보다 높았으며, 특히 소나무의 虫癭葉에서 높게 나타난 것은 솔잎혹파리侵入의 刺戟으로 인한 含量의 增加이며 따라서 가을에 虫癭部位가 赤褐色으로 變한다고 생각된다.

Introduction

Along with pine caterpillar (*Dendrolimus spectabilis* Buttler) and pine bark beetle (*Myelophlus piniperda* L.), pine gall midge (*Thecodiplosis japonensis* Uchida et Inouye) is one of the severely injurying insects to *Pinus densiflora* in Korea.

Since the time of the first discovery of this insect in the areas of Seoul and Mokpo in 1929,[37] the area of pine plantation damaged by this insect has increased to be more than 290,000 hectares of *Pinus densiflora* and *Pinus thunbergii* forests.[22] Thus, the proper control measures to pine gall midge are urgently needed. Various attempts to control the midge have been made by several biological[19,20,21] and chemical methods.[3,4] One way of reducing the density of this insect population may be through plantation of trees genetically resistant to this insect: selection of phenotypically resistant trees and hybridization between them.

First, physiology of host trees attacked by insects as well as attacking behavior of the insect should be investigated prior to initiation of breeding work.

Hatched larvae infest new needles in late-May. The midges cause infested needles of susceptible *Pinus desiflora or Pinus thunbergii* to form galls at a needle base and browning of needles in late-August. This browning may be due probably to either occurrence of new pigment or changes in pigments. However, neither browning nor gall formation occurs in resistant *Pinus rigida* trees even though they are infested.[25] Also, growth of the needles attacked by the larva, whether *P. rigida* or *P. densiflora*, ceased in the middle of the growing season. Therefore, knowledge for reduction in photosynthesis and changes in pigments related to growth following the attack enables to provide a basic information about breeding resistance for pine gall midge.

The purpose of this study was to compare photosynthesis, respiration and pigments related to growth between susceptible *P. densiflora* and resistant *P. rigida* to pine gall midge before and after its attack.

Review of Literature

Trying to determine physiological factors relating to the resistance of plants to insects is helpful for breeding programs of insect resistance. The physiology of tree resistance to insects was reviewed by Hanover (1975).[11] Physiological changes in needles following pine gall midge attack were investigated by Lee (1970)[25] who mentioned that the resin duct anatomy may be associated with resistance in *P. rigida*. Furthermore, resin-like substances in the needles secreted by the stimulation of insect attack appeared to play a preventing role in further larval feeding (Lee *et al.*, 1981).[26]

Phenolic substances following pine gall midge attack were increased in the *P. densiflora* or *P. thunbergii* resestant to this insect (Lee *et al.*, 1981).[27] Gallic acid was detected in the susceptible trees of *P. densiflora* after attack. In *Citrus* trees, the free phenolics in the leaves of cultivars tolerent to *Radopholus similis* were increased from 2 to 7 percent of the total phenolics after infection (Feldman and Hanks, 1968).[6]

Trees affected by *R. similis* also show a reduction and compaction of terminal twig growth. Park (1982)[32] reported that the retardation in terminal and cambial growth of *P. densiflora* simultaneously occurred in the same year when trees were infested by pine gall midge.

Much work in investigating the effect of plant diseases upon photosynthesis, respiration or pigment contents has been done with agricultural crops, but seldom with forest trees. The photosynthetic CO_2 uptake was markedly reduced in diseased leaves of tobacco (Hopkins and Hampton, 1969)[13] and barley (Jensen, 1968)[15] as compared with healthy leaves. Respiration was rather greater in diseased than in healthy leaves.[1,5,15]

Insect injury on trees may affect both growth and yield. Apple fruits were reduced due to insect damage (Hall, 1974).[8] Significant reduction in photosynthesis was observed in apple trees simulated by insect injury one day after treatment when losses in leaf area exceeded 10 percent (Hall and Ferree, 1976).[9]

Seasonal trends in photosynthesis and respiration of *Pinus taeda* and *Pinus strobus* seedlings were reported by McGregor and Kramer (1963)[30] who found that maximum photosynthesis of *P. taeda* was attained in mid-September and that of *P. strobus* in mid-July.

Many investigators studied on seasonal changes in chlorophyll contents[18,33] and leaf chlorophyll contents in relation to photosynthetic rate or leaf age.[7,36] Total chlorophyll in virus-infected tobacco tissues was remarkably reduced (Hampton *et al.*, 1966)[10] and systemic infection with virus reduced over-all photosynthesis (Owen, 1957).[31] Barley infested with yellow dwarf virus showed a decresse in chlorophyll content and photosynthesis (Jensen, 1968).[15] However, chlorophyll contents were rather higher in *Pinus taeda* damaged scarcely by pine bark beetle than in *Pinus thunbergii* much damaged (Kurogi *et al.*, 1972).[24]

Carotenoid in plants was considered as accessory pigments in photosynthesis and as coloring matters in flowers and fruits. However, there were some evidence on variations in its content according to insect damage. For example, carotenoid contents were higher in the needles of *P. densiflora* or *P. thunbergii* damaged much by pine bark beetle than in the needles of *P. taeda* or *P. caribaea* scarcely affected (Kurogi *et al.*, 1972).[24] *P. densiflora* susceptible to pine bark beetle showed rather increased carotenoid content (Kim, 1975).[17] Thus, carotenoid contents were increased by pine bark beetle attack.

The red and blue colors of plants are mostly due to the presence of anthocyanin pigments. Hida (1958)[12] reproted that red coloration in conifer needles during the autumn was due to anthocyanins. Anthocyanins and leuco-anthocyanins were found in *Pinus lambertiana* seedlings during the fall (Krugman, 1956).[23] *Quercus ellipsoidalis* and *Corylus americana* exhibited development of anthocyanin during autumn coloration but *Populus tremuloides* did not (Sanger, 1971).[33]

As mentioned above most of studies of the relationship between photosybthesis, pigments and insect or disease injury have been conducted on the cultivated crops, but recently on forest trees. In particular, research on variations in photosynthesis, chlorophyll contents, carotenoid and anthocyanin contents after insect attack was seldom with tree species.

Materials and Methods

Materials

Three susceptible trees of *P. densiflora* and three resistant trees of *P. rigida* to pine gall midge were provided as plant materials (Table 1).

The plantation located at Pajang Dong, Suweon, Kyunggi-Do and consisted mostly of *P. densiflora* and *P. rigida on a slope facing south-eas*t has been severely attacked by this insect.

Needles were collected monthly starting from June 15th through October 15th. In order to reduce the measurement variation, the experimenters divided the crown of each tree into the upper part, the middle part, and the low part. Larvae in soil were collected in March and April. Larvae in galls of the needles were collected in September by separating from galled tissues.

Measurement of needle growth

Hundred needles and 100 fascicles per each species were collected in October 20th for length and weight measurements, respectively. Average length was measured with a ruler to the nearest 0.1cm and dry weight measured with a balance after drying at 80°C in an oven for 72 hours.

Photosynthetic ability and respiration rate measurement

Photosynthetic ability and respiration rate of the needles were measured using Oxygen Electrode and Meter (YSI Co., Ohio, USA).[14] The 0.2g of the needles sliced into small pieces were put

Table 1. General description for sample trees of *P. densiflora* and *P. rigida* used in this study

Species	Age (yrs)	Height (m)	D.B.H. (cm)	Infestation rate (%)
P. densiflora	10-15	5.6	14.0	50.2
P. rigida	10-15	5.2	9 5	35.3

into a cell and exposed under a tungsten lamp (300 μE m^{-2} sec^{-1}) at a temperature varying between 26°C and 27°C. Four *mi* of 50mM potassium-phosphate buffer (pH 7.2) solution containing 0.5mM $MgCl_2$, 0.1mM $CaSO_4$ and 2*ml* of 0.625M $NaHCO_3$ solution were used as reaction solution.

Extraction and determination of chlorophylls

Pine needles were sliced into small segments. Eighty percent aceton was used to extract chlorophylls from 0.5g of fresh needles. The chlorophyll content was determined by the method described by Arnon (1949)[2] who measured optical densities at 645nm and 663nm using spectrophotometer. According to the Arnon's formula, the amounts of total chlorophyll, chlorophyll *a* and chlorophyll *b* were calculated as follows: total chlorophyll $(mg/l) = 8.02A_{663} + 20.20A_{645}$, chlorophyll *a* $(mg/l) = 12.7A_{663} - 2.69A_{645}$, and chlorophyll $b\,(mg/l) = 22.9A_{645} - 4.68A_{663}$.

Extraction and determination of carotenoids

The sliced needles or larvae were ground in a mortar and then added with 10 to 15*ml* of acetone-ethanol (1:1 v/v) solution. The extract was carefully moved with a pipette into a flask. This process was repeated until no more pigment was extracted. The combined extracts were centrifuged at 8,000 rpm for 10 minutes. The extract was evaporated up to 30*ml* and then added with 3*ml* of 60% KOH. The mixed solution was stored at room temperature for 2 hours in a dark room. After saponification, the aqueous phase was extracted with 33*ml* of distilled water and then with 66*ml* of ether. The lower phase was extracted again with 34*ml* of ether. The combined extracts were washed twice with distilled water. The ether extract was concentrated through N_2 gas and added with 50*ml* of acetone.

The absorbance of the extracts was measured using spectrophotometer. The amount of total carotenoids in the extract was calculated according to the following equation:[28] $C = D \times V \times f \times 10/2500$, where C, D, V and f stand for total carotenoid in miligrams, optical density, total volume in milliliters, and dilution factor, respectively. The value, 2500, indicates average extinction coefficient for carotenoids.

Extraction and determination of anthocyanin

The method described by Mancinelli (1975)[31] was adopted for the extraction of anthocyanin. Anthocyanins were extracted from the 3g of fresh needles sliced into small pieces with 20*ml* of 1% HCl in methanol (w/v) for 3 days at a refrigerator. The extracts were filtered and their absorbances were measured at 530nm and 657nm using spectrophotometer. The amount of anthocyanin was calculated according to the following formula:

$$\text{Anthocyanin} = A_{530} - 0.33A_{657}.$$

Results

Needle growth and photosynthesis

Needle length and dry weight were compared between the normal and the infested needles of *P. densiflora* and *P. rigida* following pine gall midge attack (Table 2). The infested needles of *P. densiflora* decreased by 48.1 percent in length compared to normal needles, while those of *P. rigida* did by 37.4 percent compared to normal needles. However, the dry weight of the infested needles of *P. densiflora* decreased by 40 percent compared to normal needles in *P. densiflora*, and that of *P. rigida* decreased by 43.6 percent.

Monthly changes in photosynthetic ability

Table 2. Needle growth and dry weight in *P. densiflora* and *P. rigida* following pine gall midge attack (measurement was done in mid-October)

Species		Needle length (cm)	Dry wt. per 100 fascicles (g)	Ratio of normal to infested needle (%)	
				Needle length	Dry wt./100 fescicles
P. densiflora	Normal	9.55	3.93	100.0	100.0
	Infested	4.96	2.36	51.9	60.0
P. rigida	Normal	10.23	7.71	100.0	100.0
	Infested	6.41	4.35	62.6	56.4

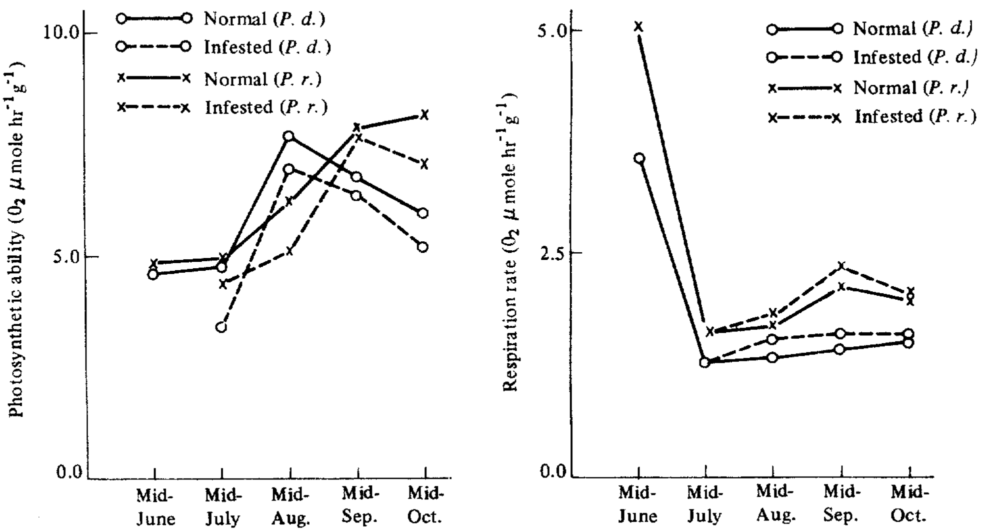

Fig. 1. Seasonal changes in photosynthetic ability and respiration rate in normal and infested needles of *P. densiflora* and *P. rigida* following pine gall midge attack.

and respiration rate per unit needle weight were shown in Fig. 1. The photosynthetic abilities of the two species were alike until mid-July. After that, *P. densiflora* showed higher photosynthetic ability than *P. rigida* and its maximum value was observed in mid-August. However, *P. rigida* showed steady increase in photosynthetic ability and then its maximum value was observed in mid-October. When photosynthetic ability was compared between normal and infested needles, both species showed higher photosynthetic ability in normal needles than in infested needles.

In contrast to that, respiration rate of infested needles was higher than that of normal needles in both species (Fig. 1).

The maximum values in respiration rate of the two species were observed in mid-June, and rapidly decreased thereafter.

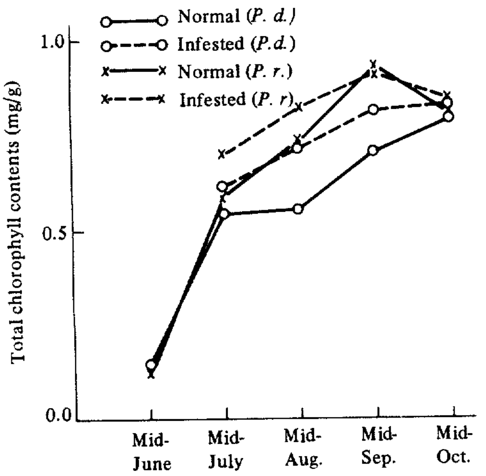

Fig. 2. Seasonal changes in total chlorophyll contents in normal and infested needles of *P. densiflora* and *P. rigida* following pine gall midge attack.

Chlorophyll contents in needles

Seasonal variations in chlorophyll *a*, chlorophyll *b* and total chlorophylls of the two species were shown in Table 3. The total chlorophyll contents of both *P. densiflora* and *P. rigida* increased with increasing dates (Fig. 2). In comparison between the two species, *P. rigida,* in general, showed higher chlorophyll contents than *P. densiflora.* The total chlorophyll contents of both species were higher in infested needles than in normal needles. Similar trends were observed in contents of both chlorophyll *a* and chlorophyll *b* of the two species.

The ratio of chlorophyll *a* to chlorophyll *b* is shown in Fig. 3. The ratio was higher in normal needles than in infested needles. *P. densiflora* had higher ratio than *P. rigida* during the growing season.

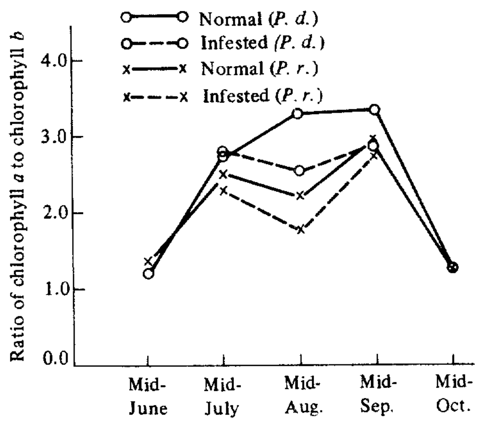

Fig. 3. Seasonal changes of the ratio of chlorophyll *a* to chlorophyll *b*.

Table 3. Chlorophyll *a* (Ch. *a*), chlorophyll *b* (Ch. *b*), and total chlorophylls (Chl.) contents (mg g^{-1} fresh wt.) in normal and infested needles of *P. densiflora* and *P. rigida* following pine gall midge attack

		Mid-June			Mid-July			Mid-August			Mid-September			Mid-October		
		Ch.*a*	Ch.*b*	Chl.	Ch.*a*	Ch.*b*	Chl.	Ch.*a*	Ch.*b*	Chl.	Ch.*a*	Ch.*b*	Chl.	Ch.*a*	Ch.*b*	Chl.
P. densiflora	Normal	0.078	0.064	0.142	0.398	0.144	0.542	0.424	0.128	0.552	0.541	0.160	0.701	0.446	0.351	0.797
	Infested	–*	–	–	0.448	0.160	0.608	0.494	0.213	0.707	0.603	0.205	0.808	0.446	0.361	0.827
P. prigida	Normal	0.069	0.051	0.120	0.418	0.168	0.586	0.500	0.225	0.725	0.679	0.235	0.914	0.456	0.355	0.811
	Infested	–	–	–	0.486	0.219	0.698	0.527	0.289	0.816	0.660	0.240	0.900	0.462	0.366	0.828

*No data available

Total carotenoid contents in needles and larvae

Fig. 4 shows seasonal variations in total carotenoid contents of *P. densiflora* and *P. rigida*. Total carotenoids were generally higher in *P. rigida* than in *P. densiflora* during the growing season. Normal needles of *P. densiflora* showed higher carotenoid than infested needles, whereas infested needles of *P. rigida* did higher cartenoid contents until mid-August and thereafter normal needles showed higher carotenoid.

The total carotenoid contents (0.094mg g^{-1}) in larvae collected from soil were similar to those (0.092mg g^{-1}) in larvae from galled tissue (Table 4).

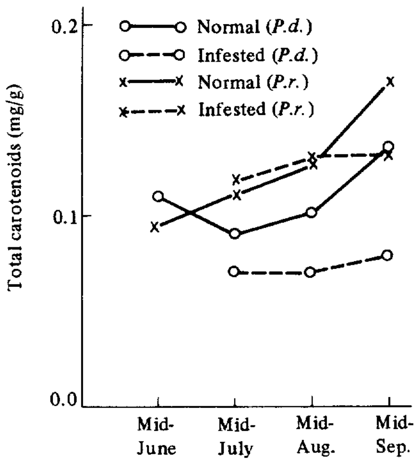

Fig. 4. Seasonal changes in total chrotenoid contents in normal and infested needles of two species following pine gall midge attack.

Table 4. Carotenoid contents (mg g^{-1} fresh wt.) in larvae

Larvae in soil	0.094
Larvae in gall	0.092

Anthocyanin contents in needles and larvae

Seasonal changes in anthocyanin contents of the two species were shown in Fig. 5. Until mid-June, anthocyanins were not detected in both species. The anthocyanin contents were higher in *P. densiflora* than in *P. rigida* during the growing season except for mid-August. The anthocyanin contents

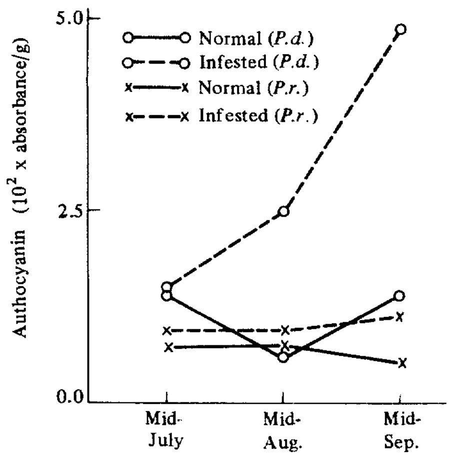

Fig. 5. Seasonal changes in anthocyanin in normal and infested needles of two species following pine gall midge attack.

in infested needles of the two species were higher than those in normal needles. This was more obvious in susceptible trees of *P. densiflora*.

Discussion

Needle growth

Needles grow by increments in its dry weights, indicating that photosynthetic organs increase in conifers.

The results that the needles infested by the pine gall midge in both *P. densiflora* and *P. rigida* decreased growth compared with normal needles were similar to those reported by Lee (1970)[25] who studied seasonal needle growth between infested and normal needles of *P. densiflora* and *P. rigida*. Similar results were also reported by Park (1982)[32] who found reduction in terminal and cambial growth after pine gall midge attack, and by Kearby and Benjamin (1964)[16] who observed that *Pinus resinosa* needles infested by red pine needle midge were shorter than the normal needles.

The higher ratio of infested to normal needles in length observed in *P: rigida* than in *P. densiflora* is probably due to the fact that death of larvae did not hinder its needle growth in *P. rigida* while in *P. densiflora* survived larvae interrupted needle growth.

Photosynthetic ability and respiration rate

Both of *P. densiflora* and *P. rigida* showed lower photosynthetic ability in infested needles than in normal needles. Such result was similar to that mentioned by Owen (1957)[31] who reported that tobacco tissues infested with virus reduced overall photosynthesis. Similar results were also observed in diseased leaves of tobacco (Hopkins and Hampton, 1969)[13] and of barley (Jensen, 1968).[15] For comparison in photosynthesis between two species, *P. densiflora* started needle opening and showed maximum photosynthesis earlier than *P. rigida*. Thus *P. densiflora* grows faster in early growing season, while *P. rigida* does faster and longer in late growing season.

In contrast to photosynthesis, both of *P. densiflora* and *P. rigida* showed higher respiration rate in infested needles than in normal needles. These results were similar to those resported by Allen (1942)[1], Daly *et al.* (1961)[5] and Jensen (1968)[15] who found increased respiration in diseased leaves than in healthy leaves. After mid-August, the photosynthesis in infested needles of *P. rigida* rapidly increased up to level of normal needles. It appears that photosynthetic activities in the infested needles of *P. rigida* have been recovered after the larval death.

Chlorophyll contents in needles

The involvement of chlorophylls in absorption of radiation and participation in photosystems transforming the absorbed radiant energy into chemical energy have been well known.

Chlorophyll contents in normal needles of *P. densiflora* or *P. rigida* were lower than those of *P. radiata* (Wood, 1974).[38] The higher ratios of chlorophyll *a* to chlorophyll *b* in the normal needles of both species than in infested needles probably support increased photosynthesis in normal needles. Higher chlorophyll contents in *P. rigida* resistant to pine gall midge than in susceptible *P. densiflora* were similar to those shown by Kurogi *et al.* *(1972)*[24] who observed that chlorophyll contents were higher in *P. taeda* scarcely damaged by pine bark beetle than in *P. thunbergii* much damaged. The chlorophyll contents of the two species were higher in infested needles than in normal needles during the growing season. These results were contrasting to those reported by Hampton *et al.* (1966)[10] who observed remarkable reduction of total chlorophyll contents in virus infested tobacco tissues and by Jensen (1968)[15] who found chlorophyll contents decrease in barley leaf infested with dwarf virus.

In general, a strong relationship between photosynthesis and chlorophyll contents was reported.[34,35] In this study, however, chlorophyll contents per unit fresh weight were higher in infested needles than in normal needles, whereas photosynthesis was lower in infested needles than in normal needles. On the other hand, if calculated based on weight per needle, chlorophyll contents and photosynthetic ability would be higher in normal needles than in infested.

Carotenoid contents in needles and larvae

The higher carotenoid contents exhibited in *P. rigida* resistant to pine gall midge than in susceptible *P. densiflora* were not similar to the result reported by Kurogi *et al.* 1972).[24] They found that the carotenoid contents were higher in *P. densiflora* or *P. thunbergii* damaged much by pine bark beetle than in *P. taeda* or *P. caribaea* scarcely affected. Reduced carotenoid contents observed in the infested needles of *P. densiflora* may be due to a possible interruption of carotenoid synthesis by larvae. The fact that higher contents observed in normal needles of *P. rigida* than in infested after mid-August could be due to normal synthesis of carotenoids after larval death.

The small amount of carotenoid in larvae from soil or galled tissues may be possibly originated from the infested needle tissues, not from themselves.

Anthocyanin contents of needles

The higher anthocyanin contents shown in galled needles of *P. densiflora* than either in normal needles or in both infested and normal needles of *P. rigida* might be due primarily to its active formation stimulated by larval attack. This was supported by the fact that reddish-brown coloration or fall browning occurred only in galled needles of *P. densiflora* susceptible to this insect.

Conclusion

Needle growth, photosynthesis, respiration, needle chlorophyll contents, total carotenoid and anthocyanin contents in needles were compared between susceptible trees of *P. densiflora* and resistant trees of *P. rigida* following pine gall midge attack. The results obstained were as follows: 1) The length of the infested needles of *P. densiflora* decreased by 48.1 percent compared with the normal needles, while that of *P. rigida* did by 37.4 percent. However, the dry weight in the infested needles of *P. densiflora* decreased by 40 percent compared with the normal needles, while that of *P. rigida* decreased by 43.6 percent. 2) When photosynthetic ability was compared between normal and infested needles, both species showed higher photosynthetic ability in normal needles than in infested needles. In contrast to that, respiration rate of infested needles was higher than that of normal needles in both species. The respiration rate of *P. rigida* showed higher than that of *P. densiflora*. The maximum photosynthetic ability of *P. densiflora* was observed in mid-August, while that of *P. rigida* was in mid-October. 3) *P. rigida* had higher total chlorophyll contents than *P. densiflora*. The total chlorophyll contents in infested needles were higher than those in normal needles of both species. 4) Total carotenoid contents were generally higher in *P. rigida* than in *P. densiflora* during the growing season. Normal needles of *P. densiflora* showed higher carotenoids than infested needles, whereas infested needles of *P. rigida* did higher carotenoid until mid-August, and thereafter normal needles were higher. Thus, low carotenoids in infested needles of *P. densiflora* may be due to insect attack. The total carotenoid contents (0.094 mg g^{-1}) in larvae from soil were similar to those (0.092 mg g^{-1}) in larvae from galled tissues. 5) Anthocyanin contents were generally higher in *P. densiflora* than in *P. rigida* during the growing season. Infested needles of both species showed higher anthocyanin contents than normal needles. Higher anthocyanins in galled needles were due primarily to its active formation stimulated by larval attack. Thus, reddish-brown coloration occurred only in galled needles of *P. densiflora*.

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