Relation between the Natural Fall of Calyptra and the Set of Seeded Berries in 'Kyoho' and 'Pione' Grapes

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'巨峰'および 'ピオーネ'における花冠の離脱と
有種子果の結実との関係
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The flower which remained covered by calyptra after bloom (called hereafter as abnormal flower) tended to occur in 'Kyoho' and 'Pione', and the rates of its occurrence were higher in vigorous shoots showing reciprocal relation to the set of seeded berries. Though the shoot spraying with SADH before bloom promoted the set of seeded berries, the abnormal flowering was not inhibited by the treatment. The cluster dipping with 4-CPA before bloom induced almost complete seedlessness accompanied by the increment of the abnormal flowers. When the normal and abnormal flowers were compared as for the set of seeded berries in the strong shoots, in which the abnormal flowering occurred at relatively high rates, they did not differ significantly each other in all treatments except in the SADH. The number of pollen tubes detected on the cross section of style was smaller in the abnormal flowers than in the normal ones, but no significant difference was found between them in the number of ovules which a pollen tube penetrated. Further, the abnormal flowers were shorter in the length of style, which was a favorable character for self pollination in such a condition. Thus, self pollination is possible in the abnormal flower, and there seems to be little causal relation between the abnormal flowering and the set of seeded berries in 'Kyoho' and 'Pione', eventhough these two phenomena show often a reciprocal relationship.

Introduction

It has been known that in the mature inflorescences of 'Kyoho' and 'Pione', poor setting 4 n grape cultivars, some flowers remain covered by the calyptra which does not fall spontaneously but becomes progressively brownish and finally decays still adhering to the stigma. ¹⁾²⁾ has shown that the abnormal flowering may occur in any cultivar during cold or rainy weather, and the persisting calyptra greatly reduces berry setting but does not appear to prevent it. Lombardo *et al.* have

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described that in a normal producing cultivar, 'Verduzzo', the pistil appears quite short compared with the whole bud length, and the floral morphology allows self pollination even in flowers still provided with calyptra.

The purpose of this study was to clarify that to what extent the abnormal opening of flowers observed in 'Kyoho' and 'Pione' affected the pollination, fertilization and berry set, and following experiments were conducted in 1987. 1) The comparison of the occurrence of the abnormal flowering among 5 leading grape cultivars in Japan involving 'Kyoho' and 'Pione'. 2) The survey on the effects of shoot vigour and the prebloom application of a growth retardant, SADH and an auxin, 4-CPA on the occurrence of the abnormal flowering and the set of seeded and seedless berries in 'Kyoho' and 'Pione'. It has been well documented in these cultivars that the more vigorous the shoot is, the worse the set of seeded berries $\frac{4,5}{1,5}$ and that the prebloom application of SADH by either shoot spraying or cluster dipping promotes the fertilization and the set of seeded berries. Further, it has been reported that the prebloom application of 4-CPA by cluster dipping increases the number of abnormal flowers in 'Muscat Bailey A' and the treatment induces markedly seedlessness in 'Kyoho'. 3) The observation on the size of female organs at bloom and the elongation of pollen tubes in pistil in the normal and abnormal flowers in 'Kyoho'.

Materials and Methods

Comparison of the occurrence rates of the abnormal flowers among cultivars. Each one adult vine of 'Delaware', 'Campbell Early', 'Muscat Bailey A', 'Kyoho' and 'Pione' grown in a plastic house of Shimane University was used. They extended long arms with a cane pruning on over-head pargola, each occupying about 70 m² cropping area. Ten shoots of moderate vigour were selected in each vine, and the number of total flowers and the abnormal flowers per cluster were counted during bloom. Before bloom, the wings of clusters were removed in all cultivars, and the basic several branches and the apical end of the rachis were cut off leaving 12 branches in 'Muscat Bailey A', 'Kyoho' and 'Pione'.

Survey on the effcts of shoot vigour and the prebloom application of SADH and 4-CPA on the occurrence of the abnormal flowers and the berry set. Each one adult vine of 'Kyoho' and 'Pione' grown in the same plastic house was used. The cropping area of a vine was devided into 2, south and north areas. The shoots of the south area were sprayed with SADH (succinic acid -2, 2-dimethylhydrazide) solution and those of the north remained untreated. Each 10 of strong (about 45 cm in length) and weak (about 25 cm) shoots were tagged in both the areas on the day of the treatment. In 'Kyoho' alone, each 10 of the strong and weak shoots were additionally selected in the SADH-untreated north area and their clusters were dipped with 4-CPA (4-chlorophenoxy acetic acid) solution. The SADH solution was applied at 5,000 ppm 23 days before full bloom (May $\frac{9}{6}$). Atlox BI was added at

1,000 ppm as a wettable agent. The 4-CPA solution was applied at 5 ppm 9 days before full bloom. Atlox BI was added at 1,000 ppm. Clusters except one on each shoot were removed on the day of the SADH treatment. One week before bloom, wings, branches and the apical end of rachises were cut off as descrided above. Shoot length was measured 23 days before full bloom (the date of the SADH application, May 6), at the beginning of bloom (May 26) and 14 days after full bloom (June 13). The number of flowers per cluster was counted until bloom. The pedicel of the abnormal flower which remained covered by the calyptra after bloom was tied by a short silk fiber as a mark and the number of the abnormal flowers per cluster was recorded. The number of set berries per cluster was counted about 3 weeks after full bloom after classifying berries by their flower type (normal or abnormal) and seed development (seeded or seedless).

Morphological observation on the normal and abnormal flowers. Thirty of the normal flower just after calyptra fall and the same number of the abnormal flower estimated at the similar stage were sampled from untreated clusters of the 'Kyoho' vine. The lateral and longitudinal length of ovary and the length of style were measured with microscopy. Further, 30 of the normal flower 3 days after calyptra fall and the same number of the abnormal flower estimated at the similar stage were sampled from the same clusters, and they were fixed and embedded following procedures previously described. Traverse sections from stigma to the bottom of ovary were stained in aniline blue and pollen tubes in the sections were observed by means of fluorescence.

Results

Cultivar difference was found in the occurrence of the abnormal flowers. None of them appeared in any examined clusters of 'Delaware' and 'Campbell Early', whereas they appeared at the rates of about 10% in those of 'Kyoho', 'Pione' and 'Muscat Bailey A' (Table 1).

The occurrence of the abnormal flowers was also affected by shoot vigour and the application of growth regulator. The occurrence rates of the abnormal flowers in the strong shoots without growth regulator were 12.0% in 'Kyoho' and 8.4% in 'Pione',

Cultivar	Date of ful	ll bloom	Occurrence rate of abnormal flower %		
'Campbell Early'	May	21	0.0 a		
'Delaware'	May	24	0.0 a		
'Kyoho'	May	27	12.7 b		
'Pione'	May	27	8.4 b		
'Muscat Bailey A'	May	29	7.9 b		

Table 1.Occurrence rates of flower remained covered by calyptra after
bloom (abnormal flower) in 5 leading cultivars in Japan.

z. Mean separation within column by Duncan's multiple range test, 5% level.

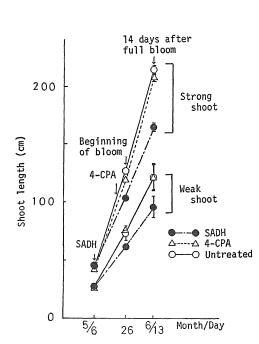
Cultivar	Shoot		Occurrence rate of abnormal	Rate of berry set %			Number of berries per cluster		
vigour	regulator	flower %	Seeded	Seedless	Total	Seeded	Seedles	s Total	
	Strong	SADH	9.3 cd	24.2 a	25.6 b	49.8 a	35.6 a	33.9 с	69.5 a
	Weak	SADH	4.8 d	30.8 a	3.6 d	34.4 bc	44.7 a	4.6 d	49.3 bc
'Kvoho'	Strong	4-CPA	53.7 a	0.6 d	42.3 a	42.9 ab	0.7 c	61.2 a	61.9 ab
ityono	Weak	4-CPA	24.4 b	1.1 d	37.3 a	38.4 b	1.6 c	53.5 b	55.1 b
	Strong	Untreated	12.0 c	6.9 c	20.5 b	27.4 cd	9.4 c	30.8 c	40.2 c
	Weak	Untreated	1.7 d	13.6 b	9.4 c	23.0 d	21.0 b	13.6 d	34.6 c
	Strong	SADH	10.0 a	12.8 b	56.0 a	68.7 a	18.7 b	83.0 a	101.7 a
'Pione'	Weak	SADH	3.5 b	19.3 a	20.5 c	39.8 bc	28.8 a	30.7 c	59.5 b
110110	Strong	Untreated	8.4 b	5.3 c	40.8 b	46.2 b	7.7 c	58.5 b	66.2 b
	Weak	Untreated	2.2 c	11.9 b	20.4 c	32.3 c	16.7 c	27.4 c	44.1 c

Shoot length (cm)

Table 2. Effects of shoot vigour, shoot spraying with SADH and cluster dipping with4-CPA on calyptra fall and berry set in 'Kyoho' and 'Pione' grapes.

z. Flower remained covered by calyptra after bloom.

y. Mean separation within columns by Duncan's multiple range test, 5% level.



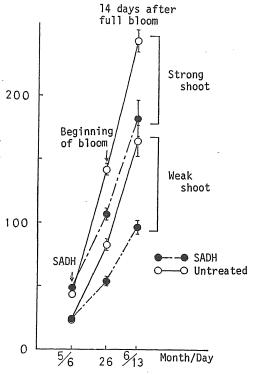
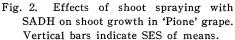


Fig. 1. Effects of shoot spraying with SADH and cluster dipping with 4-CPA on shoot growth in 'Kyoho' grape. Vertical bars indicate SES of means.



Cultivar	Type of flower	Set rate	%	
		SADH-treated	4-CPA-treated	Untreated
'Kyoho'	Abnormal	17.9 b	0.5 a	7.9 a
	Normal	25.3 a	0.8 a	6.8 a
'Pione'	Abnormal	10.1 a		4.3 a
	Normal	12.8 a		5.3 a

Table 3. Set rates of seeded berries in the normal and abnormal flowers of the strong shoots in 'Kyoho' and 'Pione' grapes.

z. Abnormal flower was that remained covered by calyptra after bloom.

y. Mean separation within columns by T test, 5% level.

which were significantly higher than those in the weak ones (1.7% and 2.2%) (Table 2). The shoot spraying with SADH significantly depressed shoot growth at and after bloom (Fig. 1 and 2), but the treatment did not affect significantly in 'Kyoho' or slightly increased in 'Pione' the occurrence rates of the abnormal flowers (Table 2). The cluster dipping with 4-CPA, which was applied in 'Kyoho' alone, remarkably promoted the occurrence of them without any effect on shoot growth and the rate in the strong shoots reached more than 50 % (Fig. 1 and Table 2).

As was expected from previous reports, the set rate of seeded berries and the number of seeded berries per cluster were significantly higher in the weak shoots than those in the strong ones in both 'Kyoho' and 'Pione' and they were significantly increased by the shoot spraying with SADH. In contrast, the cluster dipping with 4-CPA in 'Kyoho', also as expected, inhibited significantly the set of seeded berries

Type of flower	Pistil length mm	Style length mm	Ovary length mm	Ovary width mm
Abnormal	5.2 b	1.2 b	4.0 a	2.8 a
Normal	5.6 a	1.5 a	4.0 a	2.4 b

Table 4. Sizes of style and ovary in the normal and abnormal flowers in 'Kyoho' grape.

z. Abnormal flower was that remained covered by calymtra after bloom.

y. Mean separation within columns by T test, 5% level.

Table 5. Pollen tube elongation in floral tissues in the normal and abnormal flowers in 'Kyoho' grape.

	Number of pollen tubes detected at				
Type of flower	Style		Ovule		
	Style	Upper part	Middle part	Lower part	
Abnormal	5.4 b	3.8 b	3.8 b	3.0 b	1.9 a
Normal	24.1 a	9.3 a	6.9 a	6.4 a	2.7 a

z. Abnormal flower was that remained covered by calyptra after bloom.

y. Numbr of ovule which a pollen tube penetrated.

x. Mean separation within columns by T test, 5% level.

and promoted the set of seedless berries (Table 2), and the seedlessness ratios in the strong and weak shoots were extremely high, 98.9% and 97.1%, respectively.

To examine the effects of calyptra fall on pollination and fertilization, the set rates of seeded berries were obtained separately from the normal and abnormal flowers using the clusters of the strong shoots, in which the abnormal flowers occurred at relatively high rates (Table 3). The rates in the abnormal flowers did not differ significantly from those in the normal ones in all the treatments including untreated controls in 'Kyoho' and 'Pione' except in the SADH treatment in 'Kyoho', in which the rate in the abnormal was significantly lower than that in the normal.

For the same purpose, the flowers of the strong shoots in 'Kyoho' were morphologically observed. The abnormal flowers were significantly larger in width of ovary and shorter in the lengths of style and pistil than the normal ones (Table 4). The number of pollen tubes detected on the cross section of style in the abnormal flowers was significantly smaller than that in the normal ones. However, the difference in the number between them and the numbers themselves became smaller as a point of observation in ovary came down, and no significant difference was found between them in the number of ovules which a pollen tube penetrated (Table 5).

Discussion

The abnormal flowers were observed limitedly in the clusters of 'Kyoho', 'Pione' and 'Muscat Bailey A' but the rates of their occurrence were lower than those we presumed, probably due to favorable temperature conditions at bloom in the plastic $\frac{2}{10}$ Vallania has described that a tetraploid plant and a 2n-4n periclinal chimera plant of 'Barbara' grape show difficulties in calyptra fall of flowers at bloom. 'Kyoho' and 'Pione', too, are tetraploid cultivars, but whether or not the abnormal flowering inclines to take place in tetraploid grape cultivars is to be ascertained further.

The occurrence of the abnormal flowers was significantly affected by shoot vigour; its rate was much higher in strong shoots than weak ones in both 'Kyoho' and 'Pione'. Further, the cluster dipping with 4-CPA before bloom promoted extremely the abnormal flowering in 'Kyoho' as shown by Ueda in 'Muscat Bailey A'. From these facts, it is suggested that a high level of auxin in florets before and during bloom may be a promotive factor for the abnormal flowering of grapes.

According to the morphological observation of this research, self fertilization occurred even in the abnormal flowers, though the chance of pollination in them seems smaller than that in the normal. In fact, the set rate of seeded berries in the abnormal did not differ significantly from that in the normal in both 'Kyoho' and 'Pione'. The abnormal flowers in 'Kyoho' had shorter pistils than the normal ones, which resulted in a longer distance between stigma and calyptra. This character may be convenient for the self pollination in the flowers enveloped by calyptra as reported by Lombardo in 'Verduzzo' grage.

Thus, at least in 'Kyoho' and 'Pione', there seems to be little causal relation between the higher occurrence rate of the abnormal flowers in the strong shoots and the lower set rate of seeded berries in them. The induction of seedlessness by the prebloom application of 4-CPA is thought to be caused mainly by the functional 11,12,13 reduction of pollen and ovules as same as in the case of gibberellin, and it seems obscure that to what extent the abnormal flowering promoted by the treatment contributes to the induction of it. The application of SADH had little effect on the occurrence of the abnormal flowers. In the clusters of SADH-treated shoots, however, the promotive effect on the set of seeded berries were smaller in the abnormal flowers than in the normal ones. The number of pollen grains adhering to stigma seems smaller in the former than in the latter so that the promotive effect of SADH on the elongation of pollen tubes in pistil tissues may be limited in the former.

In conclusion, the abnormal flowering tending to occur in vigorous shoots in 'Kyoho' and 'Pione' is thought to have little direct effect on the set of seeded berries since self pollination and fertilization are possible in them. The abnormal flowers, however, may be more susceptible to diseases such as gray mold than the normal ones for their difficulties in calyptra fall. Therefore, the occurrence of them would have detrimental effects on the berry set depending on the climatic conditions at bloom.

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摘 要

⁽巨峰'と'ピオーネ'では,花冠不離脱花が発生しやすく,とくに強い新梢では弱い新梢より発生率が高く, 有種子果の着果率と逆の関係にあった. SADH 新梢散布により有種子果の着果率は上昇したが,花冠の離 脱は促進されなかった. 4-CPA 花房浸漬は花冠不離脱花を著しく増加するとともに高い無核化作用を示し た. 不離脱花の出現率の高かった強新梢で,不離脱花と離脱花の有種子果着果率を比較すると,巨峰の SADH区を除いて,両品種のいずれの区でも差が認められなかった. 花柱内に侵入した花粉管の数は,不離 脱花が少なかったが,花粉管の侵入したはい珠の数では,離脱花と差がなかった. 不離脱花の花柱は離脱花 より短かく,閉花受粉に有利と思われる形態を備えていた. 以上のように,'巨峰'と'ピオーネ'における 花冠不離脱花の発生と有種子果の着果とは逆の関係を示すことが多いが,不離脱花でも自家受粉,受精が可 能であり,両者の直接的な因果関係はきわめて少ないように思われる.

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