Anatomy of Swollen Rachis and Its Effects on Berry Growth of 'Muscat of Alexandria' Grapes

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Summary

Abnormal swelling of the rachis of 'Muscat of Alexandria' grapevines grown in a heated glasshouse was investigated. The swelling became apparent at the peduncular node or along the rachis three to four weeks after full bloom. Berry growth was inhibited thereafter with the degree of inhibition varying among clusters. Anatomical studies revealed that a mass of small parenchyma cells divided among or around the phloem tissue two to four weeks after anthesis, which stymied further development of sieve tubes. Numerous granules (0.5 - 2.0 µm in diam.) were observed in these meristematic cells under SEM.

Introduction

Abnormal swelling of the rachis of 'Muscat of Alexandria' grape (Vitis vinifera L.) cultivated in heated glasshouses had been observed on a small scale and in a limited area of Okayama prefecture since the 1970's. Presently, this abnormal swelling occurs in most heated vines of the prefecture, becoming apparent before veraison and often inhibits berry growth and/or ripening.

Arora et al. (1974) reported the occurrence of rachis swelling accompanied by cracking in 16 grape cultivars out of 87 planted in a research station in Bangalore, India. Berry quality, however, was not affected by the malady. In Japan, a similar abnormality occurred in a few vinifera cultivars such as 'Pizzutello Bianco' and 'Rish Baba' on rare occasions and the berry growth was not inhibited.

This paper reports the time of occurrence of rachis swelling and its anatomical development in 'Muscat of Alexandria' grapes.

Materials and Methods

Three mature vines of 'Muscat of Alexandria' showing severe rachis swelling in recent years and growing in commercial glasshouses were selected for this study. Located in Mitsu-cho in Okayama Prefecture, the glasshouses were heated for forcing from January or February every year. A vine, on which such abnormality had not been observed in a non-heated glasshouse at Okayama University was selected as a control vine. All vines were on Hybrid Franc rootstock, trained to a multi-cordon system, and spur pruned. They showed normal vigor and carried a normal crop level.

Twenty to forty clusters per vine were tagged at anthesis. The diameters of both the peduncular node and the rachis, mid-way between the peduncular node and the first branch of the cluster, were measured at 1- to 2-week intervals. Rachises at various stages of the swelling were sampled for anatomical studies beginning three weeks before anthesis and ending at veraison. The nodal part of the samples were fixed with FAA and dehydrated with EtOH-BuOH series and embedded in paraffin blocks. Cross sections, 14 µm thick, of the rachis were stained with basic fuchsin and alcian blue to follow the development of the xylem, phloem, and cortical tissues. The size of both sieve tubes and vessels was measured from photomicrographs using a planimeter. Samples obtained from another peduncle were fixed with 1% glutaraldehyde solution and dehydrated with EtOH-BuOH series and embedded in paraffin blocks. Cross sections, 14 µm thick, of the rachis were stained with basic fuchsin and alcian blue to follow the development of the xylem, phloem, and cortical tissues. The size of both sieve tubes and vessels was measured from photomicrographs using a planimeter. Samples obtained from another peduncle were fixed with 1% glutaraldehyde solution and dehydrated with acetone series and isoamyl acetate, dried in a critical point dryer, and coated with platinum in a vacuum evaporator. The fine structure of the abnormal cells were photographed under SEM.
Results

1. Occurrence of rachis swelling

Two types of rachis swelling were found; 1) swelling only at the node and 2) swelling along the rachis (Fig. 1). Abnormal rachis swelling began three or four weeks after full bloom and continued until about veraison in both types of rachis swelling (Fig. 2). Some rachises cracked longitudinally at the position of the node. Rachis diameter in normal bunches, on the other hand, increased gradually until the berries ripened. The rate of occurrence and the type of rachis swelling varied among vines (Table 1), but both types caused severe inhibition of berry enlargement, ranging from about 4 to 24% of the clusters. Rachises on the unheated control vine did not swell.

2. Berry growth and quality

Berry weight and TSS content at harvest were lower on the clusters with a swollen rachis than those with a normal one. Acid content of berries on swollen and normal rachises were not significantly different (Table 2).

3. Anatomy of the swollen rachis

Abnormal cell division started two weeks after anthesis in the middle part of the immature phloem, resulting in the failure of sieve tubes to develop (Fig. 3). The development of the phloem was completed three weeks after blooming. When the abnormal cell division continued after the completion of the phloem development, abnormally dividing cells were compacted between the phloem and cortex so that the phloem was compressed inwardly. Four weeks after anthesis, a mass of rectangular cells were found among and around the phloem tissue of swollen rachises (Fig. 4-C). The abnormal cells were parenchymatous and resembled the shape of phloem ray cells. The phloem cells remaining near the cambium were compressed (Fig. 4-D), whereas those which differentiated earlier seemed to press outwardly against the cortex; they had scattered and were crushed. The vessels in the xylem of swollen rachises appeared normal.

Examination of the cross sections of swollen and normal rachises at veraison revealed that the phloem of the former had fewer cells and larger...
sieve tubes, compared to those of the latter (Table 3). Concurrent to the swelling, berry growth was inhibited. There was no difference in the number and diameters of vessels between swollen and normal rachises.

4. Fine structure of abnormal cells under SEM

Round or oval granules (0.1~2 μm in diameter) were abundant in the abnormal cells and cortical cells of swollen rachises when observed under SEM (Fig. 5). Similar but fewer granules formed in the cortical cells adjacent to the phloem in normal rachises. About 40~60% (avg. 51.0%) of the cortical cells in swollen rachises contained granules, whereas 32.4% of the cells in normal rachises formed them.

Discussion

The effect of abnormal rachis swelling on berry growth varied among clusters. Berries grew normally in most of the clusters with swollen rachises, but some of them failed to ripen fully. The rachis swelling observed in this study was due to the abnormal cell division among and around immature phloem. Such cell division causes...
an anomalous development of phloem tissue with fewer cells and compressed sieve tubes. Thus, the retarded berry growth and delayed ripening are attributed to the inhibition of phloem transport of photosynthates.

The fewer sieve tubes which differentiated in the swollen rachis on which the berry growth stopped indicates that the time of occurrence of the abnormal cell division is critical for further berries development. We suppose that the berries would continue to grow if the abnormal cell division occurs three or four weeks after anthesis. Nii (1979, 1980a, 1980b) and Nii and Coombe (1983) have conducted anatomical studies about the de-

Fig. 4. Transverse sections of normal (A, ×20; B, ×200) and swollen (C, ×20; D, ×200) rachises 4 weeks after full bloom. In a swollen rachis, mass of parenchyma cells are being squeezed between the cortical and cambial cells. Phloem cells are compressed. AbC, abnormal cells; Cam, cambium; Cor, cortex; Ph, phloem; PhF, phloem fiber; PhR, phloem ray; Xy, xylem.
development of vascular system in the fruit and fruit stalk of peaches, Japanese persimmons, pears and grapes. In their studies, they noted that vascular bundles in the pedicel of grape (cv. ‘Grenache’) developed rapidly during the first few weeks, which is consistent with our observations.

It is known generally that Agrobacterium tumefaciens causes an abnormal cell division in plant bodies by causing the DNA of the host cells to produce auxins and cytokinins (Chilton et al., 1982; Draper et al., 1988). Parasitism of this bacterium on grapevines is well known (Winkler et al., 1974). Recently, Goodmann et al. (1987, 1993) reported the effect of rootstock on the development of root galls in grapevine. They also reported the occurrence of aerial galls on grape cane, but not for rachis or fruit bunches. No relationship was found between the degree of rachis swelling and location of glasshouse, vine age and vigor, and temperature around clusters. These facts seem to suggest the cause of this abnormality may not be an environmental or physiological condition of the vine. To test this idea, isolation of micro-organisms from swollen rachis would be indispensable.

Small granules found under SEM might have some association with the abnormal cell division because the population density seems to correspond with the degree of swelling. Nakano (1987) found small round granules (0.1–1 µm in diameter) in the berry and receptable of ‘Muscat of Alexandria’ grapes which showed ‘sun-scald’ disorder. He did not identify the granules but suggested the possibility of MLO (mycoplasma-like organisms) because the tetracycline treatment decreased both their population and the occurrence of the disorder. Nii and Coombe (1983), on the other hand, reported the presence of membraneless globules, presumed to be phenolic compounds, in grape pedicels, especially in the cells of cortex and vascular region. Further observations under TEM and biochemical/histochemical analyses should be done for possible identification of the granules observed in this study.

**Table 3. Development of phloem and xylem in swollen and normal rachis of ‘Muscat of Alexandria’ grape.**

<table>
<thead>
<tr>
<th>Types of rachis and berry growth</th>
<th>Phloem</th>
<th>Xylem</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No. of cells/phloem</td>
<td>Avg. area of a sieve tube (µm²)</td>
</tr>
<tr>
<td>Swollen</td>
<td></td>
<td></td>
</tr>
<tr>
<td>-Stopped</td>
<td>125.5 a</td>
<td>125.0 a</td>
</tr>
<tr>
<td>-Continued</td>
<td>228.6 b</td>
<td>117.3 a</td>
</tr>
<tr>
<td>Normal</td>
<td></td>
<td></td>
</tr>
<tr>
<td>-Continued</td>
<td>248.2 b</td>
<td>163.8 b</td>
</tr>
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* Duncan’s multiple range test (P<0.05).

![Fig. 5. Electron micrograph of granules in an abnormal cell of a swollen rachis of ‘Muscat of Alexandria’ grape.](image)

**Literature Cited**