OBSERVATIONS ON THE GERMINATION AND STORAGE OF TEA POLLEN AND SEED
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1. Introduction

With a view to future breeding work which might be undertaken it seemed desirable to obtain some more information about the germination and storage properties of tea pollen and seed.

With regard to pollen, WellesNiek noted that it can be germinated in water, but little is further known about it. With respect to seed, studies have been made in the past regarding the thickness of the shell (Gadd) and the effect of its removal on germination (Turns). In recent years some practical information has become available concerning the cold storage of seed (Hume, Noves).

In order to add to the present knowledge a few experiments have been carried out on germination and conditions for storage.

2. Pollen germination and storage

The germination of the pollen was tested in different sucrose concentrations by means of the “hanging-drop” culture. It was found that 3 to 4 hours at room temperature (about 70°F.) sufficed to obtain maximal germination in any of the media tested. The average germination percentages (6 replicates) obtained in 0, 5, 10, 15, 20 and 25% sugar solutions were 83, 87, 87, 88, 83 and 75%, respectively. These figures show that tea pollen germinates equally well in water as in sugar solutions up to 25%.

Pollen (including the stamens) collected from just opened flowers was subsequently stored in desiccators at a range of relative humidities. The latter were obtained with different concentrations of sulphuric acid. The desiccators were kept in a refrigerator at about 32°F. The viability of the pollen thus stored was tested at regular intervals by determining its germination percentage in 10% sugar solution (in triplicate). The results are presented in figure 1.

![Figure 1. The viability of tea pollen in relation to relative humidity when stored at 32°F. Germination percentage.](image-url)
It appears from figure 1 that tea pollen can be optimally stored at 40% relative humidity; the germination percentage was still above 60% three months after storage. Unfortunately, the experiment had to be discontinued because of an accident with the desiccators. However, past experience suggests that the pollen might have been kept viable for some time longer, but not for a whole year. In order to prolong the longevity for one year or more, the pollen would have to be stored at deep freeze temperatures (Visser).

3. Seed germination

It has been found with many seed species that removal of the seed coat or damaging it, accelerates the germination of the embryo. This might be due to the greater or lesser imperviousness of the seed coat for water and air, or its mechanical resistance (Tuber, Visser). As a complement to Tuber's studies it was further investigated what role the shell plays in germination with regard to the effect of pre-soaking of the seed and the cracking of the shell (by a light hammer blow).

I. In the first experiment seeds of which the shell was removed, cracked or left intact were "soaked" in water for 1, 2, 3, 5 and 7 days respectively, and subsequently germinated with the shell removed (embryo), cracked or intact. Accordingly, the tests were carried out with 6 groups of seeds:

a. Shell removed while soaked; germination with shell removed
b. " cracked " removed
c. " intact " removed
d. " cracked " cracked
e. " intact " cracked
f. " intact " intact

The seeds, were germinated in boxes with peat at room temperature. Progress of germination was checked at daily intervals; seeds showing a protruding radicle were taken as having germinated. The test was carried out with "fresh" seeds which had been kept in the room during a fortnight after collection in the tea seed garden. The results of soaking on water uptake by the seeds and subsequent germination are given in tables 1 and 2.

Table 1. Relative weight of fresh seeds submerged in water for different periods expressed in % of weight of seeds 13 days in water; figures based on 2 x 100 seeds/treatment.

<table>
<thead>
<tr>
<th>Days in water</th>
<th>0</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>5</th>
<th>7</th>
<th>10</th>
<th>13</th>
</tr>
</thead>
<tbody>
<tr>
<td>Relative weight in %</td>
<td>71.0</td>
<td>81.0</td>
<td>87.4</td>
<td>91.6</td>
<td>98.0</td>
<td>99.0</td>
<td>99.7</td>
<td>100</td>
</tr>
<tr>
<td>% Sinkers</td>
<td>0</td>
<td>33</td>
<td>78</td>
<td>91</td>
<td>98</td>
<td>100</td>
<td>100</td>
<td>100</td>
</tr>
</tbody>
</table>
Table 2. Number of days required to obtain 75% germination of “fresh” seeds submerged in water for different times and subsequently germinated with shell intact, cracked or removed; figures based on $2 \times 50$ seeds/treatment.

<table>
<thead>
<tr>
<th>Soaked with shell</th>
<th>Germination with shell</th>
<th>No. days needed for 75% germination after in water for 0 days</th>
<th>1 day</th>
<th>2 days</th>
<th>3 days</th>
<th>5 days</th>
<th>7 days</th>
</tr>
</thead>
<tbody>
<tr>
<td>a. Removed</td>
<td>Removed</td>
<td>11.0</td>
<td>9.5</td>
<td>10.5</td>
<td>10.5</td>
<td>10.0</td>
<td>12.0</td>
</tr>
<tr>
<td>b. Cracked</td>
<td>Removed</td>
<td>11.0</td>
<td>11.0</td>
<td>9.5</td>
<td>9.0</td>
<td>9.0</td>
<td>12.5</td>
</tr>
<tr>
<td>c. Intact</td>
<td>Removed</td>
<td>11.0</td>
<td>10.5</td>
<td>9.5</td>
<td>9.0</td>
<td>9.0</td>
<td>7.5</td>
</tr>
<tr>
<td>d. Cracked</td>
<td>Cracked</td>
<td>36.5</td>
<td>36.0</td>
<td>29.5</td>
<td>27.0</td>
<td>25.0</td>
<td>36.5</td>
</tr>
<tr>
<td>e. Intact</td>
<td>Cracked</td>
<td>34.5</td>
<td>28.0</td>
<td>25.0</td>
<td>22.5</td>
<td>20.0</td>
<td>33.0</td>
</tr>
<tr>
<td>f. Intact</td>
<td>Intact</td>
<td>49.0</td>
<td>50.5</td>
<td>47.0</td>
<td>41.0</td>
<td>41.5</td>
<td>55.5</td>
</tr>
</tbody>
</table>

Table 1 shows that the seeds took up most of their water during the first 5-7 days of soaking, only a little more being taken up after that. That the seeds were nearly saturated at that time is also evident from the fact that all seeds had become “sinkers.”

With regard to the influence of pre-soaking on germination it follows from table 2 that the quickest germination was obtained after 5 days of soaking (column 7). Immersion for 7 days (column 8) seemed to have had an adverse effect, as germination of such seeds was considerably delayed as compared with the former, especially for cracked and intact seeds (d, e, f). The removal or cracking of the seed shell prior to soaking gave slightly less results than soaking the seeds intact (compare a with c, d with e).

It is also apparent from table 2 that cracking of the shell and even more so its entire removal greatly accelerated the subsequent germination (compare c, e, f). It can be noted that the germination of the naked embryos (a, b, e) is little affected by pre-soaking. This is probably due to the fact that once the seed coat is removed, the rate of water uptake, respiration and mechanical resistance are no longer limiting factors.

These observations are confirmed by the experiment of Tubbs who found that the removal of the seed shell is accompanied by an increased water uptake and consumption of nitrogenous and fatty substances, indicating the increased physiological activity (respiration) of the embryo. Likewise, the favourable effect of cracking the shell beforehand can be attributed to a decreased mechanical resistance and increased respiration, as has also been shown to be the case in apple seed (Visser).

II A second experiment on the effect of soaking and subsequent cracking of the shells was carried out with seeds which had been stored for 3 months at room temperature (about 70°F). The intact seeds were soaked for 0, 2, 4, 6 and 9 days and then germinated intact, or with a cracked shell. In contrast with the former experiment the seeds were germinated out of doors in beds made up of river sand and covered by jute hessian.
Table 3. The effect of soaking in water (seeds intact) and cracking of the seed shell on the germination of 3 months old seeds (100 seeds/treatment).

<table>
<thead>
<tr>
<th>No. days soaked</th>
<th>% Germination Obtained after 30 days</th>
<th>Germination Obtained after 50 days</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Intact</td>
<td>Cracked</td>
</tr>
<tr>
<td>0</td>
<td>33</td>
<td>37</td>
</tr>
<tr>
<td>2</td>
<td>29</td>
<td>32</td>
</tr>
<tr>
<td>4</td>
<td>42</td>
<td>46</td>
</tr>
<tr>
<td>6</td>
<td>50</td>
<td>39</td>
</tr>
<tr>
<td>9</td>
<td>31</td>
<td>14</td>
</tr>
</tbody>
</table>

N.B. The germination recorded after 50 days is the final germination percentage reached; the other seeds had succumbed to rotting by that time.

It appears from the results, presented in table 3, that pre-soaking up to 6 days appreciably accelerated the germination of the intact seeds (column 2), less so that of the cracked seeds (column 3). Soaking of the seeds for more than 6 days adversely affected the germination, particularly that of cracked seeds. It made little difference to the germination (after 30 days) whether the seeds were germinated intact or cracked (compare columns 2,3). From the assessment after 50 days it can be derived (columns 4, 5) that soaking has little effect on the final germination of the seeds unless they are soaked too long. The eventual germination of cracked seeds, except for the not soaked ones, is less than that of the intact seeds. This is probably due to the fact that a greater number of the former seeds rotted as the cracked shell afforded less protection against rotting organisms.

It appears from the two germination tests that the cracking of seed coats, had little or no effect on the “old” seeds (kept for 3 months), but greatly speeded up the germination of the “fresh” seeds (kept for 2 weeks) especially if they were also pre-soaked.

This difference in response is probably largely explainable on account of the different physiological conditions of the respective seeds. Pre-soaking and cracking of the seeds would sooner realise the condition required for the initiation of germination, such as the provision of the embryo with enough moisture and the lessening of mechanical hindrances. The vitality of the fresh seeds is such that the embryo can be presumed to react as soon as these conditions are fulfilled. The older seeds, however, have lost much of their vigour and their metabolism is accordingly retarded. Consequently, these embryos are not likely to profit from the enhanced water uptake and “air flow” by cracking the shell, as it would not be matched by an equivalent increase of physiological activity, hence the difference in response between old and fresh seed.

It can be remarked that tests with embryos (shell removed) enabled a quick assessment of the germination capacity of the seed, provided the seeds were comparatively “fresh”. A prolonged storage of the seeds was found to increase the percentage of embryos which succumbed to rotting in the germination medium.

It is worthy of note that the shells can be easily cracked by putting the wet seeds in hot sunshine. It was found that between 50 and 95 per cent of the seed shells cracked if spread for only 15 minutes in the sun.

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4. Seed storage

Freshly collected seed (2 weeks dry at room temperature) was stored in desiccators at the same relative humidities and temperature as used for the pollen. Twenty seeds were removed from storage at regular intervals and germinated at room temperature in peat. The seeds were soaked for 2 days and their shells cracked prior to the germination test.

![Figure 2. The viability of tea seed in relation to the relative humidity when stored at 32°F. Germination percentage:](image)

% Relative humidity

It can be derived from the results, as presented in figure 2, that the viability of the tea seeds was maintained better as the relative humidity was higher. This is rather unusual because the great majority of seed species optimally store at humidities between 10 and 50 per cent. The contrasting findings with tea seed are probably partly explained by the fact that storage of the seeds at lower humidities dries out the embryo to such an extent that it takes a long time before enough water is taken up to allow germination. During that time lag, the cracked seeds are liable to become rotten in the germination medium.

Our findings substantiate those of Hume and Noyes who stored seed in wooden boxes at 40°F. The former obtained more than 90 per cent, the latter nearly 70 per cent germination, 6 months after storage. It would, therefore, seem that no special precautions are needed for storage of seeds at temperatures just above freezing and under conditions which allow aeration. Naturally the seeds should be reasonably dry. On the other hand, if seeds are stored in sealed containers and/or below freezing point, it is necessary to dry the seeds thoroughly beforehand in order to prevent damage due to asphyxiation or freezing. It is likely that the failure of the seeds to germinate after storage in sealed tins at 20°F in Hume's experiment is due to the fact that the seeds were not sufficiently dry. Results with other seed species (Weibull) suggest that also tea seed might profitably be stored at deep freeze temperatures if not packed airtight and reasonably dry, or thoroughly dried if packed in sealed containers.

5. Summary and conclusions

Experiments were carried out with regard to the germination and storage of tea pollen and tea seed. The following observations were made:

1. Tea pollen germinated well both in water and in sugar solutions between 5 and 25 %. 

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2. Pollen stored at relative humidities between 0 and 100% and at 32°F optimally maintained its viability at 40% humidity. At this humidity the pollen still showed nearly 70% germination 3 months after storage. Storage at temperatures below freezing point is likely to maintain viability for longer than one year.

3. Pre-soaking of the seeds in water for 3 to 5 days and subsequent cracking or removal of their shell markedly speeded up the germination of comparatively fresh seeds. Older seeds were similarly affected by pre-soaking, but the cracking of the seed shell had little effect on the germination. Accordingly, if in practice an accelerated germination is desirable, pre-soaking and cracking of the seeds (in the sun after soaking) can be resorted to. Seed growers who want to evaluate quickly the germination capacity of freshly-harvested seeds can do so by carrying out tests with seeds of which the shell is completely removed.

4. Seeds stored at the same humidities and temperature as the pollen appeared to give over 50% germination 10 months after storage at 100% relative humidity. Lower humidities gave less good results presumably due to subsequent infection by rotting organisms in the germination medium before the seeds are able to germinate. It is probable that storage at deep freeze temperatures would prolong the longevity of the seed if packed pre-dried.

References