

STUDIES ON POLLEN. III.

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Since the appearance of my second contribution on the study of pollen I have continued my investigation on this subject and have added a considerable number of plants to those already studied in ways mentioned in my previous papers. As the work has progressed certain new methods have suggested themselves as expedient or in many cases as necessary for the proper advance of the work. One former method of allowing pollen to be placed for germination in the desired solution between a slide and cover-glass is, while often giving results, unfair since certain life processes can not normally be carried out under these conditions. The use of an ordinary glass ring cell cemented to a slide and having the pollen for investigation in a hanging drop also served in many cases but allowed of no regulation of temperature by an exchange of air in the ways desired. A common gas chamber served the purpose better and obviated the difficulty just mentioned and also allowed of certain other forms of experimentation, not possible with the glass ring cell, that I hope to investigate later on. In this last type of culture cell, as here used, it was necessary to use wet filter paper in the cell or a small quantity of water to prevent the specimen from drying up. All the culture cells mentioned thus far had the disadvantage of allowing only one experiment or culture to be so arranged at one time. To offset this the glass ring cell was used in large numbers but individual cultures made in this fashion require a great deal of extra work and are not conducive either to convenience or accuracy. In some cases at first when an extra large number of cultures were to be made I supplemented the glass culture ring apparatus by cells made of filter paper and kept moist in damp air under a bell jar. These latter, however, were much less satisfactory for various evident reasons.

• Since my second contribution on this subject I have increased the number of plants whose pollen I have studied from 435 to 540. In the case of all of these 540 plants I have tried to grow the pollen in the following solutions: distilled water and cane sugar of 1%, 5%, 10%, 15%, 20%, 30%, 40%, 50% and 60%. A great many, of course, did not grow but many did. The pollen of all of these 540 plants were experimented with in this way and given a trial even though, as is well known, some few of them have not yielded results in this way. Of these 540 phanerogams only the pollen of five produced more than one tube on germination. This is very different from some pollen which produces many tubes on germination as in the case of *Malva crispa*. Of all the 540 plants I found only one whose pollen tube branched. That was the pollen of *Caladium bicolor* one of whose pollen tubes branched three times and two of whose pollen tubes branched twice. The record of this plant is as follows, viz: In distilled water ten pollen grains in one hundred germinated. In the cane sugar solutions 21 germinated in the 1%; 4 in 5%; 0 in 10%; 3 in 15%; 27 in 20%; 15 in 30%; 3 in 40%; 0 in 50%, and 0 in 60%. The pollen in this experiment was all of the same age and grown under exactly the same conditions. The first point of interest, therefore, is that in 10% none grew while just above and

below this strength of the sugar solution the pollen grew. The second point of interest is that the branching of the pollen tube which this plant showed occurred only in the 30% of cane sugar and as above noted. It will be seen that in 40% cane sugar the germination dropped off greatly as only 3 grains of pollen grew. It has been known for a long time that the pollen tube sometimes branches as it descends through the stigma of some plants. The germination of the pollen of this plant was rather slow as it required one-half an hour to begin. Its pollen lived in the ungerminated condition for eight days and it grew best in a 20% solution of the cane sugar. Most of the pollen experimented with in the cultures used in this investigation was from plants gathered in Monroe County, Indiana. Some of the plants were obtained from widely separated localities and wherever and whenever an opportunity presented itself. Some came, for example, from Texas, some from Missouri, and others from Colorado which I gathered on Pike's Peak and vicinity. Twelve plants from Pike's Peak that I tested especially as regards their longevity, lived only ten days in the longest case. Some of these I did not expect would germinate under any conditions, whether their pollen was young or old, and this was fully verified by experiment. However, these plants were in every case also subjected to the usual investigation in order to leave no doubt on the question. Still others of the plants came from Dearborn County and from Clark County, Indiana. All of the experiments in this paper were carried out, as above stated, in pure distilled water and in the different strengths of cane sugar mentioned. The distilled water and cane sugar were as pure as possible. No experiments were performed or attention given to the question of additional substances, such as gelatine, added to these solutions. Considerable work also has been done in some phases of this field. It constitutes, however, a problem in itself and merits much more extended investigation than it has yet received.