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Studies on the Functional Significance of Pollen Surface Characters[1)]

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花粉表面形質의 機能的 意義에 關한 研究

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ABSTRACT

From 32 angiosperm species, pollen surface characters such as sculpture pattern, roughness, oil abundance and stickiness were subjectively scored, and compared with size and weight of the grains, and pollination mechanisms.

There was a linear relationship between size and weight of the grains. Animal-pollinated grains showed significantly higher roughness, oil abundance and stickiness than wind-pollinated grains. In the former, the grain size was highly correlated with roughness and oil abundance. No such relationship was found in the latter.

It was suggested, therefore, that the larger the animal-pollinated grains, the greater is the stickiness required, where the stickiness depends on the surface oil abundance and roughness. No functional significance of the sculpture pattern could not be detected in this study.

INTRODUCTION

Exine has been known to function in clumping of the grains (Stebbins, 1974) and adhesion to vector (Wodehouse, 1935; Walker, 1971) owing to the bound sticky materials (Walker, 1971; Echlin, 1973), prevention of cell collapse (Payne, 1972), and storing incompatibility substances (Heslop-Harrison, 1973; 1976).

Although no cumulative datum has been provided, it is generally accepted that elaboration of the exine is characteristically found in animal-pollinated pollen (Davis and Heywood, 1963; Whitehead, 1969). The sculpturing of the exine is so variable that it has been used as one of the most important pollen characters (Faegri and Iversen, 1964; Walker, 1971; Taylor and Levin, 1975). However, few studies have been made to elucidate what the functional significances of different sculpture patterns are. From the Polemoniaceae in which the diversity of surface sculpturing is striking, it was failed to find any significant relationships between the ornamentation and either the environmental factors or pollination mechanisms (Taylor and Levin, 1975). Echlin (1968) emphasized the role of heridity in playing a part in determining the complex surface patterns of pollen grains. This study was initiated to substantiate

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he functional role of sculpture patterns if possible as well as other pollen surface characters.

MATERIALS AND METHODS

Flowers were collected on the Duke University campus and the greenhouse of the Botany Department during the spring and summer of 1976. The materials, collection dates, and places are listed as follows:

Anacardium occidentale DC. (Anacardiaceae) 5/20 Greenhouse

Antirrhinum majus L. (Scrophulariaceae) 5/19 Greenhouse

Apocynum androsaemifolium L. (Apocynaceae) 5/20 Campus

Bougainvillea buttiana Holttum & Standl. (Nyctaginaceae) 5/18 Greenhouse

Chenopodium album L. (Chenopodiaceae) 4/7 Greenhouse

Cornus florida L. (Cornaceae) 4/2 Campus

Cucurbita pepo L. (Cucurbitaceae) 6/2 Greenhouse

Cuphea micropetala Baill. (Lythraceae) 7/17 Greenhonse

Daucus carota L. (Umbelliferae) 6/15 Campus

Eichhornia crassipes Solms. (Pontederiaceae) 5/18 Greenhouse

Epilobium angustifolium L. (Onagraceae) 7/21 Greenhouse

Fuchsia sp. (Onagraceae) 3/31 Greenhouse

Hibiscus rosa-sinensis L. (Malvaceae) 3/23 Greenhouse

Hippeastrum leopoldii Leop. & Hook. (Amaryllidaceae) 3/25 Greenhouse

Lilium longiflorum Thunb. (Liliaceae) 4/6 Greenhouse

Liquidambar styraciflua L. (Hamamelidaceae) 3/26 Campus

Lobelia erinus L. (Campanulaceae) 5/17 Greenhouse

Lonicera japonica Thunb. (Caprifoliaceae) 6/16 Campus

Magnolia grandiflora L. (Magnoliaceae) 6/9 Campus

Modiola caroliana (L.) G. Don. (Malvaceae) 6/1 Campus

Nicotiana tabacum L. (Solanaceae) 4/12 Greenhouse

Plantago lanceolata L. (Plantaginaceae) 4/8 Greenhouse

Quercus falcata Michx. (Fagaceae) 4/13 Campus

Q. phellos L. (Fagaceae) 4/6 Campus

Ruellia amoena Nees (Acanthaceae) 4/1 Greenhouse

Salix nigra Marshall (Salicaceae) 4/2 Campus

Sorghum vulgare Pers. (Gramineae) 6/2 Greenhouse

Thalictrum polyganum Muhl. (Ranunculaceae) 4/28 Greenhouse

Tillandsia baileyii Rose (Bromeliaceae) 3/30 Greenhouse

Vicia faba L. (Leguminosae) 5/19 Greenhouse

Zea mays L. (Gramineae) 6/7 Greenhouse

Zephyranthes grandiflora Nichols. (Amaryllidaceae) 6/2 Greenhouse

The materials were carried into the laboratory in a Petri dish with wet filter paper. The mature anthers were opened with tweezers under a dissecting microscope. In semi-controlled room conditions, i.e. temperature 24°C and humidity 50−62%, the following measurements were made.

Stickiness of surface was subjectively determined by examining the degree of attachment of the pollen grains to the eyelash brush, which was made by inserting a single eyelash into the tip of a tapered glass tube. After the anther was opened under a dissecting microscope, the eyelash brush was gently manipulated to collect as many pollen grains as possible. Some very sticky grains (Stickiness Index, SI=5) were collected easily and a large cluster was formed, while only a few non-sticky grains (SI=1) were collected and a cluster was never formed. The medium sticky grains (SI=3) did not form a cluster, yet covered the surface of the eyelash in a single coat.

Under a compound microscope, abundance of oil droplets was also subjectively recorded by examining the pollen grains in the 8% sugar medium (Dyer, 1966). The oil abundance indices (OI) 1 to 5 were assigned to the range of oiliness, 1 when

Table 1. Selected pollen characters of various angiosperm taxa

Pollen Types and Taxa	Grain length x width (μm)	Grain diameter (μm)	Grain shape (W/L)	Sculpture pattern	Roughness Index	Oil Abundance	Stickiness Index	Pollination Vector
Monocolpate								
Eichhornia	32×82	43.8	.39	scabrate	3	3	5	animal
Hippeastrum	40×116	57.0	.35	reticulate	3	4	5	animal
Lilium	56×130	74.1	.39	reticulate	4	5	5	animal
Tillandsia	30×84	42.3	.36	reticulate	2	4	5	animal
Zephyranthes	38×93	51.2	.41	reticulate	2	4	5	animal
Tricolpate								
Anacardium	68×85	73.3	.80	baculate	3	5	5	animal
Antirrhinum	16×30	19.7	.52	scabrate	2	4	5	animal
Bougainvillea	17×28	20.1	.49	reticulate	4	4	5	animal
Lobelia	21×40	26.0	.49	psilate	2	2	4	animal
Magnolia	39×94	52.3	.42	reticulate	2	3	4	animal
Tricolporate								
Cornus	26×47	31.7	.54	verrucate	2	1	3	animal
Cuphea	22×15	19.4	1.47	striate	2	2	3	animal
Daucus	14×33	18.6	.42	scabrate	1	1	4	animal
Nicotiana	22×43	27.5	.51	striate	2	1	5	animal
Quercus phellos	21×39	25.8	.54	scabrate	3	1	1	wind
Q. falcata	22×37	26.2	.59	scabrate	1	1	1	wind
Salix	13×25	16.2	.50	reticulate	2	2	1	animal
Vicia	26×53	33.0	.49	scabrate	2	2	5	animal
Monoporate								
Sorghum	46×46	46.0	1.00	scabrate	1	2	4	wind
Zea	85×85	85.0	1.00	scabrate	2	1	2	wind
Triporate								
Epilobium	—	—	—	—	3	—	5	animal
Fuchsia	72×35	56.6	2.05	striate	3	1	3	animal
Lonicera	55×32	45.9	1.72	echinate	3	3	5	animal
Ruellia	54×54	54.0	1.00	reticulate	3	3	3	animal
Periporate								
Chenopodium	31×31	31.0	1.00	scabrate	2	1	2	wind
Cucurbita	145×145	145.0	1.00	echinate	4	5	5	animal
Hibiscus	156×156	156.0	1.00	echinate	5	5	5	animal
Liquidambar	32×32	32.0	1.00	scabrate	2	1	1	wind
Modiola	41×48	43.2	.85	echinate	4	2	5	animal
Plantago	28×28	28.0	1.00	scabrate	2	1	2	wind
Thalictrum	22×25	23.0	.85	verrucate	2	1	3	animal
Tetrad								
Apocynum	32×32	32.0	1.00	scabrate	2	1	5	animal

no oil droplets were detected to 5 when the whole surface of the pollen grains was covered by the droplets, either in the droplet form or in the fashion of a thick coating.

Roughness of surface pattern was scored by subjective judgement on the smoothest (Roughness Index, RI=1) to roughest (RI=5) scale.

The smoothest grains were those from most of wind-pollinated plants, while the roughest ones were the conspicuously echinate pollen of some animal-pollinated malvaceous species (Tab. 1).

The weight of dry pollen grains was measured with a quartz-fiber microbalance which was handmade as described in Lowry and Passonneau (1972). Here the term "dry" was used in the sense of "air-dry" (Payne, 1972) such that the grains would contain some amount of water. Such air-dried grains were embedded in the immersion oil and measured (Payne, 1972). The adjusted diameter was calculated in the conventionally adopted formula: $d'=2(a^2\times b)^{1/3}$ where a is the radius of equatorial plane and b is a half of the polar axis length.

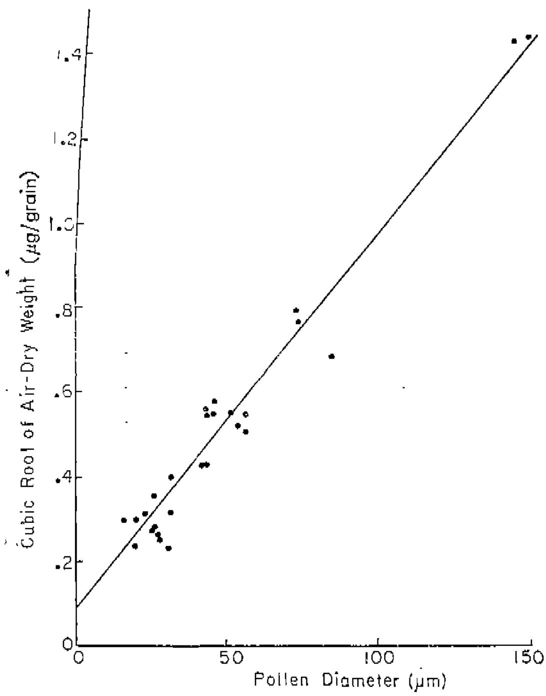

Fig. 1. Pollen weight as a function of size. The cubic root of the dry weight is linearly correlated with pollen diameter with a regression equation $Y=0.101+0.009X$ ($P<0.001$).

RESULTS

The measurements of surface and other selected characters are summarized in Table 1. From the obtained data, there was found a linear relationship between the pollen diameter and the cubic root of the weight. Not suprisingly, the data confirmed that the larger the grains, the higher was the weight attained (Fig. 1).

Table 2 shows that all the surface characters measured in animal- and wind-pollinated pollen grains are significantly different, i.e. the former is stickier, rougher and oilier than the latter. The statistical significance was greatest in the difference of stickiness.

Table 2. t-Tests of surface characters of animal vs. wind-pollinated pollen

vectors / characters	Animal	Wind	t-Test
Roughness	2.72±0.936	1.86±0.690	$P<0.05$
Oil abundance	2.70±1.444	1.14±0.378	$P<0.01$
Stickiness	4.32±1.069	1.86±1.069	$P<0.001$

Table 3 shows that the grain size of animal-pollinated pollen is more highly correlated with roughness and oil abundance than with stickiness. This result reveals that the larger the animal-pollinated grains, the higher are the roughness and oil abundance indices. Such correlations were not obtained in the wind-borne pollen. The grain size is not highly correlated with the stickiness in the former as well as in the latter.

Table 3. Correlation coefficients between the grain diameter and surface characters of animal- and wind-pollinated pollen

vectors / characters	Animal	Wind
Roughness	.721	—.082
Oil abundance	.650	.142
Stickiness	.329	.344

Table 4. Correlation coefficient matrix between pollen surface characters

	Sculpture	Roughness	Oil drops	Stickiness
Sculpture	—	.381	.430	.167
Roughness	.381	—	.624	.417
Oil drops	.430	.624	—	.649
Stickiness	.167	.417	.649	—

In the animal-pollinated pollen, the fact that the correlation coefficient between size and stickiness is low but the stickiness is high, suggests that no matter how large the pollen grains are, the grains are sticky enough to clump each other and to adhere the brush owing to the highly rough and/or oily nature of the pollen surface. On the other hand, the low indices and weak correlations in the wind-borne pollen characters suggest that for the better dispersal, they should not be highly correlated with the surface characters.

The relationships among sculpture pattern and other surface characters were summarized in the matrix of correlation coefficents between the variables (Tab. 4). In calculating the correlations, sculpture pattern was scaled in the following order: (1) psilate, scabrate, verrucate, echinate and baculate, (3) striate, and (5) reticulate. It was showed that the higher coefficients are between stickiness and oil abundance(.65), and between roughness and oil abundance (.62). The low coefficients were between sculpture pattern and oil abundance (.43), between roughness and stickiness (.42), and between sculpture pattern and roughness (.38). The weakest correlation appears between stickiness and sculpture pattern (.17). This result suggests that stickiness of pollen grains is more highly dependent on the abundance of oil than on roughness of the surface; sculpture pattern is not an important character for the adhering function.

Combining the high correlation between grain size and the oil abundance as well as roughness (Tab. 3), with the two highest correlations among the surface characters, i.e. between stickiness and oil abundance, and between roughness and oil abundance (Tab. 4), a self-evident suggestion was obtained. Larger pollen grains require greater contactability, where the contactability depends on the surface oil abundance which tends to be determined by surface roughness.

DISCUSSION

The relationships between the pollen surface characters suggests that among the characters roughness would be functionally the most important. The roughness itself would be closely related to the ability of pollen to contact the pollinator. This role would be played more efficiently owing to its ability to hold the sticky oil substances. It is not suprising to observe that higher roughness index and more abundant oil substances are associated with larger animal pollinated grains.

Collection of pollen grains by animal vectors was represented by the stickiness measured in this study. Stickiness in turn would be determined mainly by roughness, oil abundance and grain size in the pollen's part. These relationships would, in the other hand, interact the flower and pollinator characters. They could be rough and sticky too. The behavior of pollinators and the spatial arrangement of the stigma or structure of the flower would be also important factors.

As far as sculpture patterns are concerned, psilate, scabrate or verrucate pattern certainly shows less stickiness or oil abundance than baculate or echinate pattern. Yet this relationship would express roughness, i.e. the former is smoother than the latter. Striate, rugulate or reticulate pattern does not show any consistancy in the stickiness or oil abundance measurements. Besides the surface characters, sculpture pattern reveals no or weak relationships with pollination mechanism, pollen size or shape. Although reticulate pattern is exclusively found in animal-pollinated species in this study, there is contradictory statistics based on the bigger sample showing that 21 species out of 85 wind-pollinated species possess echinate or reticulate pattern (Lee, 1977).

Although baculate, echinate and reticulate patterns at least would facilitate the storing of adhering substances, the assumption that these patterns were adapted for the animal pollination is yet premature. It could be proposed that the wind-pollinated species with echinate and reticulate patterns would have been somewhat recently evolved from the animal-pollinated species which had attained these characters. Certainly *Ambrosia* having echinate grains is a recent genus adapted to wind pollination (Stebbins, 1974). Taxa closely related to wind-pollinated genera such as *Liquidambar*, *Fraxinus* and *Rumex* are still pollinated by insects (Faegri and Van der Pijl, 1966). However, many other wind-pollinated genera, e.g. *Platanus*, *Calitriche*, *Trigochin*, *Potamogaton*, *Sparganium* and *Typha* would make us await for acceptance of the assumption. Conclusively no functional importance of sculpture pattern could be suggested in this study (Taylor and Levin, 1975). It is assumed that any inheriditarily determined sculpture pattern could only modify its roughness and thereby oil abundance in response to the pollinators and flowers (Echlin, 1968).

It has been understood that the surface of wind-borne pollen is smooth whereas that of animal-borne pollen rough, and the stigma of wind-pollinated flower is long and hairy whereas that of animal-pollinated flower is more or less globose and smooth (Corner, 1964; Faegri and Van der Pijl, 1966; Whitehead, 1969). One can assume that there is a complementary or lock-and-key mechanism involved between sculpture pattern and stigmatic surface or pollinator's hair. The SEM pictures showed no indication of this mechanism but rather revealed that the dimension of roughness or sculpture pattern of the pollen grains would be completely different from that of the insect hairs and stigmatic surface (Lee, 1977). Namely, if the structure of stigmatic surface and bee hairs is called microscopic, that of the pollen is ultramicroscopic.

Thus, although the surface of pollen, pollinator's hair and stigma are adapted for the same efficient pollination, their structures are not complementary. Since these characters are closely associated with the conveyance of pollen grains from anther to stigma, they might be closely related to each other to insure the maximum efficiency of pollination and consequently seed set in a biological system.

Studies with a broad array of materials and measurement of the surface characters by mechanical means would allow us to resolve the functional significance of the exine sculpturing.

摘 要

32種의 被子植物에서 花粉을 採取하여 表面의 粗度(거친 程度), 油度(기름의 量) 및 粘度(끈끈한 程度)를 測定하여 花粉粒의 크기와 무게 및 受粉方法과 比較하였다.

花粉粒의 크기와 무게 사이에는 直線的 關係가 있었다. 動物에 依해 受粉되는 花粉粒은 바람에 依한 것에 比해 粘度, 油度, 그리고 粗度가 훨씬 높았다. 前者에서 花粉粒의 크기는 粗度 및 粘度와 높은 相關關係를 보여 주었으나 後者에서는 이런 關係는 볼 수 없었다. 따라서 動物에 依해 受粉되는 花粉은 그 크기가 클수록 粘度가 커야하며 粘度는 表面의 油度와 粗度에 依해 決定된다고 생각된다. 本硏究에서 表面무늬의 機能的 意義는 發見되지 않았다.

REFERENCES

Corner, E.J.H. 1964. The Life of Plants. Weidenfeld and Nicolson, London. 325pp.

Davis, P.H. and V.H. Heywood. 1963. Principles of Angiosperm Taxonomy. Van Nostrand, N.Y. 558pp.

Dyer, A.F. 1966. Pollen tube mitosis in culture; osmotic control and acenaphthene treatment. *Stain Tech.* 4 : 277—280.

Echlin, P. 1968. Pollen. *Sci. Amer.* 218 : 80—90.

______. 1973. Pollen development and the pollen grain wall. The role of the tapetum during microsporogenesis of angiosperms. Pages 41—61, in J. Heslop-Harrison (ed.), Pollen Development and Physiology. Butterworths, London.

Faegri, K. and J. Iversen, 1964. Textbook of Pollen Analysis. Hafner Publ. Co., N.Y. 237pp.

Faegri, K. and L. Van der Pijl. 1966. Principles of Pollination Ecology. Pergamon Press, N.Y. 291pp.

Heslop-Harrison, J. 1973. The pollen wall: structure and development. Pages 75—98, in Heslop-Harrison (ed.), Pollen. Butterworths, London.

______. 1976. The adaptive significance of the exine.

Pages 27—37, in I.K. Ferguson and J. Muller (eds.). The Evolutionary Significance of the Exine. Academic Press, London.

Lee, S. 1977. Investigations of the Functional Morphology of Angiosperm Pollen. Ph. D. thesis of Duke University. 191pp.

Lowry, O.H. and J.V. Passonneau. 1972. A flexible system of enzyme analysis. Academic Press, N.Y. 291pp.

Payne, W.W. 1972. Observations of harmomegathy in pollen of Anthophyta. *Grana.* 12 : 93—98.

Stebbins, G.L. 1974. Flowering Plants. Evolution above the species level. Balknap Press, Cambridge, Mass. 399pp.

Taylor, T.B. and D.A. Levin. 1975. Pollen morphology of Polemoniaceae in relation to systematics and pollination systems: Scanning electron mcroscopy. *Grana.* 15 : 91—112.

Walker, J.W. 1971. Pollen morphology, phytogeography and phylogeny of the Annonaceae. *Contr. Gray Herb.* 202 : 1—130.

Whitehead, D.R. 1969. Wind pollination in the angiosperms: evolutionary and environmental considerations. *Evolution.* 23 : 28—35.

Wodehouse, R.P. 1935. Pollen Grains. McGraw-Hill, N.Y. 574pp.

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