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RAGWEED POLLEN DENSITY[1]

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ABSTRACT

HARRINGTON, JAMES B., JR., and KURT METZGER. (U. Michigan, Ann Arbor.) Ragweed pollen density. Amer. Jour. Bot. 50(6) : 532–539. Illus. 1963.—The density of ragweed pollen has been measured using a Beckman gas pycnometer. Commercially dried pollen of *Ambrosia artemisiifolia* in moisture equilibrium with air at 52% relative humidity had an effective density[2] of 0.84 g/cc, while the density of the solid material in the grain was 1.32 g/cc. The density of fresh ragweed pollen varies with age and relative humidity, being 1.28 g/cc at 100% humidity and 1.05 g/cc at humidities below 52%. After dehiscence, the evaporation of water from the pollen grain is extremely rapid so that adjustment to the external humidity regime is virtually instantaneous.

RAGWEED pollen, being one of the most serious aeroallergens, has been studied extensively. Along with other pollens and spores it is sampled at most of the major hospitals in the Eastern United States as well as at several research institutions. Sampling at the hospitals is undertaken using the standard Durham sampler (Durham, 1946b), a microscope slide placed horizontally in the space between 2 circular metal plates. At the research institutions sampling is carried out using a variety of instruments of the impaction, impingement, or filtration types. In all these samplers the density of the pollen plays an important role in determining collection efficiency (Harrington, Gill, and Warr, 1959). Without an accurate measure of pollen density no reliable conversion from catch to air concentration can be made.

Durham (1946a), using a vertical settling chamber, determined the density of *Ambrosia artemisiifolia* to be 0.55 g/cc, while Crawford (1949), using an elutriator and also a permeometer, found a value of 1.30 g/cc. With these values of ragweed pollen density and assuming that ragweed pollen, diameter 20 μ, obeys Stokes' Law for a sphere falling in air, the fall speeds would be 0.39 and 0.93 m/min respectively. One can see that gross errors in the calculation of pollen concentration could be made. Since both Durham's and Crawford's experiments were very carefully undertaken the question must be asked: Could they both be right?

There is certainly no doubt that 2 samples of ragweed pollen may have quite different densities. For instance, saturated pollen will most certainly be heavier than dried pollen, which in turn should be heavier than dried and defatted pollen. Unlike many pollens which collapse when dried, the ragweed pollens have a thick, rigid, spherical shell which is rarely found in the collapsed state. Therefore, when the pollen grains decrease in weight, they also decrease in *effective density*, because the volume is constant.

When ragweed pollen is dried the loss of moisture causes the protoplasm to shrink, leaving 3 large air spaces within the grain. Polar photomicrographs of ragweed pollen grains at various humidities clearly illustrate the relative size and shape of the protoplasm and air spaces [Fig. 1(a)–(c)]. Figure 1(d) shows an equatorial view of the protoplast in its sac. Note that the sac is joined to the exine wall along 3 meridians.

The object of the research reported here was to measure the *effective density* of both fresh and commercially dried ragweed pollen at various humidities, as well as to measure the density of the solid material in the grains and the volume of the air spaces.

PROCEDURE—A new instrument, the Beckman air comparison pycnometer, was used to determine pollen density. This instrument operates very simply as follows. Two cylinders, *A* and *B* (Fig. 2), are connected by a valve and also by a null reading pressure gage. Pistons in each cylinder can be cranked forward to the stops indicated in the figure.

[1] Received for publication October 6, 1962.
[2] Effective density is the density of the whole pollen grain including its interior air spaces.

Initially, the connecting valve is open so that the pressure and temperature in the 2 cylinders are the same. The valve is then closed and piston A advanced to the stop while piston B is advanced in such a way that the pressures in the 2 cylinders are maintained equal. Due to the presence of the unknown volume V_x, piston B will displace a volume V_M less than piston A.

Indicating initial conditions by the subscript zero and final conditions by the subscript one, the number of moles of air in cylinder A will be

$$N_A = \frac{P_0 V_0}{RT_0} = \frac{P_1 V_1}{RT_1} \qquad (1)$$

and in B will be

$$N_B = \frac{P_0(V_0 - V_x)}{RT_0} = \frac{P_1(V_1 - V_x + V_M)}{RT_1} \qquad (2)$$

Dividing (2) by (1) we get

$$V_x = \frac{V_M}{1 - V_1/V_0} = \frac{V_M}{1 - n} \qquad (3)$$

where $n = V_1/V_0 = 0.5$ for the Beckman pycnometer. An indicator gives the unknown volume directly in cc's.

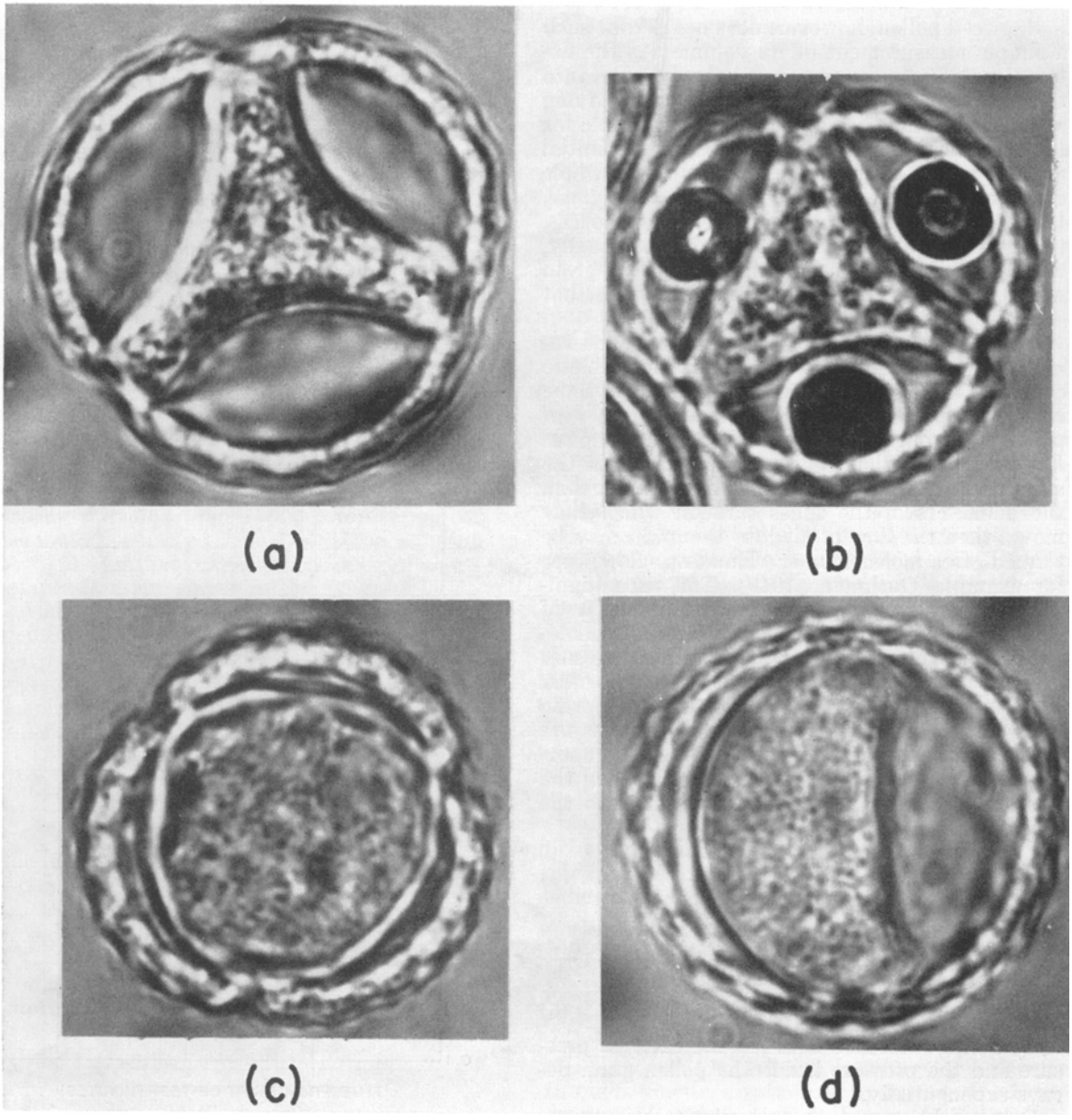

Fig. 1. Polar, cross-sectional photomicrographs of commercially dried ragweed pollen in equilibrium with air at various humidities: (a) 11%, (b) 33%, (c) 100%; (d) is an equatorial view showing the protoplasm sac.

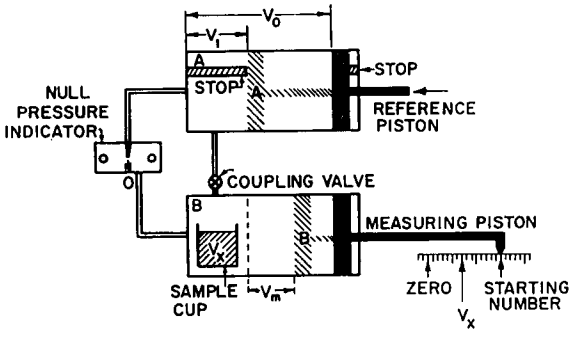

Fig. 2. Schematic diagram of the pycnometer.

Ragweed pollen, however, does not permit such a simple measurement of its volume. As the external pressure is increased, air slowly seeps into the internal air space of the pollen grain, giving a variable volume reading. If it were possible for full compression to be instantaneous, the initial reading would indicate the volume of the whole grain including the air spaces, the *effective volume*. Later, as air leaked into the interior air spaces, the indicated volume would decrease until finally, when the pressures inside and outside the grain were equal, the indicated volume would be that of the solid portion of the grain only.

By making some simple assumptions concerning the type of flow into the grain, it is possible to calculate the flow rate and even to draw some conclusions regarding the size and number of passages through which the air molecules pass. Let us suppose that the mean free paths of the air molecules are an order of magnitude larger than the diameters of the tubes through which they move; then the flow of air into the grain may be termed free-molecular or Knudsen flow; see for example Dushman (1949). The tube diameters would then have to be of the order of 0.005μ since the mean free path of air molecules at a temperature of 25 C and a pressure of 1.5 atm is about 0.05μ. As is shown in Appendix B, where the mechanics of air flow into the grain is discussed in more detail, the rate of pressure change in the pollen grain would then be proportional to the product of a constant, b (called the admittance), and the pressure gradient into the grain, i.e.,

$$\frac{dp_{at}}{dt} = b(p_1 - p_{at}) \qquad (4)$$

where p_1 is the final pressure and p_{at} is the pressure in the pollen grain air space at time t.

If we integrate eq (4) we see that

$$p_1 - p_{at} = e^{-bt}, \qquad (5)$$

so that the difference between the external pressure and the pressure inside the pollen grain decays exponentially.

One would expect that if all the hypotheses were correct, a plot of the natural logarithm of $p_1 - p_{at}$ versus time would produce a straight line with slope $-b$. The natural logarithm of $V_{xt} - V_s$ plotted against time, where V_{xt} is the measured volume of the pollen and V_s is the volume of the solid portion of the grains, should have the same slope since a simple manipulation of eq (A9) of Appendix A will show that measured volume and pressure differentials are linearly proportional. Figure 3 shows the natural logarithm of $V_{xt} - V_s$ plotted against time for a sample of commercially dried ragweed pollen.

The data of Fig. 3 indicate that when the pressure differential is large, the points do, indeed, lie on a straight line, so that near $t = 0$ we can assume that b is constant with considerable accuracy. The value of b for this sample was 0.02 sec^{-1}.

When the pressure gradient across the pollen pore becomes small, however, the slope b is no longer constant, but decreases. A possible explanation is discussed in Appendix B.

Returning to the problem of the measurement of pollen density, let us assume that the exponential law of eq (5) holds, and further, that the pressure in the pycnometer can be increased instantaneously. Were this to be true we would then expect to measure the volume and therefore the *effective density* of the grain at time zero, while after an infinite time we would expect to measure the volume of the solid portions of the grain only. A hypothetical curve of measured density versus time is shown (Figure 4, curve A).

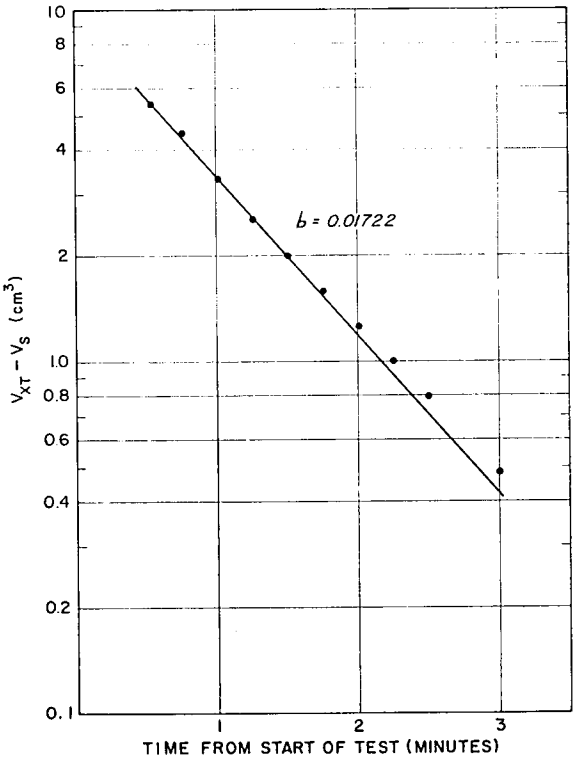

Fig. 3. The difference between measured volume and the volume of the solid part of the pollen grain ($V_{xt} - V_s$) versus time (dried commercial pollen).

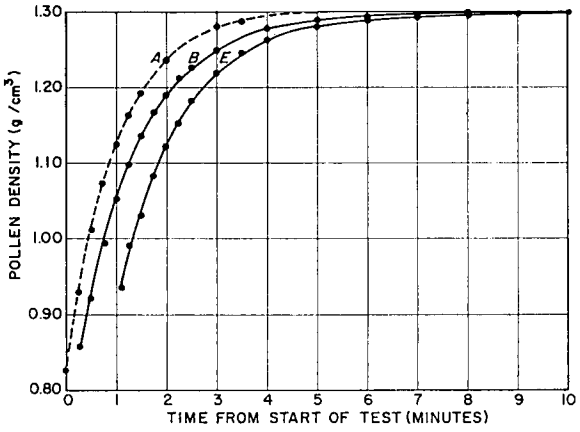

Fig. 4. Measured pollen density versus time: (A) hypothetical example assuming zero compression time; (B) compression time 22 sec; (E) compression time 66 sec.

Unfortunately, the pressure in the pycnometer cannot be increased instantaneously, so that, experimentally, curves such as B or E (Fig. 4) are obtained rather than A. To find the *effective density* we need to know the density at time zero. The problem then becomes one of finding some means of extrapolating our curves back to time zero.

Three methods of extrapolation are suggested: (1) Measure the density at the end of compression for various compression times and extrapolate the resulting points back to time zero (curves B through E, Fig. 5). (2) The measured volume and, therefore, density can be read at frequent intervals during compression and these points extrapolated to time zero. (3) The density which would be measured at any time during compression can be calculated using eq (A10) and the solution to eq (A4) of Appendix A. The results of one such computation are shown in Fig. 6.

All 3 techniques were used, each giving virtually identical results.

Results—The first sample of pollen tested had been collected in 1955 by Greer Drug Company of Lenoir, North Carolina, and had been dried to 1% moisture content but not defatted. The *effective density* was found using all 3 methods outlined in the previous section. One result of the extrapolation technique is shown in Fig. 5. The solid portion of the pollen was found to have a density of 1.31 g/cc, while the *effective density* was 0.82 g/cc. Repeated tests using all 3 extrapolation techniques completely confirm this result.

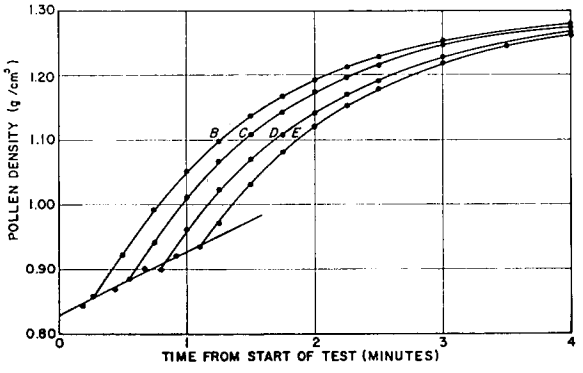

Fig. 5. Measured pollen density versus time for various compression times, showing the method of extrapolation to time zero.

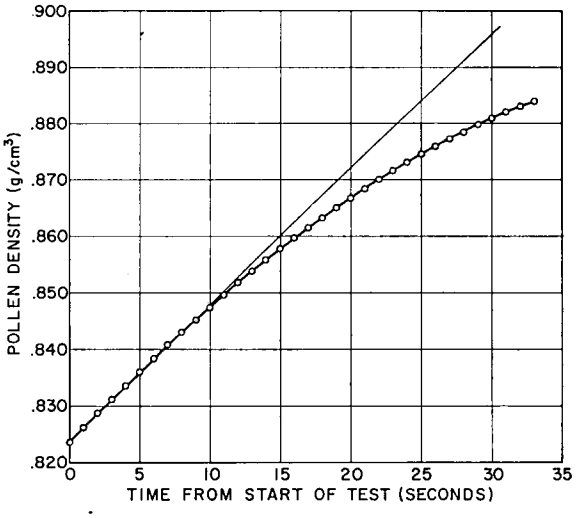

Fig. 6. Computed apparent density during compression for a commercially dried sample of 1955 *Ambrosia artemisiifolia* pollen.

The volume of the interior air space, V_a, was computed to occupy a fraction, 0.36, of the entire grain, a result which may be subjectively confirmed by a visual inspection of Fig. 1(a).

Table 1 shows the density of the same pollen after having reached equilibrium with air at various humidities. The humidities were controlled by the use of hygroscopic salts placed in sealed desiccators. The density of the pollen placed in the desiccators was measured at 2-week intervals until equilibrium was reached. Further checks were made for several months. The large volume of pollen measured (13 g) and its short exposure outside the desiccator (15 min or less) made any change in density during measurement negligible.

One unexpected result concerns the relatively constant value taken by the density of the solid portion of the grain. Were the air spaces in the driest grains to be filled with water, the new effective density would be 1.20 g/cc. This is considerably lower than the saturated density of 1.28 g/cc observed. One possible explanation for this discrepancy is that the exterior dimensions of the grain shrink when moist. A 2% reduction in diameter (0.4 μ) would produce the observed effect.

A second possible explanation is that the pollen walls and protoplasm can increase their density through kinetic sorption or hydration. Such hydration by hygroscopic polymers has been observed in cellulose (Ott, Spurlin, and Grafflin, 1954, p. 430) and in proteins (Neurath and Bailey,

TABLE 1. *Density of commercially dried ragweed pollen as a function of atmospheric relative humidity*[a]

R. H.	11%	33%	52%	75%	93%	100%
Fractional vol. of air space	0.36	0.35	0.35	0.18	0	0[b]
Density of solid portion (g/cc)	1.32	1.29	1.29	1.27	1.28	1.28[b]
Effective density (g/cc)	0.84	0.84	0.84	1.05	1.28	1.28[b]
Admittance, b (sec^{-1})	0.021	0.022	0.016	0.014	—	—[b]

[a] Pollen exposed March 2nd, density measured May 25th.
[b] The density of the pollen samples was measured several times. Pollen in saturated air reached an equilibrium density of 1.28 by April 3rd and very soon afterward began to mold.

1953, p. 559). However, the apparent lack of density change at high humidity would not be explained by hydration, since this effect takes place only during the commencement of moisture absorption.

The fine distinctions between pollen grains at various humidities which might be inferred from Fig. 1 do not appear in any of the actual measurements. Indeed, no significant difference in the grains can be measured until the humidity exceeds 52%. There is a small random variation in the size of the air spaces at any given humidity; thus the apparent decrease in air space size between 11 and 33% humidity, as illustrated in Fig. 1, was likely fortuitous or a product of wishful photography on the part of the authors.

TABLE 2. *The density of fresh ragweed pollen under various humidities*

	(a)	(b)	(c)
Relative humidity %	100.0	100.0	11.0
Fractional volume of air space	0.12	0.18	0.27
Density of solid portion g/cc	1.28	1.28	1.43
Density of whole grains g/cc	1.12	1.05	1.05
Admittance, b sec^{-1}	0.12	0.98	0.017

Fresh pollen was collected from ragweed plants grown in a greenhouse and quick-frozen until a sufficient quantity had been accumulated for accurate measurement in the pycnometer, i.e., about 5 g. It was then thawed in a desiccator at 100% humidity. After the pollen had thawed, several density measurements were made at short intervals. It became apparent that the density was changing rapidly with time. The first such measurement, shown in Table 2, Column (a), was taken less than 12 min after the pollen was removed from the desiccator. The second observation labelled (b) was made 10 min later.

An attempt was made to determine the rapidity with which natural pollen dries after dehiscence of the anther sac. Pollen was taken from extended but unopened florets, placed on 4 slides, then immersed in oil at 15-min intervals. On inspection, all the grains contained large air spaces, including those grains which had been immersed in oil almost immediately after the floret had been opened. Apparently evaporation from a fresh pollen grain exposed to unsaturated air is extremely rapid.

On hearing of these results, Dr. Willard W. Payne, of the University of Michigan Botany Department, opened new florets under oil. These had a totally expanded protoplasm sac. He then opened florets in air; the sacs were collapsed. He then opened a floret under water and watched the grains as the water evaporated. The sac remained fully expanded until the surrounding water evaporated, at which time it appeared to collapse fully in a few seconds [Fig. 7 (b) and (c)].

Findeisen (1939) developed an equation governing the rate of evaporation of a water drop in air. For drops as small as 20μ evaporation is extremely rapid, the drop disappearing in the order of a few tenths of a second at humidities below 90%.

Unlike water drops, which reduce in size as they evaporate, the ragweed pollen grain maintains a constant surface. Let us assume that this surface can be maintained in a saturated state until tht protoplast has fully collapsed, and further, thae one third of the grain is composed of evaporable water. Then, ignoring thermodynamic effects and considering diffusion only, the times taken to completely evaporate this water at various relative humidities can be computed and are shown in Table 3.

The data of Table 3 show that Dr. Payne's observations are entirely plausible; however, the rapidity of the evaporation must be questioned from another point of view. During compression

TABLE 3. *The time taken to evaporate water occupying ⅓ the volume of a ragweed pollen grain, assuming the surface is kep moist*

R H%	95	90	80	70	60	50	40	10
t sec	1.08	0.24	0.13	0.081	0.058	0.048	0.040	0.028

in the pycnometer, a period of about 51 sec must elapse before the pressure differential across moderately dry pollen grains is reduced by 50%. How then can a volume of water occupying one-third of the pollen grain leave in only a few seconds? The only explanation which presents itself is that during evaporation water is drawn to the surface by fluid flow under the forces of capillary action rather than by diffusion. The fact that evaporation takes more than a few tenths of a second may be explained by assuming that evaporation does not occur over the entire pollen grain surface, but only from the pores. Indeed, since the pores occupy about 1/120 of the surface of the grain, one might expect that at 50% humidity complete evaporation would take 5.8 sec, a figure very similar to that observed.

An attempt was made to measure the *effective density* of dried defatted pollen without success. Apparently the defatting process ruptures the membrane or pollen sac, allowing air to flow into the grain unimpeded. Under a microscope the sac, which normally encloses the protoplast, appears to have disappeared, leaving the protoplast settled in a heap inside the exine shell. The density of the dry solid material, however, was 1.31 g/cc.

Sources of error—Two different pycnometers were used during the course of the experiment. The first was used exclusively on the dried commercial pollen while the second was used for all the measurements at constant humidity and with the fresh pollen.

The limit of repeatable accuracy in reading the instruments was ±0.015 cc, providing an error of from 0.1 to 0.3%, depending on the volume of pollen measured.

Non-linearity of the null reading pressure gage over the full compression range in the second pycnometer plus distortion of the pressure element due to differential heat conduction introduced an additional error of as much as 0.04 cc or $\frac{1}{3}$–1% when using this instrument.

A third, and more important, source of error occurred in the computation of the *effective density* of *fresh pollen*. When measuring the volume of fresh pollen it was found that the admittance, b, did not remain constant over any portion of the pressure difference range. A value of the admittance appropriate to the high-pressure-difference end of the range, i.e., near time zero, could at best be estimated. Fortunately a 10% error in the admittance causes only a 0.85% error in the computation of *effective density*. Consequently it is unlikely that the computation of fresh pollen density is in error by more than 10–20%.

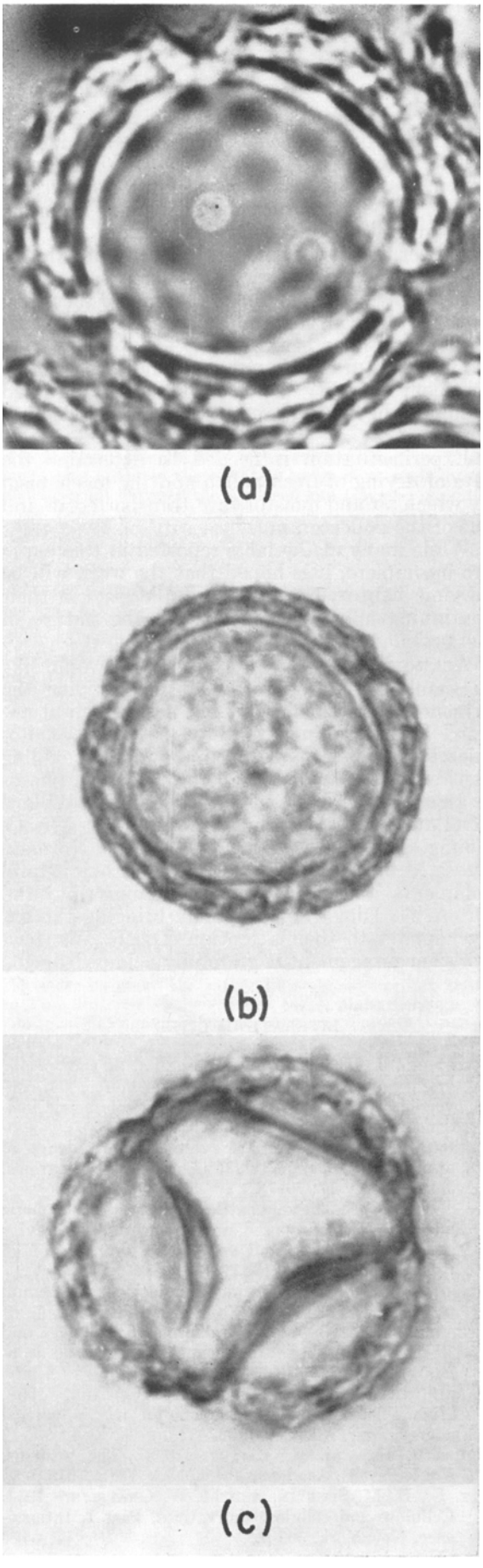

Fig. 7. (a) A fresh ragweed pollen grain, after 2 days in water, showing an empty protoplasm sac. The protoplasm itself has probably washed out by diffusion through a ruptured membrane at one or more of the pores; (b) fresh pollen undehisced; (c) fresh pollen seconds after dehiscence.

CONCLUSIONS—The gas pycnometer is ideally suited to measure the *effective density* of some pollen grains, but not of others. The *effective density* of pollens which are solid may be measured and also of those which contain air sacs, whenever the air sac is separated from the exterior air by an air-tight barrier or by small apertures. The *effective density* of pollens having air sacs open to the exterior air may not be measured using the methods outlined herein.

During future measurements using fresh pollen, the room humidity should be carefully controlled to match the humidity under which the pollen has been kept.

Experimentation is needed to determine the rate of drying of fresh pollen and the mechanism by which air and moisture are transported in and out of the pollen grain.

While many of the facts reported in this paper are incomplete, it is hoped that the work will be of some help to botanists and allergists in their continuing efforts to understand the nature of the pollen grain.

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APPENDIX A

Computation of measured pollen density during compression

Consider a steady cranking speed such that in cylinder A (Fig. 2)

$$V_t = V_0 \qquad \text{at } t = 0$$

$$V_t = V_1 \qquad \text{at } t = \tau,$$

where τ is the total compression time, so that

$$V_t/V_0 = 1 - ct, \tag{A-1}$$

where

$$c = \frac{1 - V_1/V_0}{\tau} = \frac{1 - n}{\tau} \tag{A-2}$$

is the fractional rate of volume reduction. In the small metal cylinders of the pycnometer all changes must be very nearly isothermal so that

$$p_t = p_0 v_0/v_t = p_0/(1 - ct). \tag{A-3}$$

Substituting p_t for the external pressure in Eq. (4) gives the simple differential equation

$$dp_{at}/dt = \left[\frac{p_0}{1 - ct} - p_{at}\right]b. \tag{A-4}$$

However, the solution of this differential equation is quite complex

$$p_{at} = p_0 e^{-bt}\{1 - \frac{be^{b/c}}{c}[\sum_{k=1}^{\omega}(-1)^k(b/c)^k(k \cdot k!)^{-1}(m^k - 1) + \ln(m)]\}, \tag{A-5}$$

where $m = V_t/V_0 = 1 - ct$. Rather than solve this formidable equation a simple computer solution to Eq. (A–4) was obtained using the Runge–Kutta finite-difference technique.

Once the pressure inside the pollen grain air space is known, the volume which would be indicated by the pycnometer can be computed. The computation is similar to that of Eqs. (1) and (2) except that the unknown volume V_x is now composed of 2 parts, V_s, the solid portion of the pollen grain, and V_a, the air space inside the pollen grain. The volume measured by the pycnometer consists of V_s plus a varying fraction of V_a.

As before, let N_A and N_B be the number of molecular weights of air in cylinders A and B, respectively. Then

$$N_A = p_0 V_0/RT_0 = p_t V_t/RT_t \tag{A-6}$$

and

$$N_B = \frac{P_0(V_0 - V_s)}{RT_0} = \frac{p_t(V_t - V_s - V_a + V_m)}{RT_t} + \frac{p_{at}V_a}{RT_a} \tag{A-7}$$

where the subscript a denotes conditions inside the pollen grain air space.

Dividing Eq. (A–7) by Eq. (A–6) and assuming that $T_a \approx T_t$ (the Joule–Thompson effect) we find that

$$V_m = V_a(1 - mp_{at}/p_0) + V_s(1 - m). \tag{A-8}$$

By replacing V_1/V_0 in Eq. (A–3) by V_t/V_0 and substituting for V_M in Eq. (A–8) we find

$$V_{xt} = V_a[(1 - mp_{at})/(1 - m)] + V_s, \tag{A-9}$$

where the value of p_0 has been taken as unity.

The volume of the pollen grain air space, V_a, can be found by evaluating Eq. (A–9) at time $t = \tau$ since $V_{x\tau}$ is measured and the other terms are known or have been computed.

Finally, the apparent pollen density at any time during crank-up can be calculated from the equation

$$\rho_{xt} = M/V_{xt}, \qquad \text{(A-10)}$$

where M is the measured weight of the unknown volume of pollen.

APPENDIX B

The mechanics of air flow into a pollen grain

It is possible for the flow of air into a ragweed pollen grain to behave in 1 of 2 ways, depending on the value taken by the ratio of the tube radius, a, to the mean free path of the air molecules, L. The 2 regimes are:

(1) $a/L > 100$ Hagen–Poiseuille Flow

(2) $a/L < 0.1$ Knudsen Flow

where the mean free path of a molecule is given by

$$L = 1.145 \times 10^4 (\eta/p)(T/M)^{\frac{1}{2}}, \qquad \text{(B-1)}$$

T being the absolute temperature, M the molecular mass, η the viscosity, the p the pressure, all in cgs units. For nitrogen at 25 C and 1.5 atm, L is approximately 5×10^{-6} cm.

If the tube radius exceeds 5μ, we should expect Hagen–Poiseuille flow, whereas if the radius is less than 0.005μ, we should expect pure Knudsen flow. Between these 2 values the flow should be intermediate although Knudsen flow will definitely predominate up to tube radii of 0.05μ (Dushman, 1949, p. 114).

Visual observations of ragweed pollen placed in oil or water have revealed air flowing out through the 3 pores as the water or oil flows into the grain. The pores are, therefore, quite likely the passages through which air moves into the grain during compression in the pycnometer, although this cannot be stated categorically.

The ragweed pollen pore is cone-shaped, having an outer diameter of about 2μ and an inner diameter of about 1μ The thickness of the shell and, therefore, the depth of the pore is about 1μ. Such an opening would allow much more rapid flow than is observed. However, the pore opening is further restricted by the protoplasm.

Several polar cross-sectional photomicrographs of commercially dried ragweed pollen kept at various humidities [Fig. 1, (a)–(c)] show the manner in which the protoplasm in its sac partly closes the pore opening. It will be noted that the shape of the sac depends markedly upon the humidity of the air in which the grain is kept.

Speculation as to the nature of the sac will not be attempted. However, Dr. Margaret Davis, of the University of Michigan Botany Department, has suggested that the sac consists of both the intine and intexine layers which have separated from the ektexine during dehydration. A clear picture of the sac can be seen in Fig. 7, a polar view of a typical grain of fresh pollen which has been kept in an aqueous environment for 2 days. Here it is postulated that water has entered the protoplasm through the entire surface of the sac while both water and protoplasm have diffused out through one or more pores where the membrane has been ruptured due to turgor pressure. The result is a grain in which the sac is intact and clearly visible while the protoplasm has completely disappeared.

When relatively dry, the protoplasm sac appears to fit into the pore opening like a plug, the sac being about 3μ wide at this point (Fig. (a)). Air might then enter the grain by passing around the sac either through an annulus-shaped channel, through a porous membrane, or through small apertures in the material. Water or oil also flows readily through these apertures.

When the grain is maintained at a higher humidity, the protoplasm sac expands and probably softens. We might expect that as this occurs the admittance, b, would change. The admittance was indeed a function of the humidity varying for the sample of fresh pollen from 0.12 sec^{-1} at 100% to 0.017 sec^{-1} at 11%.

The value of the admittance, b, also changed as the pressure differential across the grain was reduced (Fig. 3). This might be explained by the structure of the protoplasm sac where it plugs the pore. If the application of an external pressure can cause the sac to move very slightly then there may be a kind of valve action present which slowly tends to shut off the flow of air as the pressure gradient drops.

Having suggested the location and mechanism by which air might flow into a pollen grain, we are in a position to propose a model for the kind of flow which is taking place. Since the largest openings into the grain are 1μ in diameter, pure Hagen–Poiseuille flow is impossible. Our flow then must occur either in the Knudsen range or at some intermediate point between the Knudsen and Hagen–Poiseuille range. Let us make the assumption that flow takes place entirely in the Knudsen range.

Knudsen's equation is usually written

$$Q = F(P_1 - p_{at}), \qquad \text{(B-2)}$$

where Q is the flow in dyne cm sec^{-1} and F is the admittance in cm^3 sec^{-1}, differing from the admittance b by a factor of $1/V_a$. We, however, are concerned with the rate of pressure change inside the grain, i.e., with Q/V_a, therefore we can write

$$dp_a/dt = (F/V_a)(p_1 - p_{at}) = b(p_1 - p_{at})$$

which is just Eq. (A-4).

For N tubes of length l, and radius a, Knudsen deduced the basic relationship

$$b = \tfrac{2}{3}(\pi V_m/V_a) \cdot N \cdot (a^3/l), \qquad \text{(B-3)}$$

where V_m is the average molecular velocity given by

$$V_m = 14{,}551\,(T/M)^{\frac{1}{2}} \text{ cm } sec^{-1}, \qquad \text{(B-4)}$$

T being the absolute temperature in degrees Kelvin and M the molecular mass in grams (Dushman, 1949, p. 91).

Now let us make the rather unlikely assumption that the flow into the pollen grain takes place through 3 cylindrical passages 1μ long and of equal diameter. Introducing appropriate values for T, M, V_a, and b into Eqs. (B-3) and (B-4), we find that the passages must have a diameter of about 4 $m\mu$. This is roughly 10 times the molecular diameters of N_2, O_2, and H_2O. Molecular flow is, therefore, entirely possible.

The computed passage radius a will of course vary with the number N and length l of the passages postulated. However, from Eq. (B-3) it may be easily seen that

$$a^3 \propto l/N \qquad \text{(B-5)}$$

so that l or N must change by 3 orders of magnitude for the computed value of a to change by 1. Our estimate of pore size is therefore likely to be within an order of magnitude of the correct value and thus the flow into the pollen grain will likely be described adequately by Knudsen's formula (B-2).

It is interesting to note here that if the flow is taking place through many tiny pores in the exine wall a value of N exceeding 1000 will require a value of the pore diameter smaller than 0.4 $m\mu$, the length of the nitrogen and oxygen molecule (Dushman, 1949, p. 506).