the monkeys had been caused by toxemia was provided by a dose-response curve for monkeys challenged intravenously with 10^5 to 10^{11} spores. The dose-response curve for doses from 10^5 to 5×10^{10} spores shows that death occurred in progressively shorter times from 50 to 20 hours respectively. Three monkeys injected with 10^{11} spores, however, died within 2 hours with symptoms of toxemia. On the assumption that each multiplication of a bacillus produces a unit of toxin, it follows that 10^{11} germinating spores would produce 10^{11} units of toxin, a quantity that was rapidly lethal for monkeys. Since 10^{11} particles could be toxic themselves, the same inoculum of 10^{11} spores was reduced in viability to the equivalent of 10^{10} spores by heat. On injection of this inoculum, rapid death did not result, and the course of the disease corresponded to that for 10^{10} viable spores. We conclude that rapid death was caused by an amount of toxin suddenly released by the germination of 10^{11} spores, but the germination of 5×10^{10} or fewer spores released insufficient toxin to be lethal immediately and the disease progressed by a different course.

By four different methods we have shown that toxin is important in causing death of monkeys infected with *B. anthracis* or challenged with crude toxin. The importance of toxin in affecting the pathogenic course, and therefore the treatment of anthrax in man, is yet to be determined. It seems probable that the effect of toxin in man is the same as the effect in other animals. Since the introduction of antibiotics, treatment has been antibacterial. Our experiments suggest that treatment also be directed toward overcoming the effects of toxin or toward preventing its formation. Virtually all Russian recommendations on treatment of any type of anthrax infection put primary emphasis on use of antiserum (*9*). Since "respiratory" anthrax, if identified at all, is typically not identified until septicemia is well advanced, the development and use of specific antitoxin seems warranted. Similar conclusions were drawn by Plotkin *et al.* from their observations on the New Hampshire epidemic of inhalation anthrax (*10*).

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References and Notes

1. H. Smith, J. Keppie, J. T. Stanley, *Brit. J. Exptl. Pathol.* **36**, 460 (1955).
2. G. K. Middleton and A. C. Standen, *J. Infect. Diseases* **108**, 85 (1961).
3. J. T. Stanley and H. Smith, *J. Gen. Microbiol.* **26**, 49 (1961).
4. F. A. Beall, M. J. Taylor, C. B. Thorne, *J. Bacteriol.* **83**, 1274 (1962).
5. D. R. Hodges and M. A. Rhian, *Exptl. Med. and Surg.* **21**, in press.
6. J. Keppie, H. Smith, P. W. Harris-Smith, *Brit. J. Exptl. Pathol.* **36**, 315 (1955).
7. P. Malek, J. Kolc, F. Zak, *Zentr. Bakteriol. Parasitenk. Abt. I Orig.* **174**, 94 (1960).
8. C. B. Thorne, D. M. Molnar, R. E. Strange, *J. Bacteriol.* **79**, 450 (1960).
9. E. N. Shliakhou, *J. Microbiol. Epidemiol. Immunobiol. (U.S.S.R.)* **28**, 748 (1957), English translation (Pergamon, New York).
10. S. H. Plotkin, P. S. Brachman, N. Utell, *Am. J. Med.* **29**, 992 (1960).

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Genetic and Environmental Variation: Effect on Pigments of Selected Maize Mutants

Abstract. The chloroplast pigments of three "pastel" mutants of maize were found to vary with temperature as well as with genotype.

It has long been known that chlorophylls, carotenes, and xanthophylls are universally present in chloroplasts and also in the purple and green bacteria (*1*). Many mutations have occurred that affect the chloroplast pigments. They vary in effect from a complete lack of all of the pigments in question to a slight change in the structure or concentration of one of the pigments. In the latter class are some which appear different under different temperature conditions because of variations in the pigment concentrations. That is, the mutants are not only of a phenotype intermediate between the albino and the normal, but their phenotypes can be varied experimentally when the temperature of the environment is controlled.

For maize, Phinney has described the appearance of certain virescents after various temperature treatments (*2*), but, with one exception (*3*), no attempt has been made heretofore to describe chemically the phenotype differences between the normal plants and intermediate mutants under various treatments.

In this study, three pastel mutants which were sensitive to temperature and the offspring of two of these with allelic albino mutants were studied under conditions of controlled light and temperature. Pastel mutants are not only pale green in phenotype, but have a light yellow or white endosperm which is distinct from the yellow of the normal siblings.

The mutants used in the experiment were: pas_{8686} (pastel), its allele w_3 (white), on chromosome 2; pas_{4889}, its allele vp_9 (viviparous), on chromosome 7; and pas_{8549}, on chromosome 6. $Pastel_{8549}$ had been previously studied by Robertson and Anderson (*3*) under other conditions of light. This genetic material was obtained from the stocks maintained at Iowa State University. The mutants w_3 and vp_9 are viviparous albinos (*4*); pas_{8686} and pas_{4089} are tem-

Table 1. Pigment contents of normal (+) and mutant (m) corn seedlings as influenced by temperature.

Seedling	Temp. (°C)	Chlorophyll (mg/g fresh wt.)	m/+ (%)	Carotene (mg/g fresh wt.)	m/+ (%)	Xanthophyll (mg/g fresh wt.)	m/+ (%)
pas_{8549}							
+	22	2.551	68.1	0.0531	92.1	0.0556	81.5
m		1.737		.0489		.0453	
+	37	2.382	13.9	.0777	66.7	.0622	22.5
m		0.330		.0517		.0140	
pas_{4889}							
+	22	2.746	19.5	.0746	20.5	.0583	50.9
m		0.536		.0153		.0297	
+	37	2.717	43.9	.0755	51.8	.0731	33.2
m		1.193		.0391		.0243	
vp_9/pas_{4889}							
+	22	2.575	7.9	.0809	10.1	.0960	15.3
m		0.203		.0082		.0147	
+	37	2.577	16.7	.0873	13.9	.0586	43.3
m		0.430		.0121		.0254	
pas_{8686}							
+	22	2.477	11.1	.0658	7.9	.0407	45.0
m		0.275		.0052		.0183	
+	37	2.870	59.6	.0787	61.4	.0407	144.7
m		1.710		.0483		.0589	
w_3/pas_{8686}							
+	22	3.546	2.8	.1105	2.2	.0780	12.8
m		0.100		.0024		.0100	
+	37	2.419	22.8	.0903	14.2	.0670	45.7
m		0.552		.0128		.0307	

perature-sensitive, pale-green mutants that approach the appearance of normal plants at high temperatures (*5*); while *pas*$_{8549}$ is a pale-green mutant that approaches the normal phenotype at low temperatures and is the only one of these mutants ever observed to grow to maturity in the field (*3*).

The plants which served as the source of the pastel seeds were used as the pollen parents of the appropriate F_1 offspring to insure a more uniform background for comparisons of homozygous pastel seedlings with the heterozygous albino/pastel seedlings. Also, all comparisons of mutant to normal seedlings were made between plants from the same ear.

Mutant and normal seeds were separated on the basis of endosperm color. They were grown in rows in sand flats for 13 days at 22°C, or for 7 days at 37°C to permit harvesting at comparable stages of growth. A growth chamber with a maximum variation of ± 2°C was used. Fluorescent lamps, supplemented by 150 watt incandescent lamps, supplied the light, and the flats were placed so that the surface of the sand received 1400 foot-candles of light before the seeds sprouted. The amount of light was measured with a Weston light meter, model 756. The seedlings were harvested, weighed immediately, and stored frozen until they could be processed.

The methods used for extracting the pigments and measuring their concentrations have been described by Robertson and Anderson (*3*). A summary of the results are given in Table 1.

The error in technique was about ± 10 percent. No replications other than duplicate aliquots of the samples were used. In the case of the normal seedlings variation does occur from ear to ear, and is probably the result of variation in genetic background and different amounts of heterosis.

The mutations under investigation cause variation in all the plastid pigments. However, the mutations do not affect the pigment levels equally, and the effects vary with the locus in question, as well as with the allelic combination. There is probably not a single, common cause for the different types of variation seen at the different loci, although, for the albinos which are allelic to *pas*$_{8686}$ and *pas*$_{4889}$, production of the more unsaturated colored carotenoids is blocked while the production of chlorophyll is not blocked (*6; 7*).

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References and Notes

1. H. Strain, in *Photosynthesis in Plants*, J. Franck and W. E. Loomis, Eds. (Iowa State College Press, Ames, 1949).
2. B. O. Phinney and P. E. Kay, *Hilguardia* **23**, 185 (1954).
3. D. S. Robertson and I. C. Anderson, *J. Heredity* **52**, 53 (1961).
4. D. S. Robertson, *Genetics* **40**, 745 (1955).
5. ———, *ibid.* **46**, 649 (1961).
6. V. M. Koski and F. H. C. Smith, *Arch. Biochem. Biophys.* **34**, 189 (1951); J. H. C. Smith, L. J. Durham, C. F. Wurster, *Plant Physiol.* **34**, 340 (1959).
7. Journal paper J-4422 of the Iowa Agricultural and Home Economics Experiment Station, Ames, project No. 1381. Supported in part by G19395 from the National Science Foundation.

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Radioiodine in Milk of Cows Consuming Stored Feed and of Cows on Pasture

Abstract. During a period of 17 days, 2 months after termination of the Russian nuclear test series in 1961, iodine-131 was measured in milk. Within 2 days after the study was begun, the herd on pasture showed 270 pc iodine-131 per liter of milk. This concentration decreased with a half-life of 7 ± 1 days. No iodine-131 was found in the milk from sheltered cows that consumed stored feed.

One of the less disruptive measures which has been suggested for reducing the levels of iodine-131 and other short-lived radionuclides which, as a result of fallout, are present in milk is the placing of cows under shelter and providing them with water and hay that had been protected from recent fallout. Thus, the only sources of iodine-131 available to the cows would be inhalation or the ingestion of sheltered feed contaminated by deposition. Under these sheltered conditions, the iodine-131 in milk remained at or below 20 pc/liter while levels in other milk during the same period were as high as 270 pc/liter.

The transfer of significant concentrations of iodine-131 from fallout to milk occurs because the cow grazes over a relatively large surface area. The iodine-131 in milk is an appreciable part of that in the feed, and has been experimentally determined to be on the average as low as 0.03 percent (*1*) and as high as 0.5 percent per liter of milk (*2*). The relatively short period between milking and consumption by humans, generally from 1 to 5 days, is insufficient for extensive decay of the radionuclide to occur. In single dose experiments, the concentration of iodine-131 in milk rose to a maximum within 1 day and then decreased with a half-life of approximately 1 day (*1–5*). Continuous exposure to a single batch of contaminated feed resulted in an iodine-131 peak in milk after 2 days, followed by an exponential decrease with an iodine half-life of 8 days (*1, 4, 6*).

Within the wide range expected from such studies, similar changes of iodine-131 concentrations in milk were obtained when the feed was contaminated by tracer solutions, gaseous deposition, or air filters containing fallout particulates. Under actual grazing conditions, contamination of the grass prior to the study should result in a more rapid decrease of iodine levels in milk because of removal of foliar deposition from grass in addition to radioactive decay (*7*), while the deposition of significant additional radioiodine by rain or dry fallout would be shown by new increases in milk levels.

In the present experiment, iodine-131 levels in the milk of sheltered cows and cows on pasture were compared by gamma spectroscopy (*8*). Ten lactating cows, sheltered and fed stored hay for 2 months at the Oregon State University School of Agriculture, Corvallis, were divided into two herds; one remained sheltered and the other was placed on pasture from 2 to 18 December 1961. The cows on pasture ingested an estimated average of 37 kg of moist grass daily and gave an average of 5.6 liters of milk; the sheltered cows gave an average of 8.0 liters of milk. Cows on pasture also ate 3 kg grain mixture and 1 kg hay, whereas sheltered cows had the same amount of grain mixture and ate hay freely. A 3.5-liter sample of milk was composited daily for each group. Samples of the pasture grass, hay, grain mixture, rain, and drinking water were also analyzed by gamma spectroscopy. The gross radioactivity levels in air were obtained from the Portland station of the Public Health Service Radiation Surveillance Network.

The milk of the cows on pasture