ovum and its implantation as a blastocyst in the uterine wall varies from 6 to 10 days in the rabbit, rat, mouse, monkey, and man. The response of the embryo to damaging agents before implantation differs markedly from its response after implantation. During this period in mice, a teratogenic agent such as radiation is more likely to kill the embryo than it is to cause a congenital anomaly (*1*)

Previous research on the teratogenic properties of thalidomide in animals has been concerned primarily with its effects after implantation and during organogenesis. Several workers have found high resorption rates when the drug was administered before the implantation stage (*2, 3*). Usually much larger amounts of the drug (on a milligram per kilogram basis) were required to produce resorptions and anomalies than appears to be the case in the human (*4, 5*). We chose a dose of thalidomide close to the estimated amount required to produce human anomalies. This dose had no detectable toxic effects in the monkey, and this fact encourages us to infer that sensitivity to thalidomide teratogenicity is similar in monkeys and man.

Others have observed directly the rabbit blastocyst before implantation; thalidomide (about 125 mg/kg), when given to the pregnant rabbit, caused degeneration of the embryonic disk of the blastocyst (*3*). When the early chick embryo was treated with thalidomide "the development of the germ per se ceased or was delayed" (*4*).

No previous study of this nature has been carried out in primates. If one intends to use the rhesus monkey as a test animal for the teratogenic effects of thalidomide, a different dosage and time schedule of drug administration will be required (*6*).

Further investigation of this property of thalidomide and perhaps of related compounds should be undertaken with a more sophisticated approach designed to explore mechanisms by which pregnancy may be prevented.

JEROLD F. LUCEY
Department of Pediatrics,
University of Vermont
College of Medicine, Burlington
RICHARD E. BEHRMAN
Laboratory of Perinatal Physiology,
National Institute of Neurological
Diseases and Blindness,
National Institutes of Health,
San Juan, Puerto Rico

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Mutagenic Action of Ethyl Methanesulfonate in Maize

Abstract. *Pollen of corn plants carrying three closely linked genes (α β Sh₂) on chromosome 3 were treated by ethyl methanesulfonate in order to determine the nature of genetic changes produced. In this genetic material the loss of the β gene alone represents a discrete genetic change, possibly a point mutation, while the loss of two or more markers represents chromosome aberrations. Ethyl methanesulfonate, x-rays, and ultraviolet light all induced numerous chromosome aberrations, but only ultraviolet light and probably ethyl methanesulfonate induced discrete genetic changes.*

In barley, the frequency of mutations (as opposed to chromosome aberrations) induced by the alkylating agent diethyl sulfate is higher than that obtained by optimum doses of gamma radiation (*1*). Because of this property of diethyl sulfate, an experiment was undertaken with another alkylating agent, ethyl methanesulfonate (EMS), which is reported to induce mutations and reversions in lower organisms (*2, 3*) and mutations in barley (*4*). This alkylating agent can ethylate the guanine moiety in position 7, thereby causing guanine to pair with thymine rather than cytosine during DNA duplications (*5*). The low toxicity of EMS permits use of concentrations that may produce a relatively high frequency of genetic changes (*2*). This experiment was similar to an earlier one in which closely linked markers were used to compare the nature of genetic changes induced by x-rays and ultraviolet light in maize (*6*).

The compound locus A^b and the closely linked gene Sh_2 were used. The components of the A^b locus, α and β, and the Sh_2 locus extend across less than 0.3 map unit in the long arm of chromosome 3. The diagram below illustrates this segment of chromosome 3 with the dominant genes of the treated male parent and their recessive counterparts on the homologous segment of the chromosome of the egg parent.

♂	*α*	*β*	*Sh*
♀	a_m		*sh*

The simultaneous loss of *α*, *β*, and *Sh* is taken as evidence of chromosomal breakage. The absence of the *β* phenotype alone could be due either to the loss of *β* because of chromosomal breakage or to a mutation of the gene.

In preliminary experiments several methods for introducing EMS into pollen were tried. They included immersing cut leaves in a dilute solution, injecting the solution into young tassel shoots, or introducing a solution by means of cotton wick through a hole drilled into the stem directly below the tassel. These methods failed to produce detectible genetic changes. However, the following treatment was successful: the leaves surrounding the shoot were slit open with a razor blade 3 to 5 days before pollen shedding, the tassel branches individually were imbedded tightly in cotton, and the

Table 1. The frequency of gene losses detected as whole and fractional endosperm changes per 10^4 seeds from a^m *sh* by treated *α β Sh*.

Treatment	Population	*α β Sh* (Colorless, shrunken)	*Sh* (Colored, shrunken)	*α β* (Colorless, normal)	*β* (Dilute, normal)
		Whole endosperm loss			
Control*	11499	7	0	0	0
UV*	8888	267	10	0	11
X-ray *	8739	611	7	0	0
EMS†	12131	41	5	0	1
		Fractional endosperm loss			
Control*	11499	16	1	0	0
UV*	8888	397	91	0	12
X-ray*	8739	90	8	0	0
EMS†	12131	216	16	2	0

*Includes data from earlier experiment (*6*); ultra-violet, 30 seconds; x-ray, 1200 r. † EMS, 0.05*M*.

leaves were closed with cellophane tape. A 0.05*M* solution of EMS prepared 2 hours previously was slowly injected into the cotton with a 30 ml-hypodermic syringe until saturation was reached (15 to 30 ml per plant). The cotton was removed 6 hours after injection, together with the partially exposed and presumably less severely treated tassel-branch tips. At flowering the treated pollen containing the dominant genes α β Sh_2 was used to fertilize plants recessive for these markers (a^m sh_2). Mutations or losses of dominant genes through chromosome aberrations occurring before fertilization gave whole endosperm effects, while changes occurring during development result in fractional effects.

The data (Table 1) show that EMS, like x-rays and ultraviolet light, is effective in producing breaks in various regions of the tested chromosome segment. Direct comparisons of total frequencies are impractical because comparable dosage levels of these agents are not known. Ethyl methanesulfonate resembles ultraviolet light in action. Both produce a relatively higher proportion of fractional losses than whole seed losses and both produce losses of β alone as well as an assortment of less discrete changes. In these aspects EMS differs from x-rays, which produce a higher proportion of whole seed losses than fractional losses and which repeatedly fail to produce discrete changes of β alone (*7*).

M. G. NEUFFER
GYULA FICSOR
Department of Field Crops, University of Missouri, Columbia

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Radiation-Induced Gelation of Dilute Aqueous Pectin Solutions

Abstract. *The low-dose gamma irradiation of dilute aqueous pectin solutions after appropriate adjustment of* p*H with hydrochloric or certain other inorganic acids leads to the formation of thermoreversible gels. Maximum gel strength is attained only under optimum conditions of* p*H, concentration, temperature, radiation dose, and exposure time, and only in the absence of oxygen and a number of radiation protectants.*

Pectin, a methyl ester of polygalacturonic acid, undergoes degradation when its aqueous solutions are exposed to ionizing radiation (*1*). No evidence has previously been presented to indicate any radiation-induced coupling reactions between pectin molecules in aqueous solutions. Cross-linking of pectin molecules can be achieved without irradiation by the addition of certain compounds such as formaldehyde, epichlorohydrin, erythrodioxide, and sulfur mustard—a radiomimetric substance (*2*). The cross-linking by bi- or polyfunctional molecules produces a principal-valent gel which is stable, insoluble in water and not thermoreversible. Reversible pectic gels are formed by hydrogen bonding through the addition of acid and a dehydrating agent such as sugar, or by ionic bonding through the addition of a multivalent ion such as Ca^{++}. This report (*3*) describes some conditions under which thermoreversible gel formation can be induced by ionizing radiation without the addition of any dehydrating agent or multivalent ion.

In a preliminary irradiation experiment with a series of dilute aqueous pectin solutions within a wide range of *p*H values, the authors observed gel formation only in those solutions with a *p*H value of about 1.6 to 2.2, and then only at doses of a few hundred kilorads of gamma radiation. For further investigation of this phenomenon, aqueous solutions (2 percent) of a highly refined citrus pectin (NF) with a methoxyl content of 9.8 percent were prepared and adjusted to *p*H 1.0 to 3.0 with HCl.

The samples (in vacuum) were exposed for 90 min at 20°C to 630 krad of gamma radiation, the source of which was Co^{60}. At the end of the irradiation period, the samples were evacuated again to remove any gas bubbles which were entrapped in the gels and which would have interfered with subsequent measurements of gel strength. The viscosity, an indicator of gel strength, was measured with a Brookfield model RVT Synchro-Lectric viscometer and Helipath spindle (T-C). The viscosity measurements in Table 1 were made at 25°C with a spindle speed of 5 rev/min.

All gels which were formed between *p*H 1.4 and 2.6 were reversible; they became dispersed by treatment with mild heat or by exposure to room temperatures for several hours. Maintaining the samples at moderately high temperatures (30 to 50°C) during the irradiation period resulted in weaker gels or no gels at all; the effect was dependent on both the temperature and the length of the exposure period. After irradiation and complete liquefaction of the resulting gels by warming for a few minutes or standing at room temperature for several hours, the viscosity of the liquids thus formed (as measured with an Ostwald-Cannon-Fenske viscometer) continued to decrease over a period of 1 to 3 days. A minimum value equal to that of a solution of the same concentration which had been irradiated with the same dose but without any adjustment of *p*H before irradiation was finally reached.

The strength of the gel formed by a 2-percent pectin solution irradiated at a *p*H of 1.8 increased as the radiation dose increased above about 300 krad until a maximum viscosity was reached at approximately 1 Mrad. Beyond this value there was a decrease in gel strength with additional radiation. At any specific *p*H value, as the pectin concentration increased within the range of 0.5 to 8.0 percent, there were increases in the gel strength at any specific dose, the maximum gel strength attainable, and the dose of maximum gelation, and a decrease in the dose necessary to cause incipient gelation. At concentrations lower than about 0.5 percent, no gel was formed at any radiation dose. Solutions with a pectin concentration higher than 8 percent were too viscous for accurate adjustment of *p*H and evaluation of any gels which might have formed. In all cases of gel formation, there was little change in *p*H and methoxyl content upon irradiation.

Maximum gel strength was attained at a *p*H of approximately 1.8, regardless of the pectin concentration, and also when the *p*H was adjusted with