

Loss of coat color in cattle grazing on pastures high in molybdenum content has been observed in England (2) and in California (3), and it is now well-known that the feeding of small amounts of copper sulfate to affected animals restores the color to new growth of hair. It has also been shown by Goss (4) that the wool of black sheep that are fed an excess of molybdenum as sodium molybdate grows out colorless, but the black pigment quickly reappears when 100 parts of copper per million are added to the high molybdenum ration.

Analyses of the black wool for copper showed 17 ppm, whereas the grey portion of the same fibers after molybdenum-feeding contained only 13 ppm. However, white wool from normal sheep consistently showed more copper than black, contrary to the report of Kikkawa *et al.*

for the guinea pig, rabbit, and man. We therefore collected samples of light and dark shades of hair from a number of species and determined the copper content. The values are given in Table 1, together with the corresponding figure from the paper by Kikkawa *et al.* (1), recalculated in parts per million. Apparently their results were obtained by spectroscopic analyses of the ash obtained by combustion of the hair in a furnace at 600°C. Our results were obtained on thoroughly washed and dried hair by a "wet-ashing" method using sulfuric, perchloric, and nitric acids; the copper was estimated colorimetrically in duplicate by use of the diethyldithiocarbamate method described by Clare *et al.* (5). Except in the hog, we have found as much, if not more, copper in white or light-colored hair than we have found in black hair of the same species.

As far as the copper content of hair is concerned, our results do not agree with those reported by the Japanese authors, and we find no confirmation of the statements that black hair is associated with a high copper content, and that white hair is low in copper or contains only a trace of copper.

HAROLD GOSS  
M. M. GREEN

*Departments of Animal Husbandry and Genetics, University of California, Davis*

#### References

1. H. Kikkawa, Z. Ogita, S. Fujito, *Science* **121**, 43 (1955).
2. W. R. Muir, *Vet. J.* **97**, 387 (1941).
3. J. W. Britton and H. Goss, *J. Am. Vet. Med. Assoc.* **108**, 176 (1946).
4. H. Goss, *Proc. Soil Sci. Soc. Florida* **10**, 175 (1950).
5. N. T. Clare, I. J. Cunningham, D. D. Perrin, *New Zealand J. Sci. Technol.* **26**, 340 (1945).

18 April 1955

### Enhancement of Radiobiological Effect by Malonic and Maleic Acids

Certain biological actions of radiations are generally believed to involve chemical intermediates, mostly formed from irradiated water, and in some of these actions the intermediates are believed to be peroxides (1). These substances may attack various important reaction systems in the cell at various sites, for example, sulfhydryl and other reactive groups, especially in sulfhydryl enzymes (2).

In many of our experiments, in which cell death was the criterion of effect, we failed to enhance the radiation effect by inhibiting oxidative phosphorylation, which has recently been reported to be exceptionally sensitive to irradiation (3). These negative results may be due to masking effects or to recovery of the system. Only with malonic and maleic acids

were positive results obtained. Malonic acid inhibits oxidative phosphorylation and is highly specific as a competitive inhibitor of succinic dehydrogenase. Maleic acid, although not so specific for succinic dehydrogenase, is a universal inhibitor of sulfhydryl enzymes.

*Saccharomyces ellipsoideus*, the experimental material, was cultured in Nägeli's solution for many generations. After x-irradiation (60 kv, 1000 r/min), the cells were cultured on Nägeli agar at pH 7. Photographs were taken and microscopic counts were made at intervals for some 20 hr. Single cells and also pairs of enlarged cells, which are ascribed to death after one division, were scored as nonsurvivors; the counts of these categories do not change in the course of prolonged incubation. Five to 10 cultures were counted in each experiment, and every culture contained 400 to 500 cells.

Malonic acid in concentration  $10^{-3}M$  without irradiation produced no detectable injury. Its sensitizing action, when it was added to the Nägeli culture solution before irradiation, is demonstrated by the data in Table 1.

If fumaric or aspartic acid was added to the Nägeli agar on which the cells were cultured after irradiation, the results shown in Table 2 were obtained. It appears that the effect of each of these acids on cells pretreated with malonic acid is essentially to bring the survival back to the value obtained when malonic

Table 1. Sensitization by malonic acid applied before irradiation.

X-ray dose (r)	Percentage survival (calibrated by control)	
	Malonic acid, $10^{-3}M$	No malonic acid
0	100 ± 1.3	100 ± 1.1
10,000	45.1 ± 3.2	77.5 ± 6.5
20,000	32.8 ± 2.1	47.9 ± 3.2
30,000	17.8 ± 2.4	30.0 ± 1.2
50,000	8.6 ± 0.9	10.6 ± 0.4

Table 2. Recovery owing to fumaric and aspartic acids after 20,000 r of x-rays (calibrated by nonirradiated cells).

Treatment before irradiation	Treatment soon after irradiation	Survival (%)
None	None	48.0 ± 2.4
Malonic acid, $10^{-3}M$	Fumaric acid, $2 \times 10^{-3}M$	50.0 ± 1.1
None	Fumaric acid, $2 \times 10^{-3}M$	50.0 ± 1.3
Malonic acid, $10^{-3}M$	Aspartic acid, $10^{-3}M$	51.8 ± 2.8
None	Aspartic acid, $10^{-3}M$	51.8 ± 1.9

Table 1. Copper content of hair in parts per million of washed, dry hair.

Source	This report	Kikkawa <i>et al.</i>
Cat, black	14	
Cat, white	34	
Holstein cow, black area	12	
Holstein cow, white area	13	
Hereford cow, dark red	27	
Hereford cow, light red	30	
Hereford cow, light red	26	
Hereford calf, red area	17	
Hereford calf, white area	23	
Angus bull, red	15	
Dog, black	17	
Dog, white	25	
Guinea pig, black	11	8
Guinea pig, golden	13	5.2
Guinea pig, white (albino)	15	2.3
Hog, black	17	
Hog, white	12	
Horse, black	10	
Horse, white	15	
Man, Caucasian, 3-yr-old child, red	47	
Same, as adult	15	
Adult, red	18	
Mexican child, red	18	
Man, Mongolian, black	15	19
Man, Negroid	15	31
Rabbit, black	14	3
Rabbit, white (albino)	17	Trace
Rabbit, white (albino)	20	
Rabbit, white (albino)	19	
Rat, black	14	
Rat, white (albino)	14	
Sheep, black	17	
Sheep, black, high Mo feed	13	
Sheep, white	20	

Table 3. Recovery owing to cysteine after 20,000 r of x-rays (calibrated by non-irradiated cells).

Treatment before irradiation	Treatment after irradiation	Survival (%)
Malonic acid, $10^{-3}M$	Cysteine, $10^{-3}M$	$67.2 \pm 2.4$
None	Cysteine, $10^{-3}M$	$72.1 \pm 2.6$

Table 4. Sensitization by maleic acid applied before 20,000 r of x-rays (2000 cells were counted as a whole, and calibration was held by control).

Molar concn. of maleic acid in Nageli's soln.	Survival (%)	
	Irradiated	Non-irradiated
$10^{-3}$	22.6	85.3
$10^{-4}$	36.1	96.2
$10^{-5}$	43.4	102.1
0	47.1	100

acid was not used (compare Table 2 with third line of Table 1). This may be explained on the hypothesis that fumaric or aspartic acid removes the block produced in the tricarboxylic acid cycle by malonic acid.

In other experiments, cysteine was added to the Nageli agar to reactivate altered sulfhydryl groups in the irradiated cells (Table 3). Although there was a substantial recovery owing to cysteine (compare Table 1 with Table 3), the sensitizing action of malonic acid was not completely overcome by the cysteine; there was the statistical significance of the difference between 67.2 and 72.1 percent.

The effect of maleic acid on radiosensitivity is shown by the data in Table 4. This acid, in concentrations less than  $10^{-3}M$ , has no inhibiting influence on growth of the yeast cell but seems to sensitize the cell to radiation, the effect varying directly with concentration in the culture medium. However, its concentration and distribution in the cell are unknown.

M. KIGA  
Y. ANDO  
H. KOIKE

Department of Radiology,  
Showa Medical School, Tokyo, Japan

#### References and Notes

1. Fricke *et al.*, *J. Chem. Phys.* 6 (1938); J. Weiss, *Nature* 153, 748 (1944).
2. Barron *et al.*, *J. Gen. Physiol.* 32 (1948); H. M. Patt, *Science* 110, 213 (1949).
3. Devick, *Brit. J. Radiol.* 25, 481 (1952); D. W. Bekkum, *et al.*, *Brit. J. Radiol.* 27, 127 (1954).

28 March 1955

## Tests of a Soil Sterilant for Forestry Use

The chemical compound 3-(*P*-chlorophenyl)-1,1-dimethylurea (CMU) (1) has been tested in the sand hills of western Florida to determine whether it would kill scrub oaks and wire grass and permit reforestation with planted pines. CMU (2) is a nonvolatile, slightly acid, grayish-white powder with a very low solubility in water (230 ppm). It has relatively low flammability and mammalian toxicity.

The chemical was applied to the soil as an aqueous suspension in 10 dosages from 0 to 37 lb/acre in March 1953, each treatment being replicated four times. Mortality of woody plants was determined by actual stem count, while ground cover was surveyed by line-spot transects. The results are based on differences between pretreatment and post-treatment vegetation surveys.

A vegetation survey that was made 16 mo after application showed that dosages of 11 lb or more per acre had controlled scrub oaks of all sizes. Twenty-two pounds or more was necessary to control grass and other ground cover. Necrosis first appeared around the leaf margins of oaks and on the tips of pines and grasses. Oaks (*Quercus laevis* and *Q. incana*) that received dosages of 22 lb or more lost as many as three sets of leaves during the first growing season after treatment. Wire grass (*Aristida stricta*), the pretreatment dominant, was nearly eradicated at dosages around 7 lb/acre, but it was replaced by *Sorghastrum nutans*, *Panicum virgatum*, *Andropogon scoparius*, and *Andropogon floridanus* during the second growing season after treatment (Table 1). Ingressive grasses in treated plots were much taller and more vigorous than those in untreated border strips. Weed species that were the most resistant to CMU included cactus

Table 1. Mortality of oaks and ground cover at end of second growing season after application of CMU to the soil.

Dosage (lb/acre)	Mortality (%)			
	Oaks 8 ft and taller	Oaks 4-8 ft tall	Oaks 1-4 ft tall	Ground cover
0	0	19	13	5
1	26	0	0	31
2	23	12	19	11
4	57	20	4	14
7	87	84	13	14
11	98	99	97	3
16	100	100	99	43
22	100	100	99	74
29	100	100	100	86
37	100	94	97	79

(*Opuntia* spp.), saw-palmetto (*Serenoa repens*), sassafras (*Sassafras albidum*), and yucca (*Yucca* spp.).

Forty-nine slash pines (*Pinus elliottii* Engelm.) were planted in each plot in January 1954, 9 mo after application of the chemical. Analyses of soil samples for residual CMU were made in July 1954. Phytotoxic amounts were found even in soil that had received dosages of 4 lb/acre; pine survival counts made in September 1954 revealed that 24 percent of the trees in this treatment were dead or severely chlorotic. Pines in the 37-lb treatment suffered 98-percent mortality and injury. The persistence of CMU even at low dosages makes its value for use in forestry questionable.

FRANK W. WOODS\*

Southern Forest Experiment Station,  
Forest Service, U.S. Department  
of Agriculture

#### Notes

1. It is also known as Karmex W.
2. E. I. du Pont de Nemours and Co. supplied the chemicals that were used in this study and made analyses of soil samples.

\* Stationed at East Gulfcoast Research Center, Marianna, Fla.

13 June 1955

## Sound of Boiling

An interest in the sound of boiling liquids has been apparent for years. Chemical plant operators in charge of evaporators and reboilers sometimes judge the performance of their equipment by the noise emitted. A general superstition is that the louder the noise of boiling the better the performance of the equipment. The noise that occurs as a hot metal is quenched in a liquid has received notice. A change in pitch accompanies the drop in temperature (1). The noise accompanying the overloading and resulting burnout of electric heaters immersed in water has been reviewed (2).

Researchers also occasionally depend on the sound of boiling. When boiling data are obtained, it is important to know which type of boiling is occurring. Boiling can occur by at least three different mechanisms. These are different to the eye and the ear and also in the manner in which the heat transfer depends on the temperature driving force.

The relationship between the heat-transfer rate and the over-all temperature driving force for each type of boiling, with methyl alcohol, is shown in Fig. 1. The short crosslines indicate the boundaries between the types of boiling: nucleate, transition, and film boiling.

Visual studies of the types of boiling have been made with still photography, using exposures of  $10^{-6}$  sec, and with