observed at all three levels of pollen grain concentration with *P. rigida* and *P. echinata*. Calcium and/or boron have been associated with the self-stimulation effect in several plant species (Rosen, 1968). The relationship of nonanol or related compounds to the endogenous self-stimulating agent of pine pollen is not known.

Some species of fungal spores have been found to respond only to certain specific chemical structures (French et al., 1975a,b). Uredospores of *Uromyces phaseoli*, for example, respond only to methyl ketones, particularly β-ionone (French et al., 1977), while those of *P. graminis* var. *tritici* respond to certain alcohols, aldehydes, and ketones, particularly nonanol and nonanal. Perhaps pollen of other genera of plants may be found to respond to specific chemical germination stimulators.

The mechanism of action of stimulating germination of rust spores may be that of overcoming endogenous inhibitors, identified by Macko et al. (1970, 1971) in uredospores. Methyl ferulate was found in *P. graminis* var. *tritici* and methyl 3,4-dimethoxycinnamate in *U. phaseoli* which the appropriate chemical stimulators appear to neutralize in inducing germination. Other biochemical activity has been observed with certain germination stimulators. Sinohara (1973) has shown that 1-octanol is the most effective of the 1- to 8-carbon alcohols in inducing de novo glucose dehydrogenase synthesis in dormant spores of *Aspergillus oryzae*. He postulates a mechanism of action in which octanol expands the endoplasmic reticulum, permitting synthesis of the enzyme. Feofilova and Arbuzov (1975) have shown β-ionone to stimulate de novo synthesis of carotogenic enzymes in *Blakeslea trispora*.

In many angiosperms, flowering is accompanied by production of a scent, often composed of fungal spore stimulating compounds, such as rhodinol, geraniol, and nonanal in the rose, or ionone in acacia and boronia flowers (Guenther, 1952a; Furia and Bellanca, 1975). Similar volatile compounds occur in gymnosperms and in various turpentines made from *Pinus* species (Guenther, 1952b; Linskens, 1964; Furia and Bellanca, 1975), in which pinenes, limonenes, and even small amounts of 8-, 9-, and 10-carbon *n*-aldehydes have been reported. A great variety of such compounds have previously been shown to stimulate fungal spore germination. Perhaps some of them stimulate pollen germination in vivo and could be effective in overcoming endogenously inhibited pollen tube growth.

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Enhanced Elimination of Kepone-^{14}C in Rats Fed Liquid Paraffin

Twelve male rats were fed a diet containing 3.3 ppm Kepone-^{14}C for 3 days. After 1 day on the control diet, six animals received a ration containing 8% light liquid paraffin for 24 days. The remaining six animals served as control. Paraffin-treated rats excreted significantly more radioactivity with feces (61%) compared to controls (52%) and had significantly lower concentrations of radioactivity in 14 of 18 tissues analyzed. Excretion of radioactivity with urine was of minor importance (0.5–0.6%) in both groups.

Kepone, decachlorooctahydro-1,3,4-metheno-2*H*-cyclobuta[*cd*]pentalen-2-one, is a persistent insecticide used mainly against ants and cockroaches (Martin, 1971). In recent years great quantities of Kepone have been released into the environment of the Virginia–Maryland coastal region due to an industrial mismanagement. Seventy of 150 occupationally exposed workers had symptoms of Kepone poisoning and high levels of Kepone in blood (Sterrett and Boss, 1977). In the Netherlands Kepone was a trace component in the fly ash of a municipal incinerator (Lahaniatis et al., 1977). Previous studies in our laboratory showed paraffin feeding to considerably enhance the elimination of hexachlorobenzene (HCB) in rats (Richter et al., 1977). In the present study we have investigated the influence of paraffin treatment upon the elimination of Kepone-^{14}C in rats.

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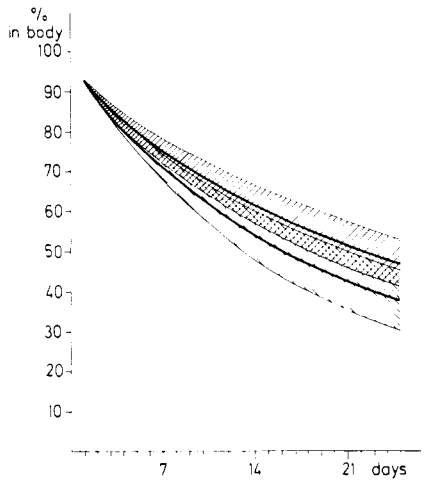

Figure 1. Elimination of Kepone-^{14}C from control (upper) and paraffin-treated (lower) rats as determined by the differences of total Kepone consumed and excretion in feces and urine ($\bar{x}$, s_x).

MATERIALS AND METHODS

^{14}C-labeled Kepone (Mallinckrodt) with a specific activity of 5 mCi/mmol was used. The purity of Kepone was greater than 98% by GC–MS (GC–MS unit LKB 9000, column length 4 m, i.d. 4 mm, filled with 1% OV-1, temperature program 190–250 °C at 5 °C/min). Kepone was dissolved in acetone and mixed with a commercial powdered rat chow (Altromin 1321) to give a final concentration of 3.3 ppm. Twelve male Sprague-Dawley rats of about 200 g body weight were housed separately in metabolism cages. After 4 days of adaptation all animals were fed the diet containing 3.3 ppm Kepone-^{14}C for 3 days. After 1 day on control diet six animals received a ration containing 8% light liquid paraffin (Merck) and the remaining six rats served as controls. Feed consumption and weekly body weight changes were recorded. Urine and feces were collected daily. Twenty-five days after discontinuing the Kepone-fortified diet, all animals were sacrificed, organ weights (liver, kidney, spleen, testes, heart, brain) determined, and 18 different tissue samples taken for analysis. Total radioactivity was determined in duplicate samples of urine, feces, and tissues and six samples of Kepone-fortified diet, as described previously (Richter et al., 1977). The two-compartment open model was calculated with an Olivetti P 6060 according to a slightly modified program of Pfeffer (1973).

Table I. Total Excretion of Radioactivity in Urine and Feces after 25 Days in Percent of the Amount Absorbed[a]

treatment	% in urine	% in feces
control	0.52 ± 0.13	52.16 ± 5.88
paraffin	0.58 ± 0.21	61.52 ± 7.21[b]

[a] Mean and standard deviation of six rats. [b] Significantly different from control ($p < 0.05$).

RESULTS AND DISCUSSION

No significant differences were seen in body weight gain and food consumption or in the absolute and relative (to body weight) weights of organs. Analyses of six samples of food gave a concentration of 3.29 ± 0.11 ppm.

During the Kepone feeding period the rats excreted a total of 15.78 ± 1.17% of the radioactivity ingested, almost exclusively in the feces (15.69 ± 1.15%). Table I shows the total excretion of radioactivity with urine and feces 25 days after discontinuing the Kepone-fortified diet. Again most of the radioactivity was found in feces. In contrast to the results of our HCB experiment (Richter et al., 1977), in the present study the concentration of radioactivity in feces was at no time significantly elevated in paraffin-treated rats. Therefore the differences in total radioactivity excreted with feces can be explained by the significantly higher output of feces in the paraffin group (11.87 ± 0.92 g/day) as compared to controls (10.01 ± 0.71 g/day). However, it cannot be determined whether this is the result of a higher biliary excretion and/or a lower degree of reabsorption of Kepone and/or metabolites. After long term exposure to Kepone, organs from female

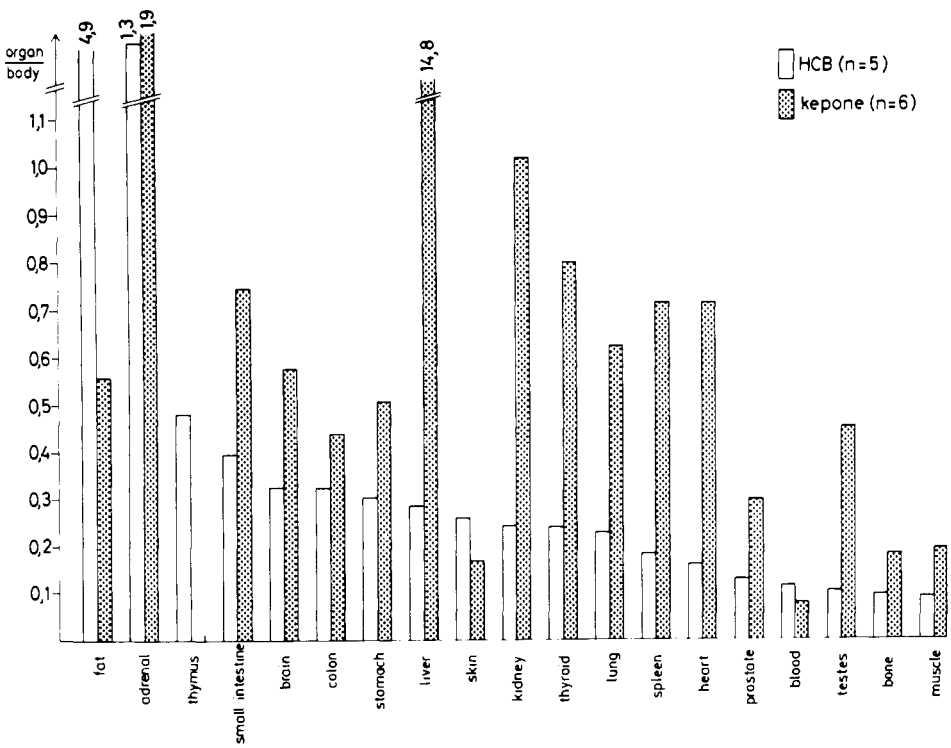

Figure 2. Ratio of ^{14}C concentration organs/body 35 (HCB) and 25 (Kepone) days after discontinuing the fortified diet. Total amount of ^{14}C in body was determined by the difference of total radioactivity consumed and excretion in feces and urine.

Table II. Elimination Constants and Half-lives ($t_{1/2}$)

treatment	t, day	A, μg	B, μg	k_1, day^{-1}	k_2, day^{-1}	$t_{1/2}$, day		SS[a]
control	1-25	28.28	157.57	0.149	0.022	4.66	31.36	3.49
paraffin		83.37	95.35	0.081	0.019	8.61	35.90	4.93

[a] SS, sum of squares.

Table III. Body Distribution of Kepone-^{14}C in Tissues of Rats (ppb Wet Weight)

tissue	control	paraffin
fat	170.7 ± 28.1	113.8 ± 25.6
adrenal[b]	570.2	445.3
small intestine	227.7 ± 26.6	158.1 ± 49.1
brain	176.0 ± 26.3	115.3 ± 21.7
colon	134.3 ± 29.9	108.9[a] ± 21.1
stomach	155.1 ± 25.1	126.1[a] ± 29.6
liver	4528.1 ± 715.2	3652.5 ± 567.3
skin	51.4 ± 7.0	40.6[a] ± 11.3
kidney	311.5 ± 63.8	179.5 ± 47.8
thyroid	244.9 ± 53.9	166.7 ± 43.3
lung	191.0 ± 34.7	118.7 ± 28.8
spleen	218.9 ± 27.7	108.8 ± 20.9
heart	218.4 ± 52.1	123.2 ± 31.8
prostate	91.1 ± 30.9	45.3 ± 7.2
blood	24.4 ± 4.2	17.1 ± 3.6
testes	139.2 ± 23.3	80.7 ± 26.4
bone	55.5 ± 7.7	33.3 ± 5.4
muscle	118.2 ± 21.8	67.9 ± 10.5

[a] Not significantly different from the controls. [b] Mixed samples.

mice lost 56% of the accumulated Kepone in 24 days (Huber, 1965). In the current investigation control rats excreted half of the radioactivity absorbed within 23.2 ± 5.1 days, treated rats within 16.9 ± 5.0 days. Figure 1 illustrates the elimination of radioactivity from rat body. The areas under the two curves are significantly different ($p < 0.05$). As can be seen from Table II the data of the elimination of Kepone from rat body fit quite well to a double exponential function of the following formula

$$C = Ae^{-k_1 t} + Be^{-k_2 t}$$

where C means the amount of Kepone in rat body in micrograms on day t of the elimination phase. Twenty-five days after discontinuing the Kepone-fortified diet the Kepone concentration in tissues was reduced in treated rats by a factor of 1.55 ± 0.25 compared with controls (Table III). As can be seen from Figure 2 the distribution pattern of Kepone is completely different from that of HCB (Richter et al., 1977). Kepone is apparently not stored in fatty tissue. The high concentrations of Kepone in liver and adrenals are of particular interest. Kepone is known to be a potential liver carcinogen (EPA Report, 1976) and to impair reproduction in mammals (Huber, 1965; Good et al., 1965). Recently, Harless et al. (1978) showed that Kepone consistently contained varying amounts of water bound at the site of the keto group. In no case, could all of the water be removed. The so-called "Kepone hydrate" is decomposed to the carbonyl form at temperatures required for vaporization of these compounds in the GC injection port. According to these results we cannot give any information about the degree of hydration of the Kepone taken up by the rats with the food. The formation of more hydrophilic derivatives of Kepone in the presence of water is possibly an explanation for the faster elimination, different distribution pattern, and the weak effect of paraffin treatment as compared to HCB (Richter et al., 1977). Whereas the half-lives, calculated by the two-compartment model, were not shortened in the Kepone experiment, with HCB a more than four times lower half-life in the postdistribution equilibrium phase was found. In contrast, with both compounds the first exponential term has a much greater importance in paraffin-treated rats. Provided that paraffin exerts the same effect in humans, this indicates that treatment of Kepone poisoning would be more useful in the first weeks after exposure.

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