0014-4754/96/010075-06$1.50 + 0.20 © Birkhäuser Verlag Basel, 1996

Acylfulvenes, a new class of potent antitumor agents

T. C. McMorris*,[a], M. J. Kelner[b], W. Wang[a], M. A. Diaz[a], L. A. Estes[b] and R. Taetle[c]

Departments of [a]Chemistry and [b]Pathology, University of California, San Diego, La Jolla (California 92093-0506, USA) and the [c]Division of Hematology, Arizona Cancer Center, 1501 N. Campbell Avenue, University of Arizona, Tucson (Arizona 95724, USA)

*Received 7 April 1995; accepted 21 April 1995***

Abstract. Acylfulvene, derived from the sesquiterpene illudin S by treatment with acid (reverse Prins reaction), is far less reactive to thiols than illudin S. However, it is reduced readily to an aromatic product, in the same way as illudin S. This may explain its greatly improved therapeutic index compared to that of the parent compound.

Key words. Acylfulvene; illudin S; sesquiterpene; antitumor agents.

Illudin S (**1**) and illudin M (**2**) are sesquiterpenes produced in cultures of the basidiomycete *Omphalotus illudens* (formerly *Clitocybe illudens*)[1,2]. Illudin S has also been isolated from closely related mushrooms by us[3] and by other workers[4,5]. The compounds are extremely toxic and are believed to be responsible for poisoning that occurs when *Omphalotus* is mistaken for an edible mushroom[6,7].

Illudins were evaluated for antitumor activity by the National Cancer Institute, in a variety of rodent tumor models, many years ago but were found to have a low therapeutic index, particularly in solid tumor systems[8]. Some time ago, we began an investigation of illudins to determine the reasons for their toxicity. We hoped that clarification of the mechanism of toxicity would enable us to design analogs which would be less toxic but still retain antitumor properties[9–12].

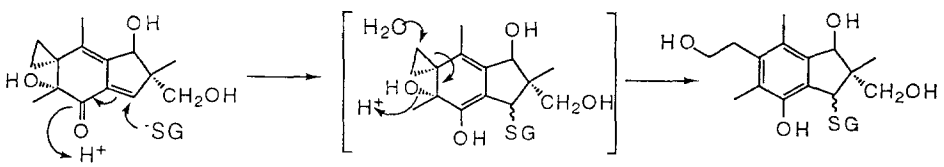
At low pH, illudins have been found to behave as bifunctional alkylating agents, but at physiological pH, they do not react with oxygen or nitrogen nucleophiles[9]. However, they react spontaneously with sulfur nucleophiles such as glutathione or cysteine, at an optimum pH of about 6, and toxicity to myeloid leukemia (HL 60) cells can be modulated by altering glutathione levels in cells[11]. The reaction of illudin S with glutathione (GSH) is illustrated in scheme 1. Michael-type addition to the $\alpha\beta$-unsaturated ketone gives a cyclohexadiene intermediate, an extremely reactive alkylating agent, which is rapidly converted to a stable aromatic product[12]. It is reasonable to assume that illudins will react similarly with enzymes containing thiol groups and this will contribute to their toxicity if vital enzymes, e.g., glyceraldehyde 3-phosphate dehydrogenase or ribonucleoside diphosphate reductase, are involved.

We have therefore sought illudin analogs which possess the key features, $\alpha\beta$-unsaturated ketone and cyclopropylmethyl carbinol, required to trigger alkylating action but which are less reactive to thiols. Two analogs of illudin M, deoxyilludin M (**3**) and dehydroilludin M (**4**) have been found to be less reactive to thiols at

Table 1. IC50 values for illudin analogs when tested in HL-60 cells*.

Compounds	nM
Illudin S or M (**1**, **2**)	3 ± 1 (0.8 ng/mL)
Deoxyilludin M (**3**)	31 ± 4 (7 ng/mL)
Dehydroilludin M (**4**)	296 ± 8 (73 ng/mL)
Acylfulvene (**5**)	415 ± 31 (90 ng/mL)
Bis-acylfulvene (**6**)	880 ± 150 (391 ng/mL)
6-Bromoacylfulvene (**7**)	410 ± 20 (120 ng/mL)
6-Iodoacylfulvene (**8**)	290 ± 10 (99 ng/mL)
6-Nitroacylfulvene (**9**)	180 ± 9 (47 ng/mL)

*For cytotoxicity tests the compounds were dissolved in DMSO (1 mg/mL stock solution) and the solutions diluted in 20% DMSO/phosphate buffered saline just prior to addition to cultures of HL 60 cells. Control cells received equal amounts of the DMSO/phosphate buffered saline. After incubation for 48 h the cells were washed, trypan blue was added, and the cells were counted. These values correlate closely with those determined by colony forming assay[8].



Scheme 1.

* Corresponding author: T. C. McMorris, Department of Chemistry, La Jolla; Fax +1619 534 4864.

** Unfortunately, publication was held up due to postal delay.

physiological pH and correspondingly less toxic than illudin M to HL 60 cells (table 1). However, dehydroilludin M was considerably more effective than illudin M when tested on Molt-4 (human myeloid leukemia) xenografts established in 4-week-old athymic Balb/c nu/nu mice[12]. These results have encouraged us to prepare further analogs and in this communication we report on the toxicity and antitumor activity of analogs for which we propose the name acylfulvenes.

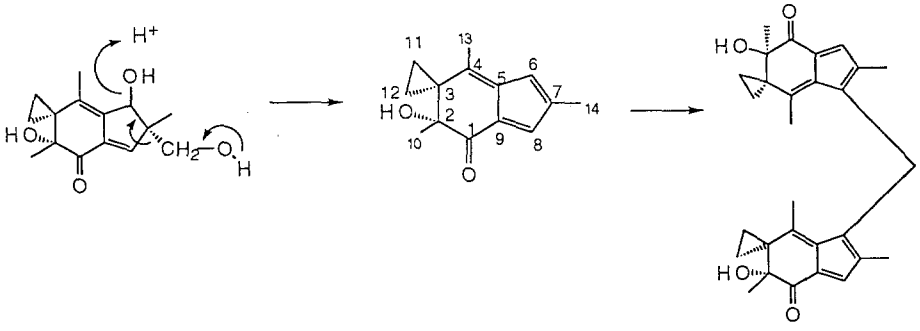
Results and discussion

Acylfulvene (**5**) is formed in a reverse Prins reaction when an aqueous solution of illudin S is acidified with dilute H_2SO_4 at room temperature (scheme 2). It reacts rapidly with the byproduct formaldehyde, in the presence of acid, to give bisacylfulvene (**6**). The structure of **6** has been confirmed by X-ray crystallographic analysis (fig. 1).

Several derivatives of acylfulvene have been prepared. For example, 6-bromoacylfulvene (**7**) was obtained by reacting **5** with N-bromosuccinimide in acetonitrile at 0 °C. The corresponding 6-iodoacylfulvene (**8**) was prepared by reacting **5** with I_2 and silver trifluoroacetate in methylene chloride at 0 °C. Treatment of acylfulvene with dilute nitric acid or nitronium tetrafluoroborate gave 6-nitroacylfulvene (**9**). All these derivatives of acylfulvene result from substitution at C-6 rather than C-8. This is consistent with the chemical shift of the C-6 proton (δ 6.43) compared to that at C-8 (δ 7.16) indicating greater electron density at the former carbon.

The fulvenes have been found to be less toxic than illudin S to HL 60 cells (table 1). As was found in the case of illudin M, deoxy illudin M and dehydroilludin M (ref. 12), the toxicity of acylfulvene correlates with reactivity to thiol.

Thus at pH 6.0, the pseudo first-order rate constant for reaction of **1** with a 10-fold excess of methyl thioglyco-



Scheme 2.

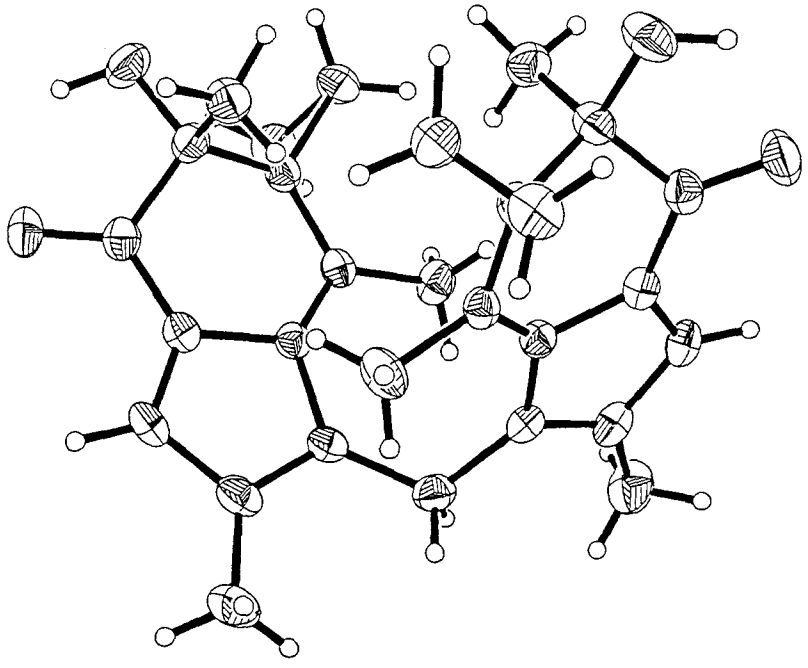
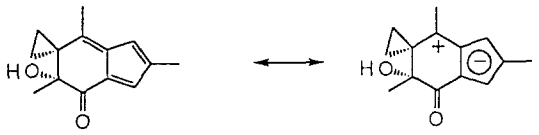


Figure 1. ORTEP view of X-ray molecular structure of bisacylfulvene **6**.



Scheme 3.

late, in methanol-water (1:19) at 25 °C, was $k = 6.5 \times 10^{-3}\ min^{-1}$ (the product has structure **10**). The corresponding value for acylfulvene was $k = 1 \times 10^{-4}\ min^{-1}$ (the product has structure **11**). This decrease in reactivity towards thiol, despite a more favorable steric situation[13], can be explained by the resonance structure shown (scheme 3).

The efficacy of acylfulvene as an antitumor agent far surpasses that of illudin S. In tests with HL 60 xenografts established in 4-week old athymic Balb/c nu/nu mice, tumor growth was greatly inhibited (>90%) in treated animals compared to control animals. The doses, 3 mg/kg IV, six doses in 14 days, were well tolerated by the animals. A dose-response effect was observed with lower doses.

Acylfulvene also inhibited growth of solid tumors. When tested on xenografts of human lung adenocarcinoma MV 522 (ref. 14), which is nonresponsive to conventional anticancer agents, substantial increase of the mean life span was observed as indicated in table 2. For comparison, *cis* platinum was also tested and found to have only a slight effect. Illudin S at a dose of 2.5 mg/kg IP 3 ×/week was extremely toxic, and all the animals died after three doses. No increase in life span was observed with sublethal doses (0.25 mg/kg 3 ×/week, 0.5 mg/kg 3 ×/week) of illudin S. Full details of these biological studies are reported elsewhere[15].

It appears that selective toxicity to tumor cells over normal cells is enhanced in acylfulvene, possibly as a result of decreased reactivity toward thiols. This raises the intriguing question of the mechanism of toxicity of acylfulvene in tumor cells. A possible answer is that bioreductive activation of the compound occurs giving a reactive intermediate which is a potent alkylating agent. Illudin S itself has recently been reported to be metabolized to aromatic products (**12** and **13**) by a rat liver cytosol preparation with NADPH in phosphate buffer containing $MgCl_2$ (ref. 16). The highly reactive cyclohexadiene intermediate (cf. scheme 1) behaves as a potent alkylating agent. Presumably, alkylation of macromolecules occurs as well and there is some evidence for this[8]. Therefore, illudins may also act as prodrugs and be activated by a bioreductive process somewhat reminiscent of the bioreductive activation of mitomycin C[17].

Acylfulvene might be activated similarly since it can be reduced readily to aromatic products in the same way as illudin S (ref. 1). For example, on catalytic hydrogenation (Pd/C), acylfulvene is rapidly converted to indanol (**14**) a result of addition of hydrogen to the conjugated ketone and cleavage of the cyclopropane ring.

When acylfulvene is dissolved in aqueous acetone with sodium chloride and acetic acid and zinc dust is added, rapid reaction occurs giving the chloroindenol **15.** Under the same conditions, illudin S is converted to a chloroindantriol which on heating with acid, readily undergoes reverse Prins reaction leading to isomeric chloroindenol **16**. In the zinc reactions, an electron is probably donated to the carbonyl oxygen followed by protonation at the δ-carbon for (**5**) and β-carbon for (**1**). Enzymatic reduction, however, might involve nucleophilic attack by hydride equivalent from NADPH at the β-carbon and protonation of the carbonyl oxygen.

Although acylfulvene (**5**) is not a natural product it is interesting to note the recent report of isolation of leaianafulvene (**17**), an orange-yellow pigment from the bright orange agaric *Mycena leaiana* (Berk.) Sacc. (ref. 18). This fulvene may well be derived biosynthetically from an illudin S-like precursor but with the opposite absolute configuration.

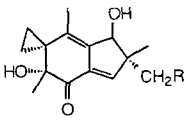
Experimental procedures

^1H and ^{13}C NMR spectra were obtained at 300 or 500 MHz and 75 or 125 MHz, respectively. Spectra were taken of solutions in $CDCl_3$ with Me_4Si as internal standard. High resolution mass spectra were determined at the University of Minnesota Mass Spectrometry Service Laboratory. Column chromatography was carried out with silica gel (Davisil 100–200 mesh and 230–425 mesh, Fisher Scientific). Analytical TLC was carried out

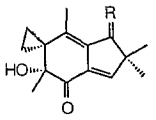
Table 2. Efficacy of acylfulvene in the human lung adenocarcinoma MV522 metastatic tumor model.

Drug	Dose*	Mean life span
DMSO/normal saline	IP 3 ×/wk (3 weeks)	100 ± 8%
Acylfulvene	6.4 mg/kg IP 3 ×/wk (3 weeks)	229 ± 50%
	12.8 mg/kg IV 3 × wk (3 weeks)	230 ± 21%
	12.8 mg/kg IP 3 ×/wk (3 weeks)	294 ± 48%
	6.4 mg/kg IV 1 ×/day (5 days)	226 ± 9%
Cis platinum	3.2 mg/kg IP 3 ×/wk (3 weeks)	137 ± 9%

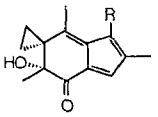
*MV522 cells[15] were injected subcutaneously at 10 million cells per animal and treatment was delayed for 7 days, until the tumors were palpable. The drug was administered intraperitoneally (IP) or by intravenous injection (IV).



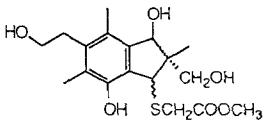
1 (R=OH)
2 (R=H)



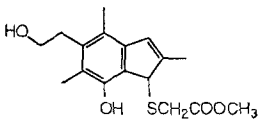
3 (R=2H)
4 (R=O)



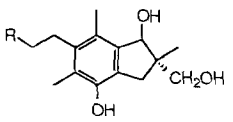
5 (R=H)
7 (R=Br)
8 (R=I)
9 (R=NO_2)



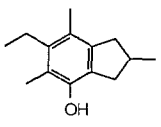
10



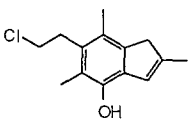
11



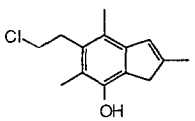
12 (R=OH)
13 (R=Cl)



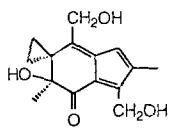
14



15



16



17

on Whatman 4410 222 silica gel plates. Reactions were routinely monitored by TLC.

Preparation of illudin S (1). This compound was isolated from cultures of *O. illudens* (formerly *C. illudens*) as described previously[1].

Acylfulvene (5). Illudin S (2 g, 9.2 mmol) was dissolved in 700 mL water and 236 mL 4N H_2SO_4 was added. The resulting solution, which was stirred at room temperature, gradually turned to an orange color and became cloudy during the first hour. An orange precipitate formed which adhered to the sides of the flask. After 20 h ethyl acetate (400 mL) was added to dissolve the precipitate. The aqueous phase was separated and further extracted with ethyl acetate (2 × 200 mL). The combined ethyl acetate extracts were washed with saturated $NaHCO_3$ solution (2 × 300 mL) to remove all acid, then with brine, and dried ($MgSO_4$). The mixture was filtered and the solvent removed leaving an orange residue which was chromatographed on silica gel with hexane-ethyl acetate (10:1) as eluent. Acylfulvene was eluted first (TLC:rf 0.35, hexane-ethyl acetate, 10:1); yield 800 mg (50%); mp 50–54 °C; ^{1}H NMR δ 0.72 (ddd, 1H) 1.09 (ddd, 1H), 1.30 (ddd, 1H), 1.50 (ddd, 1H), 1.38 (s, 3H), 2.00 (s, 3H), 2.15 (s, 3H), 3.95 (br s, OH), 6.43 (s, 1H), 7.16 (s, 1H); ^{13}C NMR 9.69, 14.3, 15.26, 16.94, 27.87, 37.30, 76.58, 120.70, 126.59, 136.13, 140.79, 142.76, 159, 197.84; HRMS m/z 216.1148 (M^+), calcd for $C_{14}H_{16}O_2$ 216.1151; λ_{max} (ethanol) 235, 325 nm (ε 16.6 × 10^3 and 8.3 × 10^3 respectively) with tailing to 480 nm; α_D (ethanol-606°, c = 0.078); ν_{max}($CHCl_3$) 3450, 1650, 1600 cm^{-1}.

Bisacylfulvene (**6**) was eluted next; yield 400 mg, mp 196–198 °C;[19] ^{1}H NMR δ (0.64) (ddd, 1H), 1.07 (ddd, 1H), 1.25 (ddd, 1H), 1.48 (ddd, 1H), 1.35 (s, 3H), 1.86 (s, 3H), 1.89 (s, 3H), 3.9 (br s, 1H), 4.07 (s, 1H), 7.09 (s, 1H). This spectrum is consistent with the symmetrical structure. The structure has been confirmed by an X-ray crystallographic analysis (fig. 1).

6-Bromoacylfulvene (7). Acylfulvene (60 mg, 0.28 mmol) was dissolved in 9 mL acetonitrile at 0 °C. N-bromosuccinimide (50 mg, 0.28 mmol) was added and the mixture was stirred at that temperature for 3.5 h during which the color of solution became darker. Water was added to the reaction mixture and the product was isolated by extraction with ether. The ethereal solution was washed with water and brine and dried over $MgSO_4$. Removal of the organic solvent gave a red gum which was purified by chromatography with hexane-ethyl acetate to yield 6-bromoacylfulvene as orange crystals (77 mg, 94%); mp 92–94 °C (recrystallized from ethyl acetate-hexane); ^{1}H NMR δ 0.75 to 1.55 (m, 4H), 1.40 (s 3H), 2.12 (s, 3H), 2.33 (s, 3H), 3.89 (s, 1H), 7.15 (s, 1H). MS m/z 296 (M^++2), 294 (M^+), 268, 266 (M^+-CH_2CH_2), 253, 251 (M^+-CH_2CH_2-CH_3), 215 (M^+-Br).

6-Iodoacylfulvene (8). To a solution of acylfulvene (60 mg, 0.28 mmol) in 15 mL methylene chloride was

added silver trifluoroacetate (63 mg, 0.29 mmol) and the solution was cooled to 0 °C. A solution of iodine (70.5 mg, 0.28 mol) in 8 mL CH_2Cl_2 was added dropwise at 0 °C. The mixture was stirred at that temperature for 3 h during which the color of the solution became dark red. The reaction mixture was then filtered through celite and eluted with ether. Concentration of the filtrate gave 6-iodoacylfulvene as a red gum (73 mg, 77%); ^{1}H NMR δ 0.76 to 1.54 (m, 4H), 1.38 (s, 3H), 2.14 (s, 3H), 2.36 (s, 3H), 3.87 (s, 1H), 7.16 (s, 1H). MS m/z 342 (M^+), 314 (M^+-CH_2CH_2), 299 (M^+-CH_2CH_2-CH_3), 296 (M^+-CH_2CH_2-H_2O), 215 (M^+-I). This fulvene appeared to be unstable on attempted chromatography with silica.

6-Nitroacylfulvene (9). Acylfulvene (99 mg, 0.46 mmol) was dissolved in methylene chloride (20 mL) and nitronium tetrafluoroborate (141 mg, 1.1 mmol) was added to the solution (nitrogen atmosphere). A dark brown precipitate formed; the mixture was stirred for 4 h, more nitronium tetrafluoroborate was added (53 mg) and stirring continued for 2 h. Water (5 mL) was added and the mixture was extracted with methylene chloride (3 × 25 mL). The combined extracts were washed with saturated $NaHCO_3$ solution, water, then dried over $MgSO_4$. Removal of solvent and chromatography of the residue with hexane-ethyl acetate gave the nitro compound (**9**) as a yellow solid (30 mg); ^{1}H NMR δ 0.90 (ddd, 1H), 1.23 (ddd, 1H), 1.50 (ddd, 1H), 1.69 (ddd, 1H), 1.46 (s, 3H), 2.02 (s, 3H), 2.34 (s, 3H), 6.97 (s, 1H). MS m/z 261 (M^+), 246 (M^+-CH_3), 244 (M^+-OH), 215 (M^+-NO_2).

Reaction of illudin S with methyl thioglycolate. Illudin S (50 mg, 0.19 mmol) was dissolved in tetrahydrofuran (1.5 mL) and water (15 mL) was added. To this solution was added methyl thioglycolate (0.4 mL, 4.5 mmol). The solution was kept overnight then extracted with ether. The extract was washed with brine then dried ($MgSO_4$). Removal of solvent and chromatography of the product gave **10** as a mixture of isomers which could not be separated. A sample of **10** was dissolved in tetrahydrofuran-water and a little dil. HCl was added. On heating the solution to boiling for 20 min, reverse Prins reaction occurred and the indenol **11** was formed which had the same properties as the compound obtained from reaction of acylfulvene with methyl thioglycolate (see below).

Reaction of acylfulvene with methyl thioglycolate. Acylfulvene (50 mg, 0.23 mmol) was dissolved in tetrahydrofuran (10 mL) and water (10 mL) and to this solution was added methyl thioglycolate (0.04 mL, 0.46 mmol). The solution was stirred overnight, more methyl thioglycolate (0.04 mL) was added, and the stirring continued for seven days with daily addition of methyl thioglycolate (0.04 mL, each time). The solution was then extracted with ethyl acetate and the extract was dried ($MgSO_4$) and the solvent removed. TLC showed some starting material, and one major product (**11**) which was isolated by chromatography with ethyl acetate-hexane; yield 15 mg, mp 96–100 °C; ^{1}H NMR δ 2.10 (s, 3H), 2.27 (s, 3H), 2.29 (s, 3H), 2.90 (dd, 2H), 2.98 (t, J = 7.2 Hz, 2H), 3.59 (s, 3H), 3.74 (t, J = 7.2 Hz, 2H), 4.19 (s, 1H), 6.58 (s, 1H).

Hydrogenation of acylfulvene. To a solution of acylfulvene (101 mg, 0.47 mmol) in acetone (14 mL) and water (26 mL) was added Pd/C (10%, 50 mg) and the mixture was subjected to hydrogenation at 42 psi at room temperature for 1.5 h. The catalyst was removed by filtration and the filtrate was extracted with ethyl acetate. The extract was dried ($MgSO_4$), concentrated and chromatographed with hexane-ethyl acetate to give the main product (**14**, 22 mg) with the following properties: mp 101°; ^{1}H NMR δ 1.09 (t, J = 7.5 Hz, 3H), 1.17 (d, J = 7.0 Hz, 3H), 2.15 (s, 3H), 2.20 (s, 3H), 2.45 (m, 2H), 2.60 (q, J = 7.5 Hz, 2H), 2.99 (m, 2H), 4.15 (br s, OH).

Reduction of acylfulvene. The fulvene (105 mg, 0.49 mmol) and sodium chloride (100 mg) were dissolved in water (1 mL) and acetone (10 mL). To this solution glacial acetic acid (6 mL) followed by zinc dust (0.66 g) were added and the mixture was stirred at room temperature for 1 h. The mixture was partitioned between water and ethyl acetate and the aqueous phase was separated and further extracted with ethyl acetate. The combined extracts were washed with saturated $NaHCO_3$ solution, brine, and dried over $MgSO_4$. After removal of solvent the solid product was purified by sublimation (100 °C, 0.1 mm Hg) yielding the chloroindenol (**15**, 40 mg) mp 145–145.5 °C; ^{1}H NMR δ 2.16 (s, 3H), 2.26 (s, 3H), 2.28 (s, 3H), 3.16 (t, J = 7.8 Hz, 2H), 3.22 (s, 2H), 3.52 (t, J = 7.8 Hz, 2H), 4.49 (s, 1H), 6.52 (s, 1H). MS m/z 238.0927 (M^++2), 236.0967 (M^+), 187.1123 (M^+-CH_2Cl). Calcd for $C_{14}H_{17}ClO$, 236.0969.

When illudin S (127 mg, 0.48 mmol) was reacted with zinc dust, acetic acid and sodium chloride in the same way as the fulvene, the expected aromatic chloroindantriol was obtained (98 mg). The crude product was dissolved in H_2O and the solution was heated with a drop of dil. HCl which resulted in reverse Prins reaction. The product was purified by sublimation (100 °C, 0.1 mm Hg) giving the isomeric chloroindenol (**16**, 41 mg) mp 135–136 °C; ^{1}H NMR δ 2.16 (s, 3H), 2.27 (s, 3H), 2.33 (s, 3H), 3.16 (t, J = 7.8 Hz, 2H). 3.20 (s, 2H), 3.53 (t, J = 7.8 Hz, 2H), 4.49 (s, 1H), 6.56 (s, 1H). MS m/z 238.0924 (M^++2), 236.0970 (M^+), 187.1123 (M^+-CH_2Cl).

Kinetics of reactions of illudin S and acylfulvene with methyl thioglycolate. A series of water-methanol (19:1) buffer solutions were prepared by using sodium acetate-acetic acid (for pH values in the range of 4.0–5.4), 2(N-morpholino)ethanesulfonic acid (MES, pKa 6.15, for pH values in the range 5.6–6.4), and piperazine-

N,N′-bis(2-ethanesulfonic acid) (PIPES, pKa 6.8, for pH values above 6.5). The pH of the solutions was measured with a pH meter. In all cases the buffer concentration was 50 mM. Illudin S and acylfulvene were dissolved in the buffer solutions to give in each case a concentration of 0.2 mM and to each 10 mL solution was added methyl thioglycolate (1.8 μL final concentration 2.0 mM). The reactions were allowed to proceed at room temperature (25 °C) and monitored by measuring the decrease of the long wavelength absorption band (λ_{max} 318 nm, for illudin S and λ_{max} 325 nm for acylfulvene) with time. For illudin S the pseudo first-order rate constant was greatest at pH 6.0 ($k = 6.5 \times 10^{-3}\ min^{-1}$) and decreased at lower or higher pH values. Acylfulvene gave a much lower rate constant which increased gradually over the pH range 6.2–5.0. At pH 6.0, $k = 1 \times 10^{-4}\ min^{-1}$.

Acknowledgements. This investigation was supported in part by grant 1-KT-02 from the Tobacco-Related Disease Research Program of the University of California. X-ray crystallographic analysis was carried out by Dr. Peter Gantzel, Department of Chemistry, University of California, San Diego.

1 McMorris, T. C., and Anchel, M., J. Am. chem. Soc. *85* (1963) 831.
2 McMorris, T. C., and Anchel, M., J. Am. chem. Soc. *87* (1965) 1594.
3 McMorris, T. C., Moon, S., and Ungab, G. J. nat. Products *52* (1989) 380.
4 Nakanishi, K., Ohashi, M., Tada, M., and Yamada, Y., Tetrahedron *21* (1965) 1231.
5 Matsumoto, T., Shirahama, H., Ichihara, A., Fukuoka, Y., Takahashi, Y., Mori, Y., and Watanabe, M., Tetrahedron *21* (1965) 2671.
6 Maretić, Z., Russell, F., and Golobic, V., Toxicon *13* (1975) 379.
7 French, A. L., and Garrettson, L. K., Clin. Toxic. *26* (1988) 81.
8 Kelner, M. J., McMorris, T. C., Beck, W. T., Zamora, J. M., and Taetle, R., Cancer Res. *47* (1987) 3186.
9 McMorris, T. C., Kelner, M. J., Chadha, R. K., Siegel, J. S., Moon, S., and Moya, M., Tetrahedron *45* (1989) 5433.
10 Kelner, M. J., McMorris, T. C., and Taetle, R., J. natl Cancer Inst. *82* (1990) 1562.
11 McMorris, T. C., Kelner, M. J., Wang, W., Moon, S., and Taetle, R., Chem. Res. Toxic. *3* (1990) 574.
12 McMorris, T. C., Kelner, M. J., Wang, W., Estes, L. A., Montoya, M. A., and Taetle, R., J. Org. Chem. *57* (1992) 6876.
13 Fahey, R. C., Myers, P. A., and DiStefano, D. L., Bioorganic Chem. *9* (1980) 293.
14 Varki, N. M., Tseng, A., Vu, T. P., and Estes, L. A., Anticancer Res. *10* (1990) 637.
15 Kelner, M. J., McMorris, T. C., Estes, L. A., Montoya, M. A., Starr, R., Samson, K., and Taetle, R., Cancer Res. (1995) in press.
16 Tanaka, K., Inoue, T., Kadota, S., and Kikuchi, T., Xenobiotica *22* (1992) 33.
17 Fisher, J. F., and Aristoff, P. A., Prog. Drug Res. *32* (1988) 441.
18 Harttig, U., Anke, T. Scherer, A., and Steglich, W., Phytochemistry *29* (1990) 3942.
19 Weinreb, S. M., McMorris, T. C., and Anchel, M., Tetrahedron Lett. *38* (1971) 3489.