was quantitative. The filtered solution was evaporated and the residue was recrystallized from aqueous methanol to give 18.7 g. (87%) of the colorless product, m.p. 140–142°.

Anal. Calcd. for $C_{14}H_{16}O_2$: C, 77.75; H, 7.46. Found: C, 77.68; H, 7.57.

Preparation of Penicillins.—The methods used are described in the previous paper of this series.[1] In almost all cases the chlorides of the side chain acids were coupled with 6-aminopenicillanic acid in aqueous acetone in the presence of sodium bicarbonate (method A), and the penicillins were isolated as their alkali metal salts. Departures from these procedures are reported in the footnotes to the Tables.

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New Antiviral Compounds with Considerable Activity *in Vivo*. IV. Aromatic α-Keto Aldehydes

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The antiviral activity in tissue culture, in chick embryo, and in mice of α-keto aldehyde derivatives of biphenyl, diphenylmethane, diphenylethane, stilbene, diphenyl ether, diphenyl sulfide, and diphenyl sulfone was studied. Several substances were found active in chick embryo against A-PR8 virus, in tissue culture against poliomyelitis virus, adenovirus, and vaccinia virus. All substances were active in mice against MHV3 virus and nonactive against Columbia SK virus. Some of them were also active in mice against A-PR8 virus. The biphenyl, diphenylethane, and diphenyl sulfide derivatives showed the best antiviral activity.

In previous papers the synthesis and the antiviral properties of 4-biphenylglyoxal, of 4,4′-bisbiphenylglyoxal, and of several derivatives[2–4] have been described. The glyoxal derivatives of biphenyl proved active as antiviral agents *in vivo* and also in human therapy.[5,6] It appeared interesting to synthesize additional substances formed from diglyoxals in several ring systems; some of these compounds were found to display good antibacterial activity.[7,8] Mono- and bis-α-keto aldehydes were introduced into the 4- and 4,4′-positions of diphenylmethane, diphenyl ether, diphenyl sulfide, diphenyl sulfone, diphenylethane, and of stilbene, and into the 2,2′-positions of biphenyl.

Experimental[9]

The aromatic α-keto aldehydes listed in Table I were prepared by two methods: (A) by oxidation of the corresponding aryl methyl ketones with selenium dioxide in aqueous dioxane; (B) by reaction of the corresponding α,α-dichloromethyl aryl ketones with hydrochloric acid. The keto aldehydes were isolated as hydrates or as monosodium bisulfite addition products. In a few cases the anhydrous α-keto aldehydes were obtained by vacuum distillation.

In order to confirm their structure, quinoxaline derivatives were prepared by condensation of *o*-phenylenediamine with the keto aldehydes. All the α-keto aldehydes reduced Tollen's reagent. 4-Acetyldiphenylmethane, 4-acetyldiphenylethane, 4,4-bisacetyldiphenylethane, 4,4′-bisacetylstilbene, 4-acetyldiphenyl ether, 4,4′-bisacetyldiphenyl ether, 4-acetyldiphenyl sulfide, and 4,4′-bisacetyldiphenyl sulfide were prepared by Friedel–Craft's reactions according to the literature.[10–15] 4-Acetylstilbene was prepared by the Meerwein reaction from 4-aminoacetophenone and cinnamic acid,[16] 2,2′-bisacetylbiphenyl by oxidation of 9,10-dioxo-9,10-dimethyldihydrophenanthrene,[17] and 4-acetyldiphenyl sulfoxide and 4-acetyldiphenyl sulfone by oxidation of 4-acetyldiphenyl sulfide with hydrogen peroxide.[18]

The α,α-dichloromethyl aryl ketones used for the preparation of the keto aldehydes according to method B were prepared by Friedel–Craft's reaction or by chlorination of the corresponding methyl aryl ketones.[8]

Preparation of α-Keto Aldehydes (Table I). A.—The aryl methyl ketone (0.1 mole), dissolved in 120 ml. of warm dioxane, was added to a solution of 0.15 mole of SeO_2 in 50 ml. of 30% aqueous dioxane at 50–60° (for the preparation of the bisketo aldehydes 0.3 mole of SeO_2 was used). The mixture was refluxed gently for 10 hr. The selenium which separated was filtered hot. The solution after standing for some days in the sunlight was filtered again and water was added in order to crystallize or precipitate the keto aldehyde hydrate which was filtered and recrystallized from aqueous dioxane or water. When this procedure was not convenient, the solvent was evaporated under reduced pressure and the residue was dissolved in anhydrous ethanol, filtered with charcoal, and again evaporated. The crude ethyl hemiacetal obtained was distilled under reduced pressure to prepare the anhydrous keto aldehyde.

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TABLE I

α-KETO ALDEHYDES, RCOCHO

Compound no.	R	Method	Yield, %	M.p., °C.	Solvent of cryst.[a]	Formula	% Calcd. C	% Calcd. H	% Calcd. S	% Found C	% Found H	% Found S
I	*o,o'*-$C_6H_4C_6H_4$	A	65	129-131		$C_{14}H_{10}O_4$	72.18	3.79		72.37	3.71	
						$C_6H_9O_4 \cdot 2NaHSO_3$			13.53			13.92
II	*p*-$C_6H_5CH_2C_6H_4$	A[b]	75	82-83	D-W	$C_{15}H_{12}O_2 \cdot H_2O$	74.36	5.83		74.91	5.59	
		B			A-W	$C_{15}H_{12}O_2 \cdot NaHSO_3$[f]	54.87	4.75	9.76	54.58	4.72	9.60
III	*p*-$C_6H_4CH_2C_6H_5$[c]					$C_{15}H_{14}O_2$	80.60	5.92		80.22	5.85	
		A	56	96-97	D-W	$C_{15}H_{14}O_2 \cdot H_2O$	71.98	6.29		75.10	6.08	
		B	80		A-W	$C_{15}H_{14}O_2 \cdot NaHSO_3$	55.96	4.40	9.94	56.12	4.73	9.36
IV	*p,p'*-$C_6H_5(CH_2)_2C_6H_5$	A	50	146-148	D-W	$C_{28}H_{34}O_4 \cdot 2H_2O$	65.11	5.49		65.33	5.10	
		B	75		A-W	$C_{16}H_{14}O_2 \cdot 2NaHSO_3$			12.66			12.85
V	*p*-$C_6H_5CH{=}CHC_6H_5$	A	45	141-146	D-W	$C_{16}H_{12}O_2 \cdot H_2O$	75.57	5.55		75.48	5.10	
						$C_{16}H_{12}O_2 \cdot NaHSO_3$	56.46	3.85	9.12	56.39	4.08	9.70
VI	*p,p'*-$C_6H_4CH{=}CHC_6H_4$	A	45	202	D-W	$C_{18}H_{12}O_4 \cdot 2H_2O$	65.85	4.91		65.96	5.31	
VII	*p*-$C_6H_5OC_6H_5$[d]	A	71			$C_{14}H_{10}O_3$	74.33	4.46		74.55	4.68	
		B	75	85	D-W	$C_{14}H_{10}O_3 \cdot H_2O$	68.84	4.95		68.82	4.71	
					A-W	$C_{14}H_8O_3 \cdot NaHSO_3$			9.71			9.78
VIII	*p,p'*-$C_6H_4OC_6H_4$	A	75	144-145	D-W	$C_{16}H_{10}O_5 \cdot 2H_2O$	60.38	4.43		60.21	4.52	
		B	64		A-W	$C_{16}H_{12}O_5 \cdot 2NaHSO_3$			13.08			12.90
IX	*p*-$C_6H_5SC_6H_5$[e]	A	40			$C_{14}H_{10}O_2S$	69.40	4.16	13.20	68.91	4.16	12.90
		B	65	69-70	E-P	$C_{14}H_{10}O_2S \cdot H_2O$	64.61	4.65	12.29	64.84	4.66	12.51
					A-W	$C_{14}H_{10}O_2S \cdot NaHSO_3$	48.56	3.20	18.51	48.53	3.71	18.46
X	*p,p'*-$C_6H_4SC_6H_4$	A	10	135-136	D-W	$C_{16}H_{10}O_4S \cdot 2H_2O$	57.49	4.22	9.59	57.23	4.01	9.19
		B	55			$C_{16}H_{10}O_4S \cdot 2NaHSO_3$			19.00			19.13
XI	*p*-$C_6H_5SOC_6H_5$	A	20	126-128	W	$C_{14}H_{10}O_3S \cdot H_2O$	60.87	4.38	11.61	60.82	4.46	11.77
XII	*p*-$C_6H_5SO_2C_6H_5$	A	75		A-W	$C_{14}H_{10}O_3S \cdot NaHSO_3$	44.55	2.93	16.96	44.75	3.08	17.10
XIII	*p,p'*-$C_6H_4SO_2C_6H_4$	B	22	153-154	W	$C_{16}H_{10}O_4S \cdot 2H_2O$	52.46	3.85	8.65	52.19	3.81	8.82

[a] D = dioxane, W = water, A = ethanol, E = ether, P = petroleum ether. [b] By this procedure it was not possible to isolate the pure α-keto aldehyde. We were nevertheless able to obain the monosodium bisulfite addition product pure. The yield of this substance was calculated. [c] B.p. 144–146° (0.2 mm.). [d] B.p. 136–140° (0.05 mm.). [e] B.p. 148–150° (0.1 mm.). [f] *Anal.* Calcd.: Na, 7.0. Found: Na, 6.80.

The keto aldehyde hydrates were obtained by treatment with water or aqueous dioxane and the corresponding hemiacetals were isolated by treatment with alcohols. When it was not possible to obtain the pure keto aldehyde either hydrated or anhydrous, we prepared the corresponding monosodium bisulfite addition product by filtering the separated selenium, and evaporating the solution *in vacuo*. The residue was dissolved in 500 ml. of ethanol and a solution of 52 g. of $NaHSO_3$ in 1000 ml. of water was added. The mixture was shaken for 4 hr. at 20°; the monosodium bisulfite addition products generally crystallized, but sometimes it was necessary to add water or ethanol. The products were filtered, washed with some water, and recrystallized from aqueous ethanol (see Table I).

B.—A solution of sodium methoxide, prepared from 4.6 g. (0.2 mole) of sodium and 100 ml. of anhydrous methanol (for the preparation of the bisketo aldehydes, 0.4 mole of sodium methoxide and 200 ml. of methanol was used), was added dropwise with stirring to a solution of 0.1 mole of the aryl α,α-dichloromethyl ketone in 150 ml. of anhydrous methanol at 40-50°. When all the sodium methoxide had reacted, the mixture was neutralized with methanolic hydrogen chloride. After cooling the separated sodium chloride was filtered. The solvent was evaporated under reduced pressure and the crude dimethyl acetal was dissolved in 150 ml. of acetic acid. The solution was filtered with charcoal, hydrolyzed by adding 20 ml. of 1 *N* hydrochloric acid and by warming the mixture for 2 hr. at 40–50°. Water was added in order to separate the α-keto aldehyde hydrate. The substances were filtered, washed with water, and purified by the procedures listed in Table I.

Quinoxalines.—These were prepared by heating a mixture of keto aldehyde with an equivalent amount of *o*-phenylenediamine in ethanol or in aqueous dioxane. After cooling, the quinoxalines were filtered and recrystallized. The yields, melting points, solvents of crystallization, and analytical data are summarized in Table II.

Pharmacology. I. Materials and Methods. (1) **Viruses.**—Different viruses were chosen for their size, for the type of nucleic acid, and for the fundamental biological properties (seat of cellular growth, quick and characteristic cytopathogenic effect (CPE), pathogenicity to mice, etc.). They were: poliomyelitis virus type 1, SK Columbia virus, adenovirus type 7, PR8 influenza A virus, MHV 3 mouse hepatitis virus, and vaccinia virus.

Poliomyelitis virus type 1, adenovirus type 7, and the vaccinia virus were tested in tissue cultures of monkey kidney; PR8 strain of influenza A virus was tested in chick embryo and in mice; MHW3 strain of hepatitis virus and SK Columbia virus were tested in mice only.

(2) **Tissue Cultures.**—Either primary tissue cultures of rhesus monkey kidney or 37 R.C. line of cercopitheous kidney were used. The growth medium employed was 5% hydrolyzed lactalbumin in Hanks' balanced saline solution plus 2-5% calf deactivated serum; as surviving medium, T 199 1% bovine albumin was used. The compounds were dissolved in Carbowax 200 on a boiling water bath for 15 min. Previously, the maximum tolerated dose (MTD) of each compound was determined, *i.e.*, the highest dose which does not provoke cytotoxic effects within 4 days.

(a) **Virucidal Tests.**—10 MTD of the compound was added to the viral stock and incubated at 36° for 30 min.; serial 10-fold dilutions of the mixture were effected and each dilution was inoculated into 4 or 5 tissue culture tubes and these were statically incubated at 36°. After 4 or 6 days from the inoculation the cytopathogenic effect (CPE) was read. Controls which contained no drugs were treated identically. End points were calculated according to the method of Read and Muench.[19] The activity of the compound was calculated from the significant difference between the titer of control and the titer of the treated virus.

(b) **Virustatic Tests.**—Groups of 4 or 5 tissue culture tubes were infected with 1,000–10,000 CPD_{50} of virus (cytopathogenic doses 50%); after adsorption for 2 hr. at 36°, the viral seed was removed and washed 3 times with Hanks' balanced saline solution. Two ml. of T 199 containing 1 MTD/ml. of the compound added to each tube was incubated statically at 36° for 22 hr. At the end of this period, the medium was removed and washed with Hank's balanced saline solution; 2 ml. of T 199 was added for each tube, and the tube stored at −40°. Controls which contained no drugs were treated identically. For the virus titration, the tubes were thawed, centrifuged and the supernatant fluids of each group of 4 tubes were serially diluted 10-fold. Each dilution was inoculated into groups of 4 or 5 tubes of monkey kidney cultures. The tubes were rotated and incubated at 36° for 6 days. The titer was calculated from the reading of the cytopathogenic effect. The activity of compounds was calculated according to the significant difference between the titer of control and the titer of the treated culture. Virustatic tests with Adenovirus were not done owing to technical difficulties.

(3) **Chick Embryo.**—Embryonated Leghorn hen eggs 9 or 10 days old were used. The compounds were suspended in buffered saline solution at pH 7.2–7.4, or dissolved in Carbowax 200. The dose chosen for the tested drugs has never shown any toxicity to eggs; therefore, the MTD was not calculated.

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TABLE II

QUINOXALINES

α-Keto aldehyde no.	Yield, %	M.p., °C.	Solvent of cryst.[a]	Formula	% Calcd. C	% Calcd. H	% Calcd. N	% Calcd. S	% Found C	% Found H	% Found N	% Found S
I	91.5	214–216	D–W	$C_{28}H_{18}N_4$	81.93	4.42	13.65		81.30	4.48	13.13	
II	90	122	A	$C_{21}H_{16}N_2$	85.11	5.44	9.45		85.18	5.78	9.39	
III	90	113	A	$C_{22}H_{18}N_2$	85.13	5.85	9.03		85.21	5.98	8.97	
IV	95	202–203	D	$C_{36}H_{22}N_4$	82.16	5.06	12.78		81.85	4.74	12.80	
V	71.5	164–165	A–W	$C_{22}H_{16}N_2$	85.69	5.23	9.09		85.33	5.28	8.81	
VI	67	252–253	D–W	$C_{30}H_{20}N_4$	82.54	4.62	12.84		82.08	4.72	12.74	
VII	98	117–118	A	$C_{20}H_{14}N_2O$	80.51	4.73	9.39		80.26	4.62	9.24	
VIII	95	170–171	D–W	$C_{28}H_{18}N_4O$	78.85	4.25	13.14		78.96	4.36	13.30	
IX	90	134–135	A	$C_{20}H_{14}N_2S$	76.42	4.49	8.91	10.18	76.30	4.64	8.88	10.20
X	90	185–186	D–W	$C_{28}H_{18}N_4S$	76.00	4.10	12.66	7.23	76.34	3.94	12.61	7.35
XI	76	167–168	A	$C_{20}H_{14}N_2OS$	72.72	4.27	8.48	9.69	72.62	4.42	8.20	9.68
XII	98	196	D–W	$C_{20}H_{14}N_2O_2S$	69.36	4.07	8.09	9.24	69.55	4.25	8.06	9.20
XIII	98	280	D–W	$C_{28}H_{18}N_4O_2S$	70.82	3.82	11.81	6.75	70.91	3.93	11.72	6.70

[a] D = dioxane, W = water, A = ethanol.

(a) **Virucidal Test.**—To the PR8 influenza A virus diluted 1:10 in buffered saline solution, 10^{-5} g./mole of compound was added. The EID_{50} (50% egg infection dose) should be at least 10^{-6}. The mixture was kept in water bath at 36° for 30 min. One-tenth ml. per egg of 10-fold dilutions of the mixture were then inoculated into groups of 5 eggs. The eggs were incubated at 36° for 40 hr. and the allantoic fluid of each egg was titrated for the EID_{50}. Controls which contained no drugs were treated identically. The virucidal activity was calculated according to the significant difference between the control virus titer and the treated virus titer.

(b) **Virustatic Tests.**—Groups of 4 eggs were inoculated in the allantoic sac with 0.1 ml. of serial 10-fold dilutions of PR8 strain of influenza A virus. After 1 hr. adsorption at 37°, 0.1 ml. of a dilute solution of the compound containing 1 egg dose was inoculated into the allantoic sac (10^{-5} g./mole). After incubation for 40 hr. at 35° the allantoic fluid of each egg was titrated for the EID_{50} titer. Controls which contained no drugs were treated identically. The virustatic activity of the compound was calculated according to the significant difference between the control virus titer and the treated virus titer.

(4) **Infection in Mice.**—Swiss albino mice weighing 12–15 g. obtained from ARSAL of Rome were used. The mouse diet was supplied by the same company. The temperature of the cages was kept at 18–20°. The compounds were tested on groups of 10 mice. The treatment was effected with 10^{-3} g./mole/kg./day of the compound finely suspended in buffered saline solution. The dose of the compound (0.1 ml.) was administered by stomach intubation twice daily. The LD_{50} was effected orally.

(a) **PR8 Strain of Influenza A Virus.**—An influenza virus strain adapted to mice was used and the viral stock was prepared with lungs taken from the live animals 3–4 days after intranasal infection. No stock was used if it contained less than 10^{-3} LD_{50}/0.5 ml. The animals were infected intranasally, under ether anesthesia with 25–100 LD_{50} (LD_{50} for mice on the tenth day from infection). The treatment was started 24 hr. before infection and continued till 5 days after infection, when all surviving animals were sacrificed and examined for pulmonary involvement. The lung suspension, 1:10wt./v. in broth for each animal, was made. After cold centrifugation the supernatant was collected. The pulmonary suspensions of each animal serially diluted 10-fold were titrated in embryonated eggs. The controls which contained no drugs were treated identically. The evaluation of the activity of the compounds was calculated according to the significant difference between the average viral titer of the control mice and that of treated mice.

(b) **Craig Strain of Hepatitis Virus MHV3.**—The animals were infected by subcutaneous injection of 60 LD_{50}/0.1 ml. A 10% triptose broth (wt./v.) suspension of livers of mice, sacrificed 4–5 days following infection, was used as viral stock. Only viral stocks containing more than $10^{2.5}$ LD_{50}/0.1 ml. were used. The treatment was started 24 hr. before infection and continued until 6 days after infection, when the surviving mice were sacrificed by beheading and the plasma of each animal was collected. In plasma, the sorbitol-dehydrogenase activity (SDA) was determined according to Gerlach.[20] Coltorti, *et al.*,[21] demonstrated that the variation of the SDA can be taken as an index of the infectious process and that the α-ketoaldehydes do not modify the SDA in uninfected mice. The controls which contained no drugs were treated identically. The evaluation of the activity of the compounds was calculated according to the significant difference between the average titer of the SDA in controls and the average titer in treated mice.

(c) **S. K. Columbia Virus.**—The animals were infected by intraperitoneal injection of 10 LD_{50} in 0.1 ml. The viral stock was prepared with suspensions of brains of infected and paralyzed mice. Viral stock containing 10^3 LD_{50}/0.1 ml. only was used. Treatment was started 24 hr. before infection and continued for 5 days after, recording paralyzed and dead mice. At the end the surviving animals were sacrificed and tested for the presence of hemagglutinin in the brain. The activity was calculated as % survival of healthy animals.

II. Statistical Methods. (a) **Virucidal and Virustatic Tests in Tissue Cultures and in Embryonated Eggs.**—The σ of control titers was calculated. Only a difference between treated groups and controls superior to 2-σ was considered significant.

(b) **Test in Mice Infected with Influenza Virus.**—Student's "t" was calculated, attributing conventionally to dead animals the average viral titer of the surviving controls. We excluded all experiments showing more than 10% of uninfected animals. A probability equal or inferior to 0.05 was considered significant.

(c) **Test in Mice Infected with Hepatitis Virus MHV3.**—Student's "t" was calculated according to a method which takes into account the dead and the surviving animals separately. To this end we determined the arithmetic mean (658.9 units/ml.) and the deviance ($\Sigma (x - \bar{x})^2 = 523{,}107.88$) of SDA of 12 controls dead within 6 days from infection. This value was given to the treated mice dead before the end of the experiment. Taking this into account, the "t" was calculated using the following formula

$$t = \frac{\left(\frac{N_t}{N'} - \frac{N_c}{N}\right)\bar{m} + \frac{N_c}{N}\bar{c} - \frac{N_t}{N'}\bar{t}}{\sqrt{\frac{D_m + D_c + D_t}{(N_m - 1) + (N_c - 1) + (N_t - 1)}\left(\frac{\left(\frac{N_t}{N'} - \frac{N_c}{N}\right)^2}{N_m} + \frac{N_c}{N^2} + \frac{N_t}{N'^2}\right)}}$$

where

N_m = number of controls dead within 6 days from infection
$\bar{m}$ = arithmetic mean of the values of SDA determined in the 12 control mice dead within 6 days from infection

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TABLE III

ANTIVIRAL ACTIVITY OF α-KETO ALDEHYDES

Compound no.	Embryonated eggs: Virucidal activity A-PR8	Embryonated eggs: Virustatic activity virus	Tissue culture: MDT,[a] γ-ml.	Tissue culture: Virucidal activity: Polio type 1	Tissue culture: Virucidal activity: Adeno type 7	Tissue culture: Virucidal activity: Vaccino	Tissue culture: Virustatic activity: Polio type 1	Tissue culture: Virustatic activity: Vaccino	Mice: LD_{50} mg./kg.	Mice: A-PR8 virus	Mice: MHV3 Craig virus	Mice: Columbia S.K. virus, % survival healthy animals[b]
I	+ + + + + + ▲[f]	0	25	0	0	0	0	0	1500	0.4	237▲	0
I-bis[c]	+ + + + + + ▲	+ ▲	6.2	0	0	0	0	± +	750	2.8▲	173▲	10
II-bis	+ + + + ▲	± + ▲	25	0	±	± ± ▲	± + ▲	0	1100	0.7	0	0
III	± ± ± ▲	0	12.5	+ ▲	+	± + ▲	0	−	600	2.2▲	5	0
III-bis	+ + + + ▲	0	25	0	± + ▲	+ ▲	±	± + ▲	750	0	444▲	0
IV	+ + + + ± + ▲	+ ▲	50	0	+ + ▲	± + ▲	0	0	>1500		361▲	0
IV-bis	+ + + + + ▲	+ + ▲	50	0	+ ± ▲	+ + ▲	+	0	>1500	1.7	173▲	0
V	+ ▲	0	25	0	0	± ▲	0	0	>1500		182▲	0
VI	+ + + + ▲	+	25	+ ▲	+	+ + ▲	0	0	>1500		191▲	0
VII	+ + + + + ▲	+ ± + + + ▲	6.2	0	0	+ + + + ▲	0	±	750		359▲	0
VII-bis	+ + + + ▲	0	12.5	0	0	6	0	0	750	1.6	102▲	0
VIII	+ + + + + ▲	+ + + + + ▲	25	0	+ + ▲	+ ± ± ▲	0	0	>1500	0.3	55▲	0
VIII-bis	+ + + + ▲	+ + ▲	50	0	± + ▲	+ ▲	0	+ +	>1500	1.4	213▲	0
IX	+ + + + ▲	0	50	0	+ ± ▲	+ + + ▲	0	0	>1500	2.3▲	30	0
IX-bis	+ + + + ▲	0	50	0	0	0	0	±	>1500	0	182▲	0
X	+ + + + + + ▲	+ ▲	50	0	+ + ▲	− − ▲	− + + ▲	− + ▲	>1500	3.0▲	145▲	0
X-bis	+ + + + ▲	+ ▲	50	0	+ + ▲	+ + ▲	0	+ +	>1500	0	382▲	0
XII	+ + + ▲	+ ▲	50	0	0	0	0	±	>1500		185▲	0
XIII	+ + + + ▲	+ + ▲	100	0	0	0	0	0	>1500	1.0	0	0
XIV[d]	+ + + + ▲	+ ▲	12.5	0	0	+ + + ▲	+	0	1500	0.5	151	0
XIV-bis	+ + + ▲	+	12.5	0	0	± + ▲	0	−	1200	2.3▲	360▲	10
XV[e]	+ + + + + + ▲	+ + ▲	50	+ + + ▲	+	+ ± + − ▲	0	−	>1500	0	316▲	0
XV-bis	+ + + + + + ▲	± + + ▲	50	0	+	+ + + + ▲	0	±	>1500	2.5▲	109▲	10

[a] Maximal tolerated dose. [b] % Survival healthy animals with hemoagglutination titer equal to zero. [c] Sodium bisulfite addition product. [d] 4-Biphenylglyoxal. [e] 4,4′-Bisbiphenylglyoxal. [f] ▲ Indicates statistical validity. The + sign corresponds to log unit.

D_m = deviance of values determined in the 12 control mice dead within 6 days from infection
N = number of controls
N_c = number of surviving controls
D_c = deviance of values in surviving controls
$\bar{c}$ = arithmetic mean of values in surviving controls
N' = number of treated mice
N_t = number of surviving treated mice
$\bar{t}$ = arithmetic mean of values in surviving treated mice
D_t = deviance of values in surviving treated mice

A probability equal or inferior to 0.05 was considered significant.

Discussion

Table III shows that all these keto aldehydes have marked virucidal activity against A-PR8 virus in embryonated eggs. Most of these compounds in the same system have virustatic activity against A-PR8 virus. In tissue cultures all the keto aldehydes, except I, I-bis, VII-bis, IX-bis, XII, and XIII, have virucidal activity against vaccinia virus. III-bis, IV, IV-bis, VIII, VIII-bis, IX, X, and X-bis show virucidal activity against adenovirus type 7. Only compounds XV, III, and VI show virucidal activity against polio virus type 1. As for virustatic activity in tissue cultures, only II-bis and X are active against type 1 poliomyelitis virus, and II-bis and X against vaccinia virus.

In mice all α-keto aldehydes are inactive against SK Columbia virus; almost all of them, except II-bis, III, IX, and XIII, are active against MHV_3 virus. Among the compounds tested in mice against A-PR8 virus, XV-bis, XVI-bis, I-bis, III, IX, and X proved active.

It is interesting to notice that the hydrated α-keto aldehydes and the corresponding monosodium bisulfite addition products behave biologically as two classes of different compounds and in some instances a selective activity against the viruses can be observed. Thus, monosodium bisulfite addition products of the biphenyl (XIV-bis, XV-bis, and I-bis) derivatives are mostly active in mice against A-PR8 virus. By contrast, the hydrated keto aldehydes of diphenylethane and of diphenyl sulfide (II, IX, and X) are active against A-PR8 virus.

From the data of this Table we can conclude that the α,β-dicarbonyl radical leads to antiviral activity but this activity is evident in tests *in vivo* only when the radical is joined to a suitable supporting moiety. The diphenyl sulfide derivatives are the most interesting ones because they have an activity in all the three systems tested.

As previously observed for the antibacterial activity of guanylhydrazone[8] and halomethyl aryl ketone derivatives,[9] in this series of glyoxals also, the biphenyl, diphenylethane, and diphenyl sulfide derivatives show more pronounced antiviral activity.

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