Virucidal Properties of Dimethyl Sulfoxide

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Dimethyl sulfoxide (DMSO) has been shown to possess bacteriostatic, bacteriocidal, and fungicidal properties (2, 3). The effect of DMSO on other microorganisms, such as the viruses, has

TABLE	1.	Effect	of DM	SO	(80%,	v/v)	on
		infect	tivity of	' viru	ses s		

	Treatment ^a					
Virus	Reagent	Exposure (min)	Infectivity titer 6.50 N 5.75			
R-8	Buffer ^b	30 60 30 + dialysis				
	DMSO	5 10 30 60 30 + dialysis	N N N N			
Ja p 305	Buffer DMSO	30 30	4.6 N			
NDV	Buffer DMSO	30 30	7.4 N			
SFV	Buffer DMSO	30 30	4.4 N			
vv	Buffer DMSO	30 30	4.38 N			
T ₂	Tris buffer DMSO (in Tris)	30 30	16.8 × 107 N			

^a Virus exposure to reagent at 25 C for indicated interval of time. Infectivity values are positive logarithms of titers per ml except for T₂ (plaques/ml); units are ELD₅₀/ml, except SFV (MLD₅₀/ml) and T₂ (plaques/ml); N = $<10^{1.0}$ ELD₅₀ or MLD₅₀/ml, or no plaques (T₂).

^b Buffer = M/15 Sorensen's buffered saline, unless indicated otherwise.

not been reported. Therefore, we were prompted to initiate the present study.

Four RNA viruses, influenza A (PR-8), influenza A_2 (Jap. 305), Newcastle disease (NDV), Semliki Forest (SFV), and two deoxyribonucleic

acid (DNA) viruses, vaccinia (VV), and *Escherichia coli* phage (T_2), were suspended in appropriate concentrations of pharmaceutical-grade DM-SO (Crown Zellerbach Corp., San Francisco, Calif.), in M/15 Sorensen's buffered saline (*p*H 7.2), or in tris(hydroxymethyl)aminomethane (Tris)- buffered saline (*p*H 6.8) at 25 C for specified periods of time. Residual infectivity was assayed as follows: (i) in 9-day-old embryonated hen's eggs, allantoic route, 0.2 ml/egg (PR-8, Jap. 305, NDV); (ii) in CFI-S female mice, 16 to 18 g, intraperitoneal route, 0.5 ml/mouse (SFV); (iii) in 9-day-old eggs, yolk sac route, 0.2 ml/ egg (VV); or (iv) by plaque assay on 2.5% Mac-Conkey agar at 37 C for 18 hr (T₂).

 TABLE 2. Effect of various concentrations of DMSO
 on influenza A (PR-8) infectivity^a

Treatment with PR-8 + DMSO (%)	Residual infectivity (ELD 50/ml)		
80	N		
70	N		
60	5.38		
50	6.62		
40	6.50		
Buffer	6.62		

^a Virus exposure to reagent at 25 C for 30 min. Infectivity values are positive logarithms of titers/ml; $N = \langle 10^{1.0} \text{ ELD}_{50}/\text{ml}.$

At a concentration of 80%, DMSO inactivated the infectivity of every virus tested (Table 1). In the case of PR-8, the inactivation occurred rapidly (<5 min) and was concentration-dependent (Table 2). Overnight dialysis after exposure of PR-8 to 80% DMSO could not restore infectivity. No significant change in infectivity was observed at DMSO concentrations below 50%.

The effect of DMSO on viral hemagglutinating activity (HA) was determined with the aid of two viruses, PR-8 and Polyoma (PV). A fixed volume of each virus was mixed with sufficient quantities of DMSO and Sorensen's buffered saline to achieve selected final concentrations of DMSO that ranged from 10 to 80%. The mixtures were allowed to stand at 25 C for 30 min; they were

Expt	Tı	Residual		
	virus	DMSO (%)	HA (ml)	
1	(PV	80	40	
		70	640	
		60	1280	
		Buffer	1280	
	PR-8	80	40	
		70	20	
:		60	20	
		50	320	
		Buffer	320	
2	PV	80	8	
		80 + dialysis	16	
		Buffer	128	
		80 + dialysis	128	
	PR-8	80	0	
		80 + dialysis	0	
		Buffer	128	
	11	80 + dialysis	128	

 TABLE 3. Effect of DMSO on hemagglutinating activity of polyoma and influenza A (PR-8) viruses

^a Virus exposure to reagent at 25 C for 30 min; overnight dialysis after exposure, as indicated.

then subdivided, and samples were titrated for HA activity immediately and after overnight dialysis against distilled water. For HA titration, twofold serial dilutions were followed by the addition of 0.5% washed guinea pig erythrocytes. In both instances, HA activity was adversely affected, but in a differential manner. Whereas the HA activity of PV was greatly reduced by exposure to 80% DMSO, it was relatively unaffected by lower concentrations (Table 3). By contrast, exposure of PR-8 to 60% DMSO was sufficient to denature all titratable activity. Overnight dialysis could not restore the HA activity of either virus.

Experiments designed to study the chemotherapeutic value of DMSO have shown the compound to possess no beneficial effect when administered parenterally to mice infected with influenza PR-8 or with SFV at nontoxic levels.

In addition to the multitude of uses suggested for DMSO (1), we would like to add the following uses, based partly on the fact that DMSO at concentrations of 30 to 50% is both bacteriocidal and fungicidal (2): (i) as an aid in the isolation of enteric or respiratory viruses from animal hosts [contaminating bacteria that are often found in sputum or stool samples can be eliminated by the use of a differential concentration of DMSO (about 50%), thus allowing the selective isolation of viruses; and (ii) for sterilization of inanimate objects where autoclaving or other means cannot conveniently be applied.

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