

HIGH PERFORMANCE LIQUID CHROMATOGRAPHY

No. L223

Simultaneous Analysis of 13 Synthetic Antibacterial Agents

Nowadays livestock and marine products like eggs, fishes, and meat which are served on table almost every day are mostly products obtained from domestic animals and cultured products. Moreover, the ratio of imported foods is on an increase. Under the circumstances, fact-finding survey on the safety of foodstuffs is being carried out at inspection stations, which has come to attract much attention.

Among feeds used for raising cattle or breeding fishes and among animal medicines, there are some which are harmful to the human body. Accordingly, the Ministry of Health and Welfare, the Japanese government, has designated 13 compounds contained in synthetic antibiotic agents as target compounds of "Monitoring Test of Harmful Residual Substances in Livestock and Marine Products", and has notified

prefectures and Metropolitan Tokyo to the effect that after 1991, monitoring should be carried out by unified methods using HPLC throughout the country.

Introduced here is an example of high sensitivity, simultaneous, rapid analysis for the 13 compounds contained in synthetic antibiotic agents by the application of wavelength programming that permits setting of optimum wavelenths for respective compounds and gradient elution by the LC-10AD pumps that permits complete separation of the 13 compounds in 30 minutes. Furthermore, the accuracy and reliability of qualitative measurement can be enhanced by obtaining absorption spectrum of each compound under the analytical conditions of Table 1 by the SPD-M6A photodiode array detector.

■ Analytical Conditions and Chromatograms of Standard 13 Synthetic Antibiotic Agents

Table 1 Analytical Conditions

Apparatus	: LC-10A DGH-1
Column	: L-Column
Mobile Phase	: A→B Gradient Elution Method
	A : 10mM(Sodium) Phosphate(pH2.6) ...85
	Acetonitrile ...15
	B : 100mM(Sodium) Phosphate(pH6.1) ...40
	Acetonitrile ...60
Flow Rate	: 1.0mL/min
Temperature	: 40°C
Detection	: SPD-10A 268nm
	(wavelength programmed with SCL-10A)

Table 2 Time program

#	TIME	FUNC	VALUE	#	TIME	FUNC	VALUE
0	0.01	MAKE A		10	24.00	WAVE A	283
1	4.00	WAVE A	285	11	25.00	B.CONC	100.0
2	5.00	B.CONC	0.0	12	28.00	WAVE A	350
3	6.00	WAVE A	227	13	30.00	B.CONC	100.0
4	6.85	WAVE A	267	14	30.10	B.CONC	0.0
5	8.00	WAVE A	366	15	45.00	STOP	
6	10.00	WAVE A	275				
7	16.00	WAVE A	265				
8	20.00	B.CONC	42.0				
9	21.00	WAVE A	258				

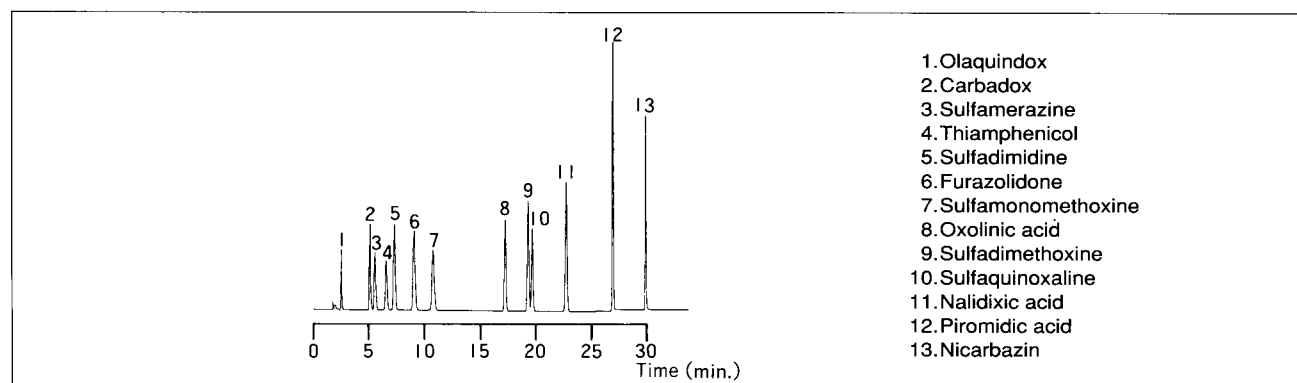


Fig.1 Chromatogram of Standard Mixture

Concentration of each compound contained in the standard sample was 20 ~ 50 ppm; Olaquinox, Carbadox, and Nicarbadin were dissolved in

acetonitrile and the others were dissolved in 50% aqueous solution.

■ Spectra of 13 Synthetic Antibacterial Agents

Shown below are absorption spectra of respective standard samples under the analytical conditions. The abscissa represents a wavelength range (200nm~380nm).

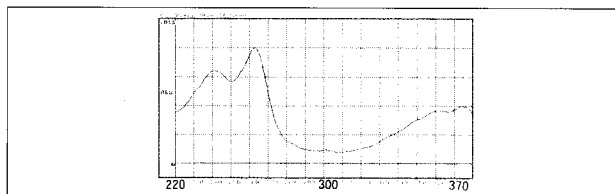


Fig.2 Olaquinox

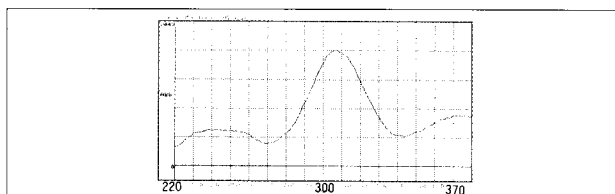


Fig.3 Carbadox

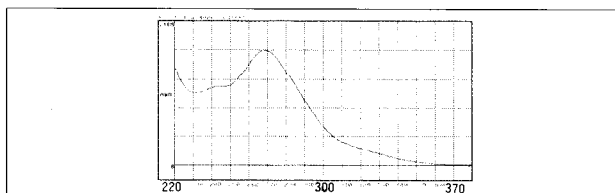


Fig.4 Sulfamerazine

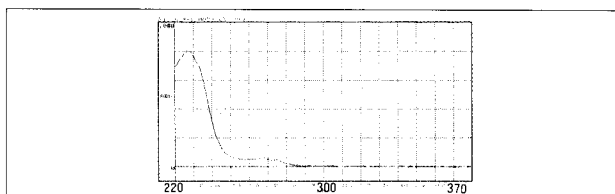


Fig.5 Thiamphenicol

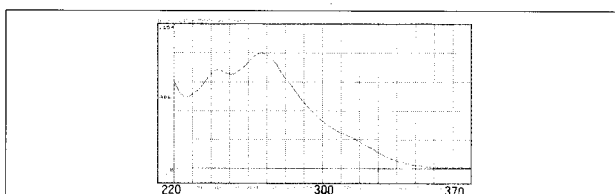


Fig.6 Sulfamidine

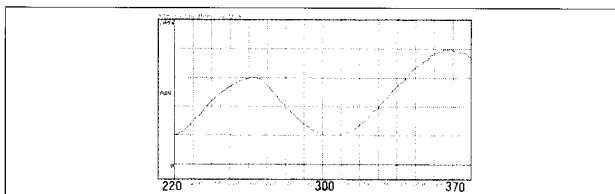


Fig.7 Furazolidone

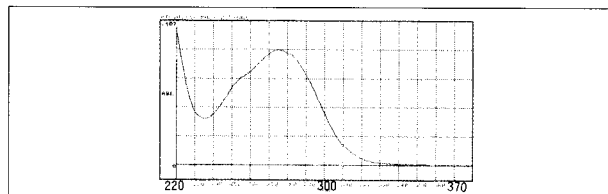


Fig.8 Sulfamonomethoxine

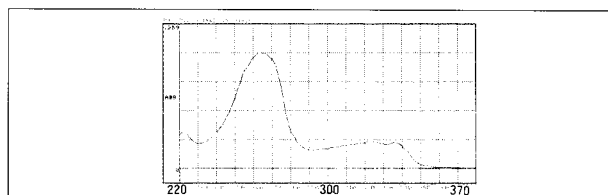


Fig.9 Oxolinic acid

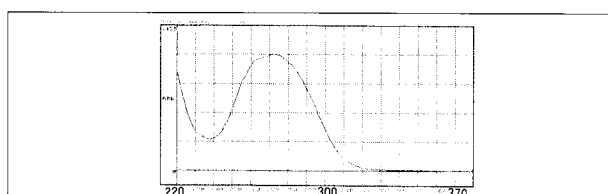


Fig.10 Sulfadimethoxine

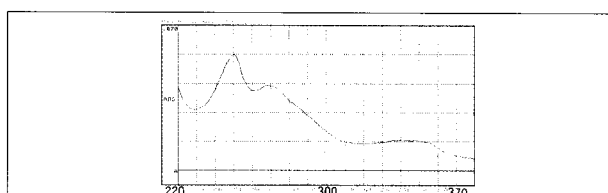


Fig.11 Sulfaquinoxaline

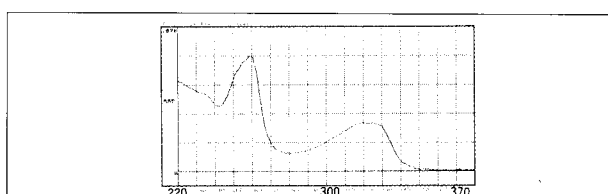


Fig.12 Nalidixic acid

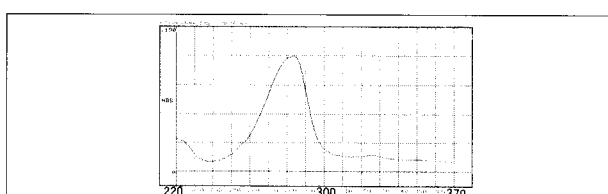


Fig.13 Piromidic acid

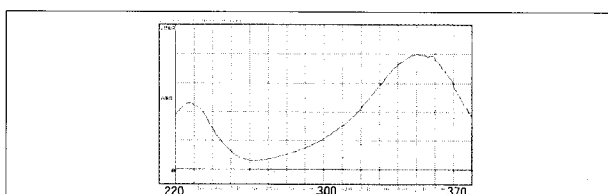


Fig.14 Nicarbadin

