

ELECTROPHORETIC STUDY OF ANTIRABIES SERUM
PRACTICAL APPLICATION

by

H. Mirchamsy, A. Korour and M. Bahmanyar*

In the immunization of animals for the preparation of antirabies serum, refractory animals must be detected at an early stage if loss of time and money is to be avoided.

Even with the mule - which is practically never refractory - it is preferable to know and keep a check on the value of each animal producing serum so that any mediocre producers may be eliminated, especially when, as in our case, an attempt is being made to obtain serum of exceptionally high titre.

With the standard serum - virus neutralization test, this work would call for a considerable number of mice, and for this reason we have tried to develop an indicative test which would at least make it possible to reduce the number of mice required for the titration.

★ ★ ★

In the course of the electrophoretic study we made in an attempt to define the "antirabies fraction" of our sera by the "paper" method which we have described, (1) we found that a fraction develops during hyperimmunization which is slight or absent in the mule in its normal state. This

★ WHO/Rabies/128,27 November 1959

1 Mirchamsy, H., Sohrab, V. & Korour, A. (1958) *Rev. Immunol. (Paris)*, 22, 553

fraction is found between the beta 1 and gamma globulins and is known as fraction T or beta 2 globulin. It increases during rabies immunization until it represents 17-25 per cent of the serum fractions in the hyperimmunized mule.

Electrophoresis also showed that there was a marked decrease in albumin as well as a slow and very slight decline in the gamma globulin fraction.

★ ★ ★

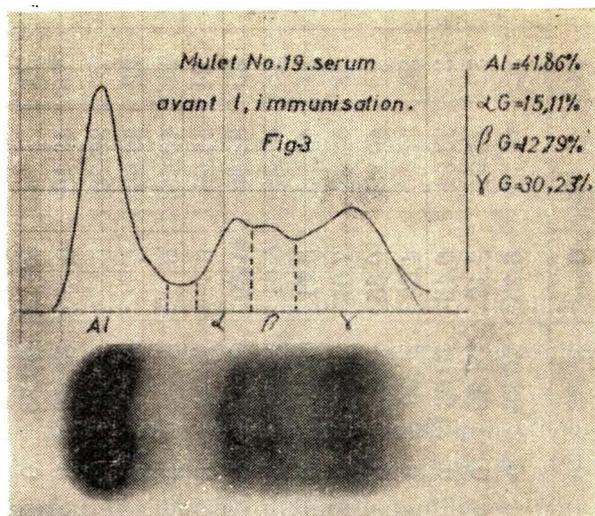
We tried to find the relationship between the increase in this beta 2 globulin fraction and the antibody increase; the results are set out in the table below.

There is therefore a constant relationship between the increase in Beta 2 globulin and the increase in rabies antibody. While there is, of course, no question of this electrophoretic method claiming to be a means for titrating rabies antibody, it nevertheless provides valuable information with respect to serum-virus neutralization titration because it makes it possible to decide on the precise dilutions of the sera to be titrated which should be used, thus economizing a considerable number of mice.

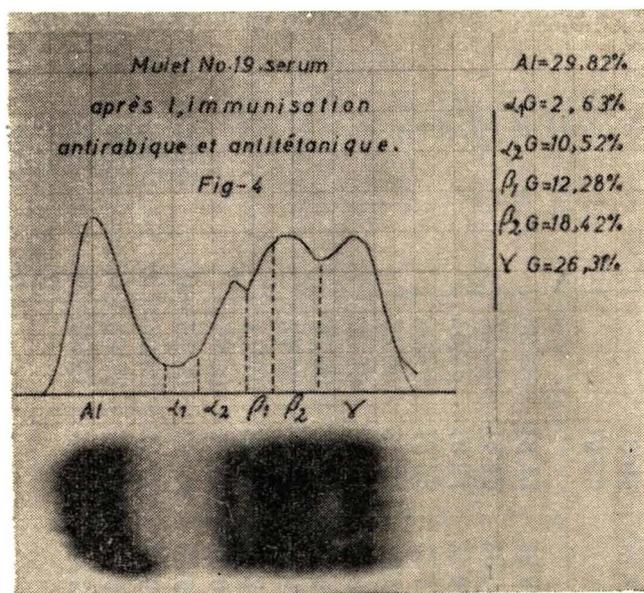
★ ★ ★

The attached curve shows how soon the electrophoretic changes occur and their appearance.

Animals	No	Before immunization				After basic immunization				After hyperimmunization			
		Alb.	Beta 2	Gamma	Rabies anti- body U/ml	Alb.	Beta 2	Gamma	Rabies anti- body U/ml	Alb.	Beta 2	Gamma	Rabies anti- body U/ml
Mules	16	32.25	7.52	34.43	0	30.00	12.20	32.17	> 40	23,18	19.56	28.98	240
	18		0	32.50	0		12.50	25.00	> 40				
	19		0	30.23	0		18.42	26.31	> 40				
	20	43.18	0	30.68	0	37.22	10.17	26.41	> 40	26,61	23.17	21.58	200
	21	36.26	0	32.96	0	31.14	10.31	32.72	> 40	23,12	23.12	29.94	320
	22	39.55	0	31.39	0	33.34	11.21	30.45	> 40	25.56	17.29	28.57	140
	23	40.86	0	27.95	0	38.56	11.47	26.92	> 40	27.39	25.34	22.60	280
	24	43.94	0	26.14	0					18.00	17.00	30.00	100
	25	38.99	11.60	27.67	0					17.91	26.86	31.34	280
	26	47.38	6.01	23.69	0					24.59	17.21	30.32	100
	27	44.76	5.71	27.61	0					23.68	21.71	28.94	200
	28	42.98	7.01	23.68	0					22.40	27.20	25.60	300
	29	40.98	10.65	15.50	0					27.53	21.01	23.18	200
	30	37.50	4.68	28.90	0					30.30	18.93	22.72	140
	31	39.51	6.45	29.03	0					29.32	16.54	27,06	100
32	43.91	8.13	24.39	0					27.73	18.48	28.57	100	
33	43.75	6.25	26.56	0					25.53	18.12	28.85	100	
Horses	4		0	33.33	0						15.32		60
	7		0	31.70	0						9.61	22.11	120
	12		7.89	27.84	0						12.50	22.42	40
	13		12.50	28.70	0						14.40	26.27	40
Donkeys	34	27.58	0	40.51	0					26.66	0	33.33	0
	36	33.33	6.66	41.66	0					24.39	9.75	34.95	40
	38	22.22	0	35.18	0					24.82	0	36.87	40
	39	28.57	11.60	35.71	0					26.71	13.74	35.14	60



Mule N. 19 serum before immunization



Mul No. 19 serum after
immunization against rabies and tetanus