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SEROLOGICAL SCREENING FOR INFECTIOUS CATTLE DISEASES I. INFLUENCE OF REPRODUCTIVE STATUS

TRIAGEM SOROLÓGICA DE DOENÇAS INFECCIOSAS I. INFLUÊNCIA DA FASE REPRODUTIVA

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SUMMARY

A cohort of 125 cows (Zebu-Holstein crosses) were screened every two months during a period of two years for IgG antibodies to 20 viral, bacterial, rickettsial and parasitic agents. The screening was performed on sera using the ELISA procedure. The IgG levels fluctuated during pregnancy in a regular and similar way for all the antigens tested. IgG reached its lowest levels in the fourth and seventh month of pregnancy, a significant increase ocurred during the last month of pregnancy and this was followed by a precipitous decline during the first month post partum. The IgG fluctuations associated with the different phases of reproductive status may interact with seasonal changes in IgG levels. The changes in IgG levels during pregnancy may have the result that an individual animal oscillates between being seropositive and seronegative. This can have an important impact on serological studies of abortion epidemiology.

Key words: cohort, serological screening, pregnancy.

RESUMO

Um estudo de coortes de 125 vacas cruza Zebu-Holandês foram monitoradas durante um período de dois anos para anticorpos do tipo IgG contra 21 agentes: vírus, bactérias, rickétsias e parasitas. A triagem sorológica foi realizada utilizando-se uma prova imuno-enzimática (ELISA). Os níveis de IgG flutuaram de forma regular e similar durante a gestação para todos os antígenos testados. Os níveis mais baixos de IgG foram encontrados no 4° e 7° mês de gestação. Houve um aumento significativo dos títulos no último mês de gestação sendo seguido por um declínio acentuado durante o primeiro mês pós-parto. A flutuação dos níveis de IgG pode estar associada com as diferentes fases reprodutivas podendo interagir com as variações estacionais dos níveis de IgG. As alterações de níveis de IgG durante a gestação pode levar a resultados onde o animal oscila entre positivo e negativo. Isso pode ter um importante impacto em estudos sorológicos na epidemiologia de abortos.

Palavras-chave: coorte, triagem sorológica, gestação.

INTRODUCTION

Serological testing for circulating antibodies is an attractive diagnostic procedure for several reasons. In certain chronic infections, like brucellosis, where there is no easy way to detect the agent, it becomes the technique of choice. Since serology treats all diseases like they were chronic-serology applied to a population measures period prevalence with the actual period in most cases undefined - it can be used to decide if an infection has occurred in the past even if the agent is no longer present. The use of serology is fairly straight forward when dealing with overt cases of disease. Generally paired samples are taken from the patient at different points in time and the diagnostician looks for an increase in serological titer. In the case of subclinical infections which generally are more common than clinical (MARTIN et al, 1987) paired samples can seldom be used and the investigator (diagnostician) will have to make a decision based on comparison of the distribution of serology titers (or percent ELISA) from test animals with those of known negative and known positive animals. Such screening procedures often work well as witnessed by successful eradication of e.g. Brucellosis and Bovine leukemia. Serological testing is, however, not without its problems and some of these have been

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pointed out by TYLER & CULLOR (1989). In this paper we will, for the first time, describe the impact of pregnancy status on IgG levels in serum.

MATERIAL AND METHODS

The study was conducted with cattle at the Center for Research, Teaching and Extension in Tropical Livestock (Centro de Investigación, Enseñanza y Extensión en Ganadería Tropical-CIEEGT) located in the North-central part of the state of Veracruz, Mexico.

Blood samples were collected bimonthly from a cohort of 125 female, crossbred Holstein-Zebu cattle during a two year period (1988-1989). Sera were separated and tested for antibodies (IgG) to viral, bacterial, rickettsial and parasitic agents using the Enzyme Linked Immunosorbent Assay (ELISA). The testing procedure and study area have been described earlier (Barajas-Rojas et al, 1993). The test at calving was performed within two weeks after parturition; the first test after calving (ITAC, Table I) was performed two months later.

Since the primary purpose of this study was to examine overall fluctuations in IgG level during pregnancy, distinctions were not made between tests deemed positive or negative but the overall percent ELISA was evaluated.

RESULTS

All the antigens tested showed similar patterns of fluctuation in the IgG level during pregnancy and the first period after calving. The average effect is shown in figure 1 which exhibits a very "damped" expression of the fluctuations since it represents ELISA responde to 20 antigens, each with its own responde level and variance.

A clear picture is gained by looking at individual responses. Figure 2, 3, 4 and 5 show ELISA res-

Table I Mean percent ELISA values for 20 infectious agents of125 cows from the tropics of Mexico according to their reproductive status (1988,1989)

	MONTH OF GESTATION (G1-G9), CALVING AND FIRST TEST AFTER CALVING										
AGENT	G1	G2	G3	G4	G5	G6	G7	G8	G9	CALVING	1TAC
CF	49.29	44.09	46.67	42.74	48.19	47.45	44.84	45.78	51.62	37.95	45.30
LH	51.80	48.63	49.01	41.20	56.71	50.27	47.94	57.16	54.70	50.72	46.44
BTV	59.50	76.54	77.36	71.52	67.93	80.92	71.08	71.14	63.03	66.04	73.34
MB	65.12	57.24	64.34	54.26	60.25	58.87	54.05	59.51	66.18	50.51	56.10
AM	53.54	57.00	56.33	63.61	52.29	65.53	50.22	56.57	56.41	54.36	61.94
PM	40.85	47.79	48.64	49.72	45.86	50.79	49.69	49.64	62.52	44.99	48.54
CB	43.92	45.28	41.57	39.37	39.51	43.89	41.44	44.20	48.58	35.92	45.61
TG	43.35	51.99	44.57	47.91	43.94	35.97	47.06	48.74	49.28	36.14	40.54
ST	40.32	52.45	50.64	39.07	53.03	48.24	49.62	44.85	60.87	46.89	54.83
SD	33.30	45.09	42.64	36.53	42.69	44.80	45.34	43.18	43.66	38.04	44.05
BRSV	58.80	58.96	49.81	51.48	56.47	51.88	58.68	58.41	59.84	56.53	53.81
CL	42.72	43.67	42.58	40.66	49.73	42.57	44.19	37.63	37.18	33.47	43.97
BVD	44.43	48.72	47.29	44.61	47.37	47.81	46.61	47.58	50.19	42.80	47.43
BA	43.82	48.73	47.15	44.89	46.59	47.61	46.50	46.79	49.81	42.14	47.51
PI3	42.51	46.41	44.63	42.68	44.81	44.83	44.45	44.76	48.71	40.15	45.36
IBR	40.63	45.51	42.99	41.71	43.52	43.66	43.65	43.53	47.25	39.29	44.47
HS	39.55	44.55	41.88	39.89	42.79	41.84	43.10	42.44	46.49	38.03	43.01
RV	39.45	44.28	41.31	39.07	42.54	41.09	42.55	41.84	45.15	37.45	42.55
LM	39.07	44.20	41.29	39.04	42.79	40.86	42.65	41.65	44.87	37.58	42.29
MP	38.72	43.55	41.02	38.30	42.69	41.27	42.28	41.05	44.50	37.70	42.44
TOTAL	43.37	47.36	45.80	43.25	46.18	46.20	45.52	46.02	49.09	41.27	46.17

CF = Campylobacter fetus
LH = Leptospira interrogans serovar hardjo
BTV = Bluetongue Virus
MB = Mycoplasma bovis
AM = Anaplasma marginale
PM = Pasteurella multocida
CB = Coxiella burnetii
TG = Toxoplasma gondii
ST = Salmonella typhimurium
SD = Salmonella dublin
BRSV= Bovine Respiratory Syncytial Virus
CL = Chlamydia psittaci-trachomatis
BVD = Bovine Viral Diarrhea Virus
BA = Brucella abortus
PI3 = Parainfluenza 3 Virus
IBR = Infectious Bovine Rhinotracheitis Virus
HS = Haemophilus somnus
RV = Rotavirus
LM = Listeria monocytogenes
MP = Mycobacterium paratuberculosis

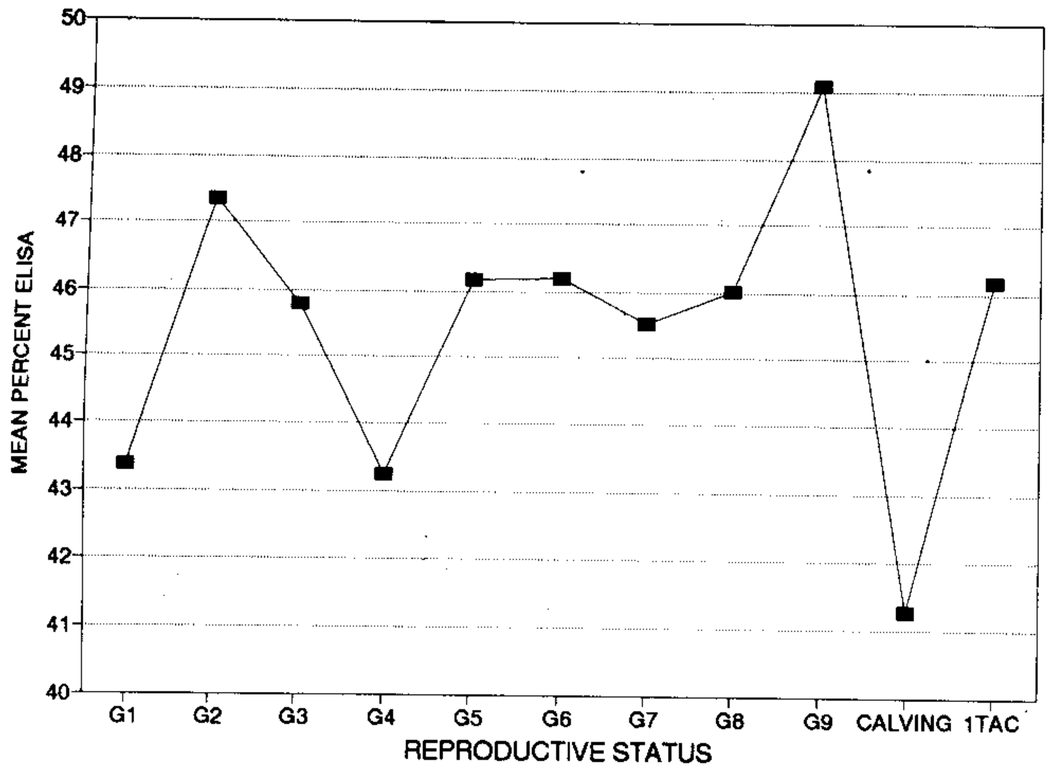

FIGURE 1 - Mean percent ELISA to 20 infectious agents of 125 cows from the tropics of Mexico by months of pregnancy, at calving and first test after calving (1988, 1989). G1 - G9 indicate month of pregnancy; 1TAC is first test after calving.

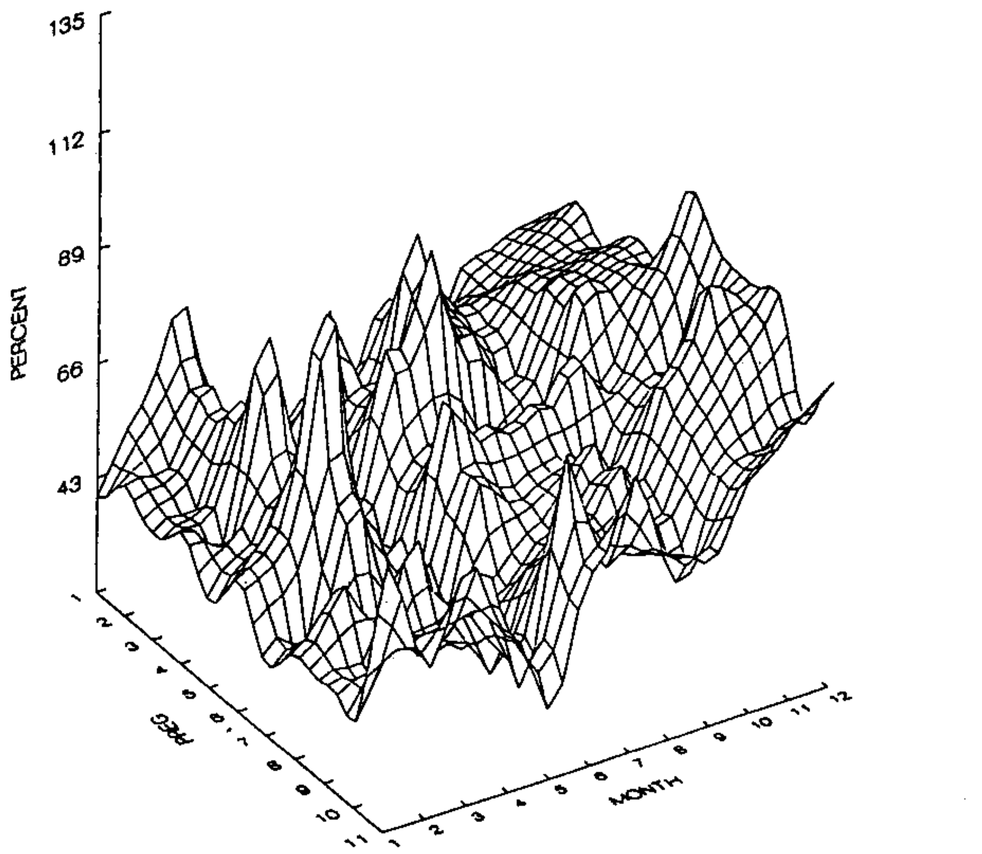

FIGURE 3 - Mean Percent **ELISA** (PERCENT) for **Anaplasma marginale** in 125 cows from the tropics of Mexico by reproductive status; 1-9 are months of pregnancy (PREG), 10 is the test immediately after calving and 11 is the second test after calving; and calendar month of the year (MONTH) 1 through 12 represent January through December (1988, 1989).

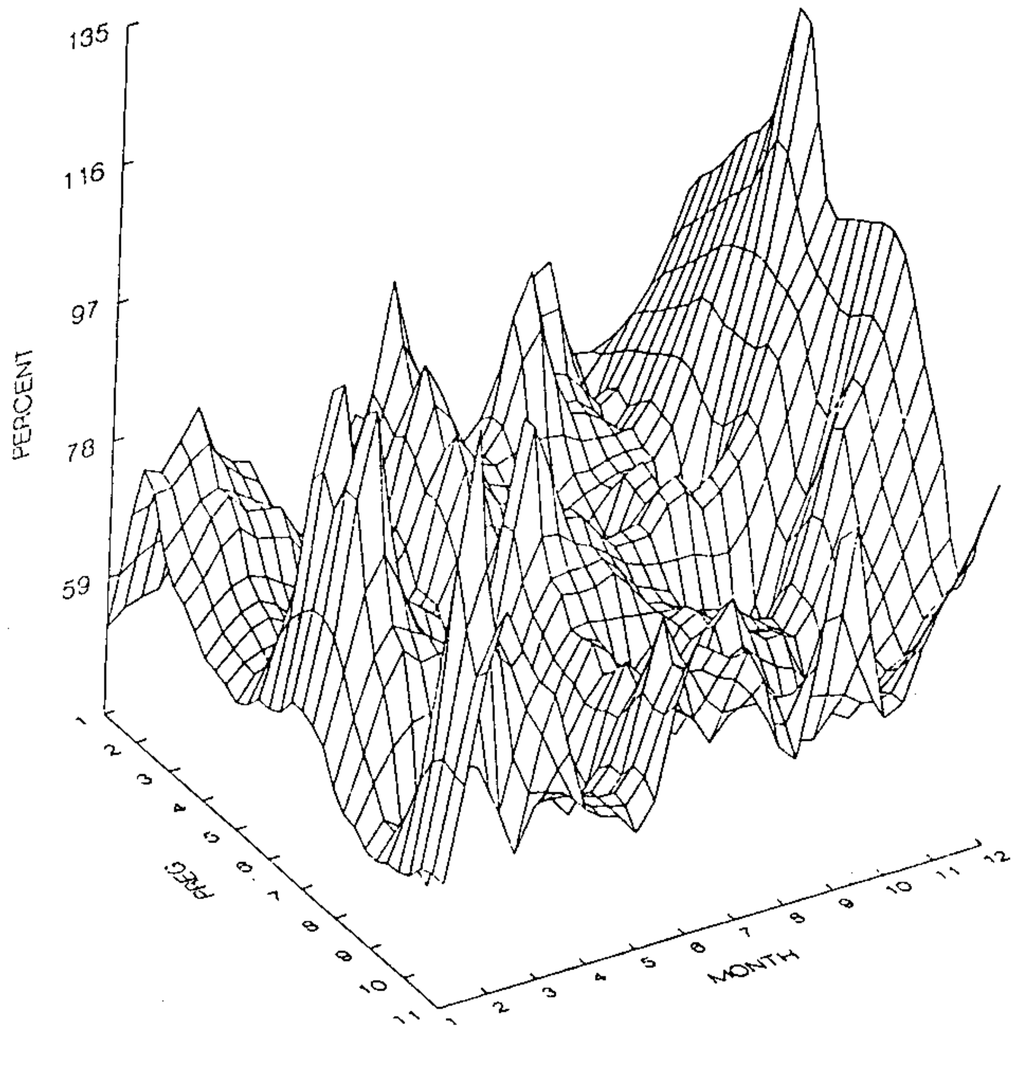

FIGURE 2 - Mean percent **ELISA** (PERCENT) for **Bluetongue virus** in 125 cows from the tropics of Mexico by reproductive status; 1-9 are months of pregnancy (PREG), 10 is the test immediately after calving and 11 is the second test after calving; calendar month of the year (MONTH) 1 through 12 represent January through December (1988, 1989).

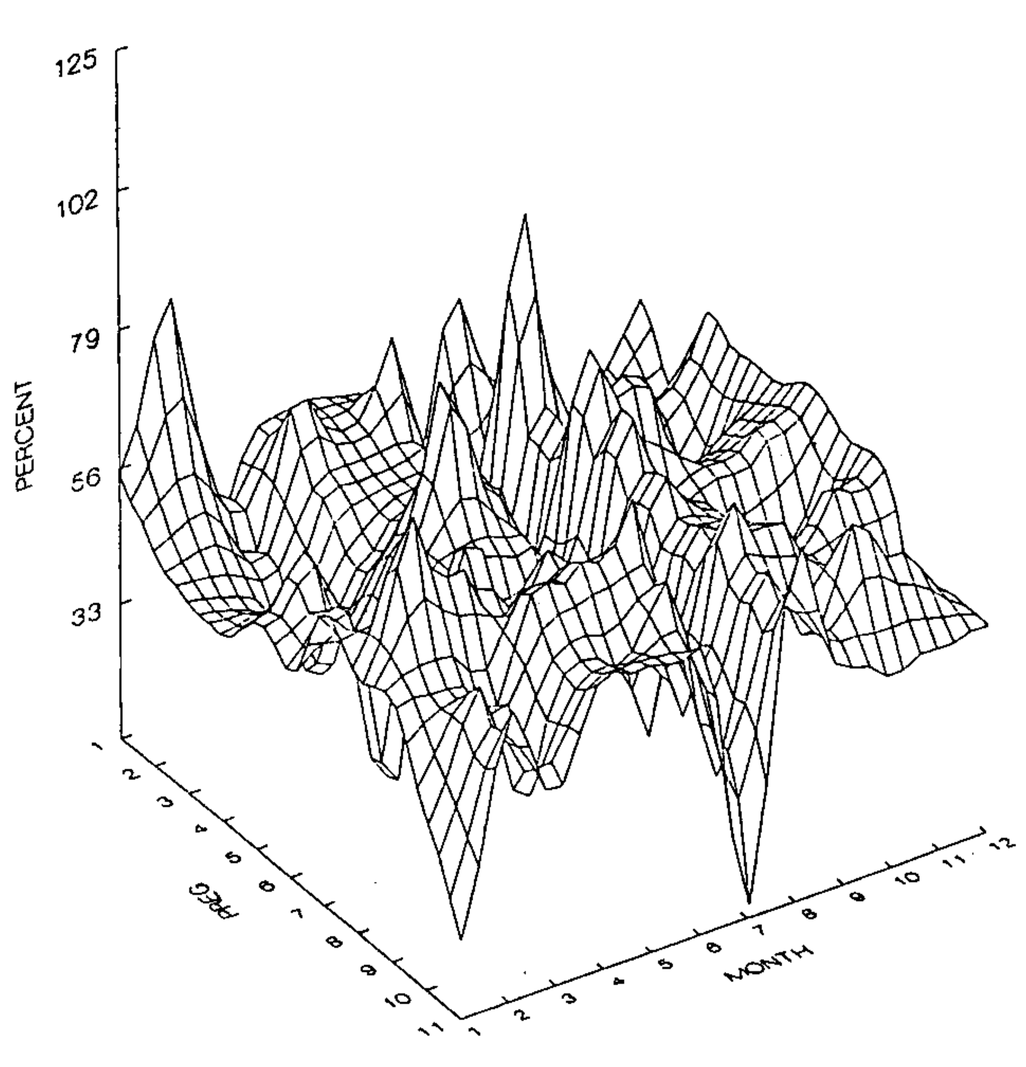

FIGURE 4 - Mean percent **ELISA** (PERCENT) for **Leptospira interrogans serovar hardjo** in 125 cows from the tropics of Mexico by reproductive status; 1-9 are months of pregnancy (PREG), 10 is the test immediately after calving and 11 is the second test after calving; calendar month of the year (MONTH) 1 through 12 represent January through December (1988, 1989).

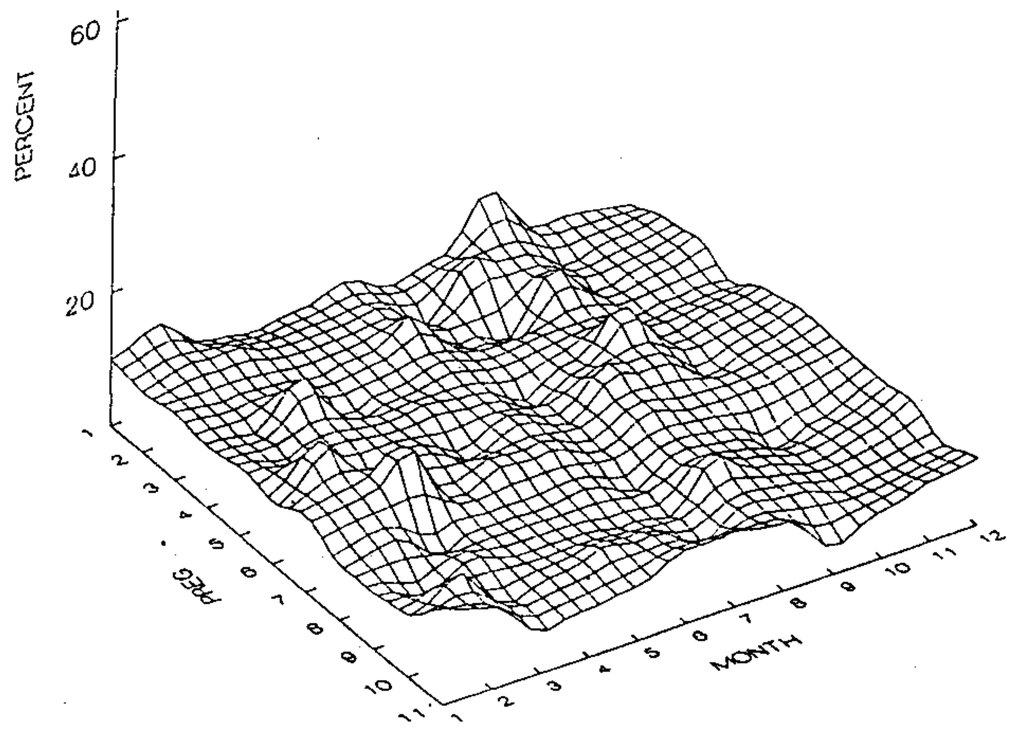

FIGURE 5 - Mean percent **ELISA** (PERCENT) for **Brucella abortus** in 125 cows from the tropics of Mexico by reproductive status; 1-9 are months of pregnancy (PREG), 10 is the test immediately after calving and 11 is the second test after calving; calendar month of the year (MONTH) 1 through 12 represent January through December (1988, 1989).

ponses for agents with high prevalence: **Bluetongue virus** (73% of tests positive) and **Anaplasma marginale** (61% of tests positive); medium prevalence: **Campylobacter fetus** (21% of tests positive) and low (in fact zero) prevalence: **Brucella abortus** (none of the tests positive). These figures show seasonal fluctuations as well as pregnancy related fluctuations. Seasonal fluctuations were discussed in a previous paper (Barajas-Rojas et al 1993). It is interesting that even antibodies to **Brucella abortus** which never reached the cut off point between negative and positive tests shows characteristic fluctuations albeit in a very reduced form. Antibody patterns to the remaining 16 antigens fall between the extremes of **Bluetongue virus** and **Brucella abortus**. The total data set for ELISA fluctuations during pregnancy is shown in Table I, not corrected for season.

DISCUSSION

The ruminant immune system has been studied by several authors (BUTLER, 1981; MORRISON, 1986). Many distinct processes of immunological nature occur in the female, such as the entrance of the spermatozoa, fertilization, implantation of the embryo, nourishment and development of the fetus, parturition and lactation; in addition cows continue to grow during the first pregnancy. All of these processes may create conditions for formation or inhibition of antibodies and other immune structures during different phases of reproduction. The allergic interrelation between the cow and the fetus show different stages of reactivity during pregnancy (BRATANOV, 1968). Fluctuations in the female hormones such as pregnanes and estranes have been reported (BUTLER, 1981; STABENFELDT & EDQVIST, 1984). It is also known that pregnant cows are partially immunosuppressed (BRATANOV, 1968). The present paper documents for the first time quantitative fluctuations in IgG levels and illustrates the regular pattern that exists independent of the nature of the infectious agent. These observations may not have great significance when herds (population) with uniformly distributed calving pattern are screened. However, they become important in seroepidemiological studies of abortion and also when infections in individual pregnant animals are diagnosed based on serology.

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