

Animal Reproduction Research Institute,
ARC. El-Haram, Giza, Egypt

CROSS REACTIVITY BETWEEN TRITRICHOMONAS FETUS AND CAMPYLOBACTER FETUS ANTIBODIES IN THE CERVICOVAGINAL MUCUS OF COWS (With 5 Tables)

By

A. ABDEL-AAL; MONA M. SOBHY and Z.M. KHOLEAF

(Received at 8/6/1997)

التفاعل المزدوج بين التريكوموناس الجنيني والكامبيلوباكتز الجنيني
في الإفرازات المخاطية المهبلية في إناث الأبقار

أحمد عبد العال ، منى صبحى ، زيدان خليف

تم تجميع عدد ١٦٠ عينة إفرازات مخاطية ترحمية مهبلية من إناث أبقار غير عشار من محافظة الفيوم . وقد تم اختبار جميع العينات سيرولوجيا لاكتشاف الأجسام المضادة للتريكوموناس الجنيني والكامبيلوباكتز الجنيني وكذلك التفاعل المزدوج بين المرضين التناسليين بواسطة اختبار التجمع الدقيق واختبار الأليزا . وكانت نتائج هذه الدراسة هي . بالنسبة لأختبار التجمع الدقيق فقد كانت نتيجة الأجسام المضادة لمرض التريكوموناس الجنيني إيجابية في عدد ٨ (٥٪) وبالنسبة للكامبيلوباكتز الجنيني هو ١٤ (٨,٧٥٪) بالمقارنة بأختبار الأليزا التي كانت نتيجة لمرض التريكوموناس الجنيني ١٩ (١١,٨٨٪) و ٣٣ (٢٠,٦٣٪) للكامبيلوباكتز الجنيني . أما بالنسبة لنتيجة التفاعل المزدوج بين المرضين فكانت إيجابية في عدد ٦ حالات (٣,٧٧٪) بالنسبة لأختبار التجمع الدقيق وكانت حالة واحدة فقط (١,٩٢٪) في اختبار الأليزا . ومن ذلك نستخلص أنه بواسطة استخدام اختبار الأليزا أمكن حل مشكلة التفاعل المزدوج بين التريكوموناس الجنيني والكامبيلوباكتز الجنيني لأنه يحدد الأجسام المناعية المضادة الخاصة لكل مرض على حدة بالنسبة و IgA و IgG في الإفرازات المخاطية المهبلية .

SUMMARY

A total number of 160 cervicovaginal mucus (CVM) samples were collected from non-pregnant cows in Fayoum Governorate. All samples were examined serologically to detect antibodies of *Tritrichomonas fetus*, *Campylobacter fetus* and cross reactivity between antibodies of the two venereal diseases were tested by using microagglutination test and ELISA

test. In the present study, the results of microagglutination test for detection of antibodies of *Tr. fetus* and *C. fetus* were 8(5%) and 14 (8.75%) respectively in comparison to 19 (11.88%) and 33 (20.63%) by using ELISA test. The cross reactivity between *Tr. fetus* and *C. fetus* was positive in 6 (27.27%) cases by using microagglutination test but ELISA test was positive only one case (1.92%). This means that using of ELISA technique solves the problem of cross reactivity between *Tr. fetus* and *C. fetus* by demonstration of the specific IgG and IgA in the cervicovaginal mucus.

Key words: *Trichomonas fetus - Campylobacter fetus - Cross reaction - Vaginal mucus-Cows.*

INTRODUCTION

Trichomoniasis and campylobacteriosis are distributed world wide and they are considered as a significant causes of economic losses. These organisms are obligate parasites of the mucosa of male and female genital organs, characterized clinically by temporary infertility, abortion and repeat breeders in female cattle and the transmission is only venereal (Mickelsen, 1983 and Goodger and Skirrow, 1986).

The tentative diagnosis of the two diseases depends upon the isolation and identification of the causative organisms, but due to the presence of rapidly growing contaminant isolation can not be easily done (Dekeyser, 1986 and Eaglesome and Garcia, 1992).

Sometimes, serological tests are used as a diagnostic method for these diseases. However, this approach can cause problems as agglutininis are demonstrated only in 50% of infected bovines and both false positive and false negative results occasionally occurs. A considerable cross reactivity between *Trichomonas fetus* and *Campylobacter fetus* antibodies were observed because they share some common antigens and the fluctuation of IgA antibodies in individual female and the possibility of false reactions occur (BonDurant, 1985 and Yule, et al., 1986).

The introduction of ELISA techniques has solved the problem of cross reactivity between *Tr. foetus* and *C. fetus* (Skirrow and BonDurant, 1988).

In the present study using ELISA test to over come the cross reactivity between the two venereal diseases in comparing to the previously microagglutination test applied on cervicovaginal mucus samples of cows.

MATERIALS and Methods

A total number of 160 cervicovaginal mucus samples were collected from non-pregnante in Fayoum Governorate. The cervicovaginal mucus samples were collected by using A.I. pipettes (Abbitt and Ball, 1978).

All samples were examined serologically to detect antibodies of *Trichomonas foetus*, *Campylobacter fetus* antibodies and show the cross reactivity between two diseases using microagglutination test (Diker and Turutglu, 1995) and ELISA test (Hum, et al., 1994). Both tests were used reference antigens of *Tr. Foetus* and *C. fetus* and also reference antibodies of *Tr. foetus* and *C. fetus* as a control positive.

RESULTS

Results are presented in Tables 1, 2, 3, 4 & 5.

DISCUSSION

It is well known that trichomoniasis and campylobacteriosis are the most important venereal diseases affecting cattle, characterized by repeat breeding,irregular oestrus cycle, endometritis and abortion at all stages of gestation resulting in a great economic loss (Roperts, 1971 and Corbeil et al., 1975,b).

In the present study, cervicovaginal mucus samples were collected from 160 non-pregnant cows using A.I. pipettes. All samples examined serologically by microagglutination test and ELISA test to study the cross reactivity between antibodies of the two venereal diseases.

A considerable cross reaction between *Tr. foetus* and *C.fetus* has been observed because they shared some common antigens among serotypes. The titer and duration of these agglutinins following infection are irregular and varied (Reece et al., 1981 and Yule et al., 1986).

Recently, it is shown that specific antigenic differences between them by using ELISA techniques demonstrated the specific IgG and IgA in the cervicovaginal mucus to be useful and helping to detect clear infection (Hawkins et al., 1986 and Hongiberg and Lindmark, 1987).

The results of microagglutination test to detect antibodies of *Tr. foetus* and *C. fetus* were 8(5%) and 14 (8.75%) respectively as compared with ELISA test 19(11.88%) positive for *Tr. foetus* antibodies and 33

(20.63%) positive for *C. fetus* antibodies (Tables, 1&2). These results agree with those of Grohn and Genigeorgis, 1985 and Hum, et al., 1994.

The present study indicates that ELISA technique appeared only in one case (1.92%) of cross reactivity between *C. fetus* and *Tr. foetus* at 1/100 end titre as compared with the microagglutination test which gave 6(27.27%) (Table 3 & 4). These results agree with those of Street *et al.*, 1982 and Yardinci, *et al.*, 1994.

The interpretation of the relative sensitivities of tests for detection of *Tr. foetus* and *C. fetus* antibodies in CVM samples indicates the efficacy of ELISA test (relative sensitivity 100%). Of these, 52 positives were obtained as shown in Table (5), compared interms of results of microagglutination test 22 positives (relative sensitivity 42.31%).

It can be concluded from the present study that the use of ELISA technique solves to a great extent the problem of cross reactivity between *Tr. foetus* and *C. fetus* in serological diagnosis in cervicovaginal mucus.

REFERENCES

- Abbitt, B. and Ball, L. (1978): Diagnosis of trichomoniasis in pregnant cows by culture of cervical-vaginal mucus. *Theriogenology*, 9,267-270.
- BonDurant, R.H. (1985): Diagnosis, treatment and control of bovine trichomoniasis. *Compendium for contiuning Education*, 7, S179-S188.
- Corbeil, L.B.; Schurig, G.G. and Bier, P.J. (1975,b): Bovine venereal vibriosis: Antigenic variation of the bacterium during infection. *Infect. Immun.* 11:240-244.
- Dekeyser, P.J. (1986): Bovine genital campylobacteriosis in current therapy in *theriogenology*. Vol.2 Cited by D.A. Morrow, V.B. Saunders Company.
- Diker, K.S. and Turutglu,H. (1995): Evaluation of immunogenicity of Campylobacter strains isolated from ovine abortions by laboratory test systems. *J. Vet. Med. B.* 42, 35-41.
- Eaglesome, M.D. and Garcia, M.M. (1992): Microbial agents associated with bovine genital tract infections and semen. PartI. *Brucella abortus*, *Leptospira*, *Campylobacter foetus* and *Trichomonas foetus*. *Vet. Bull.*, 62:8,743-775.

- Goodger, W.J. and Skirrow, S.Z. (1986):* Epidemiologic and economic analysis of unusually long epizootic of trichomoniasis in a large California dairy herd. *J. of Vet. M.Ass.* 189, 772-776.
- Grohn, K. and Genigeorgis, C. (1985):* Adaptation of ELISA for the detection of *Campylobacter* antibodies and its application in seroepidemiological studies in sheep and cattle herds. *Acta.Vet.Scandinavica*, 26:30-48.
- Hawkins, C.F.; Eaglesome, M.D. and Garcia, M.M. (1986):* Vaccination studies for the control of campylobacteriosis in Jamaican cattle. *Vet. Rec.* 119: 299-301.
- Honigberg, B.M. and Lindmark, D.G. (1987):* Trichomonads and *Giardia* in immune responses in parasitic infections, immunology, immunopathology and immunoprophylaxis. Vol. IV Ed.by E.J.L. Soulsby, CRC Press BocaRaton, pp. 99-139.
- Hum, S. ; Quinn, C. and Kennedy, D. (1994):* Diagnosis of bovine venereal campylobacteriosis by ELISA. *Aust. Vet. J.* 71: 140-143.
- Mickelsen, W.D. (1983):* Diagnosis of Trichomoniasis: Herd history and culture techniques. *Bovine Practitioner*, 15, 134-135.
- Reece, R.L.; Dennett, D.P. and Johnson, R.H. (1981):* Trichomonas foetus agglutination tests upon samples collected from cattle: Cross reactions associated with vaccination against *Campylobacter fetus* subsp. *Venerealis*. *Aust. Vet.J.* 57:352-353.
- Roperts, S.J. (1971):* Veterinary obstetrics and genital diseases. 2nd Ed. Ann. Arbor.Mich.Edwards, Brothers. Inc. pp. 427-433.
- Ruppanner, R.; Meyer, M.E. and Willeberg, P. (1980):* Comparison of the enzyme-linked immunosorbent assay with other tests for Brucellosis using sera for experimental infected heifers. *Am.J.Vet.Res.*, 41: 1320-1332.
- Skirrow, S. and BonDurant, R.H. (1988):* Bovine trichomoniasis. *Vet. Bull.* 58:591-603.
- Street, D.A.; Taylor-Robinson, D.; Ackers, J.P.; Hanna, N.F and McMillan, A. (1982):* Evaluation of an enzyme linked immunosorbent assay for the detection of antibody to trichomonas vaginalis in sera and vaginal secretions. *British J. of venereal diseases*, 58: 330-333.
- Yardinici, H.; Diker, K.S. and Akan, M. (1994):* Use of ELISA for detection of *Campylobacter* antibodies in sheep. *Turk.Vet.Ve-Hayvancilile, Dergisi*, 18: 129-133.

Yule, A.; Staat, J.; Skirrow, S.Z. and BonDurant, R.H. (1986):
 Development of enzyme immunoassays for trichomonias foetus antigenic: Investigation of unexpected cross reactivity between immune rabbit antisera and components of uninfected bovine cervical mucus and preputial scrapings. Proceeding of the 67th Ann. Conf. for Res. Workers in Animal Disease, Chicago, p.60.

Table 1 : Results of Microagglutination test for detection of antibodies of Tr. Fetus and C.fetus in CVM sample.

Disease tested	No. of tested samples	No. of positive samples	Mucus Dilution					
			1/10	1/20	1/40	1/80	1/160	1/320
1- Tr.fetus	160	8	3	2	1	1	1	-
		5%	1.88%	1.25%	0.62%	0.62%	0.62%	-
2- C. fetus	160	14	5	3	2	2	1	1
		8.75%	3.13%	1.88%	1.25%	1.25%	0.62%	0.62%

Table 2 : Results of ELISA test for detection of antibodies of Tr. Fetus and C.fetus in CVM sample

Disease tested	No. of tested samples	No. of positive samples	Mucus Dilution					
			1/100	1/200	1/400	1/800	1/1600	1/3200
1- Tr.fetus	160	19	6	6	4	2	1	-
		11.88%	3.75%	3.75%	2.5%	1.25%	0.62%	-
2- C. fetus	160	33	13	7	6	4	2	1
		20.63%	8.13%	4.38%	3.75%	2.5%	1.25%	0.62%

Table 3: Results of Cross reactivity between Tr. Fetus and C.fetus by using Microagglutination test :

Disease tested	No. of positive samples	No. of cross reactive samples	End Titre *					
			1/10	1/20	1/40	1/80	1/160	1/320
1- Tr.fetus	8	2 25%	1 12.5%	1 12.5%	-	-	-	-
2- C. fetus	14	4 28.57%	1 7.14%	1 7.14%	1 7.14%	1 7.14%	-	-
Total	22	6 27.27%	2 9.09%	2 9.09%	1 4.55%	1 4.55%	-	-

* End dilution of CVM samples

Table 4: Results of Cross reactivity between Tr. Fetus and C.fetus by using ALISA test :

Disease tested	No. of positive samples	No. of cross reactive samples	End Titre *					
			1/100	1/200	1/400	1/800	1/1600	1/3200
1- Tr.fetus X C.fetus	19	-	-	-	-	-	-	-
2- C. fetus X Tr.fetus	33	1 3.03%	1 3.03%	-	-	-	-	-
Total	52	1 1.92%	1 1.92%	-	-	-	-	-

* End dilution of CVM samples

Table (5): Comparison of the relative sensitivity of ELISA, MAT on specimens of CVM.

Test Item	No. of positive	Relative sensitivity *
ELISA	52	100 %
MAT*	22	42.31 %

* : As measured on specimens positive to ELISA

* : Microagglutination test .

$$RS = \frac{\text{No. of test positive}}{\text{ELISA positive specimens}} \times 100\%$$

(Ruppanner, et al ., 1980)