CASE REPORTS: BLOOD TEST (BTB) FOR DIAGNOSIS AND MANAGEMENT OF TB.

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CASE 1

Specificity and Sensitivity of Reactivity in a group of CCT(+) reactor stags.

Historical

The laboratory was approached (Feb 88) by a veterinarian who had been using the CCT routinely to test large numbers of deer in a herd which had a history of infection with *M. bovis* with extremely high background levels of non specific reactivity due to *M. avium*. Earlier BTB tests (Nov 86) in this herd had shown that the reactivity in 96% of 178 ST(+) Reactor hinds could be attributed to *M. avium* exposure. These BTB(-) hinds were later moved onto a separate property and all animals passed a recent whole herd ST without any residual evidence of *M. avium* reactivity after being farmed on the new property for eight months.

Current Test.

Because the veterinarian had failed to find any lesions following autopsy of 7 CCT(+) reactor animals in June 1987, he identified the prospect of limitations in the specificity of the CCT. On carrying out a CCT test on 250 adult stags in Jan 1988; 24 animals gave CCT(+) reactions. Samples from these animals were submitted for BTB testing 14 days after applying the CCT, to verify the specificity of their skin test reactivity.

Findings.

Number CCT(+)	Number with reactivity specific for *M.bovis* using BTB	Number with Lesions at autopsy
24	8	7

Conclusions.

1. Animals with high levels of non-specific (*M. avium*) reactivity can rapidly

convert to become unreactive when farmed on a property with low endemic levels of *M. avium*.

2. When animals are exposed to high levels of endemic *M. avium* False (+) CCT reactivity may occur. Such reactivity can be attributed to *M. avium* exposure rather than *M. bovis* using the higher discriminating activity of the BTB assay.

CASE 2

BTB reactivity and M. bovis in Fallow Deer (*Dama dama*).

Because the parameters for the BTB system have been established using a database obtained from red deer (*Cervus elaphus*) it was of interest to test the discrimination of the BTB in an unrelated species of cervids. Samples were submitted from four fallow deer which had reacted to ST. The herd had an earlier history of *M. bovis* infection.

Results

ST (+)	BTB (+)	*M. bovis* Lesions at Autopsy
4	4	4

Conclusions

The BTB can be used to accurately identify *M. bovis* infection in fallow deer. The level of BTB reactivity is of a similar level to that found in infected red deer for the BTB.

Two significant differences were found between the fallow deer and red deer.

1. Fallow deer showed a consistently higher level of inflammatory cell activity than is found typically in red deer. Whether this is the result of stress associated with handling or inherent differences between the species remains to be established.

2. Although the blood leukocytes from fallow deer have the same physical characteristics (size/bouyancy) the background level of lymphocyte activity in negative control cultures is higher (3,000-8,000) in fallow deer than in red deer (500-2,000cpms).

We await with interest an increase in our database for fallow deer to determine further the common denominators and differences of relevance between these species.

CASE 3

Generalised tuberculosis (GTB) at a low incidence among ST reactors.

A significant number (65) of ST reactor hinds were identified among a group of animals tested in Oct 1987. Because *M. bovis* had been isolated from three animals in this herd in 1980 and 1986 a BTB test was requested and carried out on the 65 blood samples submitted in Oct 1987.

Results

	BTB			
ST(+)	Bovine	Equivocal	Avian	Negative
65	1	9	30	25

Clear reactivity to bovine-PPD was found in one animal which also had an inflammatory response compatible with infection. All other animals with even the remotest hint of bovine reactivity were placed in the equivocal group and seperate management recommended. A confirmatory BTB test was carried out on the bovine reactor and the equivocal animals in March 1988.

Results

The equivocal group of animals retested clear and the bovine reactor gave a repeat BTB(+) bovine reaction of increased intensity. A third BTB test was carried out on this animal in April 1988 at which it showed a further dramatic increase in bovine reactivity and a massive increase in inflammatory reactivity.

The diagnosis suggested by the laboratory was that the reaction in this animal was specific for *M. bovis* and the increasing levels of immunological and inflammatory activity suggestive of serious disease. On autopsy the animal was found to have generalised tuberculosis (GTB) which was confirmed by laboratory diagnosis. A repeat ST skin test was carried out on this group of

animals in June 88 with one animal ST(+). A subsequent BTB carried out on this animals confirmed avian reactivity.

Conclusion

1. There may be a low incidence of diseased (*M. bovis*) deer amongst a large cohort of non-specific reactors.
2. Had the CCT been used to check these animals it would have identified non-specific activity due to *M. avium* but it may have failed to identify the *M. bovis* infected animal.
3. Recent studies in our laboratory suggest that up to 40% of Tb infected ST(+) animals, give a repeat ST(-) reaction, if retested using the skin test.

CASE 4
Identification of an *elusive* False(-) ST reactor.

A herd (563) of red deer and red deer/wapiti hybrids, first established in 1979, was largely maintained as a closed herd, apart from the introduction of accredited stock. At the first whole herd test carried out in 1986, no reactors were evident. A second whole herd test was carried out in July 1987 and 22 ST reactor stags were identified. These animals were slaughtered at the DSP with 17 showing lesions, amongst which 5 yielded *M. bovis* cultures. Because of the sudden and severe onset of disease the farmer was concerned about spread of disease within the herd, and he approached the laboratory with a view to carrying out the BTB on 'in-contact' animals. He received independent advice, which suggested that a cost saving might be effected by slaughtering 50 x 18month old velveting stags, considered to be at some risk from infection. On slaughter no lesions were found among these 50 animals.

In November 1987, the BTB was carried out on 39 breeding females, 14 x 2year old stags and 16 adult (4-12year old) stags. Results of BTB showed no evidence of *M. bovis* reactivity in any of the 69 ST(-) animals tested. This evidence of low disease prevalence from the BTB assays complimented the slaughter result showing no lesions in the 50 young stags.

In March 1988 samples were submitted from a further 35 ST(-) animals. Results showed 34 animals clear to BTB with one animal showing significant *M. bovis* reactivity. A confirmatory test carried out on the BTB(+) animal 14 days later showed a repeat reaction specific for *M. bovis*. Autopsy was suggested and a lung lesion diagnosed, which was histologically and microbiologically positive for *M. bovis*, following slaughter at a DSP. This 5yo stag had been purchased three years earlier from property on which *M. bovis* has been diagnosed in recent years.

Conclusions.

1. BTB! would it have pre-empted slaughter?

Had the BTB been carried out prior to the slaughter of the 50 x 18month old velveting stags, our advice would have been that there was not a significant risk from *M. bovis* as the exposure had been at a very low incidence generally within this herd. Valuable genetically selected stock could have been retained using that approach.

2. Did we find the chronically diseased animal?

It is debatable that the False(-) ST animal, found to harbour lesions and which was identified as BTB(+), had aquired the infection recently or had come on to the property harbouring *M. bovis* infection. The recent history of the herd of origin would suggest that this animal may have been infected much earlier.

3. Did the BTB justify the cost?

Even though it was necessary to screen a large number (114) of animals to identify the False(-) ST animal the effort may have been well justified considering the prospect of pulmonary excretion of *M. bovis* and further infectious spread within the herd. The recent history suggests that this farmer has made significant progress in identifying and containing *M. bovis* infection over a short period using a combination of ST and BTB. The costs incurred could have been underwritten by savings effected had he retained the 50 young males sent unnecessarily for slaughter.

CASE 5

Management of Tuberculosis in a herd using ST and BTB

Background and Test Schedule

A large (220) mixed herd of red deer first found 3 ST(+) reactors in a sale test in 1985. Following a whole herd test a further 7 reactors were found, which first alerted this farmer to the prospect of Tb infection. Tb was diagnosed following autopsy of reactors, and a large number of reactors (47) were found at the next whole herd test. With the impending prospect of uncontrolled spread of Tb within his herd, the farmer questioned his ability to farm through the problem using a 'test and slaughter' policy.

Test history

27/05/85	Sale Test - 3 Reactors
31/05/85	Whole herd Test - 7/196 Reactors
04/06/85	10 Reactors autopsied - 1 *M. bovis* abscess diagnosed
13/08/85	Whole herd test - 47 Reactors
05/06/86	47 Reactors killed at DSP - 13 *M. Bovis* lesions (10GTB)
Aug 86	Herd split into 5 groups on separate properties as outlined in Figure 1.
Jan-Dec 87	ST and BTB carried out on the different properties
27/03/87	40 Stags processed through DSP - 40 NVL
03/07/87	35 Reactor (ST or BTB) animals autopsied at DSP - 23 Tb
30/09/87	3 Reactor (BTB+) animals autopsied at DSP - 2 Tb
Jan-Jun 88	ST and BTB carried out - 9 Reactors
June 88	9 Reactors autopsied - 3 Tb lesions

Management Regime 1986 - 1988

In 1986 - 261 deer were divided into groups and sent to be farmed separately on five separate properties as follows;

Property H - 80 ST(-) BTB (-) Hinds
6 ST(-) BTB (-) Stags
32 'At risk' equivocal* animals (ST±, BTB±)

Property T - 23 equivocal* Hinds and 20 weaner stags regarded as 'high risk' animals, with equivocal reactor status

Property N - 40 ST(-) BTB(-) Clean hinds

Property S - 40 ST(-) BTB(-) Clean weaner hinds

Property W - 20 ST(-) BTB(-) Weaner stags

* Animals which showed positive reactivity in the BTB and/or the ST.

Test and Slaughter 1987

During 1987, 35 equivocal (ST+ or BTB+) animals selected from H and T and regarded as at risk from *M. bovis* infection were slaughtered at the DSP - 23 showed *M. bovis* lesions. Of 20 stags taken from property H and T for slaughter at DSP four showed *M. bovis* lesions. Of the 20 stags slaughtered from property W, no lesions were found at the DSP.

Test and Slaughter 1988

ST and BTB tests carried out early in 1988 showed very little *M. bovis* reactivity, except at a low incidence on property H & T.

Test and Slaughter June 88

	Property			
	H	T	N	S
ST	0/165	5/50	1/68	0/56
BTB	2/165	5/50	0/68	1/56
Autopsy	2 NVL	3 - Tb	-	1 - NVL
Residual Stock	163	47	68	55
Status	No DCP	DCP	No DCP	No DCP

Conclusions

1. Using the ST and BTB to segregate and isolate stock into clean and equivocal groups soon after slaughtering a large number of tuberculous animals, this farmer has been apparently successful in reaching disease free status for 286 animals inside two years. There also appears to be a low incidence of disease in the equivocal herd (T), so there is the likely prospect of salvaging the majority of the 47 animals on this property.

2. False (-) ST animals would have persisted in herds H and T, had they not been identified by the BTB. In fact one ST negative but BTB(+) animal was found out of the three diseased animals slaughtered, on property T at the final test in June '88.

3. Repeat skin tests (ST) carried out on these animals were totally inconsistent. Unequivocal ST(+) status was evident in no more than 50% of the diseased animals. Herein lies a major note of caution for veterinarians using the CCT on ST(+) reactors in herds which harbour *M. bovis* infection.

4. The farmer has been restricted in sale of his female progeny but he has continued to slaughter male stock, and salvage the carcase value of many of the reactor animals through DSP slaughter.

5. Whereas he faced the prospect of depopulation in 1985 he currently farms 286 animals and the foundation breeding females in his herd are largely intact. The low incidence of disease on the 'high-risk' out-farm would suggest that he may be disease free on this property within the near future.

Figure: Animal movements on "Farm-H"