Bovine brucellosis eradication - the end game

All students of epidemiology ponder the problems of selecting appropriate diagnostic tests for the final phases of disease eradication schemes.[1] On one hand, it is clearly critical that the last residue of infection is found and dealt with firmly. On the other, as virtually all tests yield some false-positives, despite there being no more infection, there will always be test-positive animals to deal with. Managing this balance between sensitivity and specificity is an important issue.

The nature and extent of the false-positive problem in New Zealand became clear from studies[2,3] of brucellosis breakdowns between 1983 and 1988. It was found that many so-called "breakdowns" were, in fact, the result of persistent *B.abortus* strain 19 infection in adult animals. Further, virtually all the test-positive animals from which virulent *B.abortus* was not isolated had been vaccinated as calves. As a result of this work, a policy of attempting to isolate *Brucella* species from reactors in all breakdowns was adopted. At slaughter retropharyngeal, submaxillary, parotid and supramammary lymph nodes, and milk were collected and cultured as described by Corner and Alton.[4]

In November 1989 the final herd was released from movement control restriction. No *B.abortus* cases have been identified since then, but there have been four "testing incidents". This article describes these cases and results of the laboratory investigations.

Incidents since November 1989

In Table 1, data from each of the episodes have been summarised.

Herd A: This was a small South Auckland dairy herd. In November 1989 the bulk-milk test[5] was positive. Blood samples were collected from all animals and one cow was found to have a very high complement fixation (CF) titre. The cow was seven years old, and had been vaccinated with strain 19 as a calf. The herd was not known to have been infected previously, nor had any of the neighbouring herds.

The animal was slaughtered immediately and tissues taken for culture. Strain 19 was isolated from biphasic medium after ten days of incubation.

Herd B: This beef herd was in the Gisborne district. Nine cows reacted to the intradermal brucellin test[6] during the routine triennial surveillance test in May 1990. Blood samples were taken from all animals. One "aged" cow was found to have a very low CF titre (50% lysis at 1:4). The animal had been vaccinated as a calf. There was no history of brucellosis in this herd, nor in the neighbouring herds.

The cow was retested after 102 days. The test results were similar to those recorded previously. The animal was slaughtered and selected tissue cultured as above. *Brucella* species were not isolated.

Herd C: Two animals in a beef herd in the Blenheim district reacted to the brucellin test. One was found to have a CF titre of 1:4. This was an aged cow which had been vaccinated as a calf. There was no history of brucellosis in this area.

The cow was retested after 32 days; similar test results were reported, and so she was slaughtered. At necropsy multiple coalescing abscesses in the lung and liver were found. A pure growth of *E.coli* was isolated from a lung abscess. *Brucella* species were not isolated from the lymph nodes.

Herd D: A pedigree bull in the Waikato district was tested prior to entering an artificial insemination station. A CF titre of 1:8 was reported. Two weeks later the bull was retested. The CF test was negative; there was still a moderate aglutination titre which became negative after rivanol treatment. There was no recent history of brucellosis in the herd of origin. The herd was blood tested with negative results.

Semen from the bull was cultured for *Brucella* species on two occasions, with negative results. The bull was subjected to a full clinical examination. No abnormalities of the urinogenital system were found. There were shallow ulcers in the mouth. On auscultation of the chest, moist rales were apparent. A blood sample was taken and found to have a bovine virus diarrhoea titre of greater than 1:256.

Discussion

During the final phases of a disease eradication scheme there is no easy answer to the problem of achieving both high sensitivity and specificity. The solution lies in developing a coordinated comprehensive management plan. This may involve many issues in addition to the testing procedures used. For example, in brucellosis eradication the withdrawal of strain 19 vaccination is a key strategic issue.

In New Zealand, the testing policy has been to maintain sensitive but cost-effective general surveillance systems, and to achieve high specificity by intensive follow-up. The results to date have been very successful. Since 1983 many herds have been subject to such an investigations and in most cases it was resolved that they were not true breakdowns. In all of the herds cleared in this manner, there has been no further evidence of infection.

The cases reported in this article are similar to many seen during the last five years. In the herd involving an animal with strain 19 infection, of note is the high CF titre and the old age of the animal.

Table 1:. Summary of data from herds where there have been test-positive animals since November 1989.

	A	B	C	D
Brucellin test	ND	Pos	Pos	ND
No. brucellin positive	-	9	2	-
No. CF positive	1	1	1	1
First CF titre	1:128	<1:4	1:4	1:8
Second CF titre	ND	1:4	1:8	Neg
First SA titre	>1:80	1:20	1:20	1:80
Second SA titre	ND	1:40	1:20	1:80
First RT SA titre	>1:80	1:10	Neg	Neg
Second RT SA titre	ND	1:20	Neg	Neg
First BCT	+++	+	+++	++
Second BCT	ND	+++	+++	ND
Days from first to second test	-	102	32	14
Strain 19 vaccinated	Yes	Yes	Yes	?
Age (years)	7	Aged	Aged	5
Post-mortem	Yes	Yes	Yes	No
Culture result	Strain 19	Neg*	Neg*	Neg**

Abbreviations:

CF : Complement fixation
SA : Serum agglutination
RT SA : Rivanol treated serum agglutination
BCT : Brucellosis card test
ND : Not done
? : Not known
Neg* : Negative culture of lymph nodes and milk (see text)
Neg** : Negative culture of semen

Milk from this herd had been tested regularly for years with negative results. One presumes that this is a case of activation of a long standing latent infection. Vaccination ceased in late 1986 and, therefore, sporadic similar cases can still be expected for a number of years.

The use of various treatments or tests[7,8] to separate IgM and IgG activity (e.g. rivanol treatment) is a controversial aspect of brucellosis serology. Of interest in the cases reported here is the lack of effect of rivanol treatment in the strain 19 infected animal. In contrast, with the sera from animals in herds C and D, a marked effect was observed. These cases involved animals with recent acute infection. The rivanol effect is consistent with the hypothesis that the *Brucella* titres were merely non-specific effects of a recent immune stimulation. In earlier investigations animals with acute systemic infections and/or allergies were also observed.

In conclusion, we can state that we are confident none of the cases reported here was due to virulent *B.abortus* infection. For the foreseeable future this policy of intensive follow-up where there is any hint of brucellosis infection will be continued. In New Zealand, brucellosis in cattle is now regarded as an exotic disease[9,10], and all such cases will receive careful and urgent attention.

References

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T J Ryan
MAF Quality Management
Ruakura.