*J. Hyg., Camb.* (1984), **92**, 401–410
*Printed in Great Britain*



# The use of the microagglutination technique to determine the antibody status of healthy New Zealanders to *Brucella abortus*

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(*Received* 12 *December* 1983; *accepted* 6 *January* 1984)

## SUMMARY

Sera from healthy blood donors from different parts of New Zealand, collected between 1977 and 1980, were analysed by the microagglutination technique for antibodies against *Brucella abortus*. Populations from both urban and rural areas were studied. The technique was shown to be capable of handling the 3351 sera studied and thus to be a useful screening test to assess the immune status of large populations. The results demonstrated some of the effects on the human population of the successful bovine brucellosis eradication programme.

## INTRODUCTION

In New Zealand (NZ), human brucellosis has been due to infection by *Brucella abortus* of bovine origin. The NZ Department of Health notifications from 1974 till 1982 suggest that the incidence of human brucellosis is declining (NZ Department of Health monthly notifications). A major contributing factor to this decline is the large decrease in the incidence of bovine brucellosis brought about by the NZ Ministry of Agriculture and Fisheries national eradication programme.

Earlier serological studies (Metcalfe *et al.* 1979, 1981) used sera from healthy blood donors to determine antibody levels to *Br. abortus*. These sera are part of the ongoing collections of the National Serum Bank (NSB), jointly administered by the Department of Health and the Blood Transfusion Services.

In the earlier studies the standard agglutination (SAT) and anti-human globulin (AHGT) tests were performed by the generally accepted tube methods. These are very labour-intensive and require the use of large amounts of reagent. A microagglutination method originally developed by C. P. Beaton (pers. comm.) for the SAT and AHGT was compared with tube methods (Bettelheim, Maskill & Pearce, 1983) and found to be a suitable rapid method for screening large survey populations. This study reports on the use of this microagglutination method to determine the antibody levels to *Br. abortus* in eight collections of sera from throughout New Zealand between 1977 and 1980.

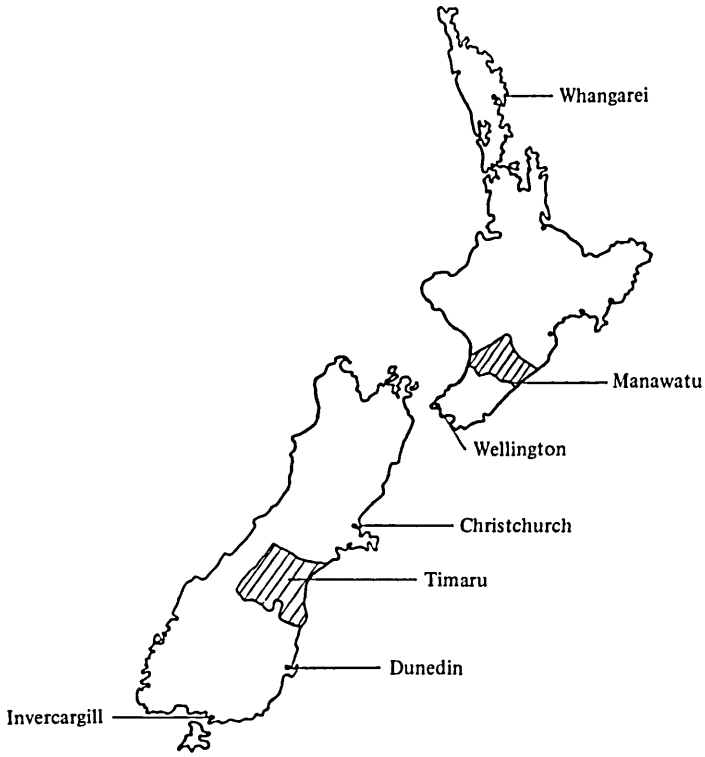


Fig. 1. Locations where serum bank collections were made.

## MATERIALS AND METHODS

*The sera*

The New Zealand National Serum Bank (NSB) is a collection of sera obtained in co-operation with the New Zealand Blood Transfusion Services. This collection is maintained at the National Health Institute. Each serum sample is tested by the Blood Transfusion Services for hepatitis B surface antigen, and only non-reactors are accepted into the bank. It is the intention for each collection from one area to comprise 50 sera from each of the age groups < 20, 20–9, 30–9, 40–9 and > 50 for both males and females. However, this ideal cannot always be achieved, particularly for the < 20 and > 50 age groups.

Sera are collected annually from the Wellington region and also from at least one other major centre of population. Where possible, rural areas are also included, and these have been particularly emphasized in this study. The areas from which the sera in this study have been collected were Dunedin, 1977; a combination of Dunedin and Invercargill, 1978; Wellington, 1978 and 1979; Whangarei, 1979; Christchurch, 1979; Timaru, 1980; and Manawatu, 1980. The locations of these areas are shown on the map (Fig. 1). While there was a rural component in all the collections, those from Timaru and Manawatu were predominantly rural.

Table 1. *Numbers of sera from males of the different collections giving microagglutination reactions*

| Area | Collection date | Age group | No. of sera giving a microagglutination titre of: < 40 | 40 | 80 | 160 | 320 | 640 | > 640 | Total |
|---|---|---|---|---|---|---|---|---|---|---|
| Dunedin | 1977 | < 20 years | 35 | 3 | 1 | — | — | — | — | 39 |
| Dunedin/Invercargill | 1978 | | 22 | 3 | 1 | — | — | — | — | 26 |
| Wellington | 1978 | | 9 | — | 1 | — | — | — | — | 10 |
| Whangarei | 1979 | | 25 | 5 | — | — | — | — | — | 30 |
| Christchurch | 1979 | | 17 | 2 | — | — | — | — | — | 19 |
| Wellington | 1979 | | 11 | 1 | — | — | — | — | — | 12 |
| Timaru | 1980 | | 10 | — | — | — | — | — | — | 10 |
| Manawatu | 1980 | | 46 | 3 | — | 1 | — | — | — | 50 |
| Total (< 20 years) | | | 175 | 17 | 3 | 1 | — | — | — | 196 |
| Dunedin | 1977 | 20–9 years | 101 | 17 | 1 | 1 | — | — | — | 120 |
| Dunedin/Invercargill | 1978 | | 54 | 7 | — | — | — | — | — | 61 |
| Wellington | 1978 | | 49 | 6 | — | — | — | — | — | 55 |
| Whangarei | 1979 | | 50 | 7 | — | — | — | — | — | 57 |
| Christchurch | 1979 | | 49 | 9 | — | — | — | — | — | 58 |
| Wellington | 1979 | | 56 | 3 | — | — | — | — | — | 59 |
| Timaru | 1980 | | 39 | 1 | 1 | — | — | — | — | 41 |
| Manawatu | 1980 | | 37 | 8 | 4 | — | — | — | — | 49 |
| Total (20–9 years) | | | 435 | 58 | 6 | 1 | — | — | — | 500 |
| Dunedin | 1977 | 30–9 years | 60 | 20 | 2 | 1 | 1 | — | — | 84 |
| Dunedin/Invercargill | 1978 | | 66 | 12 | 1 | — | — | — | — | 79 |
| Wellington | 1978 | | 40 | 11 | — | 1 | — | — | — | 52 |
| Whangarei | 1979 | | 36 | 14 | 2 | 1 | — | — | — | 53 |
| Christchurch | 1979 | | 57 | 20 | — | 1 | — | — | — | 78 |
| Wellington | 1979 | | 68 | 6 | — | — | — | — | — | 74 |
| Timaru | 1980 | | 38 | — | — | — | — | — | — | 38 |
| Manawatu | 1980 | | 29 | 14 | 4 | 3 | — | — | — | 50 |
| Total (30–9 years) | | | 394 | 97 | 9 | 7 | 1 | — | — | 508 |
| Dunedin | 1977 | 40–9 years | 39 | 12 | 2 | — | — | — | — | 53 |
| Dunedin/Invercargill | 1978 | | 38 | 8 | 2 | — | — | — | — | 48 |
| Wellington | 1978 | | 38 | 9 | — | — | — | — | — | 47 |
| Whangarei | 1979 | | 34 | 23 | — | — | — | — | — | 57 |
| Christchurch | 1979 | | 44 | 16 | — | — | — | — | — | 60 |
| Wellington | 1979 | | 46 | 7 | 1 | — | — | — | — | 54 |
| Timaru | 1980 | | 46 | 2 | 1 | — | — | — | — | 49 |
| Manawatu | 1980 | | 26 | 17 | 6 | 1 | — | — | — | 50 |
| Total (40–9 years) | | | 311 | 94 | 12 | 1 | — | — | — | 418 |
| Dunedin | 1977 | > 50 years | 19 | 10 | 1 | 1 | — | — | — | 31 |
| Dunedin/Invercargill | 1978 | | 25 | 11 | — | — | — | — | — | 36 |
| Wellington | 1978 | | 28 | 7 | — | 1 | — | — | — | 36 |
| Whangarei | 1979 | | 23 | 11 | — | — | — | — | — | 34 |
| Christchurch | 1979 | | 46 | 17 | — | — | — | — | — | 63 |
| Wellington | 1979 | | 45 | 10 | — | 1 | 1 | — | — | 57 |
| Timaru | 1980 | | 48 | 3 | — | — | — | — | — | 51 |
| Manawatu | 1980 | | 31 | 10 | 7 | 2 | — | — | — | 50 |
| Total (> 50 years) | | | 265 | 79 | 8 | 5 | 1 | — | — | 358 |
| Total (all males) | | | 1580 | 345 | 38 | 15 | 2 | — | — | 1980 |

*The microagglutination method*

This was performed in rigid V-bottomed microtitre plates exactly as described by Bettelheim *et al.* (1983). Both the micro version of the SAT (MSAT) and the AHGT (MAHGT) were performed on each serum sample. The same control sera described for the earlier study were employed.

Table 2. *Numbers of sera from females of the different collections giving microagglutination reactions*

| Area | Collection date | Age group | Microagglutination titre of | | | | | | | Total |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | < 40 | 40 | 80 | 160 | 320 | 640 | > 640 | |
| Dunedin | 1977 | < 20 years | 24 | 6 | — | 1 | — | — | — | 31 |
| Dunedin/Invercargill | 1978 | | 20 | 4 | — | — | — | — | — | 24 |
| Wellington | 1978 | | 8 | 1 | — | — | — | — | — | 9 |
| Whangarei | 1979 | | 24 | 5 | — | — | — | — | — | 29 |
| Christchurch | 1979 | | 21 | — | — | — | — | — | — | 21 |
| Wellington | 1979 | | 13 | 1 | — | — | — | — | — | 14 |
| Timaru | 1980 | | 1 | — | — | — | — | — | — | 1 |
| Manawatu | 1980 | | 36 | 14 | — | — | — | — | — | 50 |
| Total (< 20 years) | | | 147 | 31 | — | 1 | — | — | — | 179 |
| Dunedin | 1977 | 20–9 years | 44 | 11 | 1 | — | — | — | — | 56 |
| Dunedin/Invercargill | 1978 | | 39 | 9 | — | — | — | — | — | 48 |
| Wellington | 1978 | | 30 | 11 | 1 | — | — | — | — | 42 |
| Whangarei | 1979 | | 26 | 8 | 1 | — | — | — | — | 35 |
| Christchurch | 1979 | | 51 | 10 | 1 | — | — | — | — | 62 |
| Wellington | 1979 | | 22 | 4 | 2 | — | — | — | — | 28 |
| Timaru | 1980 | | 14 | 1 | — | — | — | — | — | 15 |
| Manawatu | 1980 | | 39 | 7 | 4 | — | — | — | — | 50 |
| Total (20–9 years) | | | 265 | 61 | 10 | — | — | — | — | 336 |
| Dunedin | 1977 | 30–9 years | 35 | 13 | — | 2 | — | — | — | 50 |
| Dunedin/Invercargill | 1978 | | 58 | 17 | — | 1 | — | — | — | 76 |
| Wellington | 1978 | | 27 | 6 | — | — | — | — | — | 33 |
| Whangarei | 1979 | | 29 | 10 | — | — | — | — | — | 39 |
| Christchurch | 1979 | | 46 | 9 | — | — | — | — | — | 55 |
| Wellington | 1979 | | 35 | 6 | — | — | — | — | — | 41 |
| Timaru | 1980 | | 20 | — | — | — | — | — | — | 20 |
| Manawatu | 1980 | | 41 | 8 | 1 | — | — | — | — | 50 |
| Total (30–9 years) | | | 291 | 69 | 1 | 3 | — | — | — | 364 |
| Dunedin | 1977 | 40–9 years | 20 | 11 | — | — | — | — | — | 31 |
| Dunedin/Invercargill | 1978 | | 36 | 4 | 4 | — | — | — | — | 44 |
| Wellington | 1978 | | 22 | 6 | 1 | — | — | — | — | 29 |
| Whangarei | 1979 | | 20 | 13 | 3 | — | — | — | — | 36 |
| Christchurch | 1979 | | 29 | 7 | 1 | — | — | — | — | 37 |
| Wellington | 1979 | | 35 | 8 | — | — | — | — | — | 43 |
| Timaru | 1980 | | 14 | 1 | — | — | — | — | — | 15 |
| Manawatu | 1980 | | 37 | 9 | 4 | — | — | — | — | 50 |
| Total (40–9 years) | | | 213 | 59 | 13 | — | — | — | — | 285 |
| Dunedin | 1977 | > 50 years | 13 | 2 | 2 | — | — | — | — | 17 |
| Dunedin/Invercargill | 1978 | | 20 | 2 | — | — | — | — | — | 22 |
| Wellington | 1978 | | 15 | 6 | — | 1 | — | — | — | 22 |
| Whangarei | 1979 | | 22 | 9 | 1 | — | — | — | — | 32 |
| Christchurch | 1979 | | 18 | 11 | — | — | — | — | — | 29 |
| Wellington | 1979 | | 25 | 7 | — | — | — | — | — | 32 |
| Timaru | 1980 | | 11 | 1 | — | — | — | — | — | 12 |
| Manawatu | 1980 | | 31 | 5 | 3 | 1 | 1 | — | — | 41 |
| Total (> 50 years) | | | 155 | 43 | 6 | 2 | 1 | — | — | 207 |
| Total (all females) | | | 1078 | 263 | 30 | 6 | 1 | — | — | 1371 |

*Results*

In all it was possible to examine 3351 sera – 1980 from men and 1371 from women – among the eight collections. The results, which have been grouped by test (MSAT or MAHGT), sex and age group of donor, and area and year of collection, are presented in Tables 1–4.

Table 3. *Numbers of sera from males of the different collections giving microanti-human globulin reactions*

| Area | Collection date | Age group | Microanti-human globulin titre of | | | | | | | Total |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | < 40 | 40 | 80 | 160 | 320 | 640 | > 640 | |
| Dunedin | 1977 | < 20 years | 36 | 2 | 1 | — | — | — | — | 39 |
| Dunedin/Invercargill | 1978 | | 22 | 2 | 1 | — | 1 | — | — | 26 |
| Wellington | 1978 | | 9 | — | — | 1 | — | — | — | 10 |
| Whangarei | 1979 | | 26 | 4 | — | — | — | — | — | 30 |
| Christchurch | 1979 | | 17 | 2 | — | — | — | — | — | 19 |
| Wellington | 1979 | | 11 | 1 | — | — | — | — | — | 12 |
| Timaru | 1980 | | 10 | — | — | — | — | — | — | 10 |
| Manawatu | 1980 | | 46 | 3 | — | 1 | — | — | — | 50 |
| Total (< 20 years) | | | 177 | 14 | 2 | 2 | 1 | — | — | 196 |
| Dunedin | 1977 | 20–9 years | 95 | 18 | 6 | 1 | — | — | — | 120 |
| Dunedn/Invercargill | 1978 | | 52 | 9 | — | — | — | — | — | 61 |
| Wellington | 1978 | | 48 | 6 | 1 | — | — | — | — | 55 |
| Whangarei | 1979 | | 50 | 6 | — | 1 | — | — | — | 57 |
| Christchurch | 1979 | | 49 | 9 | — | — | — | — | — | 58 |
| Wellington | 1979 | | 49 | 9 | 1 | — | — | — | — | 59 |
| Timaru | 1980 | | 35 | 4 | — | 2 | — | — | — | 41 |
| Manawatu | 1980 | | 35 | 13 | 1 | — | — | — | — | 49 |
| Total (20–9 years) | | | 413 | 74 | 9 | 4 | — | — | — | 500 |
| Dunedin | 1977 | 30–9 years | 61 | 16 | 3 | 2 | 2 | — | — | 84 |
| Dunedin/Invercargill | 1978 | | 61 | 14 | 3 | — | 1 | — | — | 79 |
| Wellington | 1978 | | 38 | 8 | 4 | — | 1 | — | 1 | 52 |
| Whangarei | 1979 | | 31 | 16 | 2 | 3 | 1 | — | — | 53 |
| Christchurch | 1979 | | 56 | 21 | — | 1 | — | — | — | 78 |
| Wellington | 1979 | | 61 | 11 | 2 | — | — | — | — | 74 |
| Timaru | 1980 | | 34 | 3 | 1 | — | — | — | — | 38 |
| Manawatu | 1980 | | 30 | 14 | 4 | 1 | 1 | — | — | 50 |
| Total (30–9 years) | | | 372 | 103 | 19 | 7 | 6 | — | 1 | 508 |
| Dunedin | 1977 | 40–9 years | 40 | 10 | 2 | 1 | — | — | — | 53 |
| Dunedin/Invercargill | 1978 | | 34 | 10 | 3 | 1 | — | — | — | 48 |
| Wellington | 1978 | | 31 | 10 | 5 | — | — | — | 1 | 47 |
| Whangarei | 1979 | | 25 | 30 | 1 | 1 | — | — | — | 57 |
| Christchurch | 1979 | | 40 | 18 | 1 | — | — | — | 1 | 60 |
| Wellington | 1979 | | 42 | 11 | — | — | 1 | — | — | 54 |
| Timaru | 1980 | | 38 | 3 | 5 | 3 | — | — | — | 49 |
| Manawatu | 1980 | | 26 | 18 | 5 | — | 1 | — | — | 50 |
| Total (40–9 years) | | | 276 | 110 | 22 | 6 | 2 | — | 2 | 418 |
| Dunedim | 1977 | > 50 years | 18 | 11 | — | 1 | — | 1 | — | 31 |
| Dunedin/Invercargill | 1978 | | 27 | 8 | 1 | — | — | — | — | 36 |
| Wellington | 1978 | | 26 | 8 | — | 2 | — | — | — | 36 |
| Whangarei | 1979 | | 24 | 9 | — | 1 | — | — | — | 34 |
| Christchurch | 1979 | | 43 | 17 | 3 | — | — | — | — | 63 |
| Wellington | 1979 | | 40 | 10 | 1 | 2 | 3 | 1 | — | 57 |
| Timaru | 1980 | | 44 | 6 | 1 | — | — | — | — | 51 |
| Manawatu | 1980 | | 20 | 15 | 11 | 4 | — | — | — | 50 |
| Total (> 50 years) | | | 242 | 84 | 17 | 10 | 3 | 2 | — | 358 |
| Total (all males) | | | 1480 | 385 | 69 | 29 | 12 | 2 | 3 | 1980 |

## DISCUSSION

The economy of reagents and the ease of automation make the micro methods particularly useful for screening large populations for antibodies to *Brucella*. The earlier study involving this technique (Bettelheim *et al.* 1983) showed that the micro methods did not give false negative results. Moreover, where a difference

Table 4. *Numbers of sera from females of the different collections giving microanti-human globulin reactions*

| Area | Collection date | Age group | Microanti-human globulin titre of | | | | | | | Total |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | < 40 | 40 | 80 | 160 | 320 | 640 | > 640 | |
| Dunedin | 1977 | < 20 years | 26 | 4 | — | 1 | — | — | — | 31 |
| Dunedin/Invercargill | 1978 | | 22 | 2 | — | — | — | — | — | 24 |
| Wellington | 1978 | | 8 | 1 | — | — | — | — | — | 9 |
| Whangarei | 1979 | | 18 | 9 | 2 | — | — | — | — | 29 |
| Christchurch | 1979 | | 17 | 4 | — | — | — | — | — | 21 |
| Wellington | 1979 | | 14 | — | — | — | — | — | — | 14 |
| Timaru | 1980 | | 1 | — | — | — | — | — | — | 1 |
| Manawatu | 1980 | | 33 | 15 | 2 | — | — | — | — | 50 |
| Total (< 20 years) | | | 139 | 35 | 4 | 1 | — | — | — | 179 |
| Dunedin | 1977 | 20–9 years | 43 | 12 | 1 | — | — | — | — | 56 |
| Dunedin/Invercargill | 1978 | | 36 | 10 | 1 | 1 | — | — | — | 48 |
| Wellington | 1978 | | 31 | 10 | — | — | 1 | — | — | 42 |
| Whangarei | 1979 | | 25 | 8 | — | 2 | — | — | — | 35 |
| Christchurch | 1979 | | 49 | 12 | — | 1 | — | — | — | 62 |
| Wellington | 1979 | | 20 | 5 | — | 2 | 1 | — | — | 28 |
| Timaru | 1980 | | 12 | 2 | 1 | — | — | — | — | 15 |
| Manawatu | 1980 | | 33 | 13 | 4 | — | — | — | — | 50 |
| Total (20–9 years) | | | 249 | 72 | 7 | 6 | 2 | — | — | 336 |
| Dunedin | 1977 | 30–9 years | 37 | 9 | 1 | 2 | 1 | — | — | 50 |
| Dunedin/Invercargill | 1978 | | 56 | 18 | 1 | — | 1 | — | — | 76 |
| Wellington | 1978 | | 22 | 7 | 3 | 1 | — | — | — | 33 |
| Whangarei | 1979 | | 24 | 13 | 2 | — | — | — | — | 39 |
| Christchurch | 1979 | | 42 | 12 | 1 | — | — | — | — | 55 |
| Wellington | 1979 | | 29 | 10 | 2 | — | — | — | — | 41 |
| Timaru | 1980 | | 18 | 2 | — | — | — | — | — | 20 |
| Manawatu | 1980 | | 36 | 11 | 3 | — | — | — | — | 50 |
| Total (30–9 years | | | 264 | 82 | 13 | 3 | 2 | — | — | 364 |
| Dunedin | 1977 | 40–9 years | 19 | 10 | 2 | — | — | — | — | 31 |
| Dunedin/Invercargill | 1978 | | 28 | 11 | 3 | 1 | — | 1 | — | 44 |
| Wellington | 1978 | | 19 | 8 | 1 | 1 | — | — | — | 29 |
| Whangarei | 1979 | | 17 | 13 | 3 | 3 | — | — | — | 36 |
| Christchurch | 1979 | | 25 | 7 | — | 4 | — | — | 1 | 37 |
| Wellington | 1979 | | 27 | 13 | 2 | 1 | — | — | — | 43 |
| Timaru | 1980 | | 12 | 2 | 1 | — | — | — | — | 15 |
| Manawatu | 1980 | | 34 | 9 | 4 | 3 | — | — | — | 50 |
| Total (40–9 years) | | | 181 | 73 | 16 | 13 | — | 1 | 1 | 285 |
| Dunedin | 1977 | > 50 years | 12 | 3 | 1 | 1 | — | — | — | 17 |
| Dunedin/Invercargill | 1978 | | 18 | 2 | 2 | — | — | — | — | 22 |
| Wellington | 1978 | | 15 | 6 | — | — | 1 | — | — | 22 |
| Whangarei | 1979 | | 18 | 13 | — | 1 | — | — | — | 32 |
| Christchurch | 1979 | | 21 | 6 | 2 | — | — | — | — | 29 |
| Wellington | 1979 | | 17 | 14 | 1 | — | — | — | — | 32 |
| Timaru | 1980 | | 11 | 1 | — | — | — | — | — | 12 |
| Manawatu | 1980 | | 30 | 5 | 3 | 2 | 1 | — | — | 41 |
| Total (> 50 years) | | | 142 | 50 | 9 | 4 | 2 | — | — | 207 |
| Total (all females) | | | 975 | 312 | 49 | 27 | 6 | 1 | 1 | 1371 |

of more than one dilution step occurred between the micro methods and the traditional tube methods, the titres obtained by the micro methods were higher in almost all (97·4 %) cases. Therefore it was considered appropriate to continue the analysis of serum bank collections by means of the microagglutination methods.

As Tables 1 and 2 show, there are only two out of 1,980 males and only one out of 1,371 females with titres of $\geqq 1:320$ in the MSAT. It is generally considered

https://doi.org/10.1017/S0022172400064627 Published online by Cambridge University Press

Table 5. *Percentage of sera giving a titre of* ≧ 80 *in the microagglutination and microanti-human globulin reactions from all collections*

| Area | Collection date | Sex | Percentage of sera giving micro-agglutination titres of ≧ 80 of the different age groups | | | | | All age groups | Percentage of sera giving micro-anti-human globulin titres of ≧ 80 of the different age groups | | | | | All age groups |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | < 20 | 20–9 | 30–9 | 40–9 | > 50 | | < 20 | 20–9 | 30–9 | 40–9 | > 50 | |
| Dunedin | 1977 | Males | 2·6 | 1·67 | 4·8 | 3·8 | 6·5 | 3·4 | 2·6 | 5·8 | 8·3 | 5·7 | 6·5 | 6·1 |
| Dunedin/Invercargill | 1978 | | 3·8 | — | 1·3 | 4·2 | — | 1·6 | 7·7 | — | 5·1 | 8·3 | 2·8 | 4·4 |
| Wellington | 1978 | | 10·0 | — | 1·9 | — | 2·8 | 1·5 | 10·0 | 1·8 | 11·5 | 12·8 | 5·6 | 7·0 |
| Whangarei | 1979 | | — | — | 5·7 | — | — | 1·3 | — | 1·8 | 11·3 | 3·5 | 2·9 | 4·3 |
| Christchurch | 1979 | | — | — | 1·2 | — | — | 0·4 | — | — | 1·3 | 3·3 | 4·8 | 2·2 |
| Wellington | 1979 | | — | — | — | 1·9 | 3·5 | 1·2 | — | 1·7 | 2·7 | 1·9 | 12·2 | 4·3 |
| Timaru | 1980 | | — | 2·4 | — | 2·0 | — | 1·1 | — | 4·9 | 2·6 | 16·3 | 1·9 | 6·3 |
| Manawatu | 1980 | | 2·0 | 8·1 | 14·0 | 14·0 | 18·0 | 11·2 | 2·0 | 2·0 | 12·0 | 12·0 | 30·0 | 11·7 |
| All males | | | 2·0 | 1·4 | 3·3 | 3·1 | 3·9 | 2·8 | 2·6 | 2·6 | 6·5 | 7·6 | 8·9 | 5·8 |
| Dunedin | 1977 | Females | 3·2 | 1·8 | 4·0 | — | 11·8 | 3·2 | 3·2 | 1·8 | 8·0 | 6·5 | 11·8 | 5·4 |
| Dunedin/Invercargill | 1978 | | — | — | 1·3 | 9·1 | — | 2·3 | — | 4·2 | 2·6 | 11·4 | 9·1 | 5·1 |
| Wellington | 1978 | | — | 2·4 | — | 3·4 | 4·5 | 2·2 | — | 2·4 | 12·1 | 6·9 | 4·5 | 5·9 |
| Whangarei | 1979 | | — | 2·9 | — | 8·3 | 3·1 | 2·9 | 6·8 | 5·7 | 5·1 | 16·7 | 3·1 | 7·6 |
| Christchurch | 1979 | | — | 1·6 | — | 2·7 | — | 1·0 | — | 1·6 | 1·8 | 13·5 | 6·9 | 4·4 |
| Wellington | 1979 | | — | 7·1 | — | — | — | 1·3 | — | 10·7 | 4·9 | 7·0 | 3·1 | 5·7 |
| Timaru | 1980 | | — | — | — | — | — | — | — | 6·7 | — | 6·7 | — | 3·1 |
| Manawatu | 1980 | | — | 8·0 | 2·0 | 8·0 | 12·2 | 5·8 | 4·0 | 8·0 | 6·0 | 14·0 | 14·6 | 9·5 |
| All females | | | 0·6 | 3·0 | 1·1 | 4·6 | 4·3 | 2·7 | 2·8 | 4·5 | 4·9 | 10·9 | 7·2 | 6·1 |
| All males and females | | | 1·3 | 2·0 | 2·4 | 3·7 | 4·1 | 2·7 | 2·6 | 3·3 | 5·8 | 9·0 | 8·3 | 5·9 |

that a titre of ≧ 1:80 represents the presence of specific agglutinating antibodies to *Br. abortus* (Kerr *et al.* 1966). For the purposes of this discussion this cut-off titre will be taken as representing specific reactors. In Tables 1 and 2 there are 92 (2·7 %) sera with a titre of ≧ 1:80. Of these, 55 are from males and 37 from females. While these numbers are low there is a general trend of increasing numbers of reactors in the higher age groups. These results are summarized as percentages in Table 5.

There are more sera with high MAHGT antibody levels than there are with high MSAT levels: 17 sera from males with titres of ≧ 1:320, and eight from females (Tables 3 and 4). There are 199 (5·9 %) sera with titres of ≧ 1:80, and of these 115 were males and 84 were females. As summarized in Table 5, there is a more marked trend of increasing numbers of MAHGT reactors with the older age groups as was noted for the MSAT.

Since 1974 the Wellington region has been sampled every year except 1975. The figures for 1974 and 1976 (Metcalfe *et al.* 1979) and for 1977 (Metcalfe *et al.* 1981) have already suggested a downward trend in numbers of reactors. The current studies include two further Wellington collections in 1978 and 1979, and these confirm this downward trend. It is particularly noteworthy that these trends can be documented in a predominantly urban population. Although only one previous collection was made in Christchurch in 1975 (Metcalfe *et al.* 1979), a comparison of the current figures with those obtained earlier again demonstrates a downward trend. These downward trends are considered particularly noteworthy as more sensitive methods are being used.

An examination of urban centres studied in the same year, i.e. Dunedin, Dunedin/Invercargill and Wellington in 1978, and Christchurch and Wellington in 1979, shows no noticeable difference in the numbers of reactors for the MSAT. However, the AHGT for the males shows more reactors in both Wellington collections than the South Island collection performed in the same year. This difference is not as marked for the females. The collection from Whangarei in 1979, which is further north, shows a higher reactor rate for females than Wellington, 1979, for the MAHGT (Table 5).

In a study such as this one it is important to examine human brucellosis in the context of animal brucellosis in New Zealand. Of the collections of sera tested in this study, most came from urban centres although there was a rural component in all. The two predominantly rural areas were Timaru and Manawatu.

Timaru has been considered clear of brucellosis since the end of 1979. The period 1972–6 saw a reduction in the proportion of infected beef herds to 4·1 % and of dairy herds to 1·4 %. By 1979 only one beef herd still contained reactors and there was no reactor in beef or dairy herds by 1980. From 1977 the milk ring test has been consistently negative on samples of milk taken in this area.

In the Manawatu area the proportion of infected herds had been reduced to only 24·6 % for beef herds and 22 % for dairy herds by 1976. In 1979 there were still 2·0 % of beef herds containing reactors and 1·7 % of dairy herds. These were reduced to 1·9 % and 0·8 % respectively in 1980. A proportion of samples of milk from this area gave a positive milk ring test.

Both these areas are rather similar in agricultural activity, but in Manawatu there are about three times as many dairy herds as in Timaru. The numbers of

beef herds are similar. As shown in Tables 1 and 2, the MSAT demonstrates that both males and females of the Manawatu collection have a greater reactor rate (giving a titre $\geqq 1:80$) than those in Timaru.

The summary in Table 5 further demonstrates this observation. This difference between the two collections, although still present, is not as marked for the MAHGT (Tables 3 and 4).

An examination of the numbers of reactors belonging to the different age groups in the two rural collections demonstrates that there is a clear rise in numbers of reactors of both males and females in the MSAT in Manawatu which is not present in Timaru. Of all collections studied, the reactor rate for the MSAT in Manawatu was the highest and showed the most consistent increase with age, both among males and females (Tables 1, 2 and 5). It should be noted that these titres do not imply that the older age groups are more likely to have had recent infections but supports the concept of persistence of *Brucella* agglutinating antibodies as described by other workers (Reddin *et al.* 1965; Kerr *et al.* 1968; Farrell, Robertson & Hinchliffe, 1975).

A similar examination of the numbers of reactors for the MAHGT shows a greater number of sera with higher titres for all collections than the number of MSAT reactors. This is particularly noteworthy in the older age groups (Table 3), and emerges clearly in a comparison between Timaru and Manawatu (Table 5).

The MAHGT, by identifying mainly IgG antibodies, is indicative of residual antibodies from past or persisting antigen exposure. The survival of IgG antibodies in the older urban populations is therefore considered a reflection of earlier exposure.

The summary of the bovine brucellosis eradication programme of the Ministry of Agriculture and Fisheries given earlier indicates that people in Manawatu were still exposed at the time of serum collection to *Brucella*, while those in Timaru were not. The continuing exposure of the Manawatu population to *Brucella* may account for the rise of numbers of reactors with age group in the MAHGT. This exposure may act like a continuous vaccination procedure maintaining high IgG levels (Coghlan & Weir, 1967).

This study demonstrates that in order to assess the immune status of large populations to an agent such as *Br. abortus*, a rapid and inexpensive screening test such as the microagglutination methods described by Bettelheim *et al.* (1983) is the preferred alternative to the more labour-intensive and expensive tube methods. Only when the microagglutination methods became available could it be readily confirmed that the *Brucella* agglutinating antibody levels were in fact lower in a human population in an area in which the bovine brucellosis eradication programme had been successful in removing all infected cattle and hence eliminating exposure to *Brucella*. Further, only by such methods can regular monitoring of urban populations be undertaken. Thus the effectiveness of the national bovine brucellosis eradication programme can regularly be monitored in human populations.

This paper is published with the authority of the Director-General of Health, Department of Health, Wellington, New Zealand.

The supply of details of the bovine brucellosis eradication programmes by the Animal Health Division of the New Zealand Ministry of Agriculture and Fisheries is gratefully acknowledged.

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