

THE QUEST FOR 'BRUCELLA MELITENSIS'
IN MAN AND IN THE GOAT

An Historical Retrospect

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Surgeon Rear Admiral P.W. Bassett-Smith, when referring to the achievements of the Mediterranean Fever Commission of 1904, stated that "no modern scientific work had been more successful or of greater use to humanity" (1). One must, however, also add that no less than eighteen years of painful failure and frustration elapsed from the discovery by Bruce of the causative germ of Mediterranean Fever in man to its discovery in the goat by Zammit in 1905. Indeed the early story of 'Brucella melitensis' affords an admirable example of the vagaries of research work, of how contemporary medical concepts may deflect the research worker from the right path by restricting his focus of attention thus missing the connecting links in the transmission of disease, of how he sometimes rubs shoulders with the vector of disease and fails to notice it, and of how the intrusion of a chance observation may bring to a successful outcome long years of planned but fruitless investigations. It is these aspects of the story of 'Brucella melitensis' that this paper sets out to relate.

Among the most important practical acquisitions of the nineteenth century in the field of medicine were (a) the identification of various specific diseases which until then had been grouped in a large undifferentiated group of "fevers"; and (b) the birth and development of bacteriology. Thus, to cite one example, typhus had by 1860 been clinically separated from typhoid although this latter term continued to be applied not only to enteric fever as we know it today but also to that state of any feverish disease marked by great prostration of the nerv-

(1) *The Daily Malta Chronicle* of 18th February 1921, p. 8.

ous and of the vascular systems (2). By 1865 Joseph Lister had shown that septic conditions were due to the presence of bacteria although it was not until 1870 that bacteriological principles were applied to the problems of infectious diseases (3). Until then, and for a number of years afterwards, emanations from untrapped drains, effluvia from badly constructed sewers, "miasmas" and "contagion" were thought to be the causes of "fevers"; while in the case of typhoid it was even held that the disease was capable of spontaneous origin (4). Considering, therefore, the state of the medical knowledge of the time it is not to be wondered at that the aetiology and mode of propagation of Mediterranean Fever were still shrouded in mystery.

The first detailed clinical study of fevers in Malta and of Mediterranean Fever in particular was undertaken in 1860 by Assistant Surgeon J.A. Marston, M.D., Royal Artillery. His description of the clinical picture, course, duration and complications of Mediterranean Fever is masterly. That it is "accurate and exact" is due, in part, to the fact that he himself suffered from the disease at the age of twenty-eight years during his stay in Malta. Besides a lucid and comprehensive account of its morbid anatomy, he also differentiated it clearly from enteric fever and recognised that "gastric remittent fever" and "bilious fever" were the same disease, the difference being but one of degree.

With regard to its aetiology, however, Marston's speculations were quite wide of the mark. "If I were asked to indicate the cause of this disease" he says, "I would suggest the defective house-drains. Without wishing to speak at all dogmatically upon this point, I would go further and say that ... the evaporation from damp houses, defective in surface and other drainage, producing other than faecal stinks are the agents in the gastric

(2) MARSTON, J.A., *Report on Fever (Malta)*, Army Medical Department, *Medical Report for 1861*, Vol. III, London, 1863, p. 487.

(3) DALE, H., *The changing Face of Medicine 1868-1951; The Practitioner*, 1951, 167, 313 (October).

(4) MARSTON, J.A., *op. cit.*, p. 493.

remittent type" (5). In these words Marston was simply echoing the epidemiological ideas of his time.

The elucidation of the cause of Mediterranean Fever came twenty-seven years later from another Army Medical Officer in Malta. This was Surgeon (later Sir) David Bruce who was appointed to the Army Medical Service in 1883. He was in the Island in 1887 on the staff of the Station Hospital, formerly the Holy Infirmary of the Knights of St. John, at Valletta. Ninety-one British soldiers had been treated for Mediterranean Fever in this Hospital during 1886. The duration of the disease was long and its after-effects disabling but its mortality was so small that not one single death occurred among them. During the first six months of the following year, however, as many as nine deaths from this fever were registered. Bruce carried out post-mortem examinations. He observed that microscopically the condition of the spleen and other organs was very similar to what obtained in bacterial diseases. He, therefore, decided to search for the possible presence of micro-organisms in other fatal cases. His first investigation was carried out on the 26th December 1886. He prepared stained sections from the human spleen which revealed "enormous numbers of single micrococci" scattered throughout the tissues. In spite of this remarkable discovery Bruce, having gone on leave of absence, did not pursue his investigations until May 1887 when, in conjunction with the Maltese physician Dr. Joseph Caruana Scicluna, the Government Analyst, he prepared tubes of agar-agar nutrient jelly with the intention of working conjointly with his Maltese colleague at the distribution of bacteria in the air in different parts of Malta. As, however, there were many cases of Mediterranean Fever in the Station Hospital at this time, Bruce and Caruana Scicluna decided to study these patients by inoculating blood taken from them into tubes of agar-agar jelly. No definite results were obtained but when one of the patients died on the 6th July, 1887, Bruce removed the spleen from the cadaver and took it to a small room in his quarters as he had learned by experience that inoculations made in the mor-

(5) *Ibidem*, p. 511.

tuary were almost certain to become contaminated. In the same evening he inoculated eight tubes with a small portion of the spleen pulp. The following day six of the tubes were placed in the incubator in Dr. Joseph Caruana Scicluna's laboratory; the other two tubes were kept at the ordinary temperature of the air. One and the same growth of colonies appeared in all the eight tubes. On examination under the microscope, Bruce saw innumerable micrococci. "They are very active", he wrote, "and dance about -- as a rule singly, sometimes in pairs, rarely in short chains" (6). On the occurrence of another fatality on the 23rd July 1887 Bruce examined the splenic pulp directly. "The result", he says, "was I confess, somewhat startling... The field of the microscope was literally crowded with myriads of micrococci dancing in the most active manner".

Bruce had thus tracked down the germ of Mediterranean Fever with the help of Dr. Joseph Caruana Scicluna which he thus acknowledged:— "I must here thank Dr. Caruana Scicluna for the very great assistance he has given me not only in preparing sterilized fluids but also in supplying apparatus etc.; in fact without his co-operation the following results could not have been attained" (7). Bruce's experiments were confirmed in 1891 by Surgeon Captain M. Louis Hughes of the Army Medical Staff, Malta (8); and yet in 1894 a Naval Medical Officer declared at a meeting of the Malta Branch of the British Medical Association that he could not believe in the germ theory of Mediterranean Fever and that in his view the illness was due to the presence of ptomaines frequently found in the bodies of persons who did not take enough exercise; as, however, the existence of the germ could not be ignored he was prepared to admit that the germ

(6) BRUCE, D., *Note on the Discovery of a Micro-organism in Malta Fever. The Practitioner*, 1887, 39, 166 (September).

(7) BRUCE, D., *op., cit.*, p. 163.

(8) HUGHES, M.L., *Note on the Endemic Fever of the Mediterranean Medico-Chirurgical Transactions*, 1896, 79.

was present "as an effect and not as the cause of the fever" (9).

If any doubts still existed regarding the casual connection of Bruce's micrococcus with Mediterranean Fever, they were dispelled by Dr. (later Sir) Themistocles Zammit when at a meeting of the Malta Branch of the B.M.A. on the 19th May, 1899, he read a paper on the "Serum Diagnosis of Mediterranean Fever". He not only demonstrated the advantages of Widal's Method when applied to Mediterranean Fever but also showed under the microscope the coagulation of the Bruce micrococcus when treated with blood serum taken from patients suffering from the fever. He likewise proved that the germ could be grown successfully from a culture seven months old and that cultures two years old gave a clear serum reaction.

One of the enigmas of Mediterranean Fever had been definitely solved but two other questions of the utmost practical value still tormented men's minds:— (a) Where was the germ to be found outside the human body? and (b) How did it gain entrance into its human host?

The outstanding theme of the medical thought of the second half of the nineteenth century was the promotion of public health and the prevention of epidemics of cholera and scarlet fever. The establishment of drainage system and the provision of clean water supplies brought a substantial reduction in morbidity and mortality in urban areas but the ideas of the hygienists about the supposed ill-effects of sewer air and water effluvia so befogged men's minds that they were unable to shift their focus of attention from these concepts of aetiology and to explore unfamiliar ways of investigation. Hence we find, in 1893, medical men in Malta thinking that the germ of Mediterranean Fever was conveyed by man by polluted drinking water; others blaming the sea in our harbours for outbreaks of the fever on board ships (10). The suggestion was also made that the "poison" of the fever was inhaled with the air and the hope was expressed that an

(9) *Minutes of the Malta Branch of the B.M.A.*, meetings of 16th May and 28th June, 1894.

(10) *Ibidem*, meeting of the 4th April, 1893.

examination of the atmosphere would reveal the micro-organism "floating in the air of sewers ventilators" (11). Others pointed to dust from infected places as the vehicle of infection. As late as 1896, Surgeon Captain M.L. Hughes could only recommend as a prevention against the disease, the constant flushing of main and house drains, the raising of drain ventilators above and away from all windows and the avoidance of damp and of overcrowding (12). There was only one man — Surgeon Major Tidbury A.M.S. — to suggest in 1894 that the origin of the fever was to be found in water and milk. The water theory was followed up but the milk hypothesis was ruled out without being discussed or investigated, although the goat was then an ubiquitous animal in the Maltese Islands. Indeed large herds of goats daily paraded our streets in both town and village, to be milked at the customer's doorstep. It was no less a person than Hughes himself who stated that he had met no fact that favoured a causal connection between the milk supply and Mediterranean Fever (13). On another occasion, while describing a hospital outbreak of the fever among venereal patients and their attendants he did not hesitate to declare that "the milk supply was above suspicion" (14).

Milk came under scrutiny in March 1900 in connection with cases of "milk poisoning". In those days whole families were sometimes attacked with sudden and severe gastro-intestinal disturbance which occasionally proved fatal. Dr. Them. Zammit succeeded in tracing these disorders to goats' milk contaminated with the 'Bacillus enteritidis sporogenes' which found its way into the milk from the unclean state of the animals' udders, of the hands of the milker and of the containers. As a preventive measure he recommended not only the extreme cleanliness of everything that came in contact with the milk but also its boiling before drinking. He expounded these views at a meeting of the

(11) *Ibidem*, meeting of the 28th June, 1894.

(12) HUGHES, M.L., *op. cit.*, p. 39.

(13) HUGHES, M.L., *Mediterranean, Malta or Undulant Fever*, London 1897, p. 52.

(14) HUGHES, M.L., *Note on the Endemic Fever etc.*, p. 33.

Malta Branch of the B.M.A. on the 13th March, 1900, but the opinion was voiced that the boiling of milk destroyed a good deal of its nourishing properties; and Dr. Zammit did not insist on the boiling of the milk — a measure that would have killed not only the 'Bacillus enteritidis sporogenes' but also the germ of Mediterranean Fever. Thus the goat and its milk had fallen under the eyes of the expert but had eluded him as the vector of Bruce's micro-organism. The goat continued to be ignored even when in 1901, the Government Veterinary Surgeon Mr. A. M. Macfarlane, suggested that this animal was the reservoir of Mediterranean Fever (15). It took another four years before the goat came again within the ambit of observation of Dr. Them. Zammit. The occurrence was purely accidental but this time he did not miss it. It happened this way.

By 1904 the incidence of Mediterranean Fever among the garrison of Malta had increased to such a marked extent that at the request of the Admiralty, the War Office and the Colonial Office, the Royal Society sent a Commission to Malta for a detailed investigation of the disease. The Mediterranean Fever Commission, of which Bruce and Zammit formed part, started its work in the same year. Experiments were carried out to ascertain whether the disease was spread by contact, by the inhalation of dust and by mosquitoes or other blood-sucking insects but no evidence was found that such means of propagation played any prominent role (16). Meanwhile the need for laboratory animals, other than monkeys and guinea pigs, induced the Commission to carry out experiments on the goat although this animal seemed "an unlikely subject for Malta fever" to the Commission who had "little hope that an examination of these animals would yield anything" (17).

A small herd of goats was obtained but, before inoculation,

(15) Royal Commission of 1912. Minutes of Evidence. Cd. 6280 p. 197.

(16) *Reports of the Commission Appointed for the Investigation of Mediterranean Fever*, London, 1905-07, 7 vols.

(17) BRUCE, D., *Recent Researches into the Epidemiology of Malta Fever*. A paper read before the Epidemiological Society, February 15th, 1907.

their blood was examined for agglutination with the micrococcus. These tests were carried out by Dr. Them. Zammit on the 14th June, 1905. Much to the surprise of the Commission, several of them were found to react naturally to the agglutination test. On June the 18th Dr. Them. Zammit distributed blood from one of the goats in six broth-tubes and on the 25th June made passages from these tubes on to agar slopes and he recovered the micrococcus in pure culture. It was subsequently found in the urine and milk.

Thousands of goats in Malta were then examined and "the epoch-making discovery was made that 50 per cent, of the goats in Malta responded to the agglutination test and that actually 10 per cent of them were secreting the micrococci in their milk" (18).

Two months later another accidental observation was made, this time associated with the drinking of goats' milk by man. This was the case of the "S.S. Joshua Nicholson" which called at Malta on the 19th August, 1905 and shipped sixty-five goats for export to the United States of America. The milk was drunk freely by the captain and the crew with the result that practically everyone who drank the milk was struck down by Mediterranean Fever. Of the sixty goats that survived the voyage and reached America, thirty-two were found to react to the agglutination test while the micrococcus itself was isolated from the milk of several of them (19).

With the definite establishment of the goat-to-man link in the transmission of Mediterranean Fever, the medical profession turned its efforts to the practical question of prevention. But that is another story one chapter of which came to a close as recently as the 1st March, 1964 with the prohibition of the sale of non-pasteurized milk in the sister island of Gozo.

(18) *Reports of the Commission Appointed for the Investigation of Mediterranean Fever*, London, 1905, Part III, p. 83.

(19) BRUCE, D., *Recent Researches etc.*, p. 8. *Reports of the Commission Appointed for the Investigation etc.*, London, 1907, Part VII, p. 107.

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