Virological investigations in congestive cardiomyopathy

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VIRUSES and other non-bacterial agents (rickettsiae and chlamydiae) can cause acute inflammatory infections in the heart, just as they can in other organs of the body. The pericardium, myocardium and endocardium can be attacked by viruses to varying degrees and in varying ways. The heart can be affected even before birth by rubella virus, with resulting congenital defects, and a direct infection of the heart – membranes or muscle, or both – can be caused by several viruses in childhood, adolescence and adult life (Table 1). Disease of the valves is associated particularly with the rickettsiae and chlamydiae, and involves, usually, a previously damaged valve. Acute myopericarditis is associated frequently, though not exclusively, with enteroviruses, particularly with the Coxsackie B group (Grist and Bell, 1974; Grist, 1977). Typically, the course of acute myopericarditis is a short one, progressing rapidly to recovery or death, although the natural history also includes the phenomenon of recurrence or recurrences, and progression to long-term cardiac disease. Curiously, the clinical end-product seems rarely to be what may be defined as congestive (primary) cardiomyopathy (Goodwin, 1970; Olsen, 1972).

Criteria for aetiological association of viruses with acute myopericarditis

Lerner and Wilson (1973), and Lerner, Wilson and Reyes (1975), have laid down criteria for associating viruses with cardiac disease, dividing them into three orders, or degrees, of validity:

High order

This requires isolation of virus from the heart (Kuhn *et al.*, 1975) or detection of viral antigen in the heart by specific immunofluorescence. In fact, there have been, so far, only seventeen isolations of enteroviruses from the heart (Table 2) and specific immunofluorescence can be done only if there is a post-mortem or biopsy. Post-mortem material may

TABLE 1. Some viruses causing myopericarditis in man

<i>Enteroviruses</i>	Coxsackie B ₁₋₅ Coxsackie A (various) Poliovirus
<i>Arboviruses</i>	Chikungunya Dengue
<i>Rubella</i>	
<i>Others</i>	Encephalomyocarditis (EMC) virus Influenza Measles Marburg virus

TABLE 2. Isolations of enteroviruses from the heart (after Grist, 1977)

Coxsackie	Heart	Pericardial fluid
A4	2	—
A16	1	—
A23 (ECHO 9)	1	—
B2	3	1
B3	2*	2
B4	6	1
B5	2*	—
B (? type)	—	1
Total	17	5

* One cardiac biopsy in each case.

be unavailable, and biopsies are rarely performed on patients with acute myopericarditis.

Moderate order

This requires isolation from the throat or pharynx, with either a four-fold rise in antibody titre or a titre of IgM (i.e. supposedly recently produced antibody) of 1/32 or more.

Low order

This entails isolation of virus from throat or faeces, or a four-fold rise in antibody titre, or an IgM titre to a single strain of 1/32 or more.

In fact, as has been stated, only a handful of isolations direct from the heart have been recorded (Table 2), and little is known of IgM reactions to

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Coxsackie viruses. Grist (1977) has pointed out realistically that the causal role at least of Coxsackie B viruses in acute myopericarditis is well attested. It is, therefore, of interest that a high order of association is attributed by Lerner and Wilson to Coxsackie B viruses 1-5.

The subsequent course after acute infection

Relatively few cases of acute myopericarditis have been followed-up for any length of time, and by no means all of these have been virologically proved. It is generally safe to conclude that many of them must have been caused by enteroviruses, of which the most likely would be a member of the Coxsackie B group. Most of the series are small. However, at least the fact has emerged that in a minority of cases recovery is incomplete and, even after apparent clinical recovery, there may be a recurrence of the illness, sometimes more than once. Some cases may proceed to continuing cardiac disease and death, although, with very few exceptions, it would appear that there is little in the published literature to suggest that congestive cardiomyopathy is a sequel to virologically proved acute myopericarditis, at least in the overt clinical form of that infection. However, this does not necessarily mean that Coxsackie B viruses are not involved. Indeed, a mild or even clinically inapparent infection (and there must be many of these) could well be expected to be the sort of pathological event to initiate the path to a chronic and progressive disorder. It could be argued that such an infection is at least as likely to do this as an overt brisk clinical attack, perhaps more so.

Retrospective investigation of patients with primary congestive cardiomyopathy

Some patients with congestive cardiomyopathy give a history of a febrile illness less than a year before the onset of symptoms, but it is rare for this to have been investigated, and there is a need in these patients for retrospective investigation, in so far as this is possible, with particular regard to virus infections. The object of such an investigation is to seek for evidence of past infection with Coxsackie B or other viruses. The obvious way to do this is by the detection of antibodies in the serum, even though it is not necessarily these which are the cause of the pathological process, and even though the significance of the presence of antibodies must be assessed with caution. Of course, antibodies to many viruses are widely distributed in the population, and it is necessary to compare cases of congestive cardiomyopathy as to the frequency of antibody to viruses at various titres with age- and sex-matched controls.

Such an investigation has been conducted on fifty cardiologically verified cases of primary congestive cardiomyopathy, with appropriate controls

(Cambridge *et al.*, 1978). These were patients with ostensibly normal coronary arteries, and included fifteen who had had coronary angiograms. The ventricle in all patients manifested the diffuse hypokinesia characteristic of primary cardiomyopathy. It has shown the following results:

(1) Antibodies, tested by the micro-neutralization test in VERO cells, were found to all types of Coxsackie B except B₆ among the patients (Table 3).

TABLE 3. Titres (reciprocal dilutions) of antibody to Coxsackie B viruses estimated by neutralization of cytopathic effect of virus in cultures of VERO cells

Dilution ≤ :	Controls (age- and sex-matched)	Cardiomyopathy patients
64	24	20
128	12	6
256	6	5
512	7	4
1024 or greater	1	15

(2) The incidence of the presence of antibodies was not significantly different between cases and controls at levels of 1 : 128, 1 : 256 or 1 : 512.

(3) Of all 100 patients studied, i.e. fifty cases and fifty controls, sixteen had titres of 1 : 1024 to one or other Coxsackie B virus. Of these sixteen, fifteen were cases and one was a control. This contrasted sharply with the situation at titres 1 : 512 or lower, where there was not a statistical difference between cases and controls.

(4) The cases were divided into two groups, as to whether they were examined (a) within one year of onset of clinical symptoms, or (b) more than one year after onset (Table 4). In group (a) there was a higher percentage of patients with titre 1 : 1024 than in (b), and it was in this group (i.e. group (a)) that the control/patient ratio is significant.

TABLE 4. Length of clinical history of cardiomyopathy patients in relation to Coxsackie B virus neutralization titres

Dilution ≤ :	1 year or less	More than 1 year
128	9	17
256	2	3
512	2	2
1024 or greater	11	4

(5) In patients with a history of less than 6 months, there was an association of titre of 1 : 1024 with a history of febrile illness and, in fact, in one patient titres falling from 1 : 1024 were found, and this patient had had a well attested pericarditis two months before. Biopsy showed no evidence of persisting myocarditis.

It would seem, therefore, that there is at least a *prima facie* case for investigating further the question of immune status to Coxsackie B viruses in patients with primary cardiomyopathy on the premise that at least some cases of primary congestive cardiomyopathy are a late result of infection with one or other of the Coxsackie viruses B₁₋₅. This is because the antibody titres suggest that cases, if examined soon enough after onset, show evidence of infection in the recent past, leading possibly to the cardiac disease. Such a sequence of events has been shown experimentally in the mouse (Wong, Woodruff and Woodruff, 1977), and the mechanism appears to be the production and activity of cytotoxic lymphocytes. Abelman *et al.* (1975) have shown depressed myocardial function in the hamster after Coxsackie B₃ virus myocarditis.

Two future lines of research suggest themselves. First, an assessment of IgM (i.e. recently produced antibody) and second, an examination of cellular immunity in these patients, and the activity of their lymphocytes on muscle cells *in vitro*.

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References

- ABELMANN, W.H., ADESANYA, C.O., GOLDBERG, A.H., PHEAR, W.P. & YOUNG, N.A. (1975) Depressed myocardial function in subacute experimental viral myocarditis. *Recent Advances in Studies on Cardiac Structure and Metabolism*, 6, 535.
- CAMBRIDGE, G., MACARTHUR, C.G.C., WATERSON, A.P., GOODWIN, J.F. & OAKLEY, C.M. (1978) Antibodies to Coxsackie B viruses in congestive cardiomyopathy. (In preparation.)
- GOODWIN, J.F. (1970) Congestive and hypertrophic cardiomyopathy: a decade of study. *Lancet*, i, 731.
- GRIST, N.R. (1977) Coxsackie virus infections of the heart. In: *Recent Advances in Clinical Virology* (Ed. by Waterson, A.P.) vol. 1, p. 141. Churchill Livingstone, Edinburgh.
- GRIST, N.R. & BELL, E.J. (1974) A six-year study of Coxsackie virus B infections in heart disease. *Journal of Hygiene, Cambridge*, 73, 165.
- KUHN, H., BREITHARDT, G., KNIERIEM, H.-J., LOOGEN, F., BOTH, A., SCHMIDT, W.A.K., STROOBANDT, R. & GLEICHMANN, U. (1975) Die Bedeutung der endomyokardialen Katheterbiopsie für die Diagnostik und die Beurteilung der Prognose der kongestiven Kardiomyopathie. *Deutsche medizinische Wochenschrift*, 100, 717.
- LERNER, A.M. & WILSON, F.M. (1973) Virus myocarditis. *Progress in Medical Virology*, 15, 63.
- LERNER, A.M., WILSON, F.M. & REYES, M.P. (1975) Enteroviruses and the heart (with special emphasis on the probable role of Coxsackie viruses, group B, types 1-5). *Modern Concepts of Cardiovascular Disease*, 44, 7.
- OLSEN, E.G.J. (1972) Pathology of primary cardiomyopathies. *Postgraduate Medical Journal*, 48, 732.
- WONG, C.Y., WOODRUFF, J.J. & WOODRUFF, J.F. (1977) Generation of cytotoxic T-lymphocytes during Coxsackie B₃ infection. II. Characterization of effector cells and demonstration of cytotoxicity against viral-infected myofibers. *Journal of Immunology*, 118, 1165.