rest was instituted and the temperature resolved in 24 hours, while all signs and symptoms disappeared in five days. Subsequent investigations showed no evidence of streptococcal infection (a throat swab grew no pathogens and the antistreptolysin titre was < 125 units/ml both one day after admission and six weeks later). The antibody titre for psittacosis LGV was > 1/1000 on admission and six weeks later was still raised at 1/128. Inquiry later showed that before his illness he had visited regularly a neighbouring house in which a healthy, psittacine bird was kept.

Discussion

In this case there are two major and three minor criteria of rheumatic fever present, but without evidence of a recent streptococcal infection. On the contrary, the presence of a cough and constitutional disturbance associated with patchy shadowing on the chest *x*-ray film and a significant and changing antibody titre all support a diagnosis of psittacosis. The rashes described in association with this disease include "rose spots,"[2] erythema nodosum,[2] and a macular scaly rash.[3] We could find no reports of an exanthem resembling erythema marginatum. Arthritis, which may be migratory,[4] is a rare complication of psittacosis in man, though in sheep and cattle polyarthritis it is well recognised.[5]

This case shows that psittacosis may present with minimal respiratory disturbance and with a clinical picture resembling rheumatic fever with erythema marginatum and polyarthritis.

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Identification of Clostridium difficile as a cause of pseudomembranous colitis

The demonstration of a toxin[1] in the stool of patients with pseudomembranous colitis and neutralisation of the toxin by *Clostridium sordellii* antitoxin[2] has provided a valuable tool in the search for the cause of this disease. These findings also implicate *Cl sordellii*, or an organism producing an antigenically similar toxin, as the causal agent. We tested all clostridial species isolated from patients with pseudomembranous colitis for toxin production and neutralisation by *Cl sordellii* antitoxin.

Patients, methods, and results

Eight patients with pseudomembranous colitis[3] were studied together with 20 patients with postoperative diarrhoea without evidence of colitis, all of whom had undergone major gastrointestinal surgery within 10 days and had three or more bowel actions each day. Clostridia were isolated on lysed blood agar containing kanamycin sulphate (70 mg/l) or nalidixic acid (10 mg/l). Broth cultures of clostridia and faeces from patients were tested for toxin on monolayers of HeLa cells. Toxin was neutralised by incubating samples with antitoxin to *Cl perfringens* types A, B, C, D, E; *Cl oedematiens* type A (Wellcome Reagents Ltd); and *Cl sordellii* (Wellcome Research Laboratories) for one hour at room temperature.

In all patients with pseudomembranous colitis high titres of toxin were detected in faeces (see table). The toxic activity caused a diffuse rounding of the HeLa cells, which became refractile and separated from the glass. In seven patients with postoperative diarrhoea a second type of cytopathic effect was observed: irregular patchy areas of separation appeared, leaving islands of deformed cells. This effect was present only at low levels after 48 hours' incubation, in contrast to the toxin associated with pseudomembranous colitis, which was evident after 18 hours. The toxic effect of faeces from all patients with colitis was neutralised at 18 hours by *Cl sordellii* antitoxin but not by the other antisera. After further incubation up to 48 hours the second patchy type of cytopathic effect developed in several cases (see table). None of the antisera neutralised the patchy type of cytopathic effect.

Toxic cytopathic effects of faecal fluid from patients with pseudomembranous colitis

Case No	Highest toxin titre	Neutralisation by *Cl sordellii* antitoxin	2nd cytopathic effect at 48 h
1	$>5\times10^2$	+	+
2	4×10^3	+	−
3	4×10^5	+	+
4	2×10^5	+	+
5	1×10^5	+	−
6	8×10^2	+	+
7	8×10^2	+	+
8	3×10^4	+	+

We isolated *Cl perfringens*, *Cl innocuum*, *Cl paraputrificum*, *Cl tertium*, *Cl sporogenes*, and *Cl sphenoides* from faeces of patients with pseudomembranous colitis. *Cl difficile* was isolated from all patients with colitis and from six with postoperative diarrhoea. A cytopathic effect identical with that seen with faeces was produced by cultures of two strains of *Cl difficile* isolated from patients with colitis but not by other clostridia. This toxin produced by *Cl difficile* was neutralised by *Cl sordellii* antitoxin. As with the faeces, incubation for 48 hours showed the second cytopathic effect, which was also produced by cultures of three other strains of *Cl difficile*. Seven strains of *Cl difficile* were sensitive to metronidazole, sulphonamide, and vancomycin and resistant to penicillins, cephalosporins, aminoglycosides, lincomycins, tetracycline, and erythromycin.

Comment

The demonstration that the cytopathic effect of *Cl difficile* is indistinguishable from that produced by faeces and is neutralised by *Cl sordellii* antitoxin suggests that *Cl difficile* is the causative agent of pseudomembranous colitis. The production of toxin by *Cl difficile* is not a new observation[4]: on subcutaneous injection into guinea-pigs it causes oedema, respiratory arrest, and death and is inactivated by heat.[5] These properties resemble the toxic effects of faecal extract from patients with pseudomembranous colitis. The finding of a second, milder cytopathic effect produced by faeces from some patients with postoperative diarrhoea and by three strains of *Cl difficile* indicate the possibility of a second pathogenic role for this organism in postoperative diarrhoea.

Little is known about the distribution of *Cl difficile* in populations, but Hall and O'Toole[4] found it in the faeces of 40% of infants. Careful search will probably disclose small numbers of *Cl difficile* in the intestinal tract of healthy adults. Under appropriate conditions these may multiply and cause postoperative diarrhoea or pseudomembranous colitis according to their potential for toxin production.

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