Leading article

Crohn's disease: the genetic contribution

It is clear that the liability to develop Crohn's disease is influenced by a wide range of genetic and environmental factors. Few of these have been specifically identified and it is not known whether any single factor is essential for the development of Crohn's disease.

Role of autoimmunity

Crohn's disease patients and their relatives are more likely to suffer from disorders with a known or suspected autoimmune origin than members of the general population. These disorders include ankylosing spondylitis,[1 2] Behçet's disease,[3] psoriasis,[4] iritis, thrombocytopenic purpura and systemic lupus erythematosus,[5] and multiple sclerosis.[6] Aberrant HLA-DR expression in the intestinal mucosa of Crohn's disease patients[7] is consistent with an autoimmune aetiology.

Serum from Crohn's disease probands and their healthy first degree relatives may contain antibodies to gut epithelial cells,[8] and peripheral blood lymphocytes from Crohn's disease patients may be cytotoxic to these cells.[9] The sharing of antigenic determinants by germ free rat colon and a lipopolysaccharide extract of *Escherichia coli* suggests that the anticolon antibodies and associated cell mediated cytotoxicity may result from stimulation of the gut associated lymphoid tissue by cross reacting enterobacterial antigens.[5] This hypothesis is unlikely to provide a mechanism for substantial inherited variation in liability, however, since, unlike ankylosing spondylitis alone where HLA-B27 positive subjects run a risk of disease approaching 100 times that of HLA-B27 negative subjects and where cross reaction and amino acid sequence homology between the host antigen and *Klebsiella pneumoniae* have been shown,[10] the anticolon antibodies of Crohn's disease are effective against allogeneic colonic epithelial cells.[8]

Lymphocytotoxic antibodies have also been shown in Crohn's disease probands and their healthy relatives but, as these antibodies react with lymphocytes from normal subjects with a wide range of HLA specificities,[11] they are similarly unlikely to account for inherited differences of liability.

Chromosomal and monogenic disorders

Crohn's disease occurs at an unusually high frequency among patients with Turner's syndrome,[12] known for its association with autoimmunity. A disorder like Crohn's disease has also been reported in the autosomal recessive Hermansky-Pudlak syndrome,[13] a form of albinism with haemorrhagic diathesis and pigmented reticuloendothelial cells, and glycogen storage disease type Ib,[14] in which there is neutropenia and abnormal neutrophil function. Comparable bowel involvement has been described in patients with other inherited neutrophil defects,[15,16] while Crohn's disease activity has been negatively correlated with neurophil superoxide production among patients in whom the disease has no obvious monogenic basis.[17] Neutrophil function in Hermansky-Pudlak syndrome seems normal.[18] It has been suggested that susceptibility to autoimmune disease in general, including Crohn's disease, is under the control of a single dominant gene, the specific clinical effects of which are determined by other genes, hormonal influences, and environment triggers.[19]

Complex segregation analysis has indicated that about 30% of all Crohn's disease patients could be homozygous for a major recessive gene with incomplete penetrance, with no residual causes of family resemblance and a relatively high frequency of homozygosity among younger onset cases.[20] Nevertheless, the differences in fit from what would be expected of multifactorial inheritance are small[21] and the pattern of familial aggregation indicative of a recessive influence (with affected sibling pairs predominating over parent-offspring pairs) could have occurred for other reasons, as discussed below.

Twins and resemblance between other relatives

The results of most family studies of Crohn's disease have been expressed in terms of the proportion of probands with either affected first degree relatives or a positive family history over all relatives investigated. These data are of limited value in making a genetic analysis. It has been argued that a better measure of familial aggregation is the ratio of prevalence among relatives to that in the general population, but this ratio alone is also of limited value since, for a given heritability, it varies inversely with population prevalence. Rational genetic interpretation of familial aggregation requires both the absolute value of the population prevalence and the numbers of affected and unaffected relatives of probands.

Several reports have quoted numbers of affected and unaffected relatives of Crohn's disease patients, including twins. The studies differ in respect of diagnostic criteria, the age structure and method of ascertainment of the sample of probands, and the number of years between diagnosis in probands and ascertainment of affected relatives. Strict comparability is not therefore possible but pooling of results (Table) provides a summary of the overall position. Population prevalences, where given, ranged from 13 to 74 per 100 000 but were generally in the region of 50 per 100 000.

Prevalence of Crohn's disease (%) among co-twins and among first, second, and third degree relatives of Crohn's disease probands, with heritability estimates for population prevalences of 25, 50, and 100 per 100 000. (a) Each twin pair ascertained through only one affected individual; (b) all affected twins ascertained independently

	Total	*Prevalence (%)*	*Heritability (%) (mean (SEM)) for population prevalences of* $25/10^5$	$50/10^5$	$100/10^5$
Twin pairs (22–24) (a) MZ	26	50·00	88 (27)	96 (27)	104 (28)
DZ	31	3·23			
(b) MZ	26	66·67	76 (30)	82 (31)	90 (34)
DZ	31	6·25			
Parents (20, 22, 27–30, 33)	2 323	1·03	63 (4)	55 (4)	46 (5)
Sibs (20, 22, 27–30, 33)	3 281	1·86	75 (3)	68 (3)	60 (3)
Children (22, 27, 28, 30, 33)	1 060	0·38	43 (9)	35 (9)	25 (10)
First degree relatives (20, 22, 24–33)	14 579	1·42	69 (2)	62 (2)	53 (2)
Second degree relatives (30, 32, 33)	4 294	0·16	58 (13)	39 (13)	18 (14)
Third degree relatives (30, 33)	2 457	0·04*	–	–	–

*One case only.

The significantly higher prevalence in siblings than among either parents ($\chi_1^2=5{\cdot}5$, $p=0{\cdot}019$) or children ($\chi_1^2=10{\cdot}7$, $p=0{\cdot}001$) could be interpreted as evidence for an environmental or recessive genetic contribution to Crohn's disease. It seems likely, however, that the low prevalence among children is largely a consequence of inability to ascertain pre-onset cases, while among parents it could stem from the effect of Crohn's disease on marital relationships[34] with reduced fertility, either through complications of the disease in women or reversible azoospermia during sulphasalazine treatment in men.[35] The results of a collaborative European study indicate that affected women do indeed have fewer children than normal control subjects.[36]

The classical multifactorial model[37] is useful for considering these observations, even though its requirements may not be fully met. The model assumes that many genes and environmental influences contribute to an individual's position on an underlying continuous scale of liability but that disease is present only in those who fall above a critical threshold value. The model allows estimation of heritability, the proportion of the population variance in liability attributable to additive genetic variation, from proband concordance rates in twins[38] and from the prevalence of disease among other relatives of probands.[37]

Heritability estimates for Crohn's disease are given alongside the prevalence data in the Table. The estimates from twins are very high, but since the standard errors are large they make a limited contribution to the overall picture. Reduced fitness and inability to ascertain pre-onset cases are likely to have resulted in serious underestimates of heritability in parents and children. On the other hand, the estimates from siblings may be inflated because of within generation environmental effects or the influence of recessive genes. The low estimates for second degree relatives, admittedly with large standard errors, also suggest that common environment or recessive genes, or both, may have contributed to the relatively high prevalence among siblings. It may be, however, that low prevalence among more distant relatives results in part from lower levels of ascertainment. Among third degree relatives, the single affected individual gave a prevalence similar to that in the general population. Taking into account all the data for the prevalence of Crohn's disease among relatives of probands, and assuming a population prevalence of around 50 per 100 000, a heritability in the region of 50–60% seems likely. In other words, perhaps a little more than half of the variation in liability to develop Crohn's disease is a consequence of inherited differences between individuals.

Two other features of the family studies are noteworthy. Firstly, Crohn's disease is rarely found among spouses of Crohn's disease probands,[39] suggesting that family environment after marriage does not contribute substantially to the disease. Secondly, familial cases have sometimes shown a lower age at onset than sporadic cases,[20 24] which is consistent with aetiological heterogeneity.

Association of Crohn's disease with genetic markers

There is little evidence of any association with HLA-A or -B antigens in individual studies, although pooling of results from a number of centres showed a significant positive association with HLA-A2 (relative risk 1·25) and a significant negative association with HLA-A11 (relative risk 0·62).[40] Various associations with HLA-DR specificities have been reported but no consistent pattern has emerged, except for the positive association with DR4 in three Japanese studies.[41–43] These findings support the contention that host antigenic variation, through an autoimmune aetiology, is unlikely to make a substantial contribution to differences in liability to develop Crohn's disease. Nevertheless, different HLA types have been associated with granuloma positive and granuloma negative Crohn's disease[44] and with other differences in manifestation of the disorder in Japan[41] and in Italy,[45] while the F and FS phenotypes of complement component C3 have been over-represented in Crohn's disease patients whose small bowel is affected.[46] Thus, there is some suggestion of variation in the immune response within Crohn's disease. Within families, evidence for an HLA haplotype association is not convincing[47] and therefore provides little indication of linkage of an important Crohn's disease susceptibility gene with the major histocompatibility complex at 6p21·3.

Studies of association with several blood group and serum protein markers have also proved inconclusive,[48 49] while no association has been found at the population level with DNA restriction fragment length polymorphisms (RFLPs) at the T cell antigen receptor alpha and beta loci.[50] A high negative correlation between the prevalence of lactose intolerance and the incidence of Crohn's disease among different populations in several countries led to the hypothesis that lactose malabsorption results in the formation of volatile fatty acids that may inhibit multiplication of potentially pathogenic intestinal orgamisms.[51] In Jewish subpopulations from different parts of the world, however, the incidence of Crohn's disease varies while the prevalence of lactose intolerance is remarkably constant.[52] Further attempts to relate the incidence of Crohn's disease to known genetic variation between populations are unlikely to be productive since differences of incidence between ethnic groups are narrowing with time and migration.[53]

Subclinical findings

Three sets of findings have highlighted possible areas where subclinical investigation of Crohn's disease patients and their relatives may be fruitful. Two of these, serum anticolon antibodies and abnormal neutrophil function, have already been considered. The third is evidence to suggest that intestinal permeability to polyethylene glycol is increased in patients with Crohn's disease and their clinically normal relatives.[54] Increased intestinal permeability to other substances has also been reported in Crohn's disease patients but not in their unaffected relatives.[55 56] raising the possibility that at least part of the permeability defect is not an inherited abnormality but secondary to the disease process.

Future possibilities

The most pressing need in investigating the genetic contribution to Crohn's disease is the collection of multiply affected families for RFLP linkage studies, to identify individual loci that contribute to liability. The chromosomal locations of markers that have shown no association at the population level should not be ignored since the lack of this association does not rule out linkage. There are, however, at least two complicating factors. First is the possibility of inheriting a major gene for liability, but not developing the disease. A prerequisite for linkage studies would thus be a thorough search for subclinical manifestations of inherited liability. Second is the extent of genetic heterogeneity within Crohn's disease and of the genetic correlation between Crohn's disease and ulcerative colitis. The loci controlling the mucosal immune system are likely candidates for investigation, in particular, those that may have the ability to suppress the generation of an effective immune response to the multiple antigens normally present in the intestinal lumen.[57] Animal models could also be useful. Transgenic rats that express human HLA-B27 and Beta-2-microglobulin have developed spontaneous inflammatory disease in the gastrointestinal tract.[58] Genetic dissection of the immune

system in this way, or through the use of mutants with different specific defects of immune function, could prove valuable in studying the influence of individual genes in the pathogenesis of Crohn's disease. Finally, detailed studies of the chromosomal and monogenic disorders accompanied by Crohn's disease like lesions should also provide insight into the inherited basis for this disease.

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1 Hickling P, Bird Stewart JA, Young JD, Wright V. Crohn's spondylitis: a family study. *Ann Rheum Dis* 1983; **42**: 106–7.
2 Mielants H, Veys EM, Joos R, Suykens S, Cuvelier C, DeVos M. Familial aggregation in seronegative spondyloarthritis of enterogenic origin. A family study. *J Rheumatol* 1986; **13**: 126–8.
3 Zoppini A, Ciccarini V, Carotenuto P. Associazione a carattere familiare del morbo di Crohn della spondilite anchilosante e della malattia de Behcet. *Ann Reumatol* 1977; **10**: 20–30.
4 Lee FI, Bellary SV, Francis C. Increased occurrence of psoriasis in patients with Crohn's disease and their relatives. *Am J Gastroenterol* 1990; **85**: 962–3.
5 Kirsner JB, Shorter RG. Recent developments in nonspecific inflammatory bowel disease (part 2). *N Engl J Med* 1982; **306**: 837–48.
6 Minuk GY, Lewkonia RM. Possible familial association of multiple sclerosis and inflammatory bowel disease. *New Engl J Med* 1986; **314**: 586.
7 Selby WS, Janossy G, Mason DY, Jewell DP. Expression of HLA-DR antigens by colonic epithelium in inflammatory bowel disease. *Clin Exp Immunol* 1983; **53**: 614–8.
8 Fiocchi C, Roche JK, Michener WM. High prevalence of antibodies to intestinal epithelial antigens in patients with inflammatory bowel disease and their relatives. *Ann Intern Med* 1989; **110**: 786–94.
9 Kemler BJ, Alpert E. Inflammatory bowel disease: study of cell mediated cytotoxicity for isolated human colonic epithelial cells. *Gut* 1980; **21**: 353–9.
10 Schwimmbeck PL, Oldstone MBA. Klebsiella pneumoniae and HLA B27-associated diseases of Reiter's syndrome and anklyosing spondylitis. *Curr Top Microbiol Immunol* 1989; **145**: 45–56.
11 Korsmeyer SJ, Williams RC, Wilson ID, Strickland RG. Lymphocytotoxic antibody in inflammatory bowel disease. A family study. *N Engl J Med* 1975; **293**: 1117–20.
12 Price WH. A high incidence of chronic inflammatory bowel disease in patients with Turner's syndrome. *J Med Genet* 1979; **16**: 263–6.
13 Schinella RA, Greco MA, Cobert BL, Denmark LW, Cox RP. Hermansky-Pudlak syndrome with granulomatous colitis. *Ann Intern Med* 1980; **92**: 20–3.
14 Couper R, Kapelushnik J, Griffiths AM. Neutrophil dysfunction in glycogen storage disease Ib: association with Crohn's-like colitis. *Gastroenterology* 1991; **100**: 549–54.
15 Ament ME, Ochs HD. Gastrointestinal manifestations of chronic granulomatous disease. *N Engl J Med* 1973; **288**: 382–7.
16 Vannier JP, Arnaud-Battandier F, Ricour C, Schmitz J, Buriot D, Griscelli C, *et al.* Neutropénie primitive et congénitale et maladie de Crohn. *Arch Fr Pediatr* 1982; **39**: 367–70.
17 Verspaget HW, Elmgreen J, Weterman IT, Peña AS, Riis P, Lamers CBHW. Impaired activation of the neutrophil oxidative metabolism in chronic inflammatory bowel disease. *Scand J Gastroenterol* 1986; **21**: 1124–30.
18 Shanahan F, Randolph L, King R, Oseas R, Brogan M, Witkop C, *et al.* Hermansky-Pudlak syndrome: an immunologic assessment of 15 cases. *Am J Med* 1988; **85**: 823–8.
19 Bias WB, Reveille JD, Beaty TH, Meyers DA, Arnett FC. Evidence that autoimmunity in man is a Mendelian dominant trait. *Amer J Hum Genet* 1986; **39**: 584–602.
20 Küster W, Pascoe L, Purrmann J, Funk S, Majewski F. The genetics of Crohn disease: complex segregation analysis of a family study with 265 patients with Crohn disease and 5387 relatives. *Am J Med Genet* 1989; **32**: 105–8.
21 Purrmann J, Cleveland S, Hengels KJ. Untersuchungen zu verschieden Verebungsmodellen beim Morbus Crohn. *Z Gastroenterol* 1991; **29**: 276–82.
22 Weterman IT, Peña AS. Familial incidence of Crohn's disease in the Netherlands and review of the literature. *Gastroenterology* 1984; **86**: 449–52.
23 Tsyk C, Lindberg E, Jarnerot G, Floderus-Myrhed B. Ulcerative colitis and Crohn's disease in an unselected population of monozygotic and dizygotic twins. A study of heritability and the influence of smoking. *Gut* 1988; **29**: 990–6.
24 Monsen U, Bernell O, Johansson C, Hellers G. Prevalence of inflammatory bowel disease among relatives of patients with Crohn's disease. *Scand J Gastroenterol* 1991; **26**: 302–6.
25 Hammer B, Ashurst P, Naish J. Diseases associated with ulcerative colitis and Crohn's disease. *Gut* 1968; **9**: 17–21.
26 Lennard-Jones JE. Medical aspects of Crohn's disease. *Proc R Soc Med* 1968; **61**: 81–3.
27 Mayberry JF, Rhodes J, Newcombe RG. Familial prevalence of inflammatory bowel disease in relatives of patients with Crohn's disease. *BMJ* 1980; **280**: 84.
28 Weterman IT, Peña AS. Prevalence of Crohn's disease in first degree relatives of patients with Crohn's disease. *Frontiers of Gastrointestinal Research* 1986; **11**: 27–34.
29 Fielding JF. The relative risk of inflammatory bowel disease among parents and siblings of Crohn's disease patients. *J Clin Gastroenterol* 1986; **8**: 655–7.
30 Lashner BA, Evans AA, Kirsner JB, Hanauer SB. Prevalence and incidence of inflammatory bowel disease in family members. *Gastroenterology* 1986; **91**: 1396–1400.
31 McConnell RB, Shaw JM, Whibley EJ, McConnell TH. Inflammatory bowel disease: a review of previous genetic studies and the Liverpool family data. *Frontiers of Gastrointestinal Research* 1986; **11**: 1–11.
32 Orholm M, Munkholm P, Langholz E, Nielsen OH, Sorensen TIA, Binder V. Familial occurrence of inflammatory bowel disease. *N Engl J Med* 1991; **324**: 84–8.
33 Zlotgora J, Zimmerman J, Rachmilewitz D. Prevalence of inflammatory bowel disease in family members of Jewish Crohn's disease patients in Israel. *Dig Dis Sci* 1991; **36**: 471–5.
34 Gazzard BG, Price HL, Libby GW, Dawson AM. The social toll of Crohn's disease. *BMJ* 1978; **2**: 1117–9.
35 Korelitz BI. Pregnancy, fertility, and inflammatory bowel disease. *Am J Gastroenterol* 1985; **80**: 365–70.
36 Mayberry JF, Weterman IT. European survey of fertility and pregnancy in women with Crohn's disease: a case control study by European collaborative group. *Gut* 1986; **27**: 821–5.
37 Falconer DS. The inheritance of liability to certain diseases, estimated from the incidence among relatives. *Ann Hum Genet* 1965; **29**: 51–76.
38 Smith C. Concordance in twins: methods and interpretation. *Am J Hum Genet* 1974; **26**: 454–66.
39 Murray CJ, Thomson AB. Marital idiopathic inflammatory bowel disease. Crohn's disease in a husband and wife. *J Clin Gastroenterol* 1988; **10**: 95–7.
40 Biemond I, Burnham WR, D'Amaro J, Langman MJS. HLA-A and -B antigens in inflammatory bowel disease. *Gut* 1986; **27**: 934–41.
41 Fujita K, Naito S, Okabe N, Yao T. Immunological studies in Crohn's disease. I. Association with HLA systems in the Japanese. *J Clin Lab Immunol* 1984; **14**: 99–102.
42 Matake H, Naito S, Ogawara S, Iida M, Matsui T, Okabe N, *et al.* HLA study in 108 Japanese patients with Crohn's disease. *Nippon Shokakibyo Gakkai Zasshi* 1989; **86**: 1448–54.
43 Kobayashi K, Atoh M, Yagita A, Konoeda Y, Inoko H, Ando A, *et al.* Crohn's disease in the Japanese is associated with the HLA-DRw53. *Exp Clin Immunogenet* 1990; **7**: 101–8.
44 Ellis A, McKay J, Woodrow JC, McConnell RB. Tissue antigens and inflammatory bowel disease. *Frontiers of Gastrointestinal Research* 1986; **11**: 35–41.
45 Modena V, Amoroso A, Frattasio C, Pera A, Costantini P, Maiocco I, *et al.* HLA antigens and clinical manifestations in Crohn's disease. *Clin Exp Rheumatol* 1988; **6**: 221–5.
46 Elmgreen J, Sorensen H, Berkowicz A. Polymorphism of complement C3 in chronic inflammatory bowel disease. Predominance of the C3F gene in Crohn's disease. *Acta Med Scand* 1984; **215**: 375–8.
47 Colombel JF, Guillemot F, Van Gossum A, Dufossé F, Cortot A, Dupont E, *et al.* Familial Crohn's disease in multiple siblings: no linkage to the HLA system. *Gastroenterol Clin Biol* 1989; **13**: 676–8.
48 van den Berg-Loonen EM, Dekker-Saeys BJ, Meuwissen SGM, Nijenhuis LE, Engelfriet CP. Histocompatibility antigens and other genetic markers in ankylosing spondylitis and inflammatory bowel diseases. *J Immunogenet* 1977; **4**: 167–75.
49 Purrmann J, Bertrams J, Knapp M, Cleveland S, Gemsa R, Hengels KJ, *et al.* Investigation of genetic markers in patients with Crohn's disease and ulcerative colitis. *Z Gastroenterol* 1989; **27**: 366–9.
50 Katakura S, Einarsson K, Hammarström L, Smith CIE. Restriction fragment length polymorphism analysis of T-cell receptor genes in inflammatory bowel disease. *Scand J Gastroenterol* 1989; **24**: 381–4.
51 Nanji AA, Denardi FG. Primary adult lactose intolerance protects against development of inflammatory bowel disease. *Med Hypotheses* 1986; **19**: 1–6.
52 Gilat T, Grossman A, Fireman Z, Rozen P. Inflammatory bowel disease in Jews. *Frontiers of Gastrointestinal Research* 1986; **11**: 135–40.
53 Calkins BM, Mendeloff AI. Epidemiology of inflammatory bowel disease. *Epidemiol Rev* 1986; **8**: 60–91.
54 Hollander D, Vadheim CM, Brettholz E, Petersen GM, Delahunty T, Rotter JI. Increased intestinal permeability in patients with Crohn's disease and their relatives. A possible etiologic factor. *Ann Intern Med* 1986; **105**: 883–5.
55 Ainsworth M, Eriksen J, Rasmussen JW, Schaffalitzky-de-Muckadell OB. Intestinal permeability of 51Cr-labelled ethylene diaminetetraacetic acid in patients with Crohn's disease and their healthy relatives. *Scand J Gastroenterol* 1989; **24**: 993–8.
56 Katz KD, Hollander D, Vadheim CM, McElree C, Delahunty T, Dadufalza VD, *et al.* Intestinal permeability in patients with Crohn's disease and their healthy relatives. *Gastroenterology* 1989; **97**: 927–31.
57 Macdermott RP, Stenson WF. Alterations of the immune system in ulcerative colitis and Crohn's disease. *Adv Immunol* 1988; **42**: 285–328.
58 Hammer RE, Maika SD, Richardson JA, Tang JP, Taurog JD. Spontaneous inflammatory disease in transgenic rats expressing HLA-B27 and human beta 2m: an animal model of HLA-B27-associated human disorders. *Cell* 1990; **63**: 1099–112.