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Research Article

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Hereditary C7 Deficiency

DIAGNOSIS AND HLA STUDIES IN A FRENCH-CANADIAN FAMILY

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ABSTRACT The serum of a 44-yr-old woman of French-Canadian descent having a B-27 positive ankylosing spondylitis was deficient in the seventh component of complement (C7) as determined by hemolytic and immunochemical methods. No inhibitor against C7 was detected, and the levels of all other complement components were normal. No deficiency in the opsonic activity of the serum was found, and the results of basic coagulation studies of the plasma were normal. On investigation of the patient's family, two sisters were found to have the same deficiency but were otherwise in good health. The seven other siblings were heterozygous for C7 deficiency, while the paternal aunt had a normal C7 level. In the third generation, six children of the three homozygous sisters and five children of heterozygotes were available for testing. Studies of the HLA antigens in all the 22 subjects and in three spouses indicated no close linkage between the C7 deficiency and the HLA system. In addition, the simultaneous occurrence of two hereditary complement deficiencies (C2 and C7) was discovered in one family of this remarkable kindred.

INTRODUCTION

Inherited deficiencies of complement components have been described both in healthy individuals and in those having clinical problems (1). Inherited C7 deficiency seems to be a very rare condition, only two cases having been published to date (2, 3). The collection of data from cases of hereditary complement deficiency could establish whether a relationship exists between C7 deficiency and certain diseases and also a possible linkage between this complement component and the HLA system. Therefore we have studied the

complement and HLA systems in a family with a congenital deficiency of C7. This is the first reported case discovered in a woman having ankylosing spondylitis and the first in a family with more than one member homozygous for the deficiency. Furthermore, we report the first HLA studies to be made in pure C7 deficiency.

Case Summary

Proposita (F. R., no. 5 in Fig. 3). The patient, a 44-yr-old woman of French-Canadian descent, was hospitalized in June 1975 for investigation of a progressive painful disease of the skeleton. The patient's childhood and adolescence were uneventful. At age 29 yr the disease started with radiating pain in the back of the thighs, resembling sciatica. Some 5 yr later, the low back pain intensified and was accompanied by morning stiffness. The radiating pains alternated from side to side and became worse upon exposure to cold and humid weather. Nocturnal pains were sometimes relieved by walking around. Over the ensuing years, the low back pain shifted to the hips and the thoracic region. In the past few months, the shoulders and the cervical spine have been involved. Salicylates did not relieve the pain, and the patient has lost 14 lb over the last 2 yr.

On physical examination, the patient appeared tired and emaciated and had a bent-over posture. She had a waddling gait. The skin around the mouth was thinned and wrinkled. There was a loss of the normal lordosis of the lumbar region, with paraspinal muscle spasm and tenderness over the whole spine. The flexion and extension of the spine and hips were limited. No neurological deficiency was present.

Laboratory examinations. Examination of the patient showed the following: hemoglobin 10.0; hematocrit, 33; mean corpuscular volume (MCV), 81; mean corpuscular hemoglobin (MCH), 26.4; white blood cells (WBC), 6,600; polymorphonuclear leukocytes, 52; lymphocytes, 36; monocytes, 8; basophil, 1; eosino-

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phils, 3; sedimentation rate (Westergren), 63; rheumatoid factor, negative; antinuclear factor, 1:10 speckled (not significant); lupus erythematosus (LE) cells, negative; serum iron, 45 $\mu\text{g}/100\text{ ml}$; total binding capacity, 261; saturation index, 17%; uric acid, 3.6 $\text{mg}/100\text{ ml}$; creatinine, 0.7 $\text{mg}/100\text{ ml}$; BUN, 11 $\text{mg}/100\text{ ml}$. Calcium, phosphorus, glucose, serum glutamic oxaloacetic transaminase, cholesterol, triglycerides, and urine analysis were normal. Serum opsonic power and basic coagulation studies [prothrombin time (PT) and activated partial thromboplastin time (APTT)]¹ were normal. Immunoglobulins G and A were normal, IgM slightly elevated. The chest X ray and barium gastrointestinal series were normal. Radiographs of the skeleton showed complete obliteration of the sacroiliac joints, moderate diffuse osteoporosis of the whole spine, and incipient obliteration of the lumbar intervertebral joint spaces. The clinical diagnosis of ankylosing spondylitis (Strumpell-Marie) was made, and there is a possibility that the patient may develop scleroderma.

Medical history of the two homozygous sisters of the patient

L. B. (no. 6 in Fig. 3) is 38 yr old and remained free of diseases throughout her childhood and adolescence. She was treated for hyperthyroidism in her early twenties. 2 years ago she was treated for a scotoma in one eye, which was caused by a cyst. No undue tendency to infection was observed.

L. M. (no. 7 in Fig. 3) is 43 yr old. Childhood and adolescence were uneventful. She had a hysterectomy for metrorrhagia and was operated on for a benign cyst in one breast. Several years ago she had three episodes of otitis media after influenza attacks. For the past few years she has complained of pain in the thoracic spine and of morning stiffness, but X-ray examination was negative.

METHODS

Complement studies

Sera. Human serum samples were separated from freshly clotted blood specimens that had been allowed to stand at room temperature for 1 hr. All sera were then either immediately stored in aliquots at -70°C or, if transported, frozen and kept in dry ice before storing. Guinea pig sera were pooled and also stored at -70°C .

Erythrocytes and cellular intermediates. The methods of Nelson et al. (4) were used to prepare optimally sensitized sheep erythrocytes (EA), as well as the intermediates

¹ *Abbreviations used in this paper:* APTT, activated partial thromboplastin time; CH50, hemolytic whole complement assay; DGVB⁺⁺, dextrose gelatin Veronal buffer; EA, sensitized erythrocytes; GVB, gelatin Veronal buffer; PT, prothrombin time.

EAC1, EAC14, EAC142, EAC1-3, EAC1-6, and EAC1-7. EAC4 (human) was prepared as described by Rapp and Borsoos (5).

Complement reagents and hemolytic assays. Gelatin Veronal buffer was prepared according to Mayer (6) and contained 0.1% gelatin (GVB). When needed, divalent cations to a final concentration of $1.5 \times 10^{-4}\text{ M Ca}^{++}$, $0.5 \times 10^{-3}\text{ M Mg}^{++}$, and a 5% solution of dextrose were added (v/v) to prepare DGVB⁺⁺. A stock solution of 0.1 M EDTA, pH 7.4, was used to prepare GVB-EDTA at the desired concentrations. Functionally pure complement components from C1 to C9 were purchased from Cordis Laboratories, Miami, Fla. Routine CH50 titrations were performed as a 3.75-ml modification of the method of Mayer (6). This method yields mean values $\pm 2\text{ SD}$ of $96 \pm 25\text{ U/ml}$ of serum for a system of 2.5×10^8 EA cells.

A screening test for the estimation of C7 was executed in the following manner: Serial dilutions of the unknown serum in DGVB⁺⁺ (0.4 ml) were reacted with 0.2 ml of the proposita's serum (diluted 1:50), having a CH50 titer of zero U/ml. 0.4 ml of EA cells (1×10^8 cells/ml) and 1.5 ml of DGVB⁺⁺ were added to give a final volume of 2.5 ml. After incubation at 37°C for 60 min, all tubes were centrifuged and lysis determined by reading the optical density at 415 nm. The normal range for C7, as established by the titration of 20 normal donor sera, was 2,000–2,800 U/ml serum.

The functional activity of C7 was also evaluated by the method of Nelson et al. (4), with modifications suggested by Cordis Laboratories. Normal mean values $\pm 2\text{ SD}$ were $32,600 \pm 5,200\text{ U/ml}$ serum as established by titration of 30 individual normal donor sera and two pools of five known control sera each. Functional C7 was expressed as a mean of three determinations.

Complement components C1 to C5 were assayed by the methods of Nelson et al. (4), and for titration of C6, C8, and C9 the techniques of the same authors were applied on microplates. Normal mean values $\pm 2\text{ SD}$ for C1 and C2 were established by this laboratory using 50 normal donor sera, and the normal range for the other complement components, except C7, were established by testing at least 10 known control sera. Two controls were run in each experiment. All values are shown in Table II.

Anticomplement activity was assayed by the method of Dausset (7) and by hemolytic titration of mixtures of different dilutions of the patient's serum with normal serum.

Immunoassay for complement components. C3, Factor B, and C4 were qualified by single radial diffusion on immunoplates (Behring-Werke AG., Marburg/Lahn, West Germany). Ouchterlony's double-diffusion method was used to identify C7 immunochemically and for semiquantitative determinations. Specific antiserum to C7 was obtained by immunizing rabbits with a pool of functionally pure C7 preparations prepared by us according to the technique of Nelson et al. (4) and C7 from Cordis Laboratories. The antiserum was absorbed with the proposita's serum, which had zero C7 hemolytic activity. After centrifugation it was concentrated on an Amicon B15 filter (Amicon Corp., Lexington, Mass.). On immunoelectrophoresis against normal serum (5 μl), this antiserum gave only one band in the beta region. The results of the Ouchterlony analysis were expressed as the reciprocal of the highest dilution of the test serum giving a visible precipitin line and were read by four independent observers. The normal range as established by assays of two pools of 5 donor sera and 10 individual control sera was 3 to 8.

Opsonic activity. This was measured by modification of the method of Stossel (8) where oil red O was replaced by zymosan. Determination of tetrazolium blue reduction was

replaced by measuring H^2O^2 released during the phagocytosis of opsonized zymosan particles.

Basic coagulation and other studies

Activated partial thromboplastin time (APTT), prothrombin time (PT), and determination of immunoglobulins and of antinuclear antibody were performed according to standard methods.

HLA studies

25 members of this family have been typed for HLA antigens in order to evaluate the relationship between C7 deficiency and the major histocompatibility complex. Nine HLA specificities of the "A" series and 14 of the "B" series were investigated: HLA-A1,2,3,9,10,11,28,29,W30 and HLA-B5,7,8,12,13,14,18,27,W15,W16,W17,W22,W35,W40. Each antigen was tested by several mono- and multispecific sera, with the exception of A28 and B18, which were tested only with one serum. Antigens AW30 and BW22 were tested with duospecific sera. The typing was done by the lymphocyte microcytotoxic technique using antisera kindly given by the National Institutes of Health, Bethesda, Md. Analysis of the odds on linkage presented in the format of lod scores has been kindly performed by Dr. Rand Huntzinger, Division of Genetics, University of Rochester School of Medicine, Rochester, N. Y.

RESULTS

Establishment of C7 deficiency in the patient. Routine complement studies are performed as part of the investigation for all patients with arthritic diseases. The CH50 titer of the patient's serum was 0 U/ml, while values for C3 and C4 by immunoprecipitation were normal. Hemolytic titration of C1 and C2 also gave normal results. No anticomplement activity was found in the assay of Dausset (7), and incubation of normal serum with varied dilutions of the patient's serum did not diminish the CH50 titer of the normal serum, but increased it instead. The hemolytic activity of the patient's serum was restored only by adding C7.

TABLE I
Effect of Purified Complement Components on the Hemolytic Activity of the Patient's Serum

Component added individually*	Cellular intermediates (1×10^6 /ml)		
	EA	EAC14	EAC1
	% lysis after 60 min at 37°		
C1, C2, C3, C4, C5, C6, C8, or C9 100 U/ml	0	0	0
C7 100 U/ml	43	50	80
200 U/ml	98	98	96

* Serum dilution: 1:50 for EA cells; 1:25 for EAC14 and EAC1.

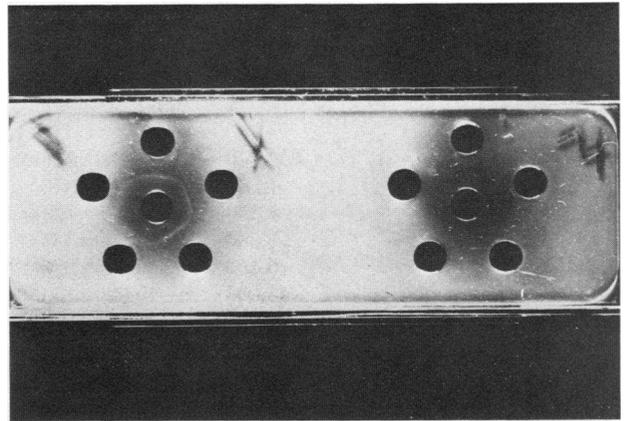


FIGURE 1 Ouchterlony pattern of the C7-deficient serum (right side) and of normal human serum (left side). The central wells contain the monospecific anti-C7 serum.

The patient's serum had no lytic effect on EAC1, EAC14, or EAC142, indicating that the deficiency was not at these stages. These intermediates were lysed when C7 was added to the patient's serum and there was a dose-dependent reactivity (Table I). The C7 titer of the proposita, when measured by Nelson's method, was 0, 6, and 10 U in three different samples taken at different times. The component could not be detected in the Ouchterlony analysis even when 15 μ l were used instead of the usual 10 μ l (Fig. 1). Moreover, immunoelectrophoresis of the patient's serum against monospecific anti-C7 gave no precipitin line (Fig. 2). Hemolytic titration of all other complement components gave normal results; C8 and C9 were found at higher levels than in the normal controls. Immunochemical quantitation of C3, C4, and C3PA were normal. All results are shown in Table II.

Genetic studies of the C7 deficiency. Members

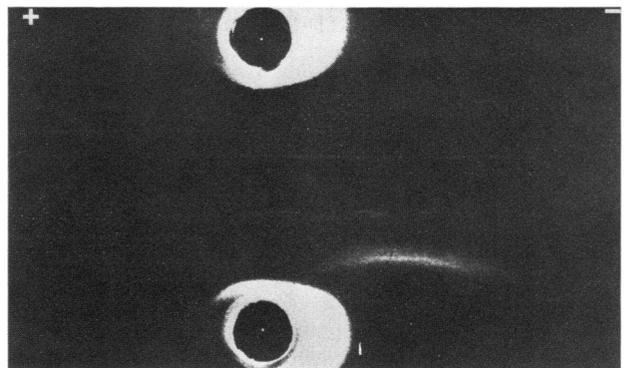


FIGURE 2 Immunoelectrophoretic pattern of the C7-deficient serum. The center trough contains the monospecific anti-C7 serum (0.1 ml), the top well contains the patient's serum (5 μ l), and the lower well contains the normal human serum (5 μ l).

TABLE II
Measurement of Individual Complement Components
in the Patient's Serum

Component	Patient F. R.	Normal range or mean \pm 2 SD*
<i>U/ml</i>		
Hemolytic assay		
C1	32,000	32,000 \pm 8,000
C2	1,760	1,430 \pm 370
C3	4,320	4,000 \pm 1,000
C4	15,500	15,000 \pm 3,000
C5	6,400	6,000 \pm 2,000
C6	43,000	45,000 \pm 15,000
C7	0	32,600 \pm 5,200
C8	512,000	160,000 \pm 96,000
C9	64,000	24,000 \pm 8,000
<i>mg/100 ml</i>		
Radial immunoprecipitation		
C3	88	65–120
C4	41	20–50
Factor B	19.2	15–30
<i>serum dilution</i>		
Ouchterlony's method		
C7	0	3–8

* Representing normal values for this laboratory (see Methods).

from three generations of the patient's family were investigated. The family pedigree is shown in Fig. 3 A, and the CH50 titer and C7 levels, functional and immunological, are listed in Table III. The proposita's parents are dead; the father died of an accident, the mother of cardiopulmonary disease. There was no known consanguinity between them. A paternal aunt (no. 1) had normal CH50 and C7 titers. Two younger sisters of the proposita were found to have CH50 titers of 6 U/ml (no. 6) and 15 U/ml (no. 7), and their C7 values were 10 and 33 U/ml, respectively. In the Ouchterlony analysis, the presence of the C7 protein could not be demonstrated. All other complement components were in the normal range, C8 and C9 showing titers above normal. Seven other siblings had CH50 titers in the lower range of the normal values; in the screening test, five of them (nos. 2, 3, 4, 8, and 9) had C7 titers ranging from 40 to 63% of the normal mean value but two siblings (nos. 10, 11) had border-

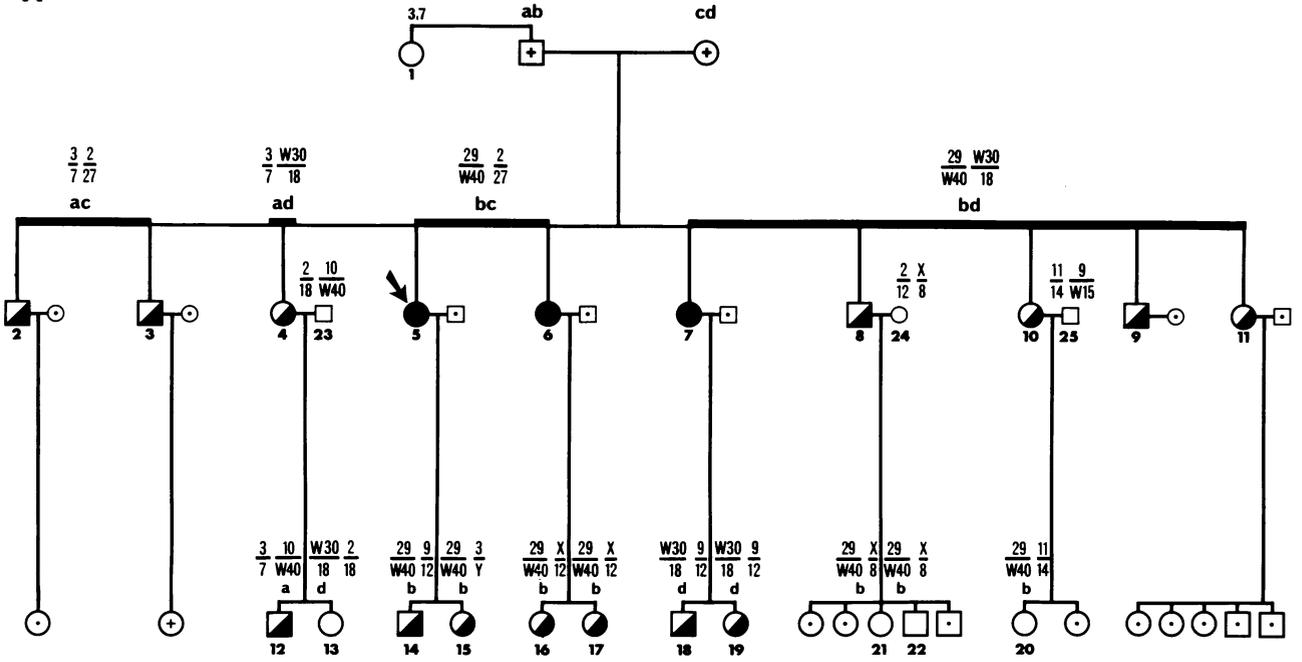
line levels. When C7 was measured by the method of Nelson, the titer of all seven siblings was well below the lower limit of 2 SD and the results are a mean value of a minimum of three determinations. In the immunoassay, the diminution of the C7 protein could be demonstrated.

In the third generation, 11 family members were available for testing. The six children of the three homozygous women (nos. 14, 15, 16, 17, 18, and 19) had low normal CH50 values. The C7 titer of these six individuals, as measured by functional and immunological methods, was about half the normal. The two available children of no. 8 had 34,000 U/ml (no. 21) and 27,000 U/ml (no. 22) of C7, respectively, and in the immunoassay the protein was in the normal range. The two children of no. 4 had CH50 values of 63 U/ml (no. 12) and 48 U/ml (no. 13), and their C7 titer, measured by the Nelson method, was 17,300 U/ml for no. 12 and 37,800 U/ml for no. 13. The C7 protein for these two children corresponded with 1.5 for no. 12 and 4 for no. 13. The low CH50 value obtained in the daughter no. 13) could not be explained by C7 deficiency. Studies of the other complement components revealed that the girl was deficient in C2, a defect she inherited from her father (no. 23), who had CH50 values of 42 U/ml and a markedly low level of C2 in his serum. Sibling no. 12, heterozygous for C7 deficiency, had a C2 level below 2 SD. The ascertainment of a true heterozygous state for C2 deficiency in this sibling warrants further study. The only available child of no. 10 (no. 20) had normal C7 values.

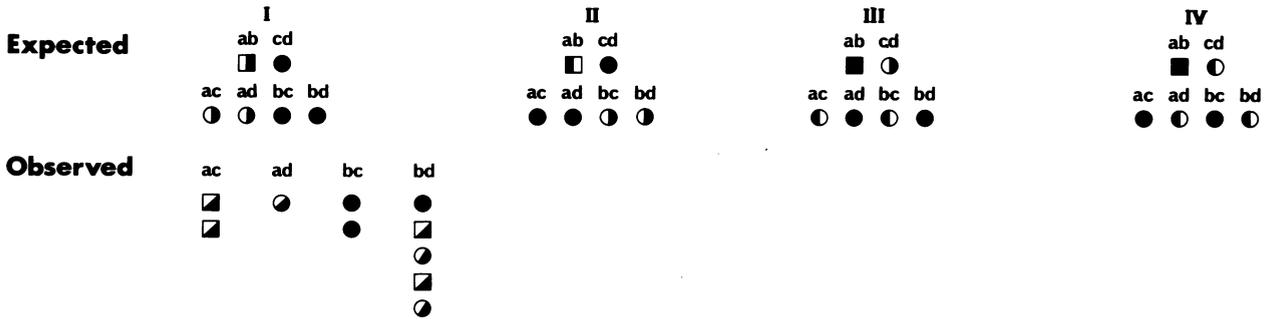
HLA typing (Table III and Fig. 3). HLA typing was performed on the proposita and 21 members of her family. In the first generation, only a living aunt was available for typing. In the second generation, all of the siblings and three of their spouses were typed. In the third generation, six children of homozygous subjects and five children of heterozygous subjects were typed. Genotypes were then determined and subjects of the second generation were distributed in the following four sib groups: *ac*: A3,B7/A2, B27; *ad*: A3,B7/AW30, B18; *bc*: A29,BW40/A2,B27; and *bd*: A29,BW40/AW30, B18. The proposita (no. 5) and her sister (no. 6), both homozygous for C7 deficiency, are in the sib group *bc*. Her second sister (no. 7), homozygous for this deficiency, is in the sib group *bd*, together with four siblings (nos. 8, 9, 10, and 11) heterozygous for this

FIGURE 3 Pedigree of the family with C7 deficiency. A. Pedigree grouped according to HLA genotypes. B. The four possible patterns of inheritance of a single normal C7 allele assuming close linkage between the HLA and C7 loci. C. Hypothetical pedigrees in the case of a normal allele for C7 being present in the first or the fourth parental haplotype. Males are indicated by squares, females by circles. Heterozygotes are shown as half-blackened symbols; homozygotes are shown as fully-blackened symbols. Each member is identified by a number. The proposita is indicated by an arrow within the symbol. Symbols enclosing dots refer to untested individuals, and a cross marks deceased members. The HLA typing is represented by letters: *a* = A3, B7; *b* = A29,BW40; *c* = A2,B27; *d* = A3,B7. The following symbols are also used: *x* = unidentified antigen; *an* = unidentified genotype (could be one of the following: A3, B7/A3,B7; A3,B7/A-,B7; A3,B7/A3,B-; A3,B-/A-, A3,B7/A-,B-); * = recombinants.

A



B



C

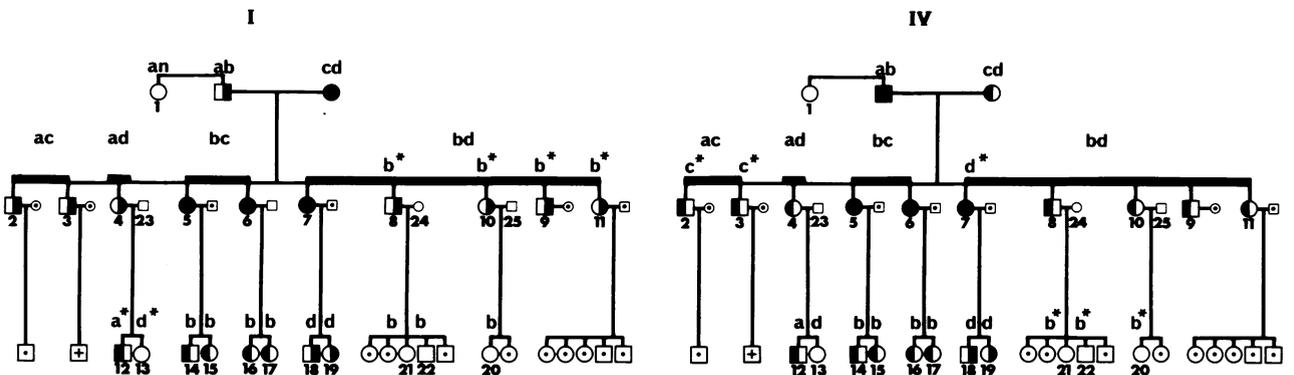


TABLE III
HLA Genotypes, Whole Complement, and C7 Levels in the Studied Family

Subject (nos. as in Fig. 3)	Genotypes	Family haplotypes	CH50	C7		
				Screening	Nelson's Method	Protein
			<i>U/ml</i>		<i>U/ml</i>	<i>Serum dilution</i>
Normal*			96±25	2,000–2,800	32,000±5,200	3–8
1st generation						
Normal						
(1)	A3,B7	<i>a</i>	77	2,200	30,000	3
2nd generation						
Homozygotes						
(5)	A29,BW40/A2,B27	<i>bc</i>	0	ND†	0	0
(6)	A29,BW40/A2,B27	<i>bc</i>	6	ND	10	0
(7)	A29,BW40/AW30,B18	<i>bd</i>	15	ND	33	0
Heterozygotes						
(2)	A3,B7/A2,B27	<i>ac</i>	71	1,090	14,000	1.5
(3)	A3,B7/A2,B27	<i>ac</i>	76	1,155	16,500	1.5
(4)	A3,B7/AW30,B18	<i>ad</i>	86	940	13,000	1
(8)	A29,BW40/AW30,B18	<i>bd</i>	86	1,129	22,600	1.5
(9)	A29,BW40/AW30,B18	<i>bd</i>	75	1,470	16,300	1
(10)	A29,BW40/AW30,B18	<i>bd</i>	73	1,827	20,700	2
(11)	A29,BW40/AW30,B18	<i>bd</i>	75	1,832	25,200	1.5
3rd generation						
Children of (5)						
(14)	A29,BW40/A9,B12	<i>b</i>	61	1,164	14,000	1.5
(15)	A29,BW40/A3,B—	<i>b</i>	79	1,350	18,300	1.5
Children of (6)						
(16)	A29,BW40/A—,B12	<i>b</i>	69	911	17,500	1
(17)	A29,BW40/A—,B12	<i>b</i>	73	1,448	16,600	1.5
Children of (7)						
(18)	AW30,B18/A9,B12	<i>d</i>	72	1,272	16,800	1.5
(19)	AW30,B18/A9,B12	<i>d</i>	71	903	16,000	1.5
Children of (4)						
(12)	A3,B7/A10,BW40	<i>a</i>	63§	720	17,300	1.5
(13)	AW30,B18/A2,B18	<i>d</i>	48	1,632	37,000	4
Husband of (4)						
(23)	A2,B18/A10,BW40		42	ND	36,400	ND
Children of (8)						
(21)	A29,BW40/A—,B8	<i>b</i>	80	ND	34,000	4
(22)	A29,BW40/A—,B8	<i>b</i>	76	ND	27,000	3
Spouse of (8)						
(24)	A2,B12/A—,B8		103	ND	37,800	3
Child of (10)						
(20)	A29,BW40/A11,B14	<i>b</i>	65	2,060	27,600	3
Husband of (10)						
(25)	A11,B14/A9,BW15		76	ND	38,000	ND

* Normal range or mean±2 SD.

† Not determined.

§ Individual having borderline C2 level.

^{||} Indicates heterozygous C2 deficiency.

deficiency. Two siblings (nos. 2 and 3) having the haplotype *ac* and one (no. 4) with the haplotype *ad* are heterozygous. In the third generation, the children of the three homozygous women inherited either the haplotype *b* (nos. 14, 15, 16, and 17) or *d* (nos. 18 and 19). The only child (no. 20) of a heterozygous sister of the group *bd* (no. 10) and two children (nos. 21 and 22) of a heterozygous brother *bd* (no. 8), who were available for testing, have all normal C7 level and all inherited the family haplotype *b*. The heterozygous sister (no. 4) having the genotype *ad* had two children. One of them (no. 12), heterozygous for C7 deficiency, inherited the family haplotype *a*, while the other (no. 13), with normal C7, inherited the haplotype *d*. In the first generation, a paternal aunt had the phenotype *an* and she was normal for C7.

Lod scores were calculated for a backcross (homozygote \times heterozygote) or intercross (heterozygote \times heterozygote) mating and are shown in Table IV.

Other studies. The opsonic activity of serum was measured in all individuals studied for complement activity and no deficiency could be detected. Coagulation studies PT and APTT were performed in the three homozygous sisters (nos. 5, 6, and 7) and in some other siblings (nos. 4, 8, 14, 15, 16, 17, 18, and 19) and were normal. The determination of immunoglobulins, performed in every individual tested for complement, revealed no deficiency of either immunoglobulin G, A, or M, and the determination of antinuclear factor gave results within normal limits for every subject.

DISCUSSION

The routine determination of complement in arthritic patients had led to the discovery of a complete absence of the seventh component of complement in a patient presenting signs and symptoms characteristic of ankylosing spondylitis. A family study revealed two additional cases of homozygous deficiency in two sisters of the proposita, whereas the seven other siblings were heterozygous for this deficiency. It is probable that one of the parents in the first generation was homozygous, while the other was heterozygous for C7 deficiency. In the third generation, seven heterozygotes were discovered.

The pattern of heredity shown in Fig. 3 conforms to an autosomal codominant character. No abnormal tendency to repeated infection, bleeding disorders, or collagen disease was found in any member of the family. In one branch of this family (no. 4) there was a coexistence of C7 deficiency with hereditary C2 deficiency; this remarkable sibship will be studied extensively later. The presence of B27 in the proposita and the known association between certain complement components and HLA (9-13) prompted us to look for a similar association in this family. The hypothesis of a close linkage between the gene for C7 and those for the HLA system allows four different combinations of these genes to be postulated, depending on which of the four parental haplotypes carries the single allele for a normal C7 level. In Fig. 3 B are depicted the four hypothetical patterns I, II, III, and IV; patterns I and IV give the best fit. However, both show some discrepancies (Fig. 3 C, patterns I and IV). In Fig. 3 C, pattern I, there are four differences between the expected and the observed situation, all in the group *bd*. Two more differences exist in the third generation. In pattern IV, Fig. 3 C, there are two differences between expected and observed in the *ac* group and one difference in a sibling in the *bd* group. Three other differences are to be found in the third generation. In pattern IV of Fig. 3 C, it is supposed that the father in the first generation would have been homozygous and the mother heterozygous. The hypothesis described in pattern IV, Fig. 3 C, would not be compatible with the presence in the paternal aunt (no. 1), normal for C7, of the phenotype A3,B7. She need not necessarily share an identical haplotype with her brother since the frequency of this haplotype in the population is high.

All the above-mentioned differences would have to be the consequence of paternal or maternal crossovers between the HLA genes and that responsible for the C7 level. Should a close genetic linkage exist between the gene for C7 deficiency and the HLA genes, an exceedingly high number of recombinants would have to be postulated, which renders the hypothesis of close linkage unlikely.

The lod score analysis for the proposed backcross mating in the first generation gives HLA:C7 deficiency linkage exclusion to 10 centimorgans. (It is generally accepted that linkage has been excluded to a specified recombination frequency when the lod score is ≤ -2.0). The possibility of an intercross mating in the first generation must be considered. Sibling no. 11 has a borderline mean functional C7 level. It is to be noted that her protein C7 values were definitely below normal. However, if sibling no. 11 were normal for C7, the lod score for an intercross mating would give an HLA:C7 linkage exclusion to about 4 centimorgans. In

TABLE IV
Lod Score Analysis*

Mating type	0.00	0.05	0.1	0.2	0.3	0.4
Backcross	$-\infty$	-3.79	-2.19	-0.84	-0.27	-0.05
Intercross	$-\infty$	-1.79	-0.70	-0.04	0.06	0.03

* Lod scores for a recombination frequency of different mating types.

any event, there certainly is no evidence in favor of linkage.

Our findings indicating independent segregation of the genes of the HLA system and those controlling the level of the seventh component of complement are in agreement with those of other workers who demonstrated genetic independence between the HLA system and deficiencies in the first (14), fifth (15), and sixth (14) components of complement. However, there is strong evidence for genetic linkage for the second (9–11) and fourth (13, 16) components of the classical pathway and for factor B (12) of the alternate pathway, whereas for C8 the reported results are equivocal (17, 18).

Complement deficiencies in man, mouse, and guinea pig may be accompanied by an increased susceptibility to infectious and immune complex diseases such as lupus erythematosus and glomerulonephritis (1), although it is to be remembered that most C2-deficient people, for example, are not clinically affected (19). When severe recurrent infections occur, as in the case of C3, C5, and C6 deficiency, the relationship between the deficiency and the infectious syndrome appears evident. However, the high incidence of collagen disorders in other complement deficiencies has raised the question (20) of a true association versus an "ascertainment artifact"; maybe the interest of rheumatologists in complement levels has introduced biased data. This phenomenon may apply in the interpretation of the simultaneous presence of ankylosing spondylitis and B27 together with a homozygous deficiency of C7 in our proposita. All the patients suffering from ankylosing spondylitis that have been tested in our service had normal or elevated CH50 titers, and to date we can find no evidence of an association between a complement deficiency and ankylosing spondylitis, except in the proposita. The two homozygous sisters of the proposita do not suffer from any ailment. In the two C7-deficient patients described in the literature, one case occurred in an apparently healthy boy (2) and the other one (3) in a female with a typical history of Raynaud phenomenon, telangiectasia, and acrosclerosis with some indication that she may develop scleroderma. Moreover, high dilutions of the serum of Wellek and Opferkuch's patient inactivated fluid phase C7 and also cell-fixed C7. Those are features that are found neither in Boyer's case (3) nor in ours. It is noteworthy that the proposita in Boyer's case and the family we are reporting have French ancestry. To ascertain the full clinical significance of this apparently rare complement deficiency, it will be necessary to compile more data. A chance finding was the discovery of a C7-deficient heterozygous sibling (no. 4) married to a C2-deficient man. This rare combination led to the simultaneous appearance of hereditary C2 and C7 deficiency in their children.

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