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PENETRATION OF HLA-LINKED PSORIASIS-PREDISPOSING GENE(S): A FAMILY INVESTIGATION

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Abstract. In the present study we have investigated a psoriatic family (26 individuals) on the basis of disease linkage to the HLA-region, including all the known alleles (HLA-A, B, C and D). Psoriasis occurred in 5 individuals. Our results seem to favour a dominant mode of inheritance of the MHS-linked disease-predisposing gene(s) though with varying clinical appearance, e.g. severe arthralgia seems to travel with the same haplotype as that associated with psoriasis. In one parent combination the individuals shared the haplotype Al B 17 but not the D-allele (MLC allele), and psoriasis was linked to only one of the haplotypes. This indicates that it is not the B 17 allele itself which is responsible for the development of disease, but other gene(s) in close proximity to the B-region.

Key words: Psoriasis; Mixed lymphocyte culture; Histocompatibility antigens

The major histocompatibility system (MHS) in humans is a chromosomal region which has recently been shown (14) to be located on chromosome no. 6 (Fig. 1). This region contains genes that besides controlling the expression of cell surface structures, HLA-antigens (HLA=human leukocyte, locus A), also encompasses genes of importance in immunological reactivity. The HLA-antigens are present on almost all nucleated cells in the organism and show great variability between individuals (27). These gene products were earlier referred to as HL-A antigens, belonging to three distinct subregions, the LA, Four and AJ subloci. These are now renamed HLA-A, B and C respectively (13) (Fig. 1).

This region also contains genes for cell surface products which stimulate reactions in mixed lymphocyte cultures, earlier called MLC products, today referred to as HLA-D products. These genes are located close to the HLA-B locus (27). Furthermore, there are in this region alloantigenic structures coded for by genes, which could be similar to or possibly identical with the HLA-D locus products. These products are distinct from the HLA-A, B and C antigens, in so far as they have a characteristic tissue distribution and also a distinct chemical structure (22).

Recently, this region has also been shown to contain genes controlling the expression of complement factors C'2 (6, 8) as well as the factor B of the alternate complement activation pathway (1). In the mouse too, the analogue of the human MHS, H-2, has been shown to contain genes for complement activity (7).

The most important genes, thought to be located within the human MHS region, are IR-genes (IR-gene=immune response gene) which control immune responsiveness to thymus-dependent antigens. In many different experimental species it has been clearly established that the MHS analogue houses IR genes, hence, their hypothetical location within the human MHS is indicated (4, 18).

It has been shown that many diseases are associated with the presence of specific HLA genes within the MHS. Thus, by analogy it is believed that associations between alleles in this region and diseases would be indirect and caused by linkage

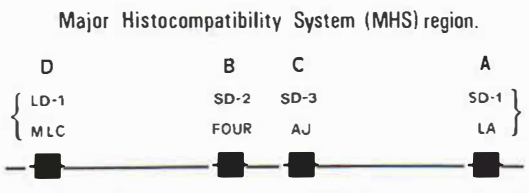


Fig. 1. Outline of the human MHS chromosomal region with the four subloci A, B, C and D (only one of the two paired chromosomal regions is shown). Previous nomenclature is given within parentheses.

disequilibrium between hypothetical IR genes and the HLA genes. Thus, disease associations with products of the HLA-B and D subregions have been clearly verified for many different diseases (26). This implies that the location of the human hypothetical IR genes would be close to the B and D subregions, and more distant from the HLA-A and C subregions.

The fact that alleles of distinct HLA subregions occur in linkage disequilibrium with each other (18), forms the basis for all studies on associations between diseases and specific alleles of the MHS-region. This implies that some combinations of genes of different subregions occur together more often or more rarely than might be expected from the frequencies of the individual gene products.

The alleles HLA-B 13, B 17 and BW 37 (previously named HL-A 13, 17 and Ty) have been shown to be more frequent in psoriatics than among unrelated healthy controls (3, 10, 16, 23, 25, 29, 30). Another interesting association to be mentioned in this context has been reported between arthritic lesions in psoriatic patients and HLA-B 27 (previously named HL-A 27) (16, 26). This latter allele has also a very strong positive association with arthritic lesions such as ankylosing spondylitis, Reiter's disease and juvenile rheumatoid arthritis, to mention a few (21, 26).

In spite of the strong associations found between psoriasis and specific HLA-B alleles it is not believed that the gene(s) which is (are) present within the MHS-region and which is (are) directly responsible for the increased risk of developing psoriasis, is identical with any of the above-mentioned alleles. Thus, many psoriasis patients have neither of the above alleles, but might of course still carry the same psoriasis-predisposing gene(s), not linked to any of the above HLA-B alleles. It is therefore impossible to predict the frequency of the MHS-linked disease-predisposing gene in a randomly

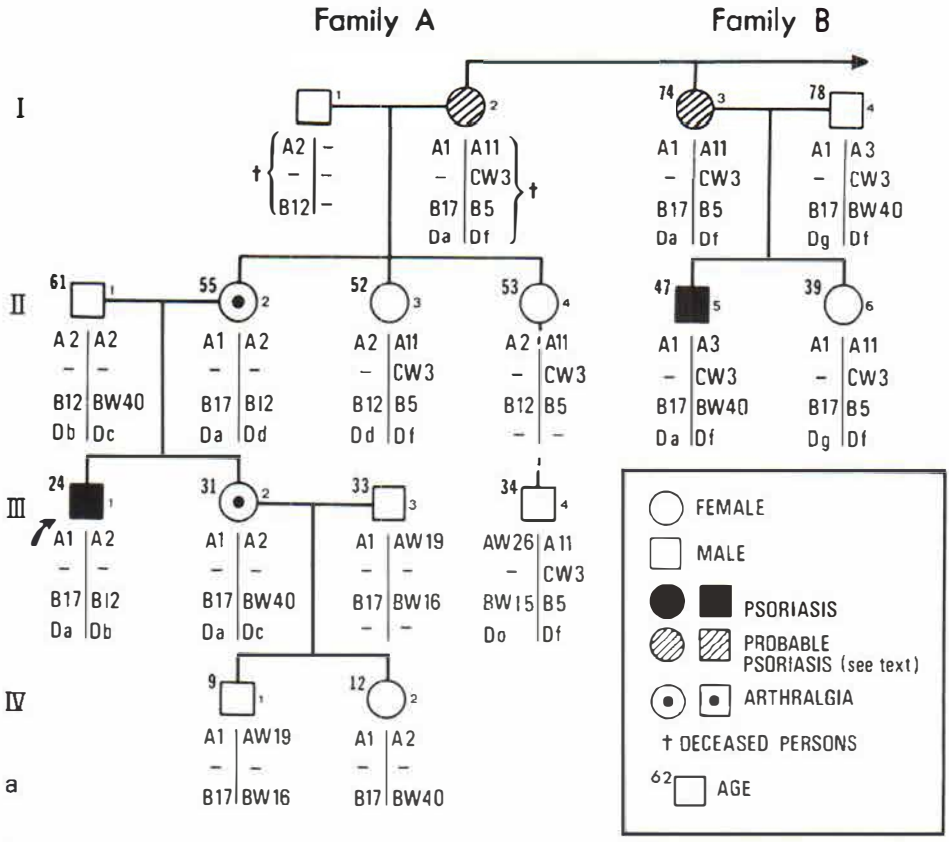


Fig. 2 a and b. Pedigree of the whole family where individuals I:2, I:3, I:5 and I:7 are siblings. The proband is indicated with an arrow. The family is subdivided into families A, B, C and D.

selected patient material. Thus, we have investigated families where several members have psoriasis. In this way only can the degree of penetration of disease in individuals carrying the MHS haplotype of the proband be evaluated. In this paper we will discuss one of the families.

MATERIAL AND METHODS

Patients

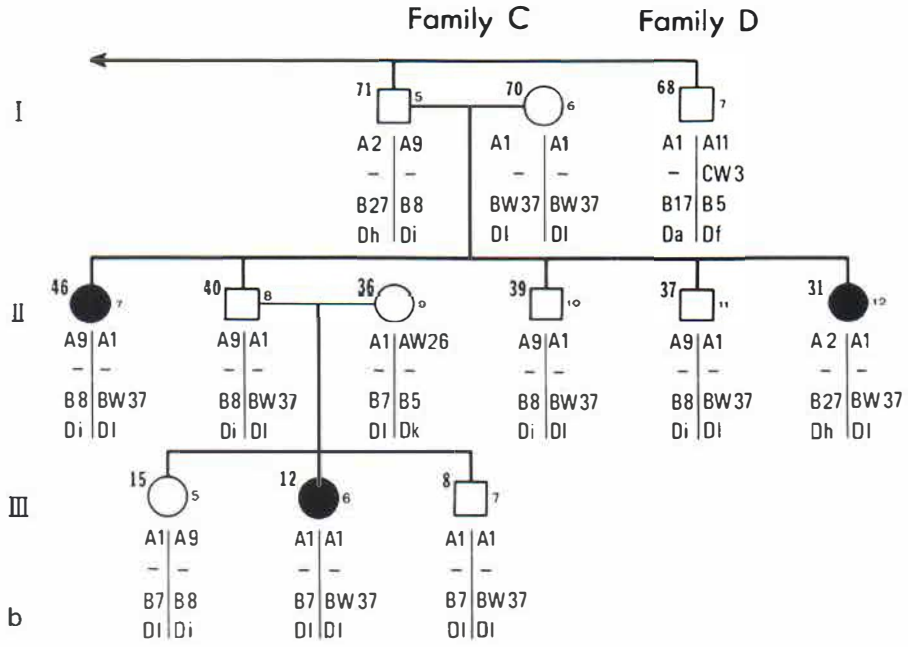
The family (Fig. 2*a*, *b*) selected for this study consisted of 26 members belonging to four generations. Five individuals had definite psoriasis (II:5, II:7, II:12, III:1 and III:6), 2 were classified as probable psoriasis (I:2, I:3) and 2 had severe joint pain (II:2, III:2). Individual I:7 has been included in this study but not his two children who according to hearsay are said to be healthy. The haplotypes of I:1 and I:2 were necessarily deduced as both of them died at the age of 70. Individuals I:2, I:3, I:5, I:7 are siblings.

The proband (III:1) is an outpatient at the Department of Dermatology, Karolinska sjukhuset, Stockholm. His parents (II:1, II:2) and the family of his sister (III:2, III:3, IV:1 and IV:2) live in Stockholm and the rest of the family is scattered in different parts of Sweden.

All members of the family have been examined by one of us (J. M.) for presence of psoriatic lesions on two occasions, with a 6–12 month interval. Individuals II:4 and III:3 have been examined once only. The whole body was carefully inspected, with special attention to the scalp, elbows, knees, nails and the buttocks. Those with unequivocal signs of psoriasis when examined are marked on Fig. 2*a* and *b*. None had seborrhoic dermatitis to such a degree that it was difficult to differentiate between that clinical entity and psoriasis.

For borderline cases the criteria formulated by Baker were adopted (2). Consequently, one individual (II:10) was not classified as psoriatic despite the fact that he had minimal subungual hyperkeratoses on four toenails. Culture for presence of dermatophytes was negative.

Those with probable psoriasis deserve further description. Individual I:3 had a period 25 years ago with scaling patches on the elbows that could have been psoriasis. However, these changes disappeared spontaneously. At the time of investigation at the age of 74 she presented no signs of psoriasis. The daughters of I:2 described how



their mother "was scratching her elbows" and had "eczema" in the submammary region. Although no definite diagnosis can be made retrospectively, the description could fit in with the diagnosis of psoriasis.

The mother (II:2) and the sister of the proband (III:2) complained spontaneously of joint pain. Therefore those two plus the proband (III:1) and his father (II:1) have been examined by Dr Alf Elman, Rheumatological Clinic, Karolinska sjukhuset, for presence of arthritic lesions. This examination was undertaken without knowledge of the results of the tissue typing and MLC test. X-ray pictures have been taken of the hands, feet and the sacroiliac joints, as described earlier (16). The sister and the mother complained of arthralgia affecting more than three peripheral joints, i.e. they fulfilled criterion no. 1 for inflammatory polyarthritis as postulated by Moll & Wright (20). However, no objective signs of arthritis could be found at the time of the investigations, i.e. criterion no. 2 according to the same authors was not fulfilled.

No radiological changes could be found. The absence

Table I. *MLC experiment with members from families A and B*

Results are given as stimulation ratios (above) and counts per minute (cpm) (below). For explanation see text

Individual	Responder cells: HLA A, C, B	Postulated D alleles	Stimulator cells: III 1 m[a]	III 4 m	III 2 m	II 1 m	II 2 m
III 1	A 1, B 17/A 2, B 12	a b	*2 600*	15 39 800	24 61 700	21 55 200	21 55 700
III 4	AW 26, BW 15/A 11, CW 3, B 5	o f	23 41 000	*1 800*	46 83 000	N.T.[b]	40 71 500
III 2	A 1, B 17/A 2, BW 40	a c	12 48 000	14 57 300	*4 000*	20 81 800	18 72 200
II 1	A 2, B 12/A 2, BW 40	b c	8 26 200	11 37 100	12 41 000	*3 400*	21 70 300
II 2	A 1, B 17/A 2, B 12	a d	19 45 200	33 78 100	18 42 000	60 143 400	*2 400*
II 3	A 2, B 12/A 11, CW 3, B 5	d f	36 79 500	14 31 500	57 125 800	55 121 500	29 63 800
II 5	A 1, B 17/A 3, CW 3, BW 40	a f	35 55 600	16 25 600	41 65 600	56 90 300	42 66 400
II 6	A 1, B 17/A 11, CW 3, B 5	g f	34 121 200	16 58 200	38 137 300	37 134 700	43 154 700

[a] m=mitomycin-treated cells. [b] N.T.=not tested.

Table II. *MLC experiment with members from family B and some members from families A and C*

Results are given as relative response and as epm (within parentheses). The boxes indicate MLC identity

Individual	Responder cells: HLA A, C, B	Postulated D alleles	Stimulator cells: II 5 m[a]	I 3 m	II 6 m	I 4 m	I 6 m	II 1 m
II 5	A 1, B 17/A 3, CW 3, BW 40	a f	*(2 100)*	0	70	45	65	112
I 3	A 1, B 17/A 11, CW 3, B 5	a f	0	*(2 000)*	72	67	73	137
II 6	A 1, B 17/A 11, CW 3, B 5	g f	48	101	*(3 300)*	20	89	215
I 4	A 1, B 17/A 3, BW 40	g f	43	59	23	*(2 200)*	79	139
I 6	A 1, BW 37/A 1, BW 37	i i	83	128	123	100	*(1 400)*	106
II 1	A 2, B 12/A 2, BW 40	b c	76	121	129	107	66	*(7 300)*
II 2	A 1, B 17/A 2, B 12	a d	40	118	141	76	107	151
I 5	A 2, B 27/A 9, B 8	h i	N.T.	100	N.T.	118	102	N.T.
C[b]	N.T.[c]		(41 600)	(73 900)	(67 100)	(39 500)	(46 800)	(72 900)

[a] m=mitomycin-treated cells. [b] C=control. [c] N.T.=not tested.

of objective signs of arthritis make us hesitant to classify the findings as arthritis but the symptoms are in fact so obvious that they deserve mention. The sheep red cell agglutination test (24) was performed on all the individuals (II:1, II:2, III:1, III:2) for estimation of rheumatoid factor. According to the norm of the laboratory any titre higher than 1/128 was considered positive. The test was negative for all except for III:2, who had a titre of 1/64.

The rest of the family was not investigated in this respect as they lived in other parts of the country.

II 3 m	II 5 m	II 6 m
32	14	30
82 200	36 900	78 300
19	12	14
33 400	21 000	25 000
24	12	24
96 700	46 900	96 000
18	12	23
60 700	42 400	77 500
26	13	37
61 300	32 000	89 800
	22	20
2 200	48 200	44 500
24		29
38 700	*1 600*	46 500
33	23	
118 800	84 000	*3 600*

II 2 m	I 5 m	C[b] m
97	N.T.[c]	(45 200)
117	119	(34 900)
178	N.T.	(44 600)
163	119	(48 700)
140	100	(44 700)
96	N.T.	(52 600)
(*2 900*)	N.T.	(49 400)
N.T.	(*3 900*)	(43 600)
(83 800)	(57 000)	(*3 200*)

Tissue typing

Lymphocytes were separated from defibrinated blood by flotation on Ficol-Isopaque as described by Böyum (5). Tissue typing was carried out by the microcytotoxicity assay of Kissmeyer-Nielsen & Kjerbye (12). Most of the antisera were obtained from the Scandia Transplant organisation. The new nomenclature for the HLA alleles will be used consistently (13).

The following specificities were tested:

HLA-A (LA, SD-1): 1, 2, 3, 10, (W 25, W 26), 11, 28, W 19, (W 29, W 30–W 31, W 32).

HLA-B (FOUR, SD-2): 5, 7, 8, 12, 13, 14, 17, 27, W 15, W 16, W 18, W 21, W 22, W 35, W 37, W 40, W 41.

HLA-C (AJ, SD-3): W 1, W 2, W 3.

Mixed lymphocyte culture (MLC)

Defibrinated blood or heparinized blood was diluted 1 : 1 with 0.9% NaCl solution and the lymphocytes were separated and purified as described above. After washing 3 times they were diluted in culture medium consisting of Eagle's minimal essential medium in Earle's salt solution supplemented with 50 IU of penicillin and streptomycin/ml and 10% inactivated (30 min in 56°C) human AB-serum. Falcon flat-bottomed microtitre plates (Falcon Plastics, Oxnard, Cal., USA) were used for cultivation of high cell concentration (1.5×10^5 cells/0.1 ml) Linbro's round-bottomed plates (Linbro Scientific Inc. Manufactures, Hamden, USA) for lower concentrations (10^5 cells/0.1 ml and 5×10^4 cells/0.1 ml). In MLC experiment no. 1 (Table I), 1.5×10^5 cells were used as responding and stimulating cells, and in test no. 2 (Table II), 10^5 cells. In all the other MLC experiments (Tables III–V) 5×10^4 cells were used. The reason for using different cell concentrations in the experiments was that, on occasion, the blood had been transported for more than 24 hours prior to purification of the lymphocytes and sometimes the yield was less than 15% of that expected. Thus the only way to carry out the experiment was to reduce the number of cells, as has been discussed previously in this type of experiment (9). The stimulator cells were produced by treatment with 0.025–0.05 mg Mitomycin (Sigma Chemical Company, St. Louis, USA) per ml of cell suspension for 45 min at a cell concentration of 2.5×10^6 cells/ml ("m" will hereafter designate Mitomycin-treated cells). The lymphocytes were afterwards washed three times in culture medium before final dilution. Each well during the culture period held 0.2 ml medium. All cell combinations were studied: A+B, A+Bm, Am+B, Am+Bm and the autologous controls, A+Am and B+Bm (A and B represent lymphocytes from 2 individuals named A and B). The combinations A+B and Am+Bm served as controls and are not reported. All the cultures were performed in quadriplicate. The controls were mostly unrelated blood donors unless healthy members of the family could serve as controls.

The cultures were incubated in a humid atmosphere of 5% CO_2 in air for 5 days; thereafter 1 μCi of ^{3}H-Thymidine (Radiochemical Centre, Amersham, England, spec. act. 5 Ci/mmol) was added to each culture and the incubation continued for another 24 hours.

Cultures were harvested on glass fibre filters using distilled water and an automatic microculture harvesting machine (Skatron, Lierbyen, Norway). Filters were dried overnight at 60°C, and thereafter added to the scintillation vials containing 5 ml of toluene-based scintillation fluid. Vials were counted in an Intertechnique (Nanoteknik) scintillation counter.

The values in MLC experiment no. 1 (Table I) are expressed as stimulation ratios (SR):

$$SR = \frac{AXm}{AAm},$$

In MLC experiments 2–5 (Tables II–V) the results are expressed as the relative response (RR), which can be derived from the following formula:

$$RR = \frac{AXm - AAm}{ACm - AAm} \times 100,$$

where X is the test cell and C is the control cell which is a double HLA-D mismatch with regard to A.

RESULTS

The results are presented with interpretation to make the reading easier. For convenience the whole family has been subdivided into four parts: families A, B, C and D.

Tissue typing

Families A and B (Fig. 2*a*). The proband (III:1) who has psoriasis carries the HLA genotype A 1, B 17/A 2, B 12. A cousin of the proband's mother (II:5) also has psoriasis and the HLA genotype A 1,

Table III. *MLC experiment with the first and second generation of family C*

Results are given as relative response and cpm (within parentheses). The boxes indicate MLC identity

Individual	Responder cells: HLA A, C, B	Postulated D alleles	Stimulator cells: *I 5 m*[a]	I 6 m	II 7 m	II 8 m	II 10 m	II 11 m
I 5	A 2, B 27/A 9, B 8	h i	(*700*)	121	109	45	138	99
I 6	A 1, BW 37/A 1, BW 37	l l[d]	96	(*600*)	50	18	37	62
II 7	A 9, B 8/A 1, BW 37	i l	98	0	(*4 600*)	0	2	0
II 8	A 9, B 8/A 1, BW 37	i l	177	2	7	(*1 200*)	9	5
II 10	A 9, B 8/A1, BW 37	i l	108	5	2	0	(*4 300*)	4
II 11	A 9, B 8/A 1, BW 37	i l	122	2	2	0	7	(*1 600*)
II 12	A 2, B 27/A 1, BW 37	h l	38	13	41	17	87	42
II 9	A 1, B 7/AW 26, B 5	l k	40	14	61	21	72	41
C[b]	N.T.[c]		(40 300)	(34 900)	(37 100)	(10 500)	(48 100)	(31 600)

[a] m=mitomycin-treated cells. [b] C=control. [c] N.T.=not tested. [d] l=DW 2 (LD 7 A).

Table IV. *MLC experiment including members from the second and third generation of family C*

Values as relative response and cpm (within parentheses). The boxes indicate MLC identity

Individual	Responder cells: HLA A, C, B	Postulated D alleles	Stimulator cells: II 8 m[a]	II 9 m	III 5 m	III 6 m	III 7 m	C[b] m
II 8	A 9, B 8/A 1, BW 37	i l[d]	(*800*)	61	3	11	6	(37 200)
II 9	A 1, B 7/AW 26, B 5	l k	26	(*1 700*)	27	31	10	(42 200)
III 5	A 1, B 7/A 9, B 8	l i	11	141	(*800*)	12	13	(16 000)
III 6	A 1, B 7/A 1, BW 37	l l	38	100	22	(*400*)	0	(45 000)
III 7	A 1, B 7/A 1, BW 37	l l	19	83	23	2	(*200*)	(49 000)
C[b]	N.T.[c]		(42 500)	(52 700)	(31 700)	(43 900)	(30 500)	(*2 800*)

[a] m=mitomycin-treated cells. [b] C=control. [c] N.T.=not tested. [d] l=DW 2 (LD 7 A).

B 17/A 3, CW 3, BW 40. The grandmother (I:2), deceased, and the sister of the grandmother (I:3) have the genotype A 1, B 17/A 11, CW 3, B 5, and they have probably had psoriasis as well. The proband's mother (II:2) with the HLA genotype A 1, B 17/A 2, B 12 and his sister (III:2) with the tissue type A 1, B 17/A 2, BW 40 both have severe arthralgia which, according to some authors, should be classified as arthritis (20). A very interesting coincidence is the fact that the maternal aunt and her husband (I:3 and I:4) of the proband's mother share the haplotype A 1, B 17 and the two children (II:5, II:6) have inherited this haplotype, but from either the mother or the father. However, the individual (II:5) who has psoriasis received his A 1, B 17-haplotype from his mother (I:3), the very same haplotype that is supposed to have transmitted the disease to the proband and which is associated with joint pain in individuals II:2 and III:2. A further interesting point is that the father (I:4) and his son (II:5) are identical with respect to HLA-A, -B, -C but not with HLA-D. This will be discussed in detail below. The same situation is to be found regarding the mother (I:3) and daughter (II:6). Thus, only one of the A 1, B 17 haplotypes seems to carry the psoriasis-predisposing gene(s).

The sister of the proband (III:2) is married to a man with the HLA haplotype A 1, B 17. The child (IV:1) has the A 1, B 17 haplotype from his mother (III:2), the particular haplotype, which is associated with psoriasis and joint pain. The child (IV:2) received the haplotype A 1, B 17 from his father who is known not to have psoriasis in his family.

Family C (Fig. 2*b*). In this family, descending from the brother of the proband's maternal grandmother (I:5) and his wife (I:6), two children have psoriasis (II:7, II:12). These two persons have the haplotype A 1, BW 37. Neither their father (I:5) nor their mother (I:6) have psoriasis. Another member of family C in the third generation is also affected with psoriasis, namely III:6. She too has the A 1, BW 37 haplotype. Obviously in this part of the family, psoriasis is associated with the A 1, BW 37 haplotype and in the latter case the disease most probably has been transmitted by her father (II:8) who is healthy. The grandmother (I:6) is homozygous with respect to HLA-A and B alleles, as well as for the D-alleles (see below). Questioning of the patient (I:6) failed to disclose any consanguinity in earlier generations.

In conclusion, psoriasis in the whole family occurs in several members carrying either A 1, B 17 or A 1, BW 37. Two members have no skin

II 12 m	II 9 m	C[b] m
22	137	(57 100)
37	N.T.[c]	(50 400)
21	79	(78 700)
52	135	(34 400)
32	77	(66 300)
38	118	(51 700)
(*1 000*)	64	(62 700)
30	(*4 000*)	(65 200)
(15 500)	(56 400)	(*800*)

Table V. *MLC identity and homozygosity between I:6, III:6 and III:7*

Values as relative response and cpm (within parentheses)

Individual	Responder cells: HLA, A, C, B	Responder cells: Postulated D alleles	Stimulator cells: I 6 m[a]	Stimulator cells: III 6 m	Stimulator cells: III 7 m	Stimulator cells: C[b] m
I 6	A 1, BW 37/A 1, BW 37	11[c]	(*300*)	2	1	(17 300)
III 6	A 1, B 7/A 1, BW 37	11	0	(*400*)	0	(41 500)
III 7	A 1, B 7/A 1, BW 37	11	0	2	(*800*)	(35 000)

[a] m = mitomycin-treated cells. [b] C = control. [c] 1 = DW 2 (LD 7 A).

manifestations but arthralgia and the haplotype A 1, B 17.

Mixed lymphocyte culture experiments (MLC)

The results are shown in Tables I–V. The postulated MLC alleles (D alleles) are designated in small letters. The results of MLC experiment no. 1 (Table I) are given in counts per min and as stimulation ratios. Knowing that sharing of one D allele gives lower responses than incompatibility with respect to two D products makes it possible to interpret the results in terms of single or double HLA-D missmatch (27). No unexpected results were obtained and no meiotic recombinations had occurred. However, a few points deserve mention:

1) The MLC alleles linked to the A 2, B 12 haplotype of II:1 and II:2 differ.

2) The MLC alleles A 1, B 17 haplotype of the proband (III:1) and the cousin of his mother (II:5) are probably identical (Table I).

As the recombination frequency between HLA-B and D is less than 1 % (27) and by the interpretation of results in the MLC experiment no. 1 (Table I), the most likely interpretation is the one given above in despite the fact hat the lymphocytes of the grandmother (I:2) were not included in the experiment.

Experiment 1 gives low values in the one-way MLC between the siblings II:5 and II:6, indicating that they must share one MLC determinant. At first glance this seems to be due to the common haplotype A 1, B 17 and the D allele linked to it. This is not the case, however, as can be seen from experiment 2 (Table II). Thus, neither the individuals II:5 and I:3 stimulate each other, nor do I:4 and II:6. If the A 1, B 17 haplotype from either parent (I:3 and I:4) were linked to the same MLC allele, then there would be no or very little stimulation between I:3 and II:6 on the one hand and between I:4 and II:5 on the other. This is not the case in this experiment, which confirms that the MLC alleles linked to the A 1, B 17 haplotype of these two parents are different.

Finally the most important point is that the A 1, B 17 haplotype associated with psoriasis does not carry the same MLC allele as the A 1, B 17 haplotype not linked to psoriasis.

If the HLA-linked gene which enhances the susceptibility to psoriasis is situated within the MHS chromosomal region of the proband's haplotype A 1, B 17, Da and if this gene is transferred to individual I:5 by crossover event between HLA-B and HLA-D, then it should be possible to demonstrate this by including I:5 in an MLC test with I:3. But the responses were too high to draw such conclusions. Individual I:6 was also included in experiment 2 (Table II) but she stimulated and responded so intensely, which excluded any similarity with respect to MLC products.

The MLC experiment 3 (Table III) including the parents I:5 and I:6 and their children II:7–II:12 shows that all the children responded weakly to their mother's (I:6) lymphocytes, thus showing that she is homozygous for HLA-D determinants. Her cells were tested against a panel of typing cells and her lymphocytes have been characterized as homozygous for DW2 (LD-7a) (28). Cells from all the other individuals within this MLC stimulated and responded as expected, with no recombination between HLA-B and D. The wife (II:9) of individual II:8 was also included in the experiment (Table III) which revealed that she shared the DW 2 allele with the grandmother, I:6. As a consequence, she (II:9) has given birth to two children (III:6 and III:7) homozygous for the D alleles. This is demonstrated in experiment 4 (Table IV). The identity between the lymphocytes of individuals III:6, III:7 and I:6 is shown in the last experiment (Table V). The individuals belonging to the family of the proband's sister (III:2, III:3, IV:1 and IV:2) has not been included in a MLC experiment, as none of the children was affected.

MLC in both directions between I:3 and I:7 gave low stimulation ratios (0.7 and 1.3), showing MLC identity.

DISCUSSION

The HLA antigens B 13, B 17 and BW 37 (previously named HLA-13, 17, TY) have been shown by several research groups to occur in a statistically higher frequency among psoriatics than among controls (3, 10, 16, 23, 25, 29, 30). In those patients who have concomitant arthritis, B 27 (HLA-27) occurs more often (16).

All these antigens are alleles of the B locus and the associations are believed to be secondary to a hypothetical disease-predisposing gene in gametic association with the specific alleles of the B locus. By including the D allele we are able to study a larger part of the MHS-chromosome region. The genetic etiology of psoriasis has been debated for many years, and various modes of inheritance have

been proposed. Watson et al. 1972 (29) suggested that psoriasis is multifactorial in its inheritance, e.g. genes interact at different loci, possibly also influenced by environmental factors. However, this analysis was based mostly on questioning of the probands regarding the presence or absence of disease among close relatives. This investigation has been criticized by Kimberling (11) who has reevaluated earlier published data by Lomholt (15) and reached the conclusion that psoriasis possibly had an autosomal dominant pattern of inheritance with various degrees of penetration. Investigations performed with the HLA antigens as genetic markers by several authors, however, tended to support the concept that two or more genes are involved in the development of psoriasis (25, 30). Furthermore there is a higher familial aggregation of psoriasis where the proband is carrying HLA-B 13, B 17 and/or BW 37, compared with those not having these HLA antigens (25, 30). Individuals carrying these antigens also have an earlier age at onset than those lacking these antigens (25, 30). Thus it is possible that the etiology of disease differs between these two groups.

In our families A and B, two individuals (III:1, II:5) have psoriasis, 2 (I:2, I:3) have probably had psoriasis, and 2 suffer from joint pain (II:2, III:2). All these members share the A1, B 17, Da haplotype with the proband. The reason why we have included joint-affected persons in this study is that we know from other reports that arthritis can precede skin symptoms (19). We also know that arthritis which is associated with psoriasis often occurs in patients who have HLA-B 27 (16, 26), and we believe that this association is revealed by linkage disequilibrium between an HLA-B gene and a gene (or genes) increasing the susceptibility to develop disease. Furthermore, as has been pointed out by Kimberling (11), we do not know whether the cutaneous manifestations of psoriasis are the only phenotypic expressions of a single genotype. The relationship between arthritis and psoriasis on the basis of the HLA-region will be discussed further in a separate communication (17). Families A and B (Fig. 2*a*, *b*) are highly susceptible to penetration by disease, which might suggest a dominant mode of inheritance by one or several MHS-linked genes. In families A and B, including individual I:7 but excluding III:3 and the young children (IV:1 and 2) in the fourth generation, out of 7 individuals carrying the same A 1, B 17, Da haplotype all but one are (or have probably been) affected by either psoriasis or arthralgia. The only individual not affected with skin manifestations of psoriasis is I:7 (joints have not been examined, as mentioned earlier), the brother of the proband's grandmother. This person, incidentally, has two healthy children, but they have not been included in the study and there is thus no further information as to penetration of disease in this part of the family. However, he (I:7) and IV:1 must be at great risk of developing psoriasis.

In family C, psoriasis seemed to be linked to another haplotype A 1, BW 37, carrying the D-allele "1", now identified as DW 2 (Ld-7a) (28). In this family the disease pattern would also indicate a dominant mode of inheritance but with variable penetration. It is worth noting that psoriasis here is associated with the haplotype A 1, BW 37 from individual I:6. No increased incidence of psoriasis was known in her family. She married I:5 from family A with a high psoriasis incidence. He was not a carrier of the proband's A 1, B 17 haplotype but might still have inherited, through a recombinatorial event, the MHS linked "pso" gene. However, this is unlikely as can be seen from the second generation in this family. Individuals II:7 and II:12 who have psoriasis have not inherited the same paternal haplotype. There is no indication of psoriasis prevalence in the family history of individual II:9 married to II:8, whom we consider a possible carrier of the gene in question, as his child (III:6) has psoriasis and the haplotype A 1, BW 37, D 1. Therefore, most probably the MHS-linked disease-predisposing gene(s) was introduced into this family by I:6. Since she is homozygous for the complete MHS haplotype, including the HLA-D allele "1", now specified as DW2, it is impossible to tell which one of their children has inherited the disease-predisposing haplotype. There is no known consanguinity in her family—hence it is possible that only one of her MHS haplotypes carries the "pso" gene. Thus, this haplotype is most probably present in II:7, 8 and 12, but could be absent from II:10 and 11, which is compatible with a relatively high disease penetration. The child in the third generation (III:7) is only 8 years old and might of course still develop the disease. Unfortunately, we have not been able to investigate this part of the family for presence of arthritic lesions, especially for presence of sacroiliitis which can occur without clinical symptom (19).

Another interesting finding in this study is that two parents, I:3 and I:4, share the haplotype A 1, B 17. Psoriasis seems only to be linked to this haplotype in person I:3. The children (II:5 and II:6) have both inherited the A 1, B 17 haplotype, but from different parents. We found that the two haplotypes differ with respect to the D-alleles, shown in MLC experiment 2 (Table II). Of these two children, only the person who inherited the mother's haplotype, known to carry the "pso" gene, has developed disease. This implies that it is not the B 17 allele itself which is involved in the development of psoriasis, but rather a gene (or genes) situated in close proximity to the B region. However, the exact charting of a responsible gene in this region can only be made if recombinational events occur involving the "pso" gene but not all other known products of the MHS. It is not very likely that the A 1, B 17 haplotype of I:4 carries the "pso" gene, as he had not developed the disease by the age of 78, no psoriasis has occurred in his earlier family and his daughter who had inherited the same haplotype by the age of 39 has not developed the disease. However, when evaluating these results one must bear in mind that the occurrence of psoriasis in families is sometimes so emotionally loaded, especially in old people, that it is very difficult to obtain relevant information. But two examinations with a 6 month interval, and case reports from the district doctor, have not revealed the disease in individual I:4.

In conclusion, our results seem to favour a dominant mode of inheritance of the MHS-linked psoriasis-predisposing gene(s), as we found an unusually high incidence of the disease, albeit of varying clinical character, in many but not all carriers of the psoriasis associated MHS haplotype. However, whether the effect of other non-MHS linked genes is of importance in the development of psoriasis, as the results of other studies might indicate (3), cannot as yet be definitely established.

Furthermore, our data might support the view that it is not B 17 gene itself but gene(s) close to the B-locus which are responsible for the development of disease. However, in order to confirm these results more such families need to be investigated.

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