

Supplementary Material

Supplementary Table 1 Variables linked to cold agglutinins

Patient no.	Sex	CA test ^a	CA titer ^b	UCT score	Hemoglobin level (g/L)	Erythrocyte count (10 ¹² /L)	Monocyte count (10 ⁹ /L)	Thrombocyte count (10 ⁹ /L)	Mean daily Temp. (°C) ^c
1	F	Neg	0	13	141	4.61	1.01	340	0
2	M	Pos	1	5	137	4.44	0.38	271	17
3	F	Pos	2	3	132	4.51	0.35	468	25
4	M	Neg	0	16	149	4.81	0.57	236	18
5	F	Pos	4	3	146	4.85	0.50	209	17
6	F	Pos	4	7	140	4.24	0.57	330	13
7	M	Neg	0	11	147	4.66	0.61	257	15
8	F	Pos	1	12	138	4.58	0.37	275	21
9	F	Neg	0	6	154	4.80	0.60	266	4
10	M	Neg	0	16	159	4.92	0.91	237	16
11	F	Pos	2	12	146	4.74	0.64	318	21
12	F	Pos	4	14	139	4.32	0.41	203	22
13	M	Neg	0	9	154	5.22	0.50	371	21
14	F	Pos	1	7	130	4.02	0.05	301	18
15	F	Neg	0	6	130	4.19	0.79	272	2
16	M	Pos	2	13	143	4.99	0.71	365	21
17	M	Neg	0	15	150	5.13	0.61	282	6
18	M	Neg	0	16	159	5.43	0.56	259	17
19	F	Neg	0	15	135	4.37	0.69	269	15
20	F	Pos	2	9	134	4.17	0.58	292	13
21	F	Pos	4	11	120	4.27	0.20	274	22
22	F	Neg	0	16	145	4.42	0.69	222	2
23	F	Pos	16	12	145	5.06	0.37	335	10
24	F	Neg	0	14	151	4.73	0.53	170	2
25	F	Neg	0	10	146	4.76	0.38	197	10
26	F	Neg	0	16	142	4.43	0.53	220	13
27	M	Neg	0	16	162	6.02	0.53	150	8
28	F	Pos	1	12	134	4.69	0.40	285	0
29	F	Pos	2	7	123	4.44	0.33	248	10
30	M	Neg	0	13	146	4.98	0.35	208	13
31	F	Pos	2	10	128	3.98	0.33	228	13
32	M	Neg	0	13	149	4.90	0.41	222	7
33	M	Neg	0	5	161	5.27	0.47	271	5
34	F	Pos	1	11	137	4.22	0.30	222	7
35	F	Neg	0	4	139	4.69	0.52	233	2

^aCold agglutinin test performed at 4°C. ^bThe numbers are inverse of the maximum serum dilution at which agglutination can be seen (i.e., CA titer 4 = agglutination at 1:4 dilution). ^cMean daily temperature on the day when blood was drawn was obtained from the national weather service website (<http://meteo.arso.gov.si/met/en/>). *Abbreviations:* CA, cold agglutinin; F, female; M, male; Neg, negative; Pos, positive; Temp., temperature; UCT, Urticaria Control Test.

Supplementary Table 2 Variables linked to cryoglobulins

Patient no.	CG test	CG conc. (mg/L)	CG Ig isotype(s)	ColdU duration (months)	Lymphocyte count (10 ⁹ /L)	Basal serum tryptase level (ng/mL)
1	Pos	137	IgG, IgM, IgA	70	2.13	3.66
2	Neg	—	—	14	1.55	3.24
3	Neg	—	—	24	2.58	3.41
4	Neg	—	—	60	2.49	11.60
5	Neg	—	—	336	1.39	8.15
6	Neg	—	—	9	1.22	15.90
7	Neg	—	—	4	1.94	5.34
8	Neg	—	—	50	1.63	5.32
9	Pos	189	IgG	169	2.89	7.57
10	Neg	—	—	120	1.52	6.19
11	Neg	—	—	228	2.49	23.00
12	Neg	—	—	156	2.68	3.57
13	Neg	—	—	60	2.44	ND
14	Neg	—	—	99	1.94	5.35
15	Pos	124	IgG, IgM	24	1.88	3.83
16	Neg	—	—	24	1.40	5.50
17	Neg	—	—	240	1.99	7.79
18	Neg	—	—	10	1.34	4.44
19	Pos	184	IgG, IgM	252	2.67	13.60
20	Neg	—	—	276	1.70	5.49
21	Neg	—	—	72	1.20	3.96
22	Neg	—	—	120	2.61	6.37
23	Pos	153	IgG, IgM, IgA	216	2.62	5.55
24	Neg	—	—	384	1.80	7.41
25	Neg	—	—	2	1.13	6.00
26	Neg	—	—	22	2.06	4.55
27	Neg	—	—	15	2.01	12.70
28	Pos	187	IgG, IgM	84	1.72	6.09
29	Neg	—	—	8	1.75	3.05
30	Pos	106	IgG, IgM	60	1.22	2.92
31	Neg	—	—	42	1.95	5.10
32	ND	—	—	8	1.47	2.90
33	Pos	138	IgG, IgM	72	1.49	6.06
34	ND	—	—	22	2.17	5.33
35	Pos	150	IgG	11	2.12	14.00

Abbreviations: conc., concentration; CG(s), cryoglobulin(s); CRP, C-reactive protein; Ig, immunoglobulin; IgA, immunoglobulin A; IgG, immunoglobulin G; IgM, immunoglobulin M; Neg, negative; ND, not done/determined; Pos, positive.

Supplementary Table 3 Characteristics of patients with a positive vs. negative cryoglobulin test

Parameter	CG test			P value (positive vs. negative)
	Total N = 33	Negative N = 24	Positive N = 9	
Female sex	22 (67)	15 (63)	7 (78)	0.681
Triggers				
Cold ambient air	27 (82)	20 (83)	7 (78)	1.000
Immersion in < 25°C water	18 (55)	13 (54)	5 (56)	0.627
Higher summer humidity levels	15 (46)	11 (46)	4 (44)	1.000
Ingestion of cold foods/drinks	18 (55)	14 (58)	4 (44)	0.697
UCT score	12 (7–15)	12 (8–16)	12 (6–13)	0.310
CST results				
Positive ice cube test	20 (61)	16 (67)	4 (44)	0.425
Positive TempTest®	12 (36)	11 (46)	1 (11)	0.107
CSTT (s)	240 (30–300); N = 18	150 (30–300); N = 14	300 (165–300); N = 4	0.182
CTT (°C)	19 (15–25); N = 12	21 (16–25); N = 11	14; N = 1	0.242
Laboratory findings				
Erythrocyte count (10 ¹² /L)	4.7 ± 0.4	4.7 ± 0.5	4.7 ± 0.3	0.685
Hemoglobin concentration (g/L)	142.6 ± 10.6	142.5 ± 11.0	142.8 ± 10.0	0.948
Hematocrit level (%)	41.7 ± 3.4	41.6 ± 3.4	41.9 ± 3.5	0.828
Thrombocyte count (10 ⁹ /L)	268.6 ± 63.2	266.0 ± 70.1	275.4 ± 42.3	0.708
Monocyte count (10 ⁹ /L)	0.5 ± 0.2	0.5 ± 0.2	0.6 ± 0.2	0.266
Leukocyte count (10 ⁹ /L)	7.0 ± 1.6	6.8 ± 1.5	7.5 ± 2.0	0.269
Neutrophil count (10 ⁹ /L)	4.3 ± 1.2	4.2 ± 1.2	4.4 ± 1.5	0.639
Lymphocyte count (10 ⁹ /L)	1.9 ± 0.5	1.9 ± 0.5	2.1 ± 0.6	0.285
Eosinophil count (10 ⁹ /L)	0.2 ± 0.3	0.2 ± 0.1	0.3 ± 0.4	0.329
Basophil count (10 ⁹ /L)	0.04 ± 0.03	0.04 ± 0.03	0.05 ± 0.02	0.094
CRP (mg/L)	1.7 (0.5–3.5)	1.5 (0.6–3.5)	2.0 (0.5–4.8)	0.984
Basal serum tryptase (ng/mL)	5.5 (4.1–7.7); N = 32	5.5 (4.4–7.8); N = 23	6.1 (3.7–10.6)	0.850
Total IgE (IU/mL)	113 (47–232); N = 29	94 (30–232); N = 21	143 (62–1101); N = 8	0.341
Mean daily Temp. on the day when blood was drawn	11.1 ± 7.6	12.4 ± 6.6	6.9 ± 9.4	0.077

Data are given as no. (%), mean ± SD, and median (IQR). If data was not obtained in all patients, patient numbers are displayed as “N” next to results. Statistical significance of differences between patient groups was calculated by Fisher’s Exact test, Mann-Whitney U test, and Independent-samples T test. No statistically significant P values were found. *Abbreviations:* CG, cryoglobulin; ColdU, cold urticaria; CR(s), cold-induced reaction(s); CRP, C-reactive protein; CST, cold stimulation test; CSTT, critical stimulation time threshold; CTT, critical temperature threshold; IQR, interquartile range; N, number of patients; s, second; SD, standard deviation; UCT, Urticaria Control Test.