DOMANDE prova orale del 25/11/2022
Allegato 1 al verbale n. 4

1) Descrivi le tecniche di isolamento per l'allestimento di colture cellulari primarie
2) Panel Citofluorimetrico: descrizione e commento
3) Quale pacchetto office utilizzeresti per la preparazione di un poster o di una presentazione

1) Descrivi la tecnica ELISA
2) Panel Citofluorimetrico: descrizione e commento
3) Quale programma del pacchetto office permette di fare un'analisi statistica e quali altri programmi conosci al di fuori del pacchetto office

1) Descrivi la tecnica di separazione e isolamento dei linfociti B
2) Panel Citofluorimetrico: descrizione e commento
3) Cosa si intende con il termine "giustificato" in un documento word

LATO APRIBILE PER L'ISPEZIONE POSTALE

LATO APRIBILE PER L'ISPEZIONE POSTALE

Autoimmune chronic spontaneous urticaria: What we know and what we do not know

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Chronic spontaneous urticaria (CSU) is a mast cell–driven skin disease characterized by the recurrence of transient wheals, angioedema, or both for more than 6 weeks. Autoimmunity is thought to be one of the most frequent causes of CSU. Type I and II autoimmunity (ie, IgE to autoallergens and IgG autoantibodies to IgE or its receptor, respectively) have been implicated in the etiology and pathogenesis of CSU. We analyzed the relevant literature and assessed the existing evidence in support of a role for type I and II autoimmunity in CSU with the help of Hill's criteria of causality. For each of these criteria (ie, strength of association, consistency, specificity, temporality, biological gradient, plausibility, coherence, experiment, and analogy), we categorized the strength of evidence as "insufficient," "low," "moderate," or "high" and then assigned levels of causality for type I and II autoimmunity in patients with CSU from level 1 (causal relationship) to level 5 (causality not likely). Based on the evidence in support of Hill's criteria, type I autoimmunity in patients with CSU has level 3 causality (causal relationship suggested), and type II autoimmunity has level 2 causality (causal relationship likely). There are still many aspects of the pathologic mechanisms of CSU that need to be resolved, but it is becoming clear that there are at least 2 distinct pathways, type I and type II autoimmunity, that contribute to the pathogenesis of this complex disease. (J Allergy Clin Immunol 2017;139:1772-81.)

Key words: *Chronic spontaneous urticaria, autoimmunity, IgE–anti-self, IgG–anti-FcεRI/IgE, causality, Hill's criteria of causality*

Abbreviations used
AAb: Autoantibody
ASST: Autologous serum skin test
BAT: Basophil activation test
BP: Bullous pemphigoid
CSU: Chronic spontaneous urticaria
dsDNA: Double-stranded DNA
SLE: Systemic lupus erythematosus
TPO: Thyroperoxidase

Chronic spontaneous urticaria (CSU) is a mast cell–driven skin disease characterized by the recurrence of transient wheals (hives), angioedema, or both for more than 6 weeks.[1] Several mechanisms have been investigated as possibly contributing to the pathogenesis of CSU, including infections, food intolerance, coagulation cascade, genetic factors, and autoimmunity.[1] Autoimmunity (ie, autoimmune mechanisms of skin mast cell activation) is held to be a frequent underlying cause of CSU. Two types of Gell and Coombs hypersensitivity reactions[2] have been postulated to be relevant in patients with autoimmune CSU.

A type I hypersensitivity to self, also called autoallergy, in which antigens crosslink the IgE on mast cells and basophils to cause release of vasoactive mediators (Fig 1), was first suggested by Rorsman[3] in 1962 to explain urticaria-associated basopenia. A role of autoallergy in urticaria was also postulated from the finding in 1999 of IgE autoantibodies (AAbs) against the thyroid microsomal antigen in the serum of a female patient with CSU.[4] This work has been confirmed and extended to propose autoallergy in the pathogenesis of both CSU and chronic inducible urticaria.[5-10]

A Type II hypersensitivity reaction in which antibodies, usually IgG or IgM, bind to antigen on a target cell (Fig 1) was originally postulated after the identification of IgG-AAbs against IgE in 3 of 6 patients with CSU.[11] The presence of these AAbs was confirmed by Grattan et al[12] in 1991 in patients whose sera induced a wheal-and-flare response when injected intradermally: the autologous serum skin test (ASST). The presence of AAbs to the high-affinity receptor for IgE on mast cells and basophils (IgG–anti-FcεRI) in a subset of patients with CSU was reported by the same group 2 years later.[13] In theory, IgG–anti-FcεRII/CD23 AAbs that were identified in sera of patients with CSU can also elicit mast cell degranulation through activation of eosinophils, with the consequent release of major basic protein and other mast cell secretagogues (Fig 1).[14]

We assessed the evidence for a role of these 2 forms of autoimmunity in patients with CSU using Hill's 9 criteria of

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TEMA 2

☐ URGENTE

COGNOME				NOME	Sesso ☐ F ☐ M	DATA ESAME
Data di nascita	gg	mm	aa	Data raccolta campione		ora arrivo
Numero accettazione esterna				Numero accettazione interna		Numero accettazione striscio midollare
ESTERNO ☐	Esenzione Sì ☐ No ☐			RICOVERATO ☐	Medico Referente	Struttura inviante

MOTIVO RICHIESTA/DIAGNOSI ______

Avvisato: ______ **da:** ______ **ore:** ______ **data:** ______

ESAMI RICHIESTI/APPROFONDIMENTI

- ☐ Immunofenotipo base
- ☐ Kappa/Lambda
- ☐ Linfoprol B
- ☐ ZAP-70
- ☐ CD38
- ☐ CD200
- ☐ CD49d
- ☐ Mieloma multiplo A
- ☐ Mieloma multiplo B
- ☐ Linfoprol NKTGD
- ☐ Linfoprol G/D
- ☐ Linfoprol T
- ☐ IF repertorio Vbeta
- ☐ MDS/LMC
- ☐ ALOT
- ☐ linfoblastica B
- ☐ linfoblastica T
- ☐ IF Liquor
- ☐ prolif. linf. PHA
- ☐ attività NK
- ☐ attività LAK
- ☐ CD40L
- ☐ Clone EPN
- ☐ Euroclass
- ☐ Rival linfoblastica B
- ☐ Rival linfoblastica T
- ☐ BAL
- ☐ clonalità T catena gamma
- ☐ clonalità B IgVH
- ☐ somatic hypermutation SHM
- ☐ repertorio TCR Vbeta Mol
- ☐ MRD CLL
- ☐ monociti classici e non
- ☐ ______
- ☐ ______
- ☐ ______

MATERIALE	quantità cc	WBC	Linf. %	Mono %	Neut. %	Eos. %	Bas. %	Cell.ottenute
☒ Sangue Periferico	20	2730 /mm³	60					26.4 · 10⁶
☐ Sangue Midollare								
☐ BAL Broncoscopista:		/ml						
☐ Altro								

IMMUNOFENOTIPO Ly 65%

4	8	20	0	73	CD3⁺ CD16⁻	32%
8	DR	72	1	5	CD3⁻ CD16⁺	1%
19	5	5	0	27	CD3⁺ CD16⁺	61%
16	3	1	62	30		
57	56	44	1	2	ESEGUITO	PANNELLO Vβ
57	3	0	44	49	Vβ 5.1	56
56	3	1	1	92		
γδ	3	0	1	92		
56	16	2	1	62		
57	16	4	41	23		

CONGELAMENTO CELLULE Vials 2

Operatore pre-analisi ______ Operatore 2° controllo pre-analisi ______ Citofluorimetrista ______

Operatore 2° controllo analisi ______ Validatore O3: ______ Compilatore scheda ______

TEMA 1

☐ **URGENTE**

COGNOME				NOME	Sesso ☐ F ☐ M	DATA ESAME
Data di nascita	gg	mm	aa	Data raccolta campione		ora arrivo
Numero accettazione esterna				Numero accettazione interna		Numero accettazione striscio midollare
ESTERNO ☐	Esenzione Sì ☐ No ☐			RICOVERATO ☐	Medico Referente	Struttura inviante

MOTIVO RICHIESTA/DIAGNOSI ______________________________

Avvisato: ____________________ **da:** ____________________ **ore:** __________ **data:** __________

ESAMI RICHIESTI/APPROFONDIMENTI

☐ Immunofenotipo base	☐ Linfoprol NKTGD	☐ prolif. linf. PHA	☐ clonalità T catena gamma
☐ Kappa/Lambda	☐ Linfoprol G/D	☐ attività NK	☐ clonalità B IgVH
☐ Linfoprol B	☐ Linfoprol T	☐ attività LAK	☐ somatic hypermutation SHM
☐ ZAP-70	☐ IF repertorio Vbeta	☐ CD40L	☐ repertorio TCR Vbeta Mol
☐ CD38	☐ MDS/LMC	☐ Clone EPN	☐ MRD CLL
☐ CD200	☐ ALOT	☐ Euroclass	☐ monociti classici e non
☐ CD49d	☐ linfoblastica B	☐ Rival linfoblastica B	☐ ________
☐ Mieloma multiplo A	☐ linfoblastica T	☐ Rival linfoblastica T	☐ ________
☐ Mieloma multiplo B	☐ IF Liquor	☐ BAL	☐ ________

MATERIALE	quantità cc	WBC	Linf. %	Mono %	Neut. %	Eos. %	Bas. %	Cell.ottenute
☒ Sangue Periferico	20	2500 /mm³	36					$23.1 \cdot 10^6$
☐ Sangue Midollare								
☐ BAL Broncoscopista:		/ml						
☐ Altro								

IMMUNOFENOTIPO Ly 35%

4	8	35	2	49
8	DR	48	4	10
19	5	9	0	85
16	3	3	2	80
57	56	26	7	2
57	3	2	31	51
56	3	3	5	77
γδ	3	0	4	79
57	16	5	3	3
56	16	30	3	1

ANALISI DELLE CELLULE CD19+ PARI AL 3.4% DEL TOT.

K	λ	41	57

LyB NAIVE	80.3%
" MARGINAL	16.1%
" MEMORY	1.8%
" ACTIVATED	6.2%
" TRANSITIONAL	0.7%
" PLASMOBLASTI	0.5%

CONGELAMENTO CELLULE ______________________________

Operatore pre-analisi ____________ Operatore 2° controllo pre-analisi ____________ Citofluorimetrista ____________

Operatore 2° controllo analisi ____________ Validatore O3: ____________ Compilatore scheda ____________

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Deciphering imprints of impaired memory B-cell maturation in germinal centers of three patients with common variable immunodeficiency

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Common variable immunodeficiency (CVID), characterized by recurrent infections, low serum class-switched immunoglobulin isotypes, and poor antigen-specific antibody responses, comprises a heterogeneous patient population in terms of clinical presentation and underlying etiology. The diagnosis is regularly associated with a severe decrease of germinal center (GC)-derived B-cell populations in peripheral blood. However, data from B-cell differentiation within GC is limited. We present a multiplex approach combining histology, flow cytometry, and B-cell receptor repertoire analysis of sorted GC B-cell populations allowing the modeling of distinct disturbances in GCs of three CVID patients. Our results reflect pathophysiological heterogeneity underlying the reduced circulating pool of post-GC memory B cells and plasmablasts in the three patients. In patient 1, quantitative and qualitative B-cell development in GCs is relatively normal. In patient 2, irregularly shaped GCs are associated with reduced somatic hypermutation (SHM), antigen selection, and class-switching, while in patient 3, high SHM, impaired antigen selection, and class-switching with large single clones imply increased re-cycling of cells within the irregularly shaped GCs. In the lymph nodes of patients 2 and 3, only limited

TEMA 5

☐ **URGENTE**

COGNOME				NOME		Sesso ☐ F ☐ M	DATA ESAME
Data di nascita	gg	mm	aa	Data raccolta campione			ora arrivo
Numero accettazione esterna				Numero accettazione interna			Numero accettazione striscio midollare
ESTERNO ☐	Esenzione Si ☐ No ☐			RICOVERATO ☐	Medico Referente		Struttura inviante

MOTIVO RICHIESTA/DIAGNOSI _____

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ESAMI RICHIESTI/APPROFONDIMENTI

☐ Immunofenotipo base	☐ Linfoprol NKTGD	☐ prolif. linf. PHA	☐ clonalità T catena gamma
☐ Kappa/Lambda	☐ Linfoprol G/D	☐ attività NK	☐ clonalità B IgVH
☐ Linfoprol B	☐ Linfoprol T	☐ attività LAK	☐ somatic hypermutation SHM
☐ ZAP-70	☐ IF repertorio Vbeta	☐ CD40L	☐ repertorio TCR Vbeta Mol
☐ CD38	☐ MDS/LMC	☐ Clone EPN	☐ MRD CLL
☐ CD200	☐ ALOT	☐ Euroclass	☐ monociti classici e non
☐ CD49d	☐ linfoblastica B	☐ Rival linfoblastica B	☐ _____
☐ Mieloma multiplo A	☐ linfoblastica T	☐ Rival linfoblastica T	☐ _____
☐ Mieloma multiplo B	☐ IF Liquor	☐ BAL	☐ _____

MATERIALE		quantità cc	WBC	Linf. %	Mono %	Neut. %	Eos. %	Bas. %	Cell.ottenute
☒	Sangue Periferico	20	3610 /mm³	37					$50.2 \cdot 10^6$
☐	Sangue Midollare								
☐	BAL Broncoscopista:		/ml						
☐	Altro								

IMMUNOFENOTIPO Ly 36%.

4	8
8	DR
19	5
16	3
57	56
57	3
56	3
38	3
56	16
57	16

39	2	44
42	3	13
10	0	82
6	2	81
22	13	3
5	30	53
6	11	72
0	3	80
9	8	2
29	6	2

ANALISI DELLE CELLULE		
CD 19⁺ PARI AL 3.4%.		
DEL TOT.		
K λ	46	52
LyB NAIVE		68.9%.
" MARGINAL		26.4%.
" MEMORY		1.8%.
" ACTIVATED		10.7%.
" TRANSITIONAL		0.4%.
" PLASMOBLASTI		0.1%.

CONGELAMENTO CELLULE _____

Operatore pre-analisi _____ Operatore 2° controllo pre-analisi _____ Citofluorimetrista _____

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Current clinical spectrum of common variable immunodeficiency in Spain: The multicentric nationwide GTEM-SEMI-CVID registry

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Common variable immunodeficiency (CVID) constitutes a heterogenic group of primary immunodeficiency disorders with a wide-ranging clinical spectrum. CVID-associated non-infectious morbidity constitutes a major challenge requiring a full understanding of its pathophysiology and its clinical importance and global variability, especially considering the broad clinical, genetic, and regional heterogeneity of CVID disorders. This work aimed to