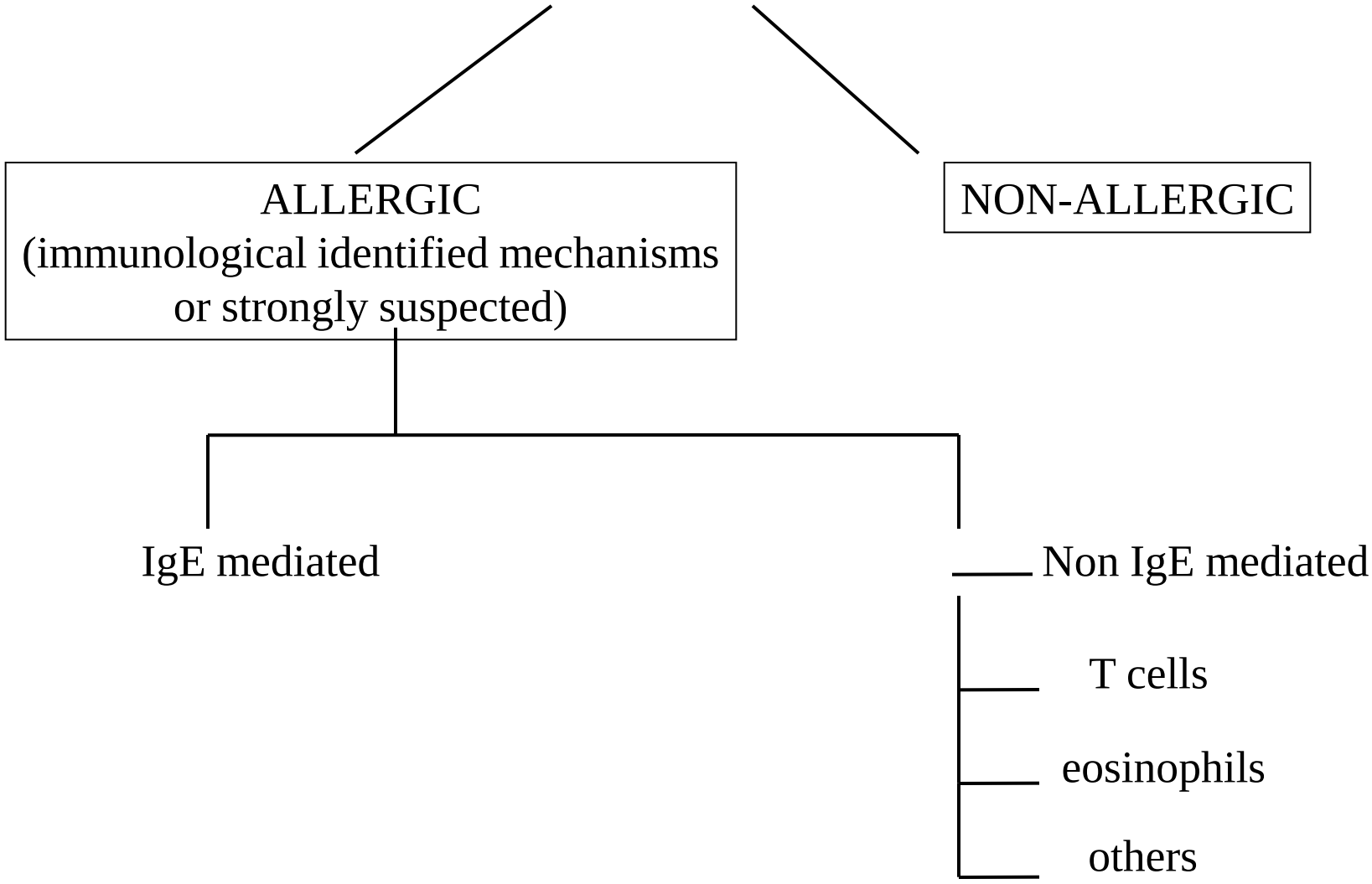
Allergia alimentare: clinica, diagnosi e terapia

Riccardo Troncone

Food hypersensitivity



EAACI Position Paper, Johansson SGO et al, 2001

Diversity of clinical manifestations and mechanisms

IgE mediated (acute onset)

- Anaphylaxis
- Urticaria angioedema
- Acure rhinoconjunctivitis, bronchospasm (wheezing)

Cell mediated (delayed onset – chronic)

- Dietary protein enterocolitis
- Dietary protein proctitis
- Pulmonary hemosiderosis (Heiner syndrome)

Mixed IgE and non IgE (delayed onset – chronic)

- Eosinophilic gastroenteropathies
- Atopic dermatitis
- Asthma

Epidemiology of food allergy

- Rising incidence in the last decade
- Children 4-6%

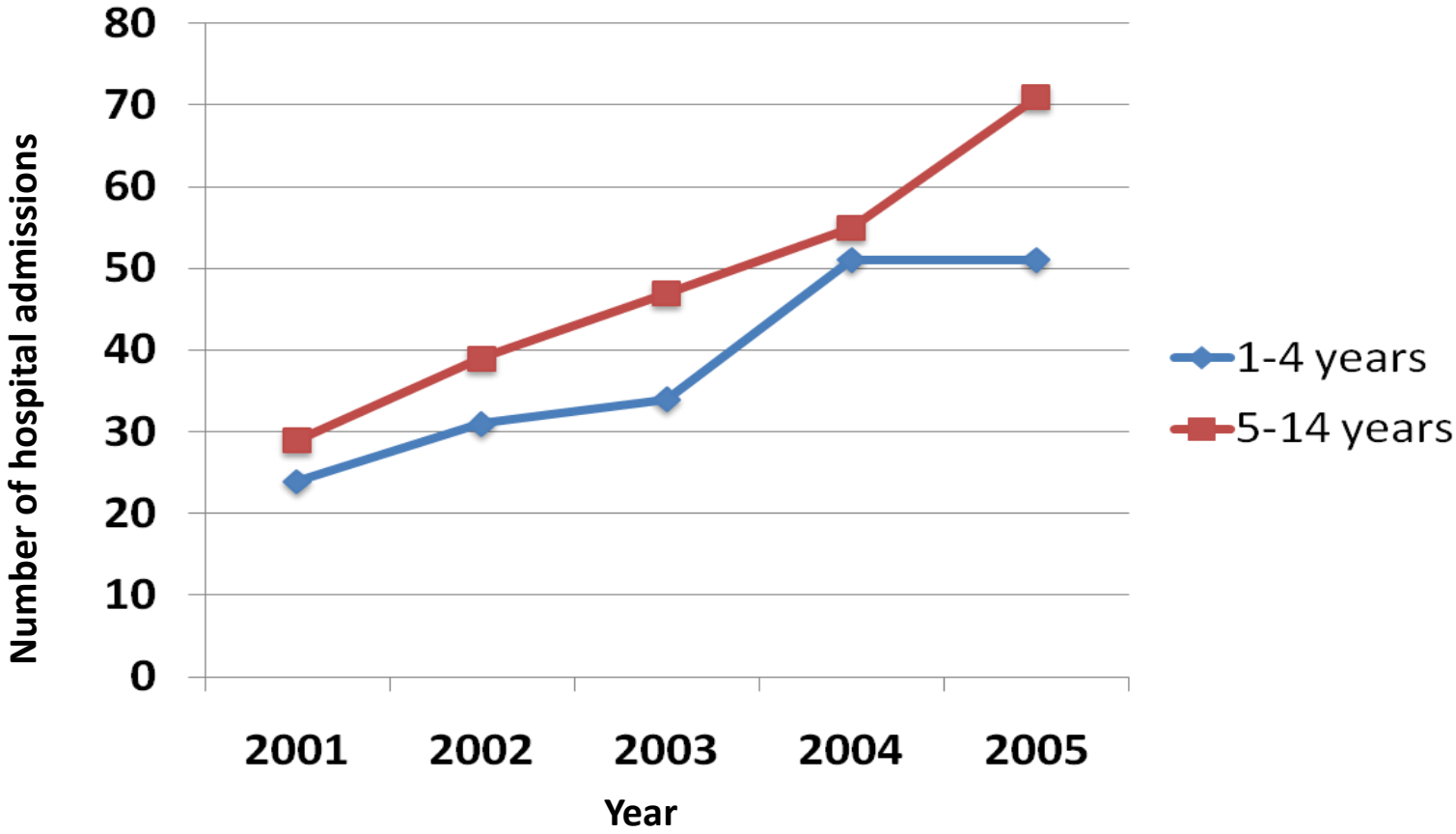
- *In the Italian pediatric practice:*

✓Family pediatricians: 44

✓Total patients enrolled: 41.958

✓Food allergy prevalence: 0.8% (age less than 3 years: 3.2%)

Food anaphylaxis in Italy



42% CMA

Ministry of Health, Health planning, essential levels of care and ethical principles of the system - Office VI.
http://www.salute.gov.it/ricoveriOspedalieri/ric_informazioni/sceltadia.jsp.

Berni Canani R et al. Clin Exp Allergy 2012, in press

Presentazioni a livello del tratto GI

IgE mediate

- Sindrome orale allergica
- Anafilassi

non-IgE mediate

- Enteropatia
- Enterocolite
- Proctite
- Esofagite eosinofila ed altre sindromi ipereosinofiliche
- Disordini della motilità (reflusso gastroesofageo, stipsi)

Allergie alimentari: sindromi cliniche non IgE-mediate

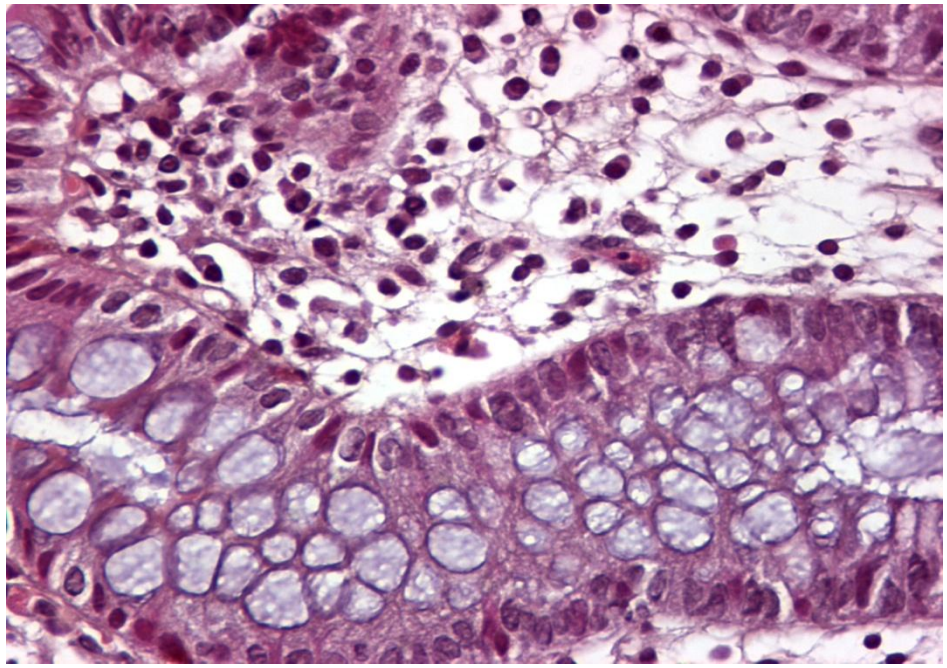
	Enteropatia	***Enterocolite***
Età (mesi)	1-18	1-6
Durata (mesi)	18-36	9-36
Presentazione	indolente	acuta
Ritardo di crescita	moderato/severo	moderato
Irritabilità	lieve	moderata
Vomito	lieve	moderato
Diarrea	moderata	severa
Sanguinamento rettale	minimo/assente	moderato
Leucociti fecali	assenti	presenti
Anemia	moderata	moderata
Protidodispersione	moderata	moderata
Alimenti responsabili	latte,soia,grano uovo,pesce	latte,soia idrolizzati

Proctocolite come manifestazione di allergia alimentare nel lattante

- Nel 50% dei casi allattati al seno, ma sono responsabili anche latte vaccino, soia, formula idrolizzata
- Inizio: 2-6 settimane
- Crescita soddisfacente
- No vomito, no distensione addominale
- Sangue nelle feci non prominente
- Raramente anemia clinicamente importante

Colite allergica

Aumento della densità di eosinofili nella lamina propria della mucosa intestinale (EE)



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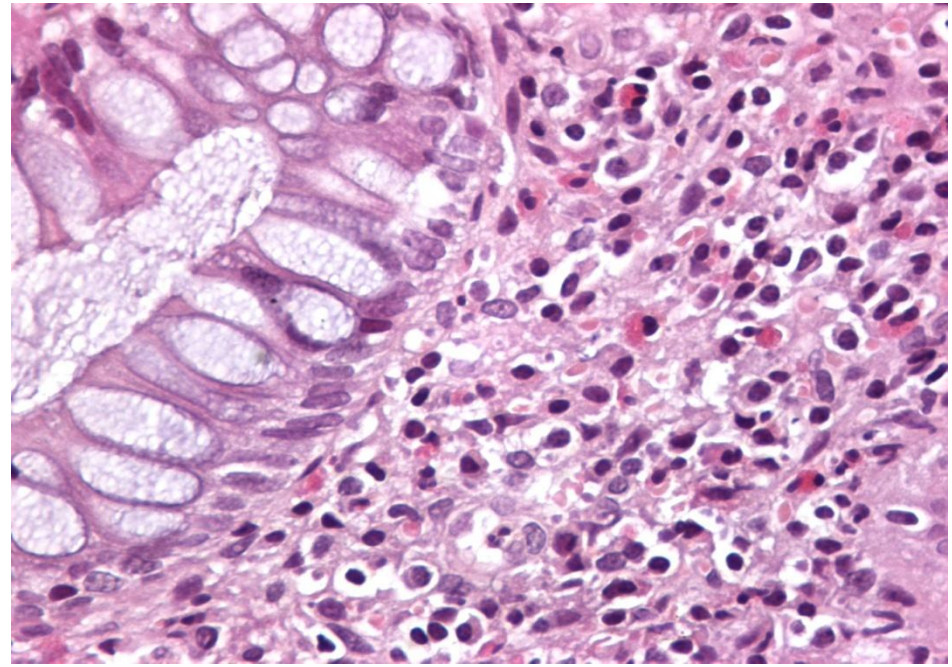
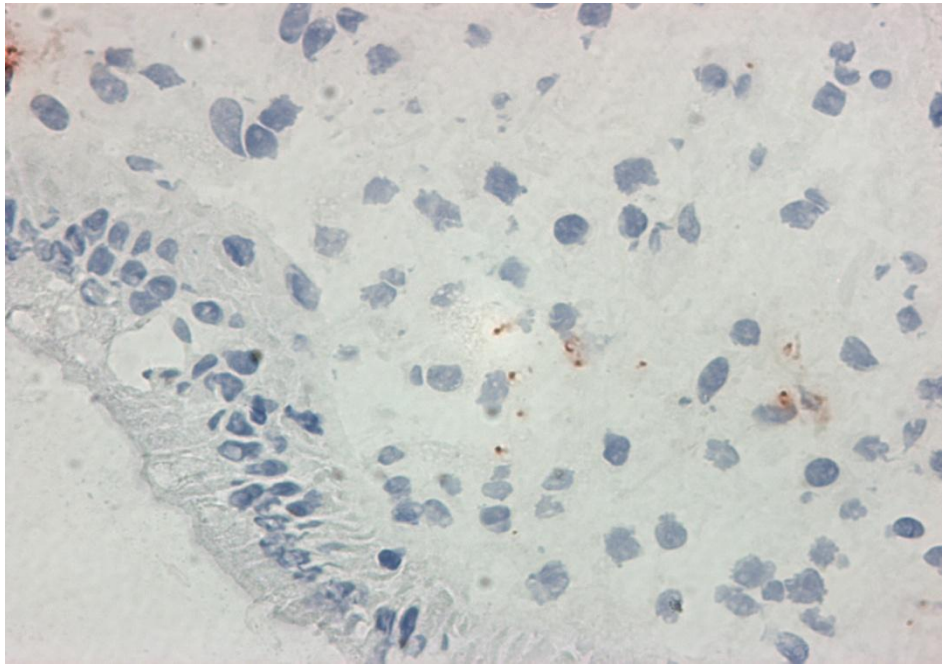


Colite allergica (400X)

Colite allergica

Aumento della densità di eosinofili nella lamina propria della mucosa intestinale (MBP-Major basic protein)



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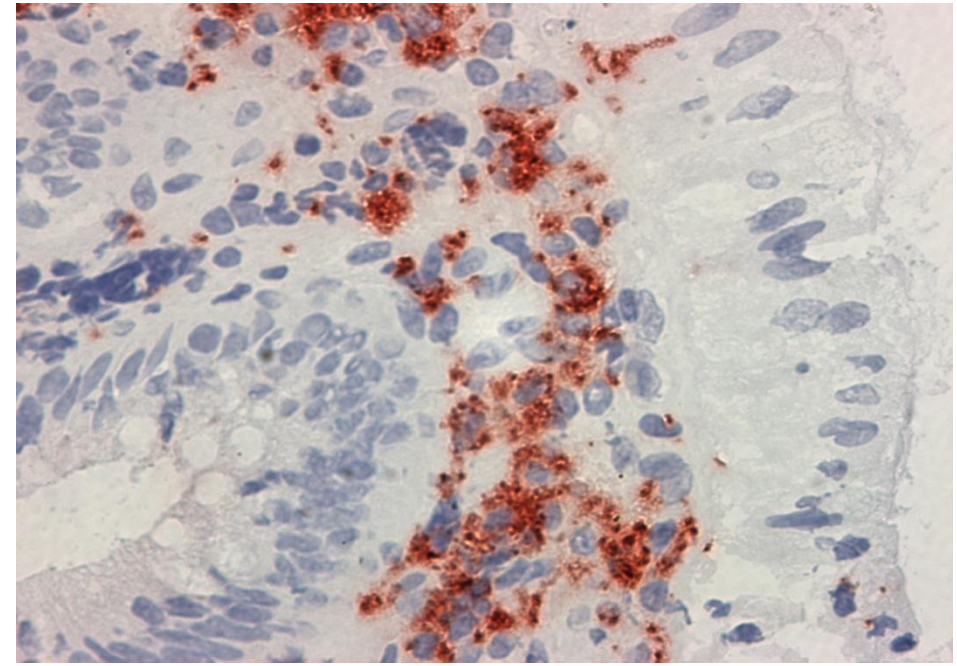
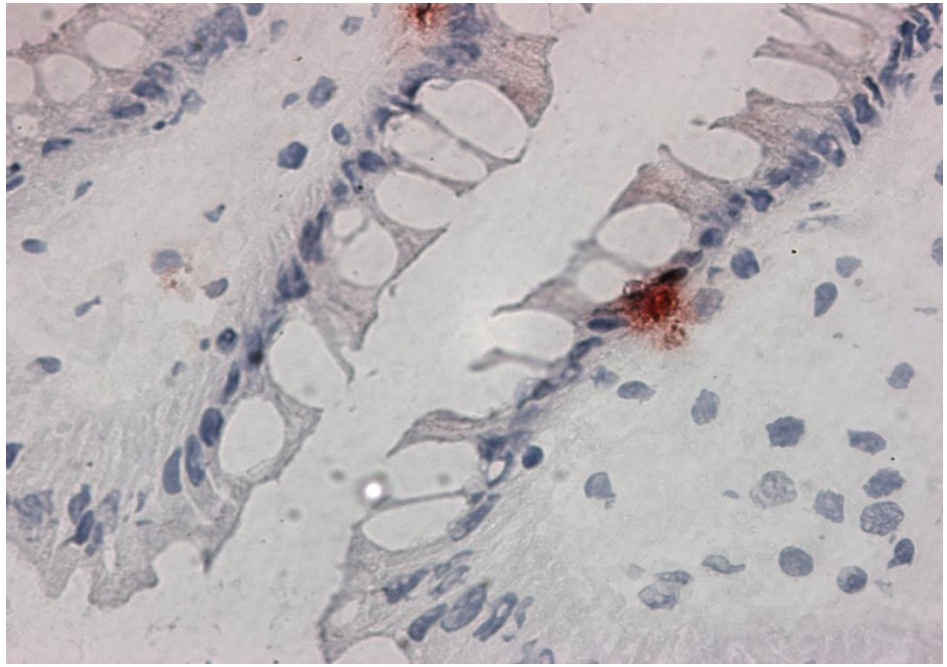


Colite allergica (400X)

Colite allergica

Aumento della densità di mastociti nella lamina propria della mucosa intestinale (triptase)



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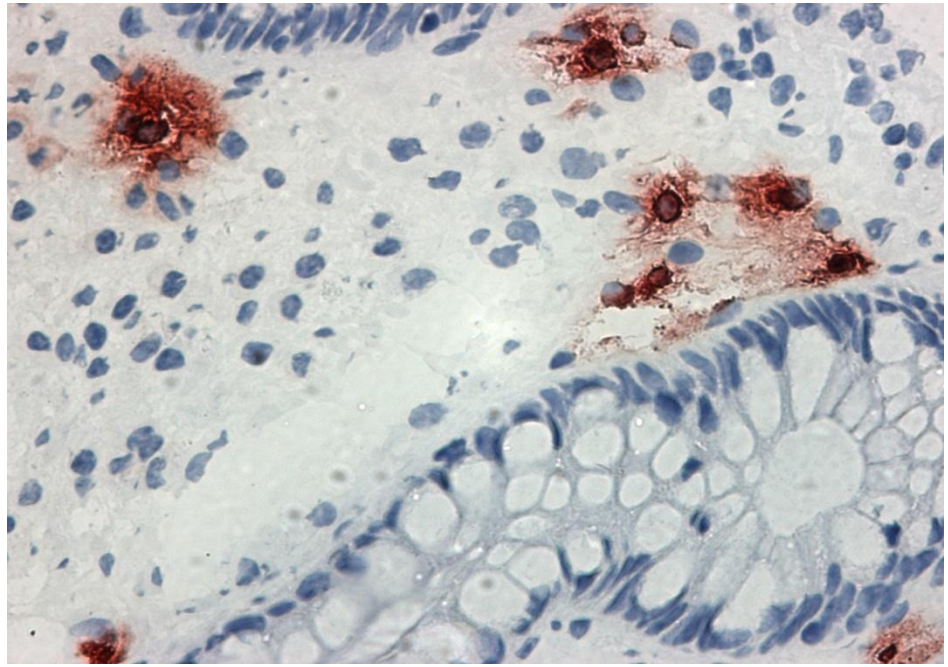
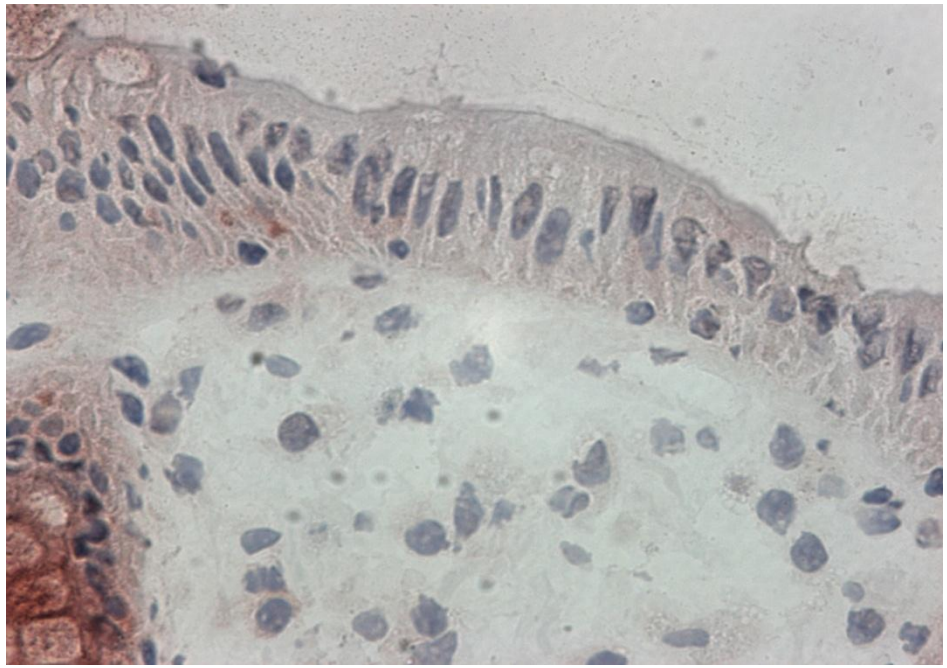


Colite allergica (400X)

Colite allergica

Aumento della espressione di eotassina nella mucosa intestinale



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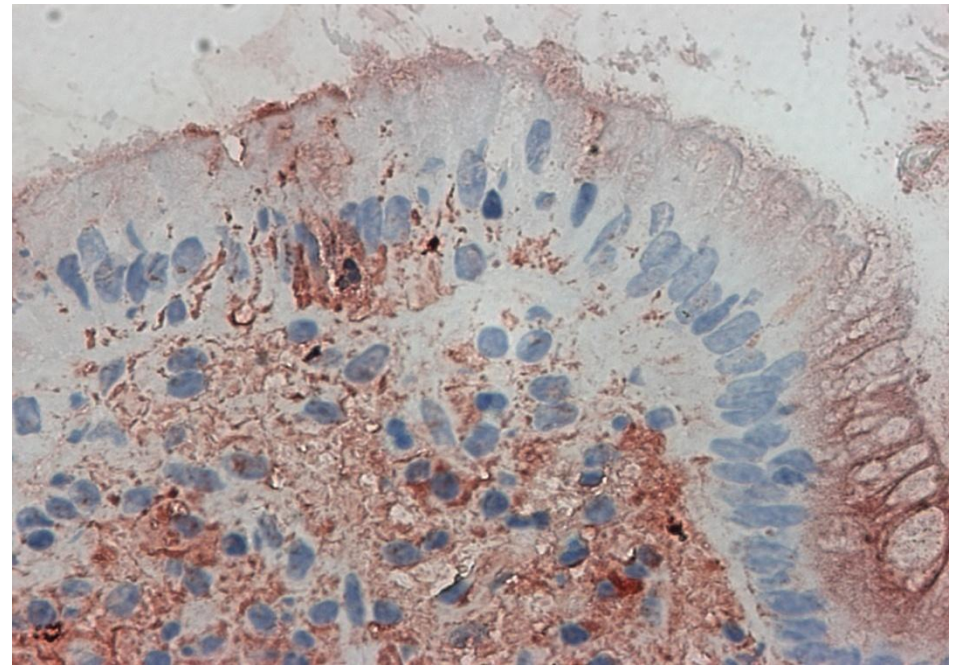
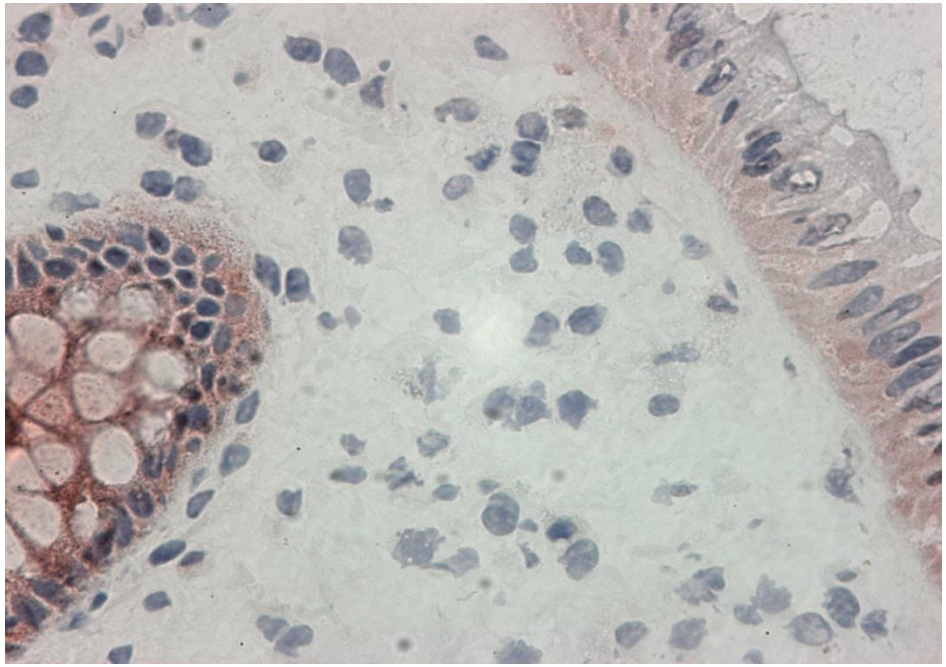


Colite allergica (400X)

Colite allergica

Aumento della espressione di CCR3 nella mucosa intestinale



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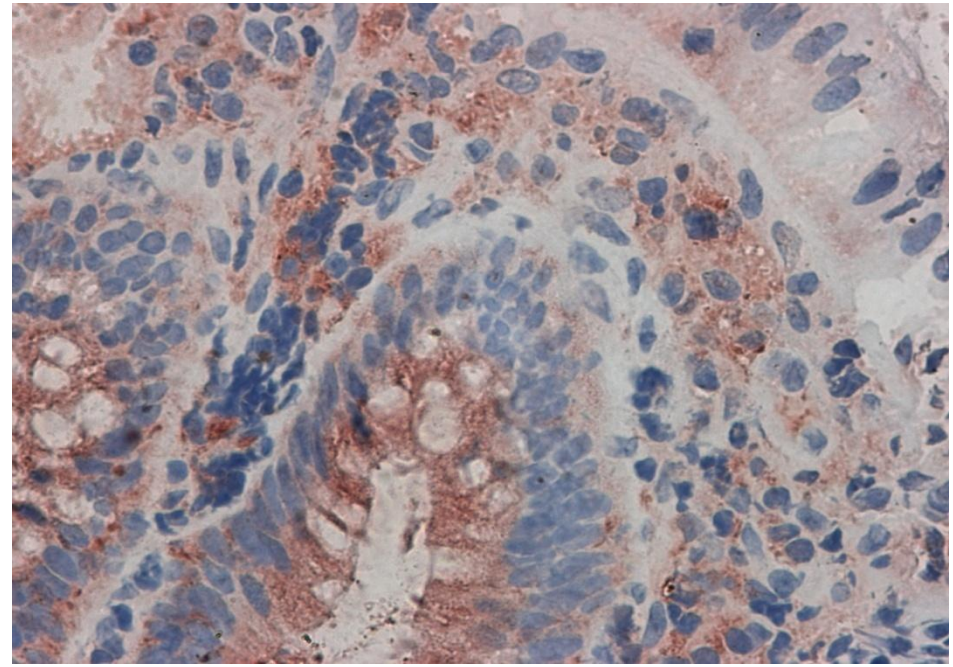


Colite allergica (400X)

Necessità di rigore diagnostico (challenge) per evitare overdiagnosi di colite allergica

84% dei pediatri americani tratta il sanguinamento rettale del lattante con dieta di eliminazione. Tuttavia solo il 14% di questi lattanti riceve una diagnosi finale di allergia al latte (Xanthacos, JPGN 2005)

•Anche nei casi severi (diagnosi finale MICI) spesso la prima diagnosi è allergia al latte (Cannioto, Eur J Pediatr 2009).

•Uno studio finlandese basato su randomizzazione suggerisce che nella maggioranza dei casi di sanguinamento rettale la dieta non ha effetto. Questa è una condizione benigna autolimitantesi (Arvola, Pediatrics 2006)

La proctite allergica non è una condizione confinata all'età del lattante

Bambini (2-14 anni) in buone condizioni generali

Persistente o ricorrente sanguinamento rettale

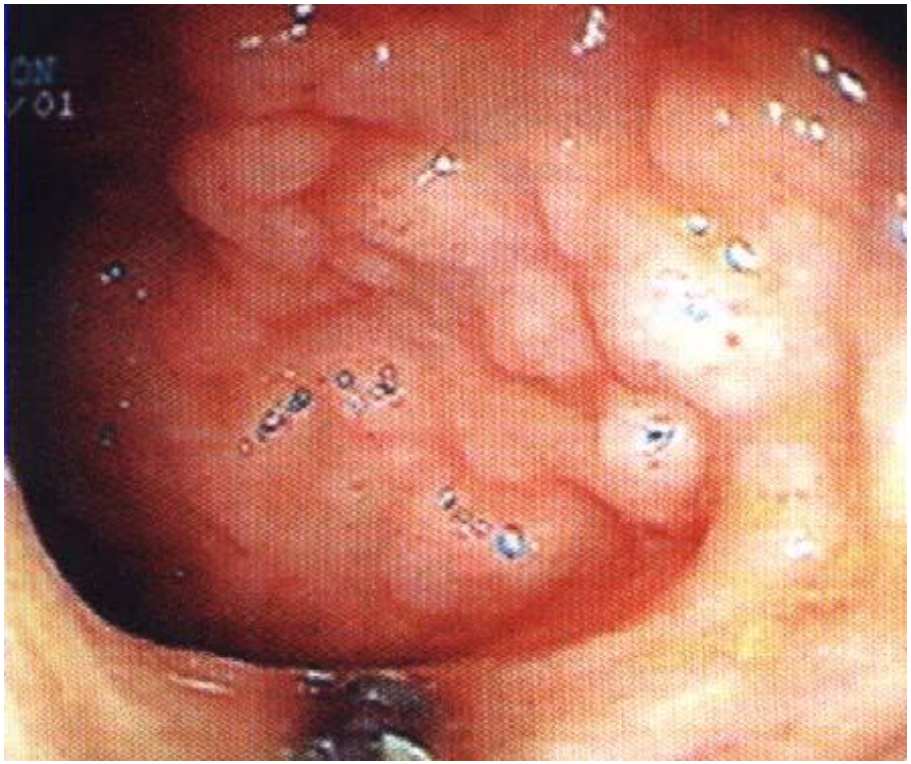
Infiltrazione di eosinofili

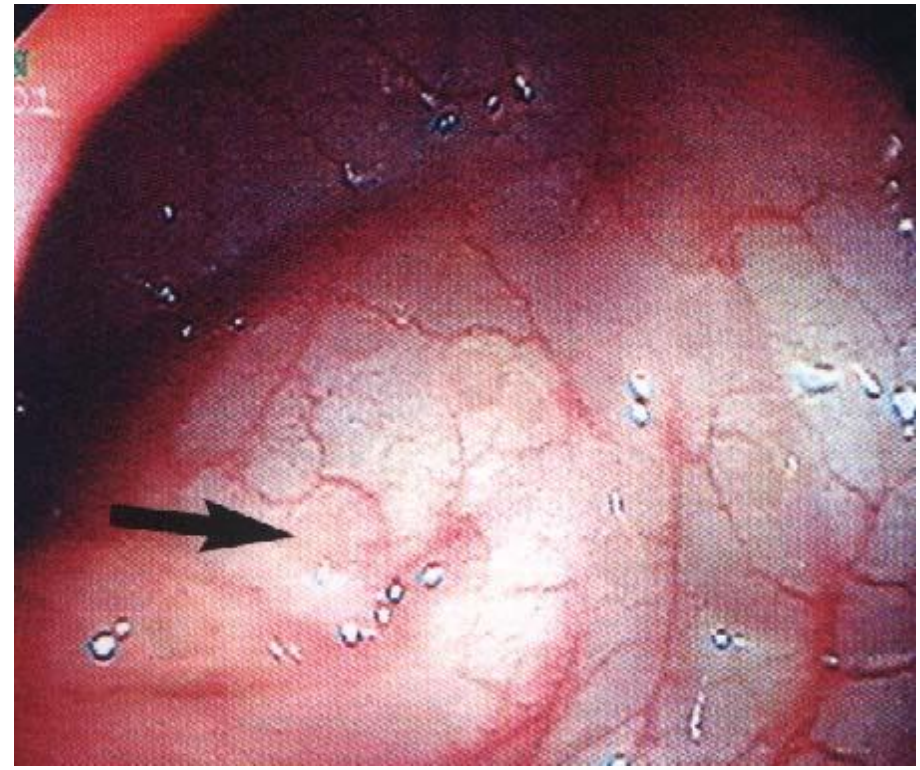
Iperplasia nodulare linfatica

Risposta clinica e istologica alla dieta senza glutine

Ravelli, Am J Gastroenterol 2008

Iperplasia nodulare linfatica





Infiltrato di linfociti gd

Iperplasia nodulare linfatica e allergia alimentare

- Riportata una associazione nel bambino (Kokkonen JPGN 1999; Iacono, Clin Gastroenterol Hepatol 2007) e nell'adulto (Carroccio, in press)

- Aumentata densità di linfociti intraepitelali gamma delta (Kokkonen)

- Solo 2/10 bambini che avevano risposto alla dieta di esclusione sono ricaduti dopo challenge (Quaglietta, Dig Liv Dis 2003)

Esofagite eosinofila

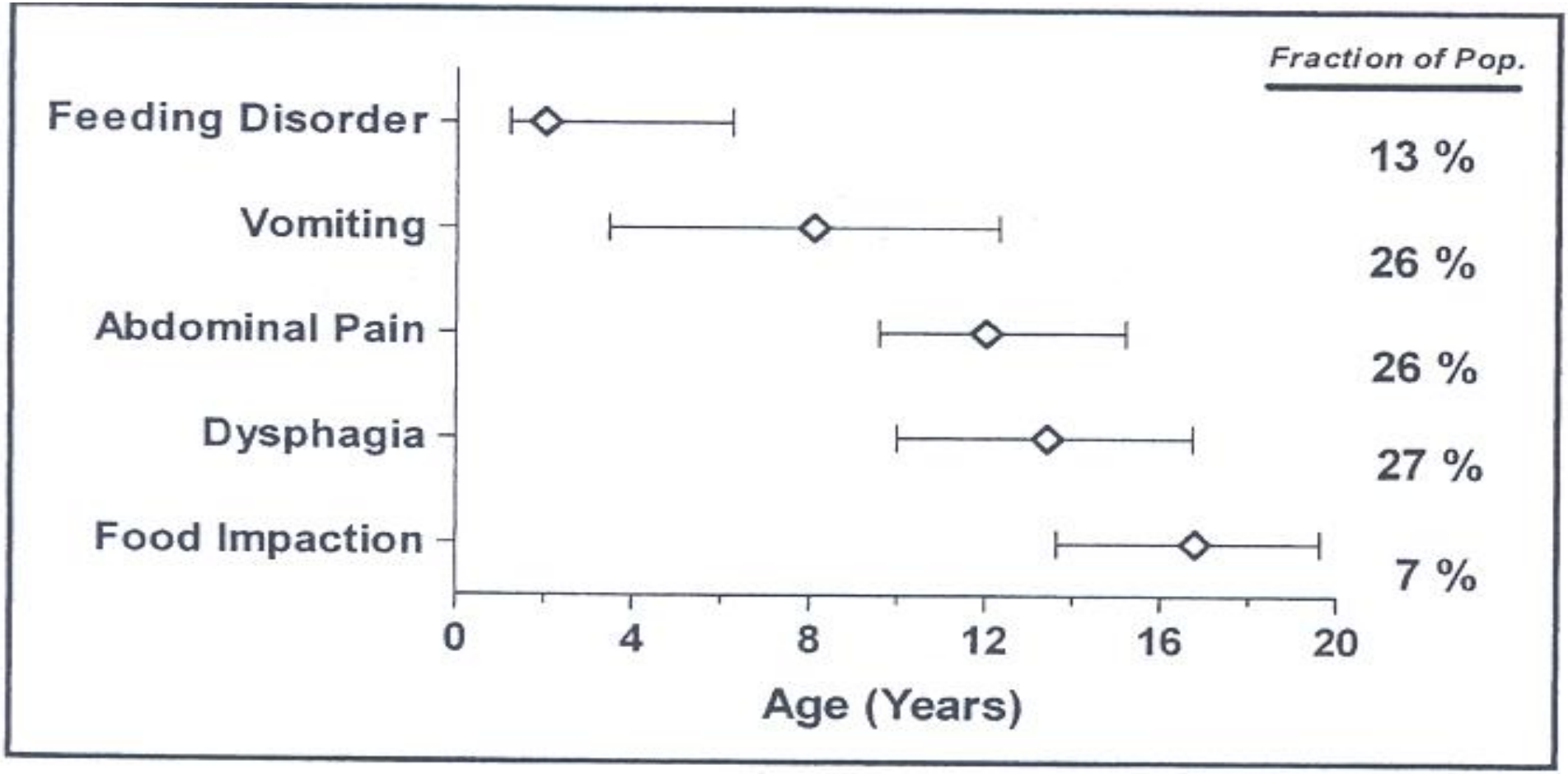
Sintomi:

- Vomito, pirosi, dolore epigastrico
- Disfagia
- Spesso basati solo su dati endoscopici

Nel 60% storia di atopia personale e/o familiare

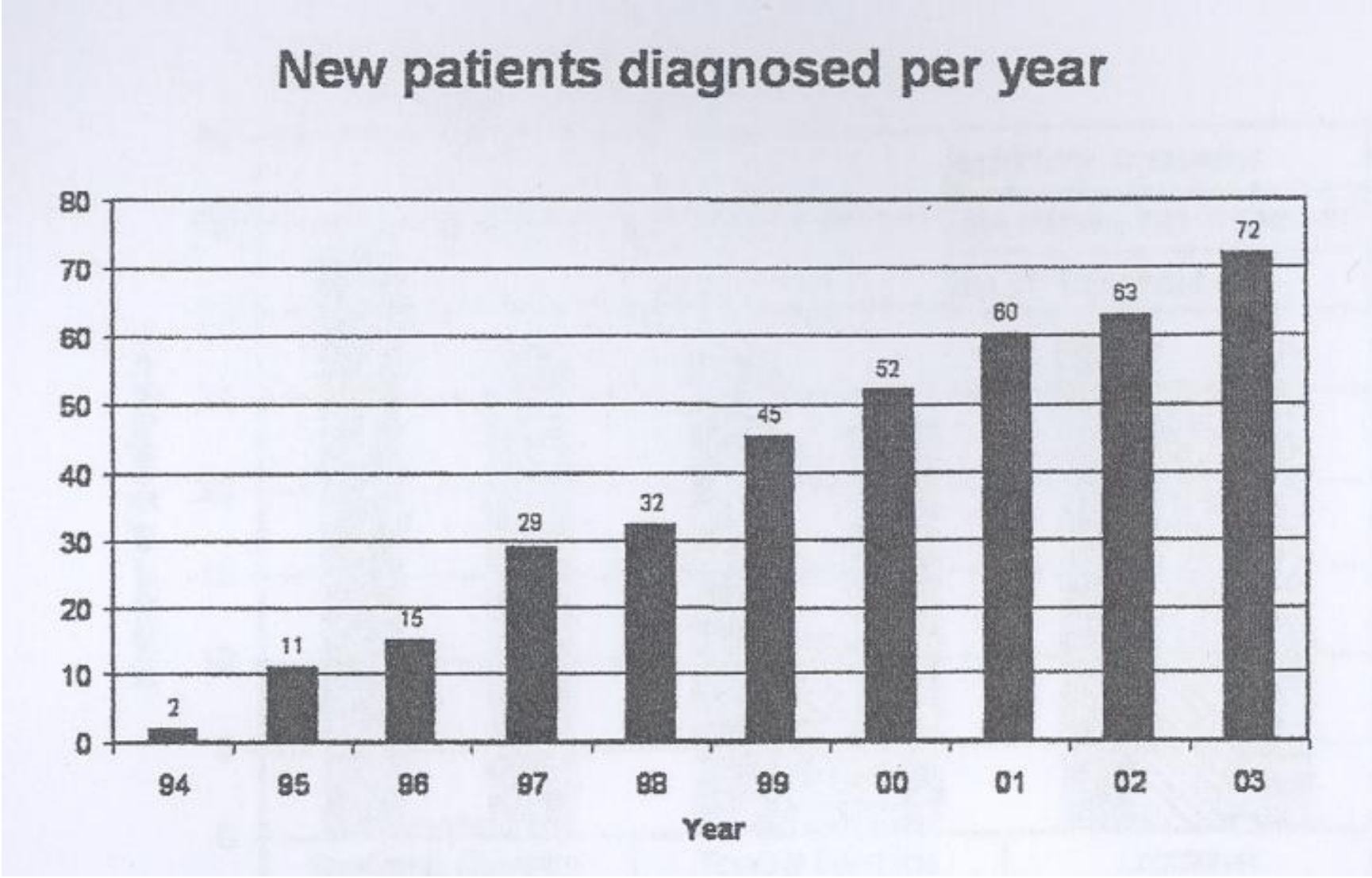
Dal 1993 in alcuni paesi c’è stato un forte aumento di incidenza

Esofagite eosinofila:
Sintomi di esordio in relazione all'età



Noel e Rothemberg, Current Opinion in Pediatrics 2005; 17: 690

Esofagite eosinofila: aumento dell'incidenza nell'ultimo decennio



Noel e Rothemberg, Current Opinion in Pediatrics 2005; 17: 690

Esofagite eosinofila

Diagnosi:

- >24 eosinofili per HPF
- Aumento di mastociti
- Cellule iperplasia, estensione delle cripte, micro ascessi eosinofili attorno alle papille
- pH-metria normale
- Nessuna risposta alla terapia anti-acida
- Risposta alle diete ad eliminazione

Esofagite eosinofila e malattia celiaca

Associazione tra esofagite eosinofila (EE) e malattia celiaca (MC)

In questi pazienti ci sono stati significativi miglioramenti dei sintomi clinici e dell'istologia esofagea con dieta senza glutine

Sono necessari ulteriori studi per comprendere la relazione tra EE e MC

Quaglietta et al, Aliment Pharmacol Therap 2007

Esofagite eosinofila

Cura:

•Dieta (amino acidi? Sulla base di prick/patch?)

•Steroidi (e.g. fluticasone)

•Montelukast?

•Mepolizumab (anti-IL5)? CAT 354 (anti-IL13)?

Scopo:

•Risoluzione dei sintomi? Risoluzione dell'infiammazione?

Allergia alimentare e disordini della motilità

Reflusso gastroesofageo

- Associazione con allergie al latte vaccino in più del 50% dei casi (Cavataio, JPGN 2000)
- Anomalie EGG dopo challenge con latte vaccino (Boulton, JPGN 1995)
- Pattern peculiare alla pH metria (Cavataio, Arch Dis Child,1985)

Allergia alimentare e disordini della motilità

Stipsi

- La stipsi cronica nei bambini potrebbe essere causata dall'allergia al latte (Iacono, NEJM 1998)

- Stipsi in atopici: un aumento della densità degli eosinofili nella mucosa rettale è associato ad un prolungato periodo di transito bocca-ano e ad un aumento del tono interno dello sfintere anale (Shah, JPGN 1999)

- Proctite in bambini con stipsi: eosinofilia nella lamina propria anale (Perlman, JPGN 2003)

Allergia alimentare e disordini della motilità

Stipsi

- Stipsi correlata ad allergia alimentare caratterizzata da proctite eosinofila (Carroccio, Scand J Gastroenterol 2005)

- Anche negli adulti stipsi refrattaria al trattamento risolta con dieta di eliminazione. Documentata proctite eosinofila (Carroccio, Scand J Gastroenterol 2006)

Stipsi cronica ed atopia

- 5113 bambini; 91 con stipsi (1,8%)
- 69 stipsi e 69 controlli (6 mesi - 6 anni)
- 12 tra quelli con stipsi (17%) e 13 nei controlli (19%) erano atopici (p=NS)
- 11/69 con stipsi erano refrattari alla cura (3 atopici). Nessun risultato dopo una dieta con latte senza proteine di 4 settimane

Simeone et al, Arch Dis Child 2008

Diagnostic tests

Antigen-specific

- Prick test
- RAST
- Patch test

Tissue damage markers

- Intestinal permeability test
- Markers of intestinal inflammation (ECP, calprotectin)
- Endoscopy/histology

Specific IgE

- Positive skin tests imply sensitisation, but do not mean need of exclusion diet!

- However, strongly positive results (e.g. prick with a wheal diameter >8 mm) indicate a greater likelihood of clinical reactivity

- Negative skin test confirm the absence of IgE mediated reactivity (negative predictive accuracy >95%)

Protein microarray technology and molecular diagnosis (or component resolved diagnostics)

- Identification of food allergic subjects (from those with sensitization based on crossreactivity)
- Prediction of severity of symptoms (es Ara h 2 for peanut allergy)
- Natural history (e.g. Bos d5.0102, k-casein, α_{s1}-casein for CMA)

Nomenclatura degli allergeni

- La versione più recente è disponibile nel sito web: www.allergen.org
- In base all'attuale nomenclatura, gli allergeni sono denominati secondo il nome tassonomico dell'organismo da cui derivano e riportano in sequenza. **3 lettere** di cui:
  - la prima maiuscola indicanti il **genere**;
  - la seconda minuscola che definisce la **specie**;
  - un numero che definisce **l'ordine successivo di identificazione** dell'allergene.

Alimento	Allergene		Genere	Specie	Numero
Arachide	Vicilina:	Ara h 1	Arachis	hypogaea	1
Soia	Profilina:	Gly m 3	Glycine	max	3
Latte	Caseine:	Bos d 8	Bos	domesticus	8
Merluzzo	Antigene M:	Gad c 1	Gadus	callarias	1

Principali allergeni di origine animale o vegetale responsabili di AA

Famiglia allergeni alimentari	Alimento	Allergene
Proteine animali		
Caseine	Latte di mammiferi	*Bos d* 8---latte vaccino
Sieroproteine	Latte di mammiferi	*Bos d* 4, *Bos d* 5, *Bos d* 7, *Bos d* 6---latte vaccino
Parvalbumine	Pesce	*Gad c* 1 --- merluzzo
Tropomiosine	Crostacei, molluschi	*Pen a* 1 --- gamberetto
Proteine vegetali		
Bet v 1	Frutta, verdura, soia	*Gly m* 4---soia; *Mal d* 1 ---mela
Cupine	Arachide, nocciola, legumi, semi	*Ara h 1*---arachide; *β-conglycinin*---soia
7S globulina	Arachide, nocciola, legumi	*Ara h* 3---arachide; g*lycinin*---soia
11S globulina	Soia, kiwi	*Gly m* 1 ---soia
Cisteine proteasi C1	Frutta, verdura, legumi	*Ara h* 5---arachide
Profiline	Cereali	*Api g* 4---sedano
Prolamine	Orzo, riso	*α*- e *γ-gliadin*---grano
Inibitori dell'amilasi/tripsina	Arachide, nocciola, semi	*Hor v* 1---orzo
2S albumine	Frutta, vegetali	*Ara h* 2---arachide
Lipid transfer proteins		*Mal d* 3---mela
		Pru p 3---pesca
		Cor a 8---nocciola

Berni Canani R, et al. PIP 2012

***Bos d* 4** epitopi conformazionali - latte trattato al calore

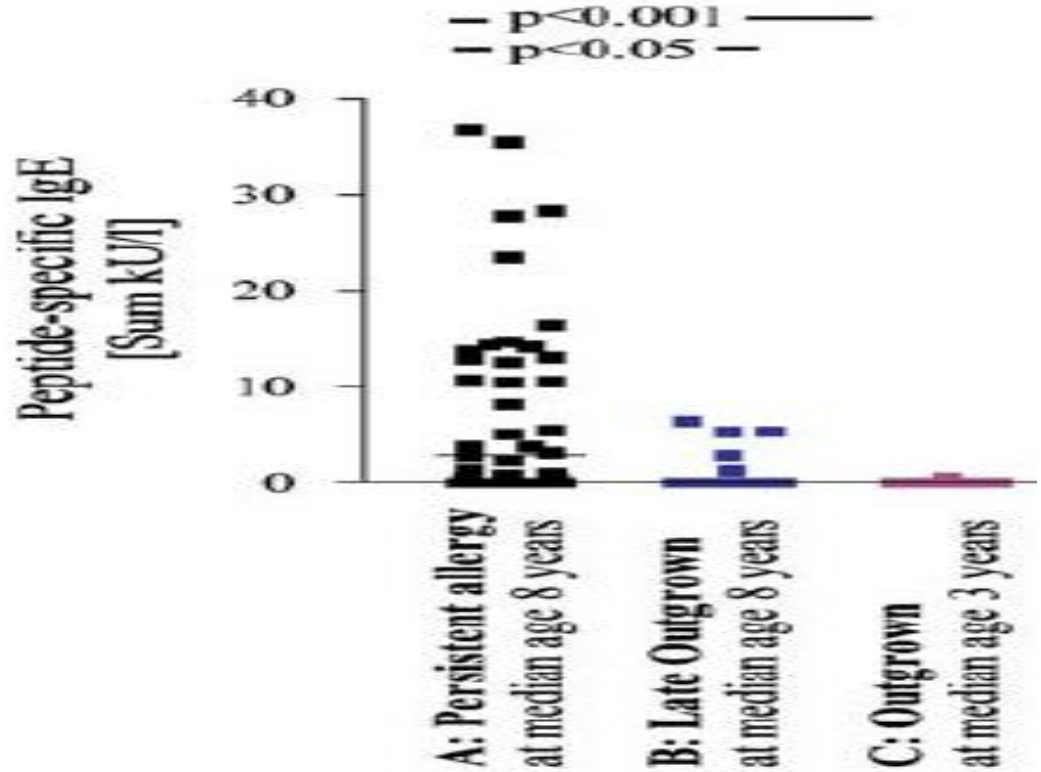
***Bos d* 5** epitopi conformazionali - latte bollito

Fraction	Protein	Allergen name	Concentration (g/l)	Total proteins (%)	Molecular weight (kDa)	Amino acids
Caseins			~30	80		
	α_{s1}-casein		12–15	29	23.6	199
	α_{s2}-casein		3–4	8	25.2	207
	β-casein	*Bos d* 8	9–11	27	24.0	209
	γ_1-casein				20.6	180
	γ_2-casein		1–2	6	11.8	104
	γ_3-casein				11.6	102
	κ-casein		3–4	10	19.0	169
Serum proteins			~5.0	20		
	α-lactalbumin	*Bos d* 4	1–1.5	5	14.2	123
	β-lactoglobulin	*Bos d* 5	3–4	10	18.3	162
	Immunoglobulin	*Bos d* 7	0.6–1.0	3	160.0	–
35-40%	BSA	*Bos d* 6	0.1–0.4	1	67.0	583
	Lactoferrin	–	0.09	Traces	800.0	703

- **Sistema ImmunoCAP**: *Bos d* 4, *Bos d* 5, *Bos d* 8
- **Sistema ISAC (microarray)**: anche *Bos lactoferrin*, *Bos d* 6

IgE specifiche per latte vaccino

La presenza di IgE specifiche verso *Bos d* 8 (allergene lineare della frazione caseinica dotato di elevata termo resistenza) predispone ad una minore tollerabilità nei confronti dell'alimento ed a un maggiore rischio di persistenza



Beyer K, et al. 2005

Fiocchi A, et al. 2011

Serological markers and prognosis

- IgE: initial levels, peak and trend
- Linear epitopes, diversity, higher affinity
- Ratio IgE/IgG1 and IgE/IgG4

Factors influencing the prognosis

- Severe symptoms
- Multiple sensitisation
- Early onset of asthma and rhinitis
- Reaction to small amount of milk
- Young age at the time of the first reaction

Allergy 2007: 62: 738–743

DOI: 10.1111/j.1398-9995.2007.01351.x

Original article

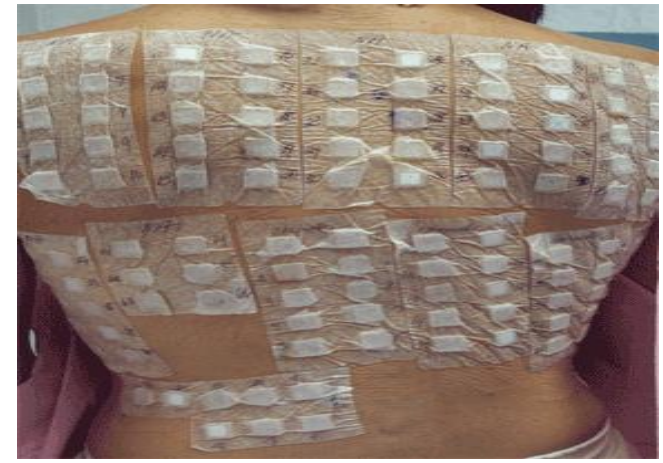
Diagnostic accuracy of the atopy patch test in children with food allergy-related gastrointestinal symptoms

Table 3. Diagnostic accuracy of the test used in CMA affected patient

	Sensitivity, % (95% CI)	Specificity, % (95% CI)	PPV, % (95% CI)	NPV, % (95% CI)	LR +/−
CM specific IgE	22.5 (0.09–0.41)	73.9 (0.51–0.89)	53.8 (0.25–0.70)	41.6 (0.26–0.57)	0.86/1.04
SPT	45.1 (0.27–0.63)	69.5 (0.47–0.86)	66.6 (0.43–0.85)	51.2 (0.30–0.66)	1.48/0.78
APT using fresh food	64.5 (0.45–0.80)	95.8 (0.78–0.99)	95.2 (0.76–0.99)	67.4 (0.49–0.82)	15.4/0.37
APT using commercial assay	6.45 (0.00–0.21)	95.6 (0.78–0.99)	66.6 (0.09–0.99)	43.1 (0.29–0.57)	1.48/0.97
APT using fresh food or SPT	87.1 (0.70–0.96)	65.2 (0.42–0.83)	77.1 (0.59–0.89)	78.9 (0.54–0.93)	2.5/0.19
APT using fresh food or SPT or CM specific IgE	90.3 (0.74–0.97)	52.1 (0.30–0.73)	71.7 (0.55–0.84)	80.0 (0.51–0.95)	1.8/0.18

APT, atopy patch test; CMA, cow's milk allergy; IgE, immunoglobulin E; LR+/−, likelihood ratio positive/negative test; NPV, negative predictive value; PPV, positive predictive value; SPT, skin prick test.

Toward a standardized reading of the atopy patch test in children with suspected cow's milk allergy-related gastrointestinal symptoms



R. Berni Canani*, A. Buongiovanni,
R. Nocerino, L. Cosenza & R. Troncone

ALLERGY Net

Table 1 Diagnostic accuracy of the atopy patch tests (APT) in children with suspected cow's milk allergy-related gastrointestinal symptoms

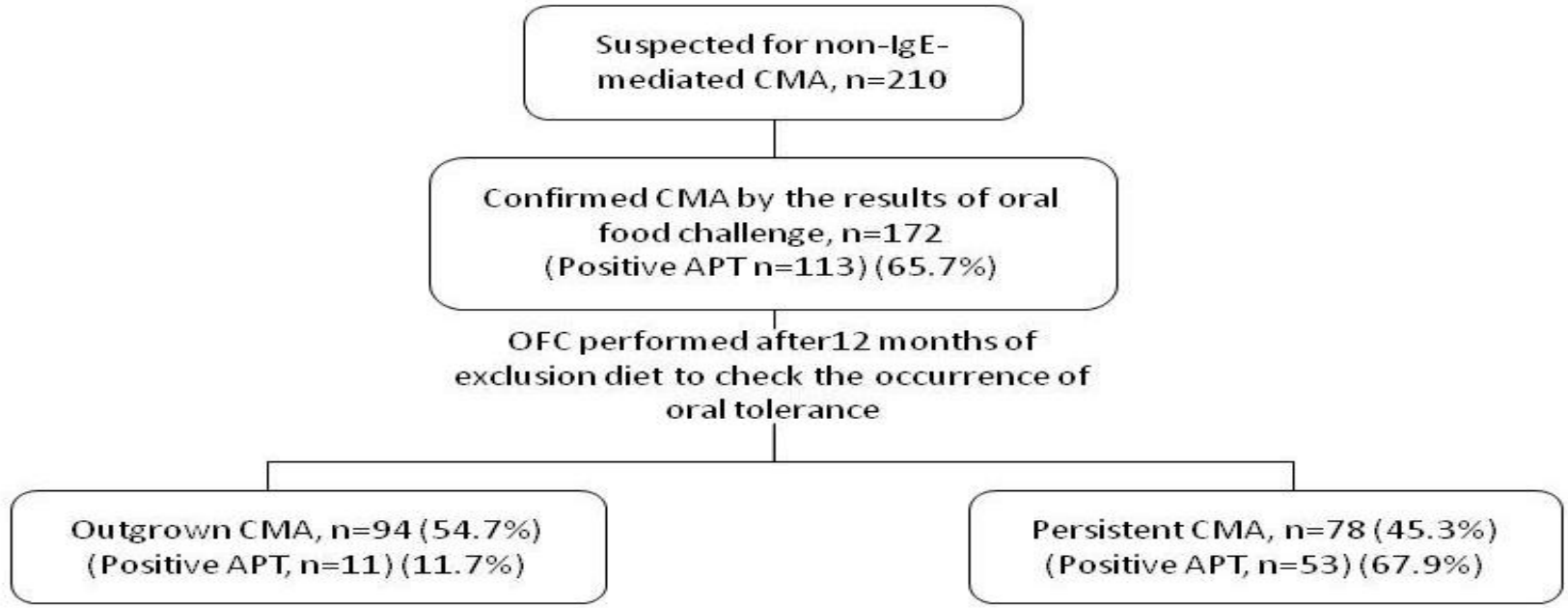
APT skin signs	Sensitivity (%) (95% CI)	Specificity (%) (95% CI)	PPV (%) (95% CI)	NPV (%) (95% CI)	LR + (95% CI)
Erythema and slight infiltration ± papules ± vesicles (≥+)	66.7 (49.8–80.9)	84.1 (69.9–93.4)	78.8 (61.1–91.0)	74.0 (59.7–85.4)	4.19 (2.1–8.6)
Erythema plus papules and/or vesicles (≥++)	53.8 (37.2–69.9)	97.8 (88.2–99.9)	95.4 (77.1–99.9)	71.0 (58.0–81.8)	24.2 (4.5–138.29)

PPV, positive predictive values; NPV, negative predictive values; LR, likelihood ratio.

Allergy
EUROPEAN JOURNAL OF ALLERGY AND CLINICAL IMMUNOLOGY
EAACI

Atopy patch tests are useful to predict oral tolerance in children with gastrointestinal symptoms related to non-IgE-mediated cow's milk allergy

R. Nocerino[1], V. Granata[1], M. Di Costanzo[1], V. Pezzella[1], L. Leone[1], A. Passariello[1,2], G. Terrin[1,3], R. Troncone[1,4] & R. Berni Canani[1,4]

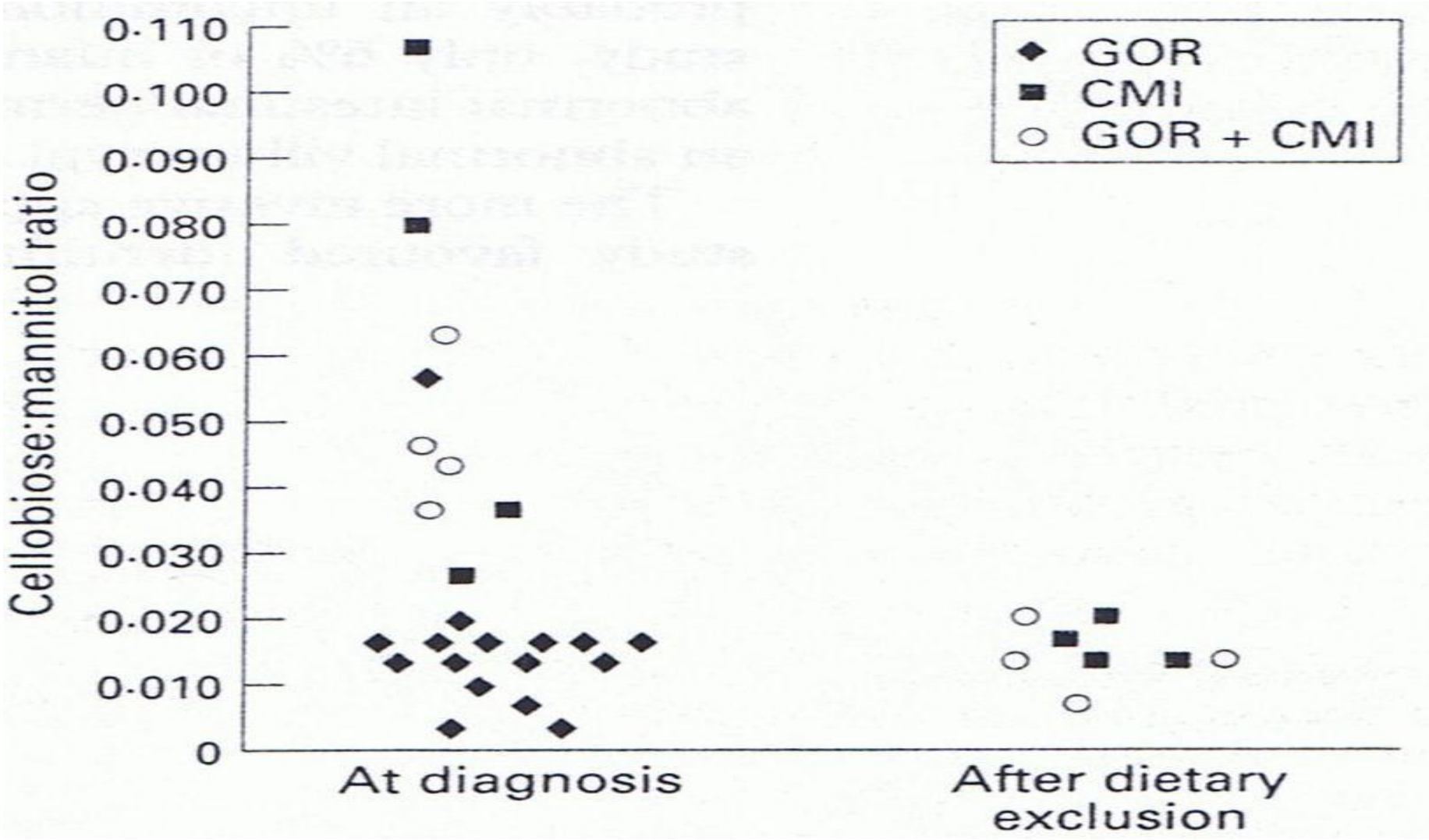


Sensitivity, % (95% CI)	Specificity, % (95% CI)	PPV, % (95% CI)	NPV, % (95% CI)	LR +/-
67.95% (95%CI 56.42-78.07)	88.3% (95% CI 80.03-94.01)	82.81% (95%CI 71.32-91.1)	76.85% (95%CI 67.75-84.43)	5.80/0.36

CMA: cow's milk allergy; APT: atopy patch test; OFC: oral food challenge

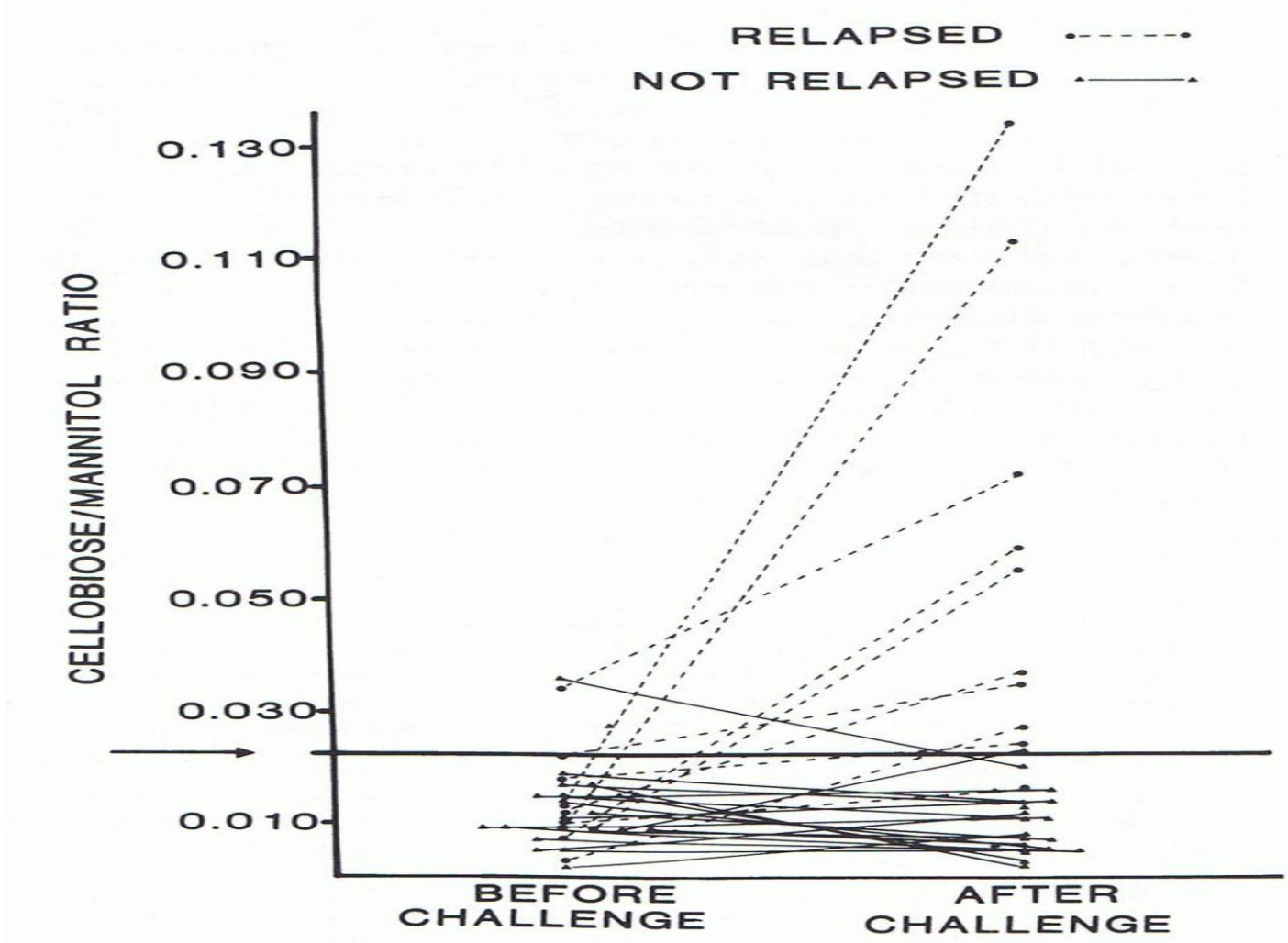
Allergy 2012

Il test di permeabilità agli zuccheri discrimina i lattanti con APLV da quelli con GER



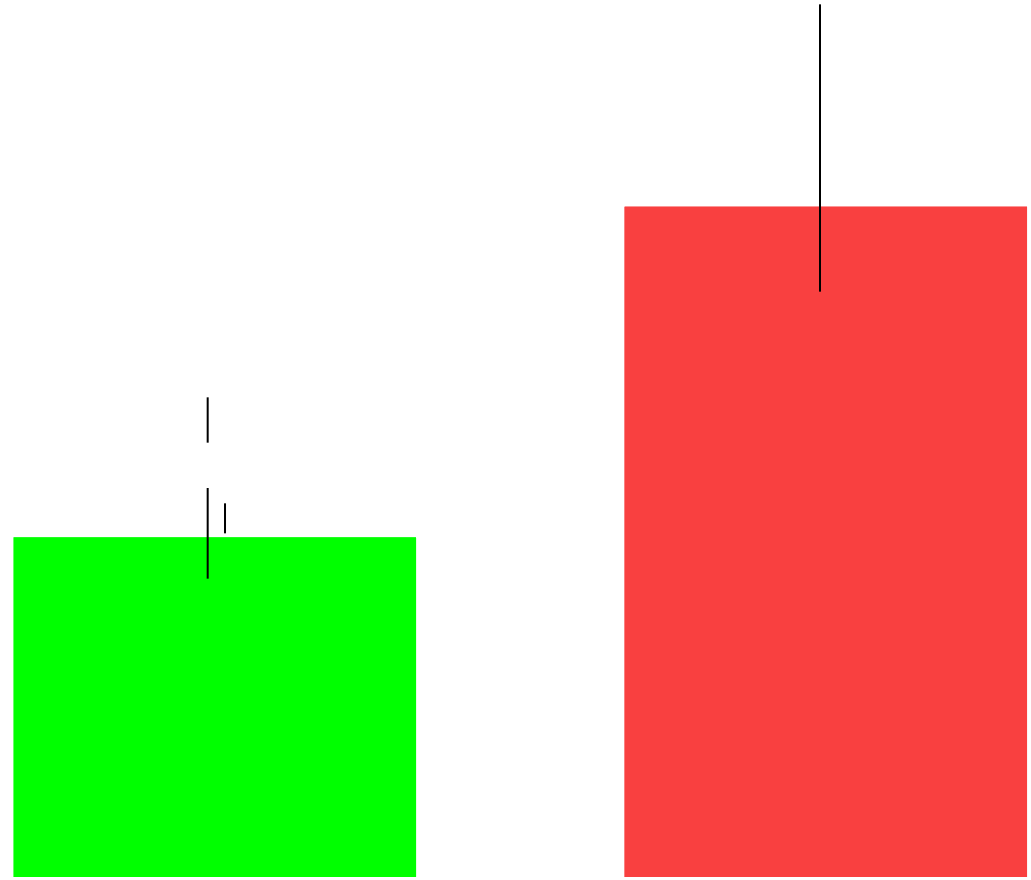
Staiano et al, Arch Dis Child 1995

Il test di permeabilità agli zuccheri si rivela utile nella valutazione dei risultati dei test di provocazione

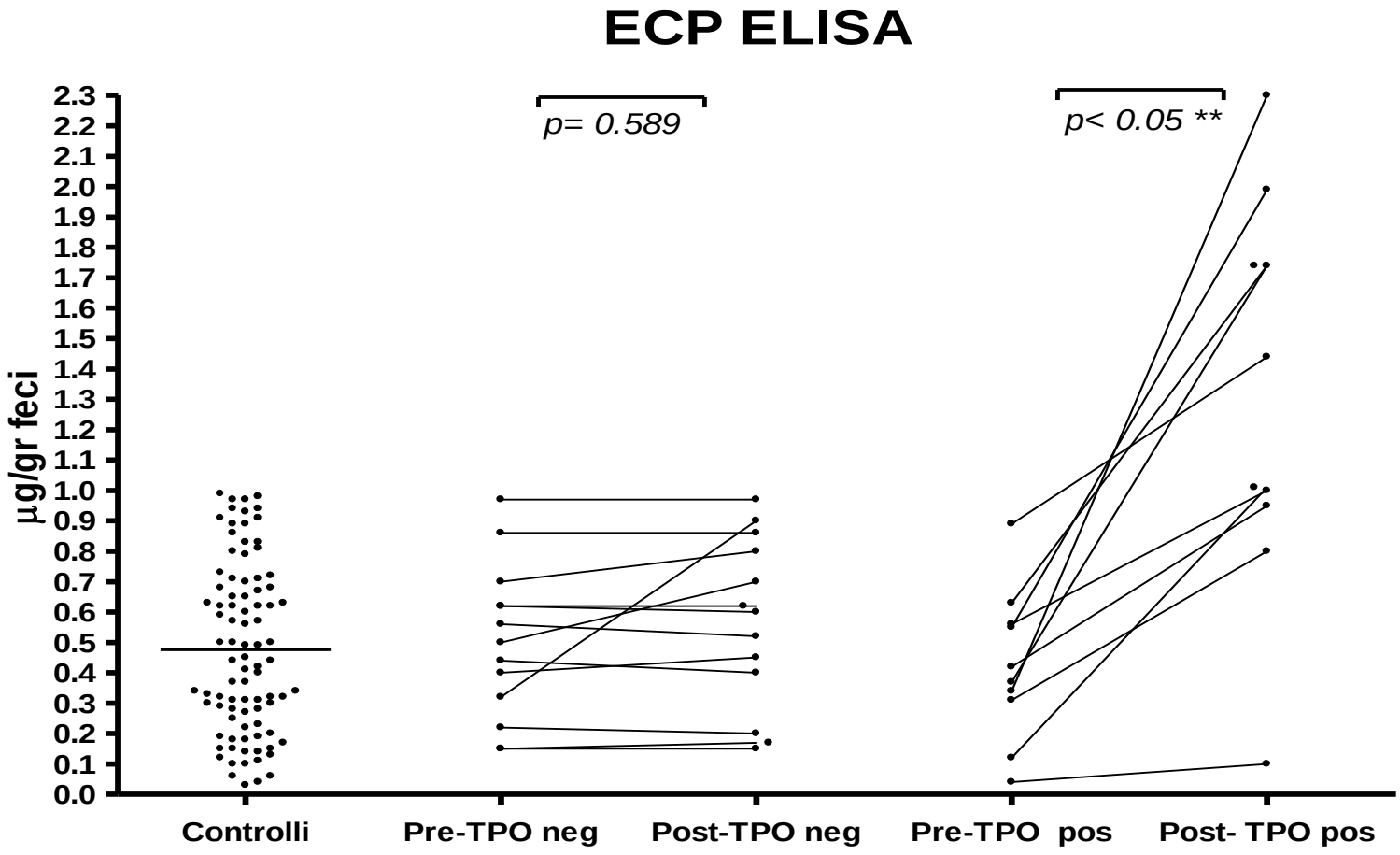


Troncone et al, Allergy 1994

Dosaggio di ECP nelle feci in bambini con allergia alimentare: risultati preliminari



PERCENTILI CONCENTRAZIONI DI ECP FECALE DETERMINATI IN BAMBINI DI ETÀ COMPRESA TRA 1-36 MESI



Cosenza L. et al. SIGENP MEETING 2008

Come viene fatta in Italia la diagnosi di APLV

- 8 Regioni (Valle d'Aosta, Piemonte, Liguria, Emilia Romagna, Marche, Lazio, Campania, Puglia)

- 44 pediatri di famiglia Totale pazienti = 41.958

- Diagnosi di AA in 340 pz (0.81%)

Sintomi (n° pazienti):

- GI 108
- respiratori 88
- cutanei 108
- sistemici 20

N° allergeni coinvolti:

- PLV 164/242
- uovo 96
- pesce 60
- pomodoro 44
- arachidi 24
- soia 18
- frumento 2

Diagnosis: DBPCFC

- The double blind placebo controlled food challenge (DBPCFC) remains the gold standard for the diagnosis of food allergy
- Clinical history and specific IgE indicate which foods should be evaluated
- Open or single blind food challenges are used to screen foods unlikely to provoke reactions
- Need for standardisation (dose, timing, drugs)

Common errors in the diagnostic work-up

Diagnosis of food allergy on the basis of :

- Allergy testing only: 28.0%
- Response to elimination diet (with or without allergy testing): 42.3%
- Oral food provocation test (open or blind): 29.7%

Probabilità di reazione ad alimenti cross reattivi

If Allergic to:	Risk of Reaction to at Least One:	Risk:
A legume* peanut	Other legumes peas, lentils, beans	5%
A tree nut walnut	Other tree nuts brazil, cashew, hazelnut	37%
A fish* salmon	Other fish swordfish, sole	50%
A shellfish shrimp	Other shellfish crab, lobster	75%
A grain* wheat	Other grains barley, rye	20%
Cow's milk*	Beef hamburger	10%
Cow's milk*	Goat's milk goat	92%
Cow's milk*	Mare's milk horse	4%

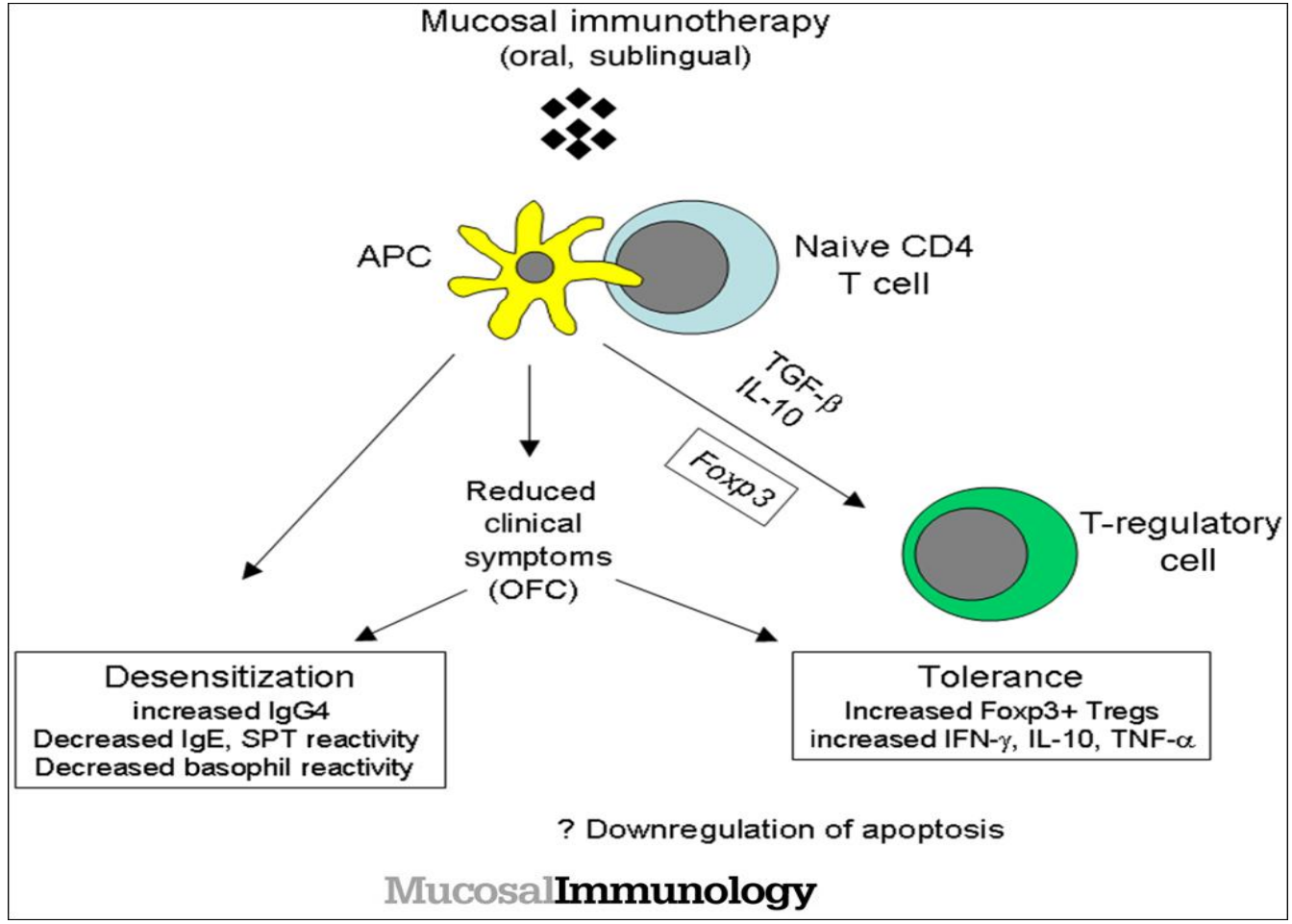
Therapy

Allergen avoidance is the mainstay of therapy

New immunotherapeutic strategies

- Anti-IgE antibodies, anti-IL5
- Modified antigens (including heat-treated)
- Adjuvants (e.g. immunostimulatory sequences such as CpG motifs or PAMPs)
- Specific oral tolerance induction (SOTI)

Desensitization and tolerance



2011

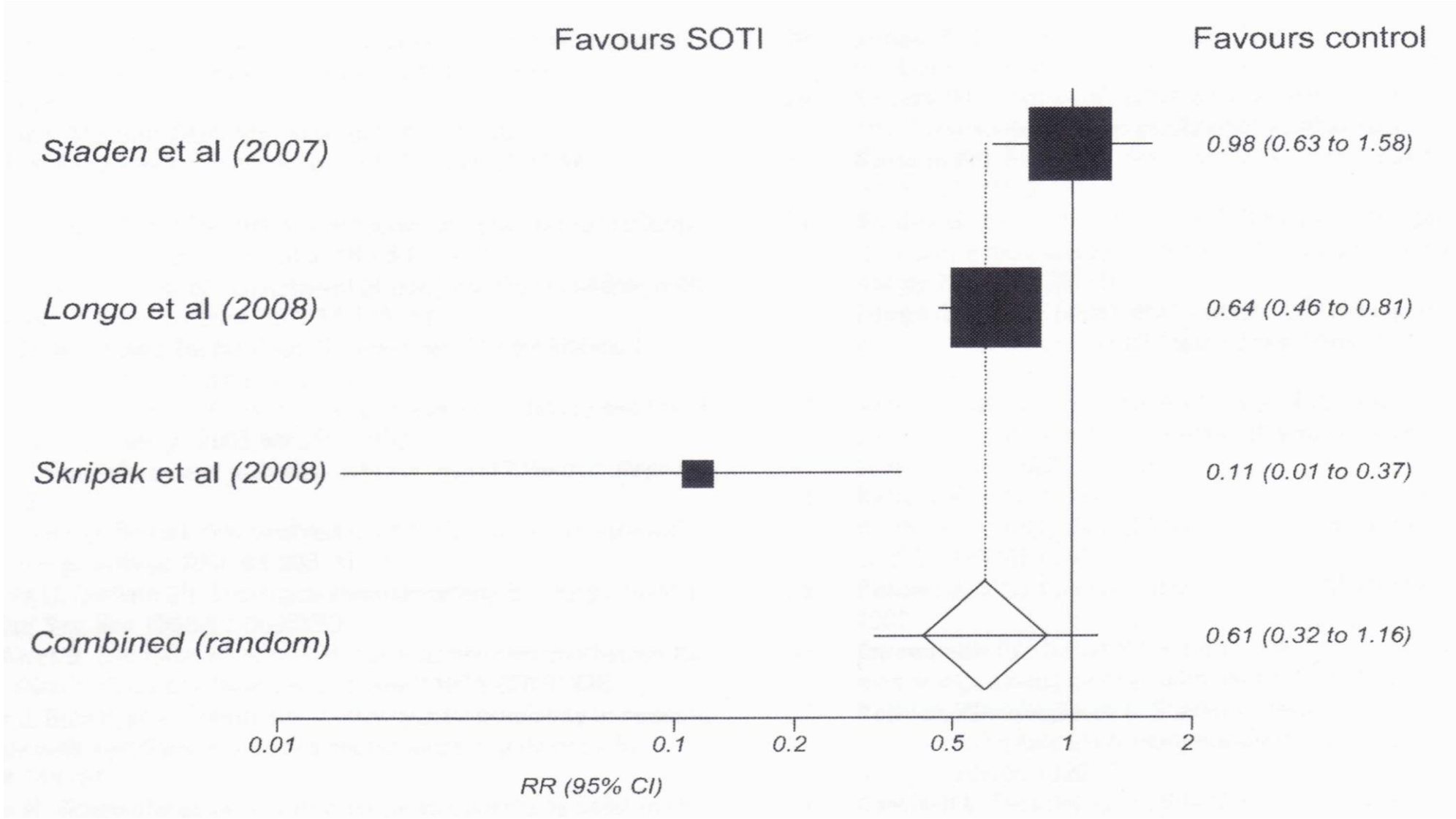
Therapy: SOTI

SOTI is the induction of tolerance that is achieved through incremental exposure to the relevant allergen

- 50-75% success
- 10-20% failure
- 10-20% partial sensitisation

- No death or life threatening events
- No predictive markers (high IgE)
- Oral sensitisation does not quickly lead to tolerance

SOTI efficacy: a metaanalysis



Fisher et al Arch Dis Child 2010;in press

SOTI: problems and perspectives

- Need for standardized and common protocols
- Predictive markers
- Compliance
- Insurance and legal aspects
- Induction of persistent tolerance
- Combination with anti-IgE

Therapy: SOTI

- SOTI cannot yet be recommended in routine practice as a means to induce tolerance in children with IgE mediated food allergy.

- Large, high quality RCT, investigating a variety of food allergens are necessary to assess the long term efficacy, safety and cost effectiveness

Strategies for prevention

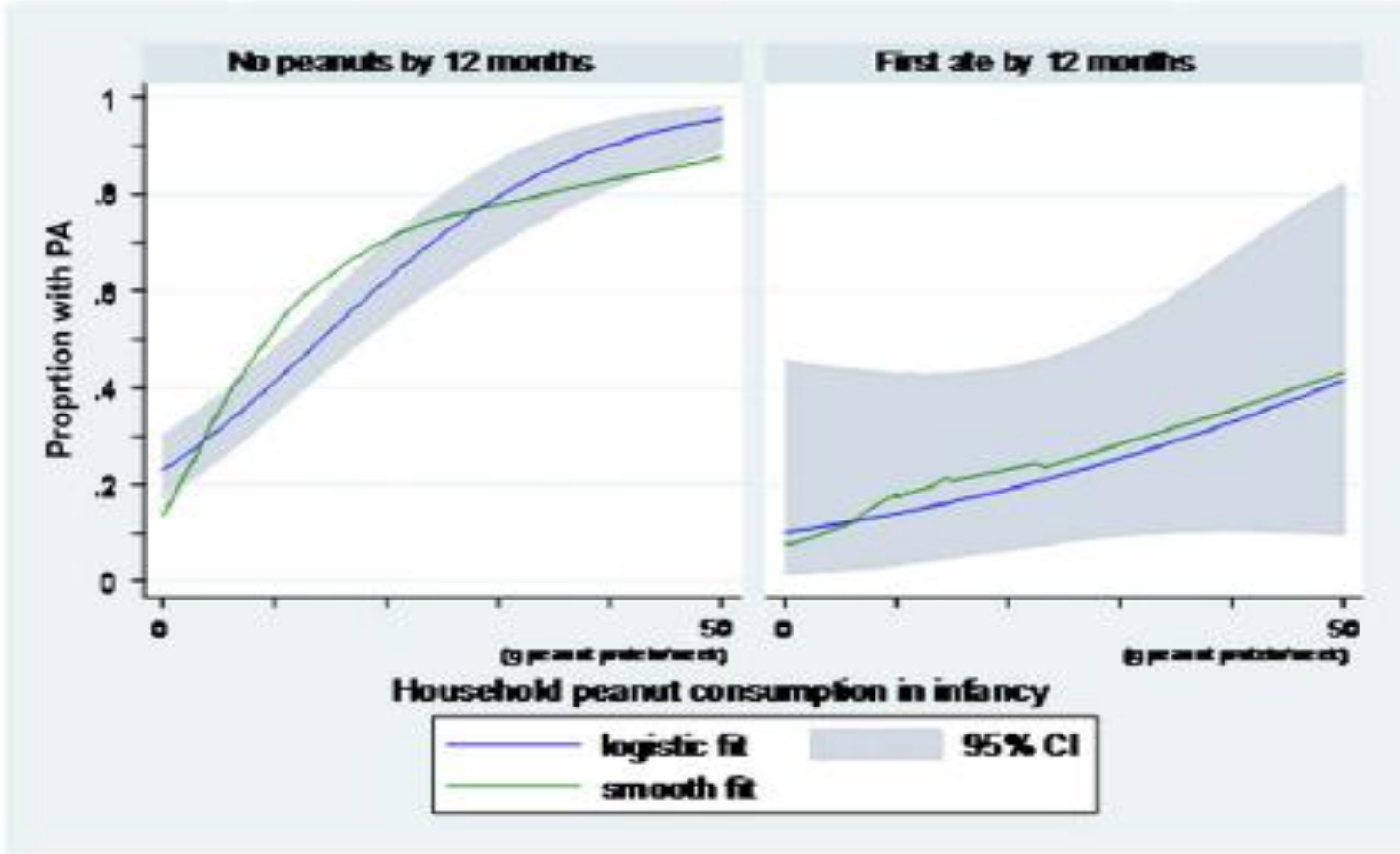
- ✓ “Prohibitionistic” approach

- ✓ Active approach, based on iummunomodulation

American Academy of Pediatrics
Prevention of allergy in at risk subjects:

- Breastfeeding for at least 4-6 months
- No exclusion diets during pregnancy and breastfeeding
- Totally or partially hydrolized formulas. No soya milk.
- No further protection if the intervention is continued after the 6th month of life
- Solid food introduction not before 4-6 months

Prevention strategies
Early administration of small amounts promote tolerance?
The example of peanuts



Fox AT et al. JACI 2009, 123, 417-423.

Early administration of small amounts promote tolerance? LEAP (Learning Early About Peanut Allergy)

640 high risk children (eczema or egg allergy: 20% risk to develop peanuts allergy)

Before 3 years: exclusion or 6 grams of peanut protein per week

Clinical evaluation, allergy tests and immunological studies

Clinical evaluation, allergy tests and immunological studies
Evaluation of allergy prevalence at 5 years of age

Celiac disease prevention in at risk children

PREVENT-CD – 36383 – FP6

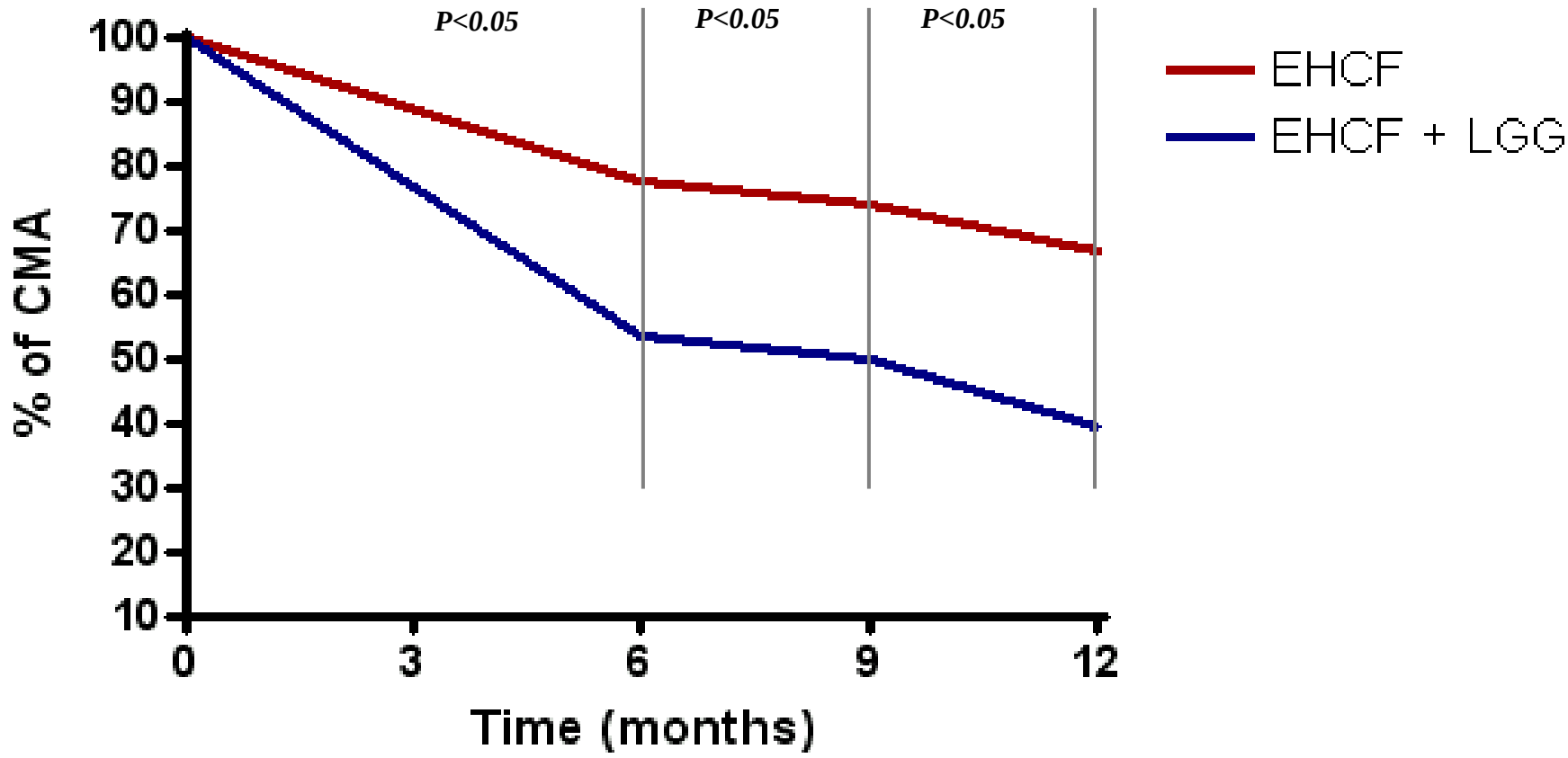
Microbiome

Innate immunity receptors signals (TLRs) play an important role in the homeostasis, Treg cells generation and allergy outcome

Specific bacterial products (polysaccharide A of the bacteroides fragile) interact with TLRs and promote Tregs, moduling the inflammation in murine IBD models (*Round et al*, *PNAS 2010; 107:12204*)

However, not conclusive results until now from trials with probiotics for allergy prevention and treatment

LGG influence tolerance recovery in CMA



Kaplan-Meyer method was used to estimate the probability of tolerance acquisition at 6, 9 and 12 month, and the resulting functions were compared with the log-rank test

NIAID, USA Guidelines

Solid food introduction not later than 4 - 6 months of age (Also potential allergenic food)

JACI 2010; 126 S1-S58

ESPGHAN guidelines

“Avoidance or delayed introduction of potentially allergenic foods, such as fish and eggs, has not been convincingly shown to reduce allergies, either in infants considered at risk for the development of allergy or in those not considered to be at risk”

JPGN 2008; 46:99–110

Future work

- Comprehension of the mechanisms underlying oral tolerance
- Therapies based on the reinduction of tolerance
- Prevention based on time and modalities of solid fod introduction in the first year of life but aso on active intervention (e.g.on the microbiome)