Coeliac disease: pathogenesis

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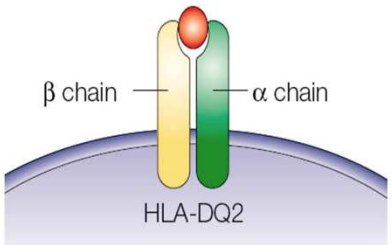
Celiac disease is a complex, multifactorial disorder

Normal intestine

Gluten proteins
(in wheat, rye, barley)

Environmental factors

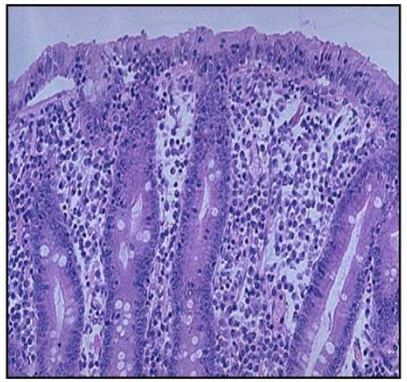
Genetic factors



+

Gene 1
Gene 2
....
Gene n

HLA-DQ2 gene
on 6p21: ~40%



Celiac disease

Definition of Celiac Disease

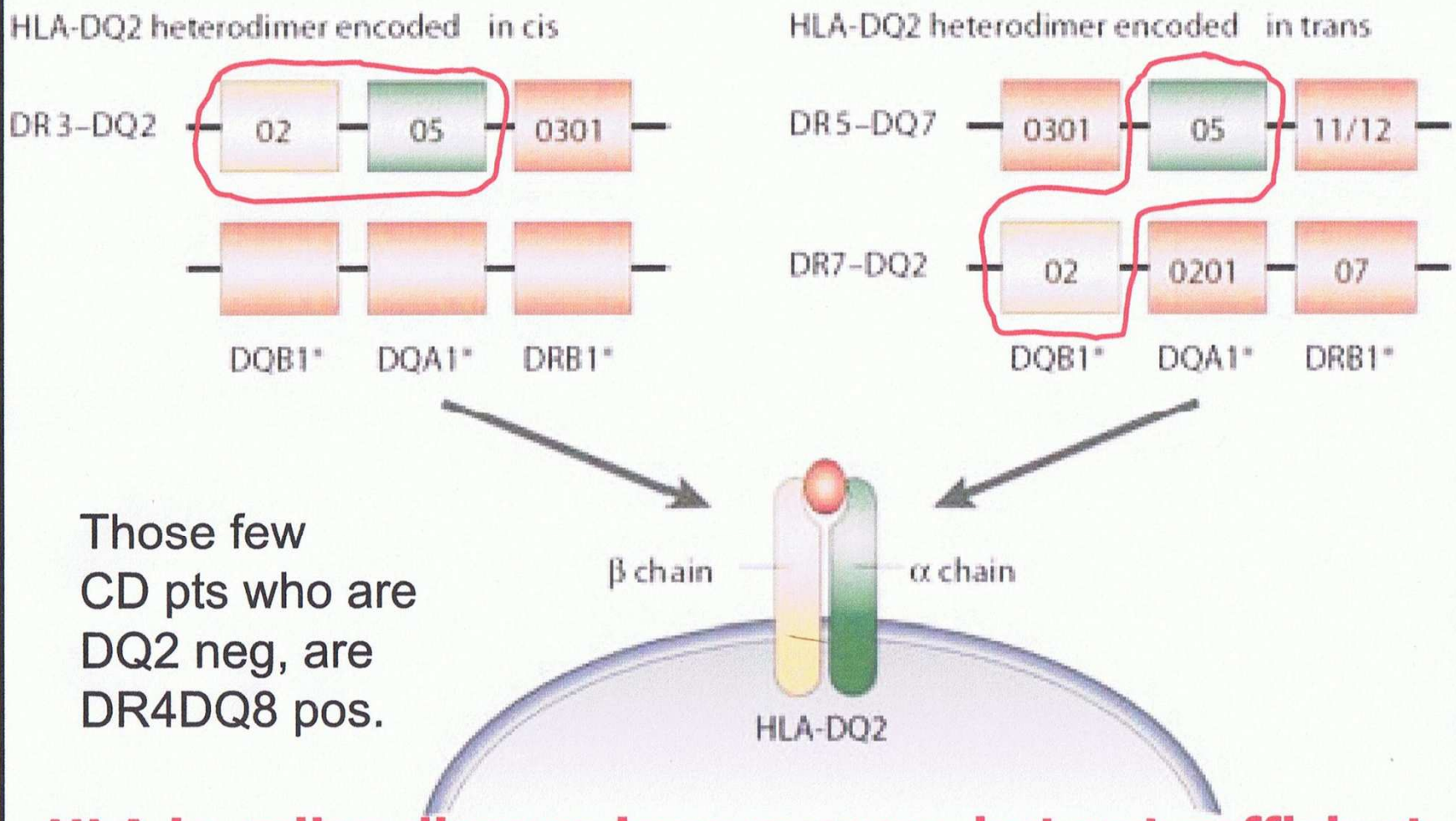
CD is an immune-mediated systemic disorder elicited by gluten and related prolamines in genetically (mainly HLA) susceptible individuals, characterized by the presence of variable combination of gluten-dependent clinical manifestations, CD specific antibodies, HLA DQ2 and DQ8 haplotypes and enteropathy

Genetics

Mechanisms

Prevention

HLA ASSOCIATION CELIAC DISEASE



Those few CD pts who are DQ2 neg, are DR4DQ8 pos.

HLA in celiac disease is necessary, but not sufficient

Risks for genotype groups in the population

Group	Genotype DR	Genotype group DQ	Risk %
H1/H1	DR3/DR3	G1 (Double DQ2)	21 %
H1/H2	DR3/DR7		
H2/H3	DR5/DR7	G2 (DQ2 in trans)	17 %
H1/H3	DR3/DR5	G3 (DQ2 in cis)	6 %
H1/H4	DR3/DR4		
H1/H5	DR3/DRX*		
H2/H2	DR7/DR7	G4 (1/2 DQ2 and/or DQ8)	5 %
H2/H4	DR7/DR4		
H4/H4	DR4/DR4		
H5	altri	G5	0,6 %

Risk for an individual to develop the disease according to his genotype group

Bourgey M. et al. Gut 2007;56:1054-9

Genetics of coeliac disease
Non-HLA genes

- T-cell development in the thymus

(THEMIS, RUNX3, TNFR SF14, ETS1)

- Immune detection of viral RNA

(TLR7-8)

- T-B costimulation

(CTLA4-ICOS-CD28, TNFR SF14, CD80, ICOS LG, TNFR SF9, TNF SF4)

- Cytokine & chemokine receptors

(2q11-12 ILR cluster (IL18 RAP), 3p21 chemokine (CCR5), 4q27 (IL2-21) , IL12A, TNFR SF18, CCR4)

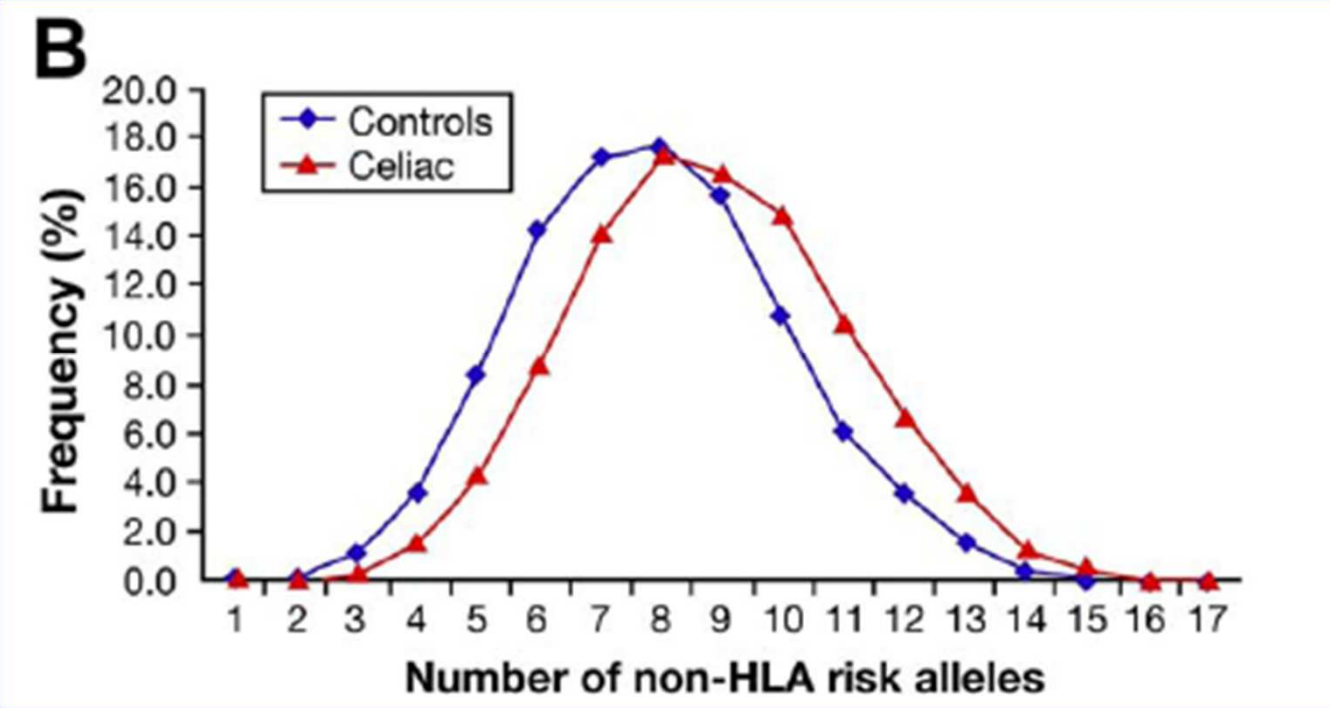
- Non-identified pathways

LPP

Dubois et al. Nat Genet 2010;42:295-302

It is possible to establish a "risk profile"

Frequency distribution of non-HLA risk alleles in Cases and Controls



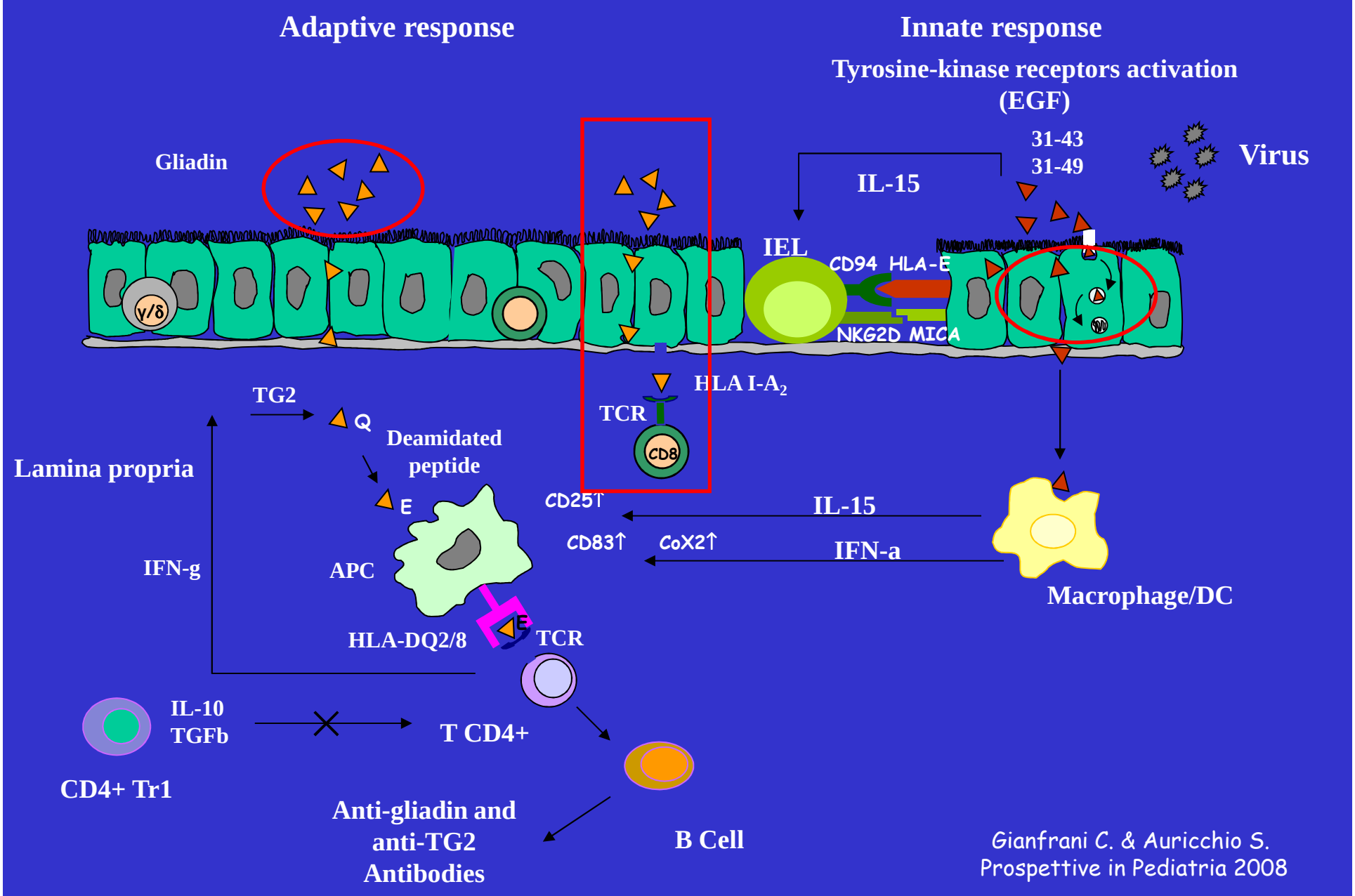
Romanos J. et al. Gastroenterology 2009;137:834-40

Genetics

Mechanisms

Prevention

Pathogenic mechanisms in celiac disease



Gianfrani C. & Auricchio S.
Prospettive in Pediatria 2008

Celiac disease: disease mechanisms

- Gliadin resistance to enzymatic digestion
- Paracellular permeability alterations
- Interference with endocytosis pathway
- Activation of innate immunity mechanisms
- Activation of lamina propria gliadin-specific CD4+ (and CD8+) lymphocytes
- Induction of autoantibodies (anti-transglutaminase) and their pathogenetic role

Gliadin resistance to enzymatic digestion

- Resistance to proteolysis by gastric, pancreatic and brush border enzymes due to high number of proline residues
- Polipeptides with high molecular weight (e.g. 33mer) final product of hydrolysis
- Efficacy of prolylendopeptidase of bacterial and fungal origin

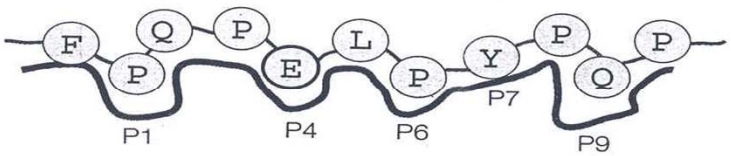
33mer (α2-gliadin 56-88)

- Prodotto finale della digestione, resistente all'idrolisi a causa dell'elevato contenuto in proline (13 su 33 residui)
- Contenente 6 copie parzialmente overlappanti di 3 epitopi T: potente stimolatore della risposta T

Khosla & Sollid, 2004

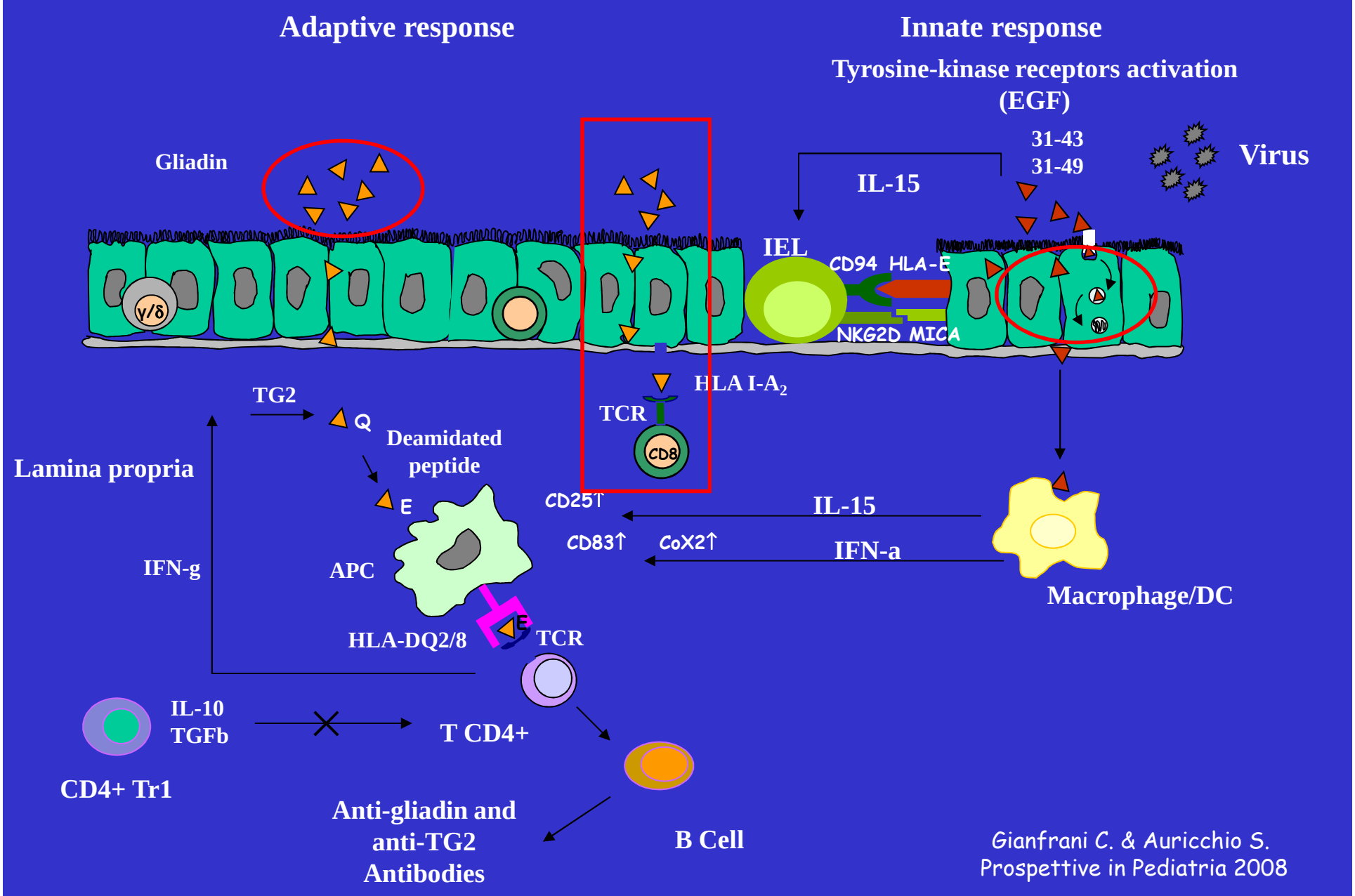
Coeliac disease: dissecting a complex inflammatory disorder
Sollid LM. Nat Rev Immunol 2002;2:647-55

Oligomerized epitopes



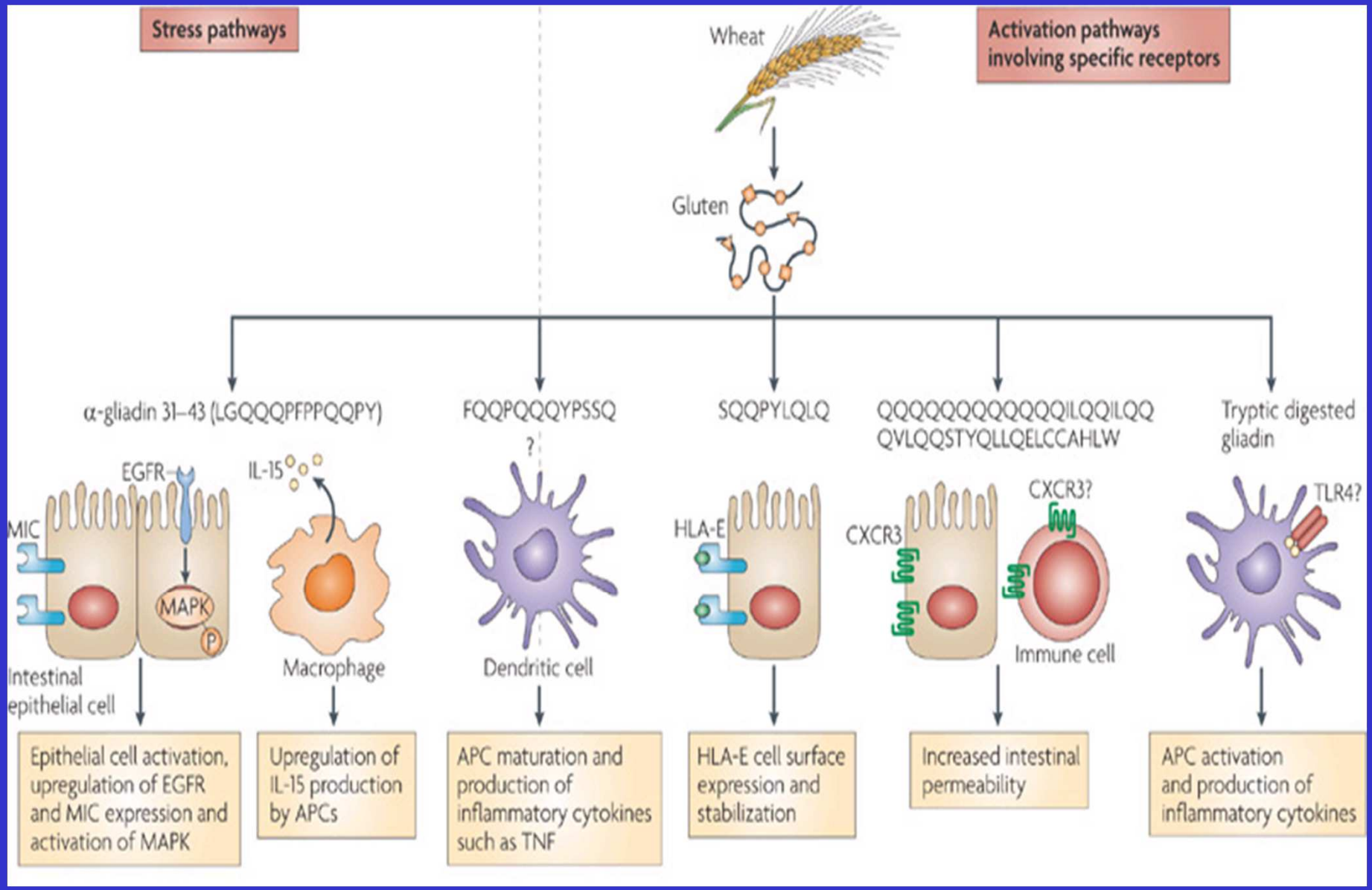
Binding to HLA-DQ2

Pathogenic mechanisms in celiac disease



Gianfrani C. & Auricchio S.
Prospettive in Pediatria 2008

Gliadin peptides and stress pathways

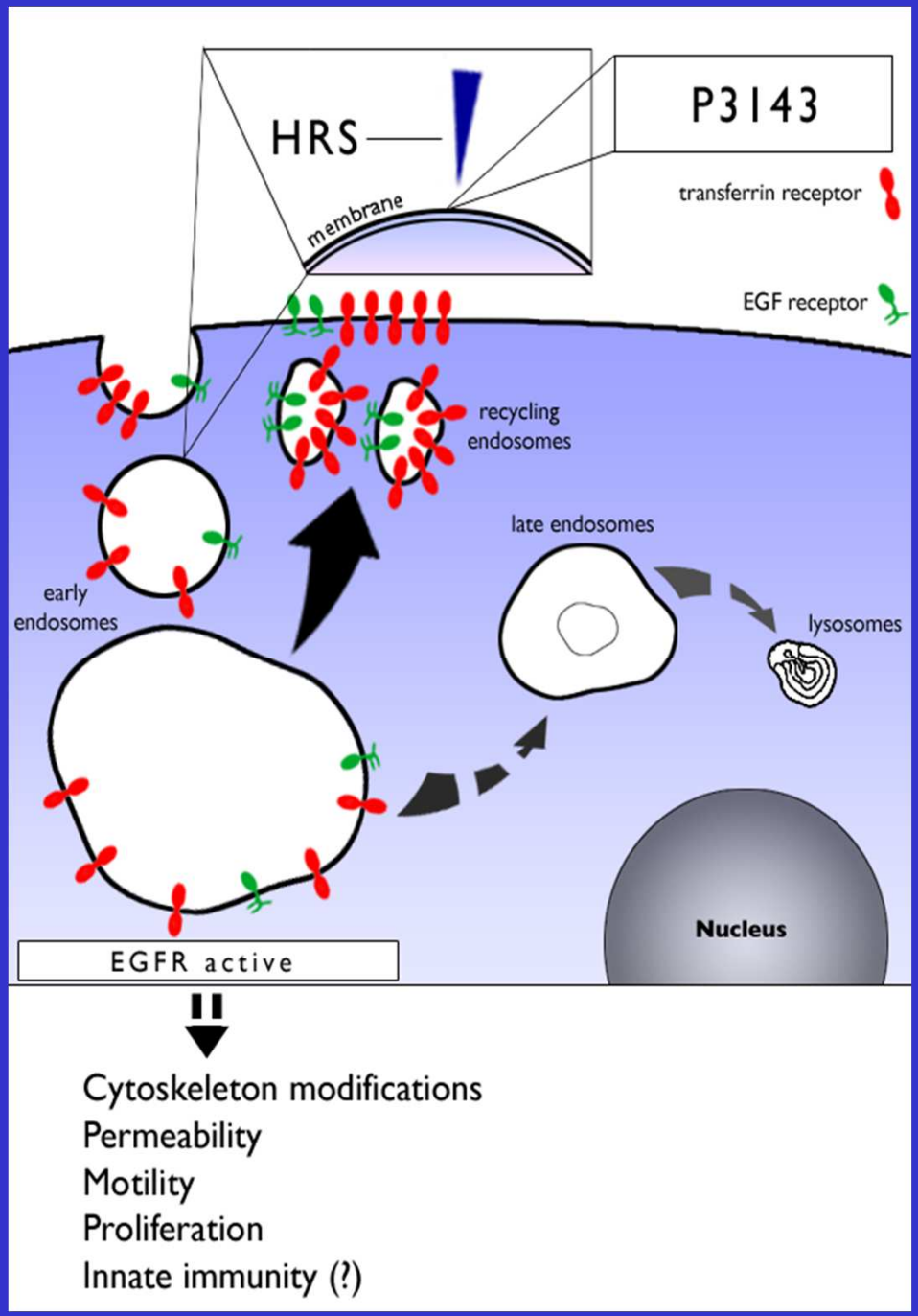


Jabri B. et al. Nat Rev Immunol. 2009;9:858-70

Possible model of P31-43 non T-cell mediated activity

P31-43 interferes with the endocytic pathway

e.g. it delays EGFR in early endocytic vesicles increasing trafficking of recycling endosomes



Interference of gliadin with endocytosis and its consequences

- Gliadin peptides interfere with endocytosis pathway delaying maturation of vesicules from “early” to “late endosomes”

- This means a longer activation of tyrosyne kinase receptors (example EGFR). The prolonged activation of EGFR causes on different cellular and tissutal types (included small intestine mucosa) different biological effects: rearrangement of actin and alterations of permeability, proliferation and tissue remodeling, probably activation of innate immunity (higher expression of IL15)

- It remains to be explained the higher susceptibility of celiac patients to these biological activities of gliadin (on a genetic basis?)

Barone et al, Gut 2007

Activation of innate immunity mechanisms

- Higher expression of IL15 at epithelial and lamina propria levels in intestinal mucosa of celiac patients (induced by p31-43)

- Higher epithelial infiltration of NK-like lymphocytes and higher molecular expression (NKG2D) that facilitate cytotoxicyty (induced by IL15)

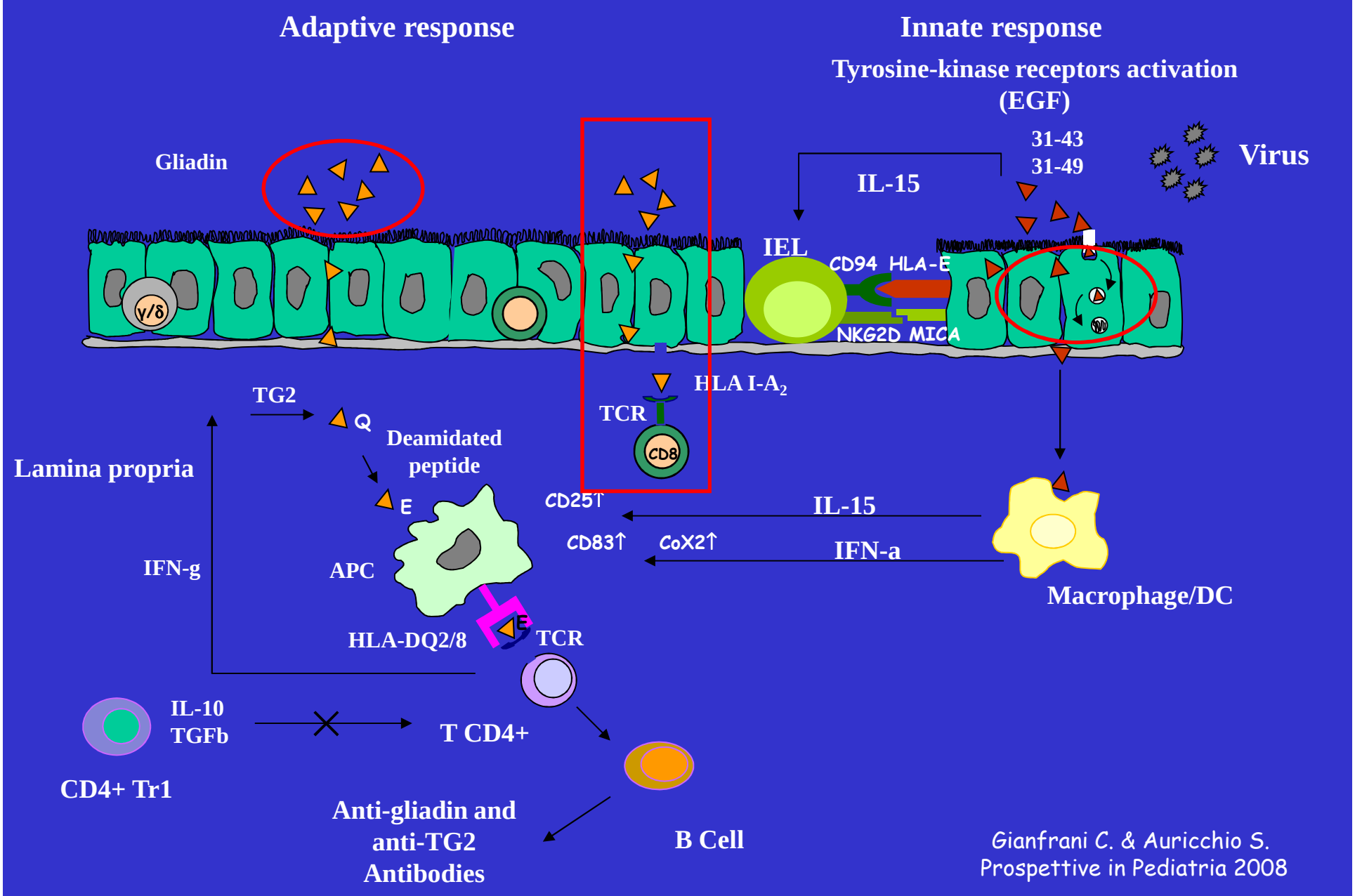
- Higher MICA expression on intestinal epithelium (induced by p31-43 and mediated by IL15). MICA is a target of NK-like TCR independent cytotoxic cells

Proinflammatory cytokines in CD

- αIFN
- IL15
- IL18

- γIFN
- IL17
- IL21

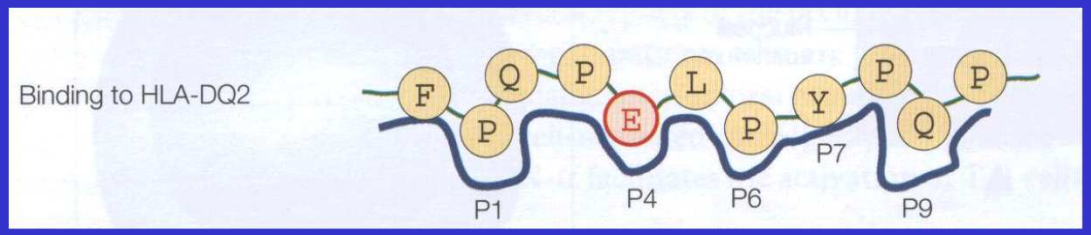
Pathogenic mechanisms in celiac disease

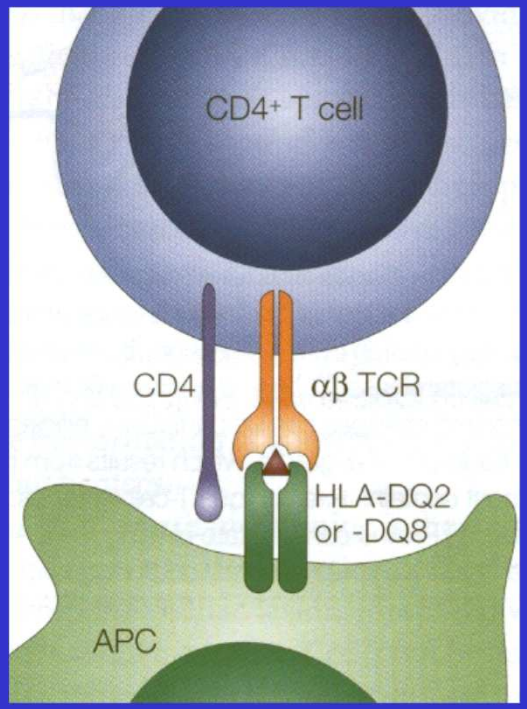


Gianfrani C. & Auricchio S.
Prospettive in Pediatria 2008

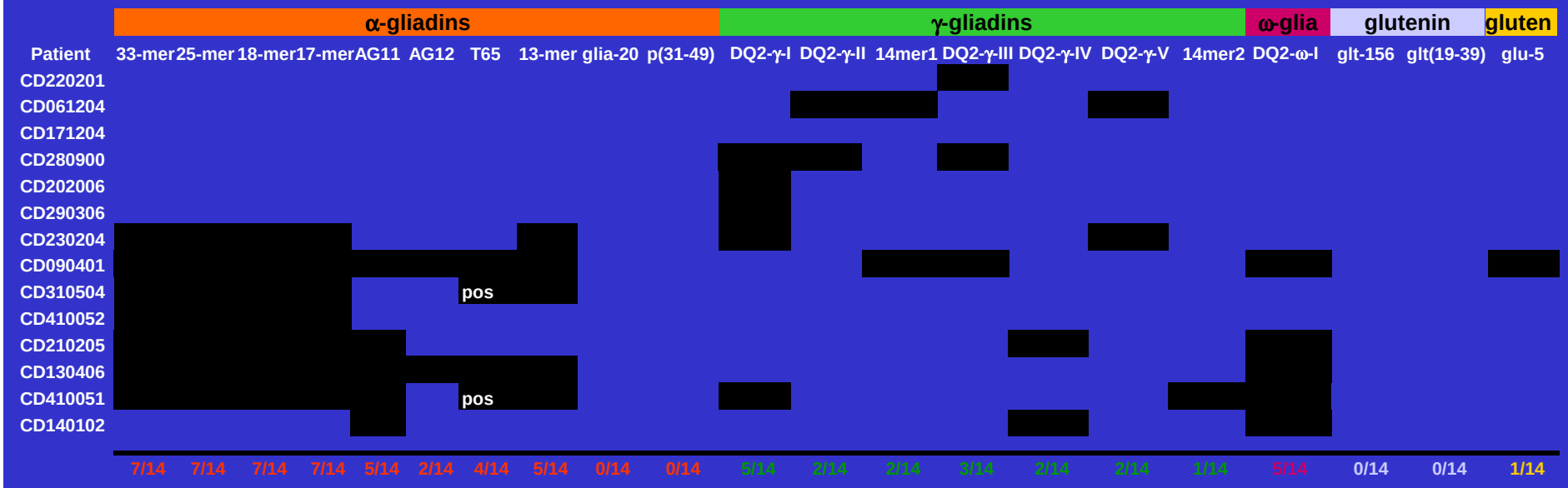
Deamidation critical step for presentation of gliadin to T cells

Deamidation of glutamine residues in glutammic acid, by tissutal transglutaminase, is critical for peptide-molecule linkage HLA (HLADQ2 - DQ8)

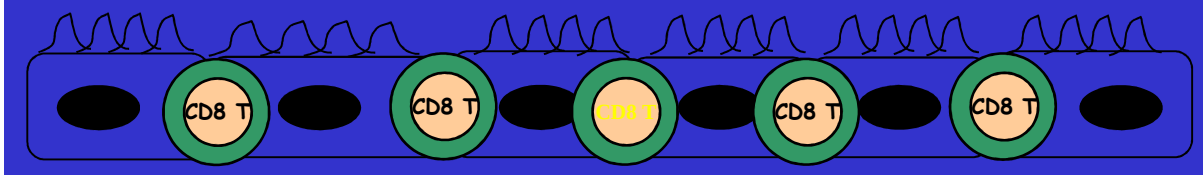




Recognition pattern of gliadin immunogenic peptides



The extensive infiltration of CD8+ T lymphocytes in the intestinal mucosa is one of the hallmarks in CD



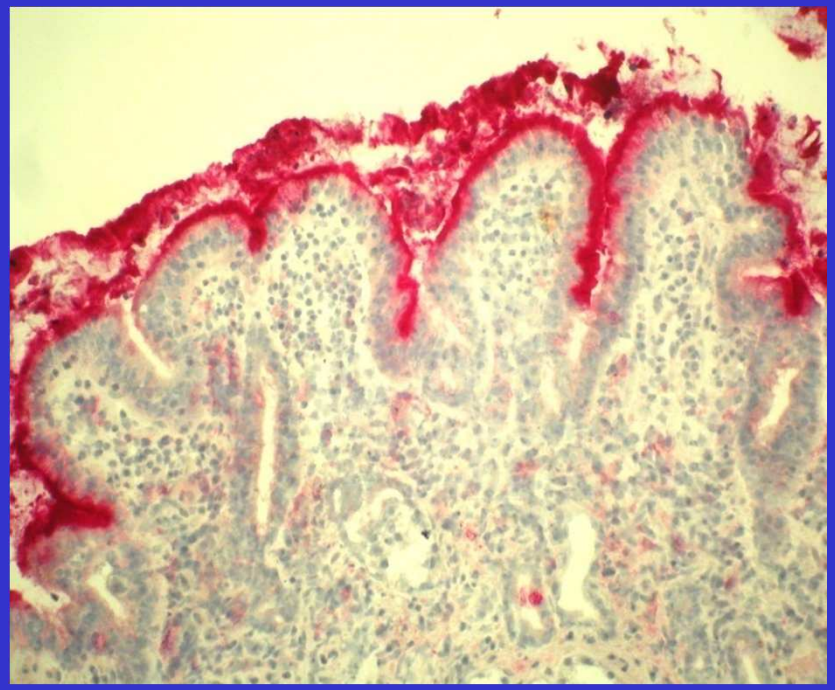
IEL

1:5 IEL:EC in normal mucosa

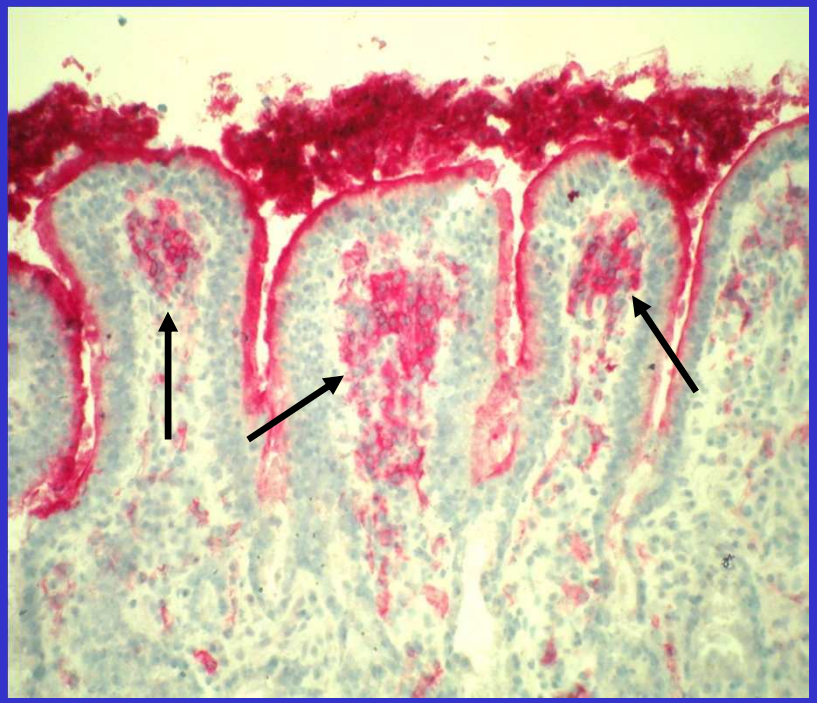
1:1 IEL:EC in CD mucosa

We have identified a peptide, A-gliadin 123-132 (QLIPCMDVVL) which is specifically recognized by HLA A2-restricted CD8+ T lymphocytes from coeliac patients **(Gianfrani et al *J Immunol* 2003; Mazzarella et al Gastroenterology 2008)**

pA2-induced CD25+ cells mainly localized under intestinal epithelium layer



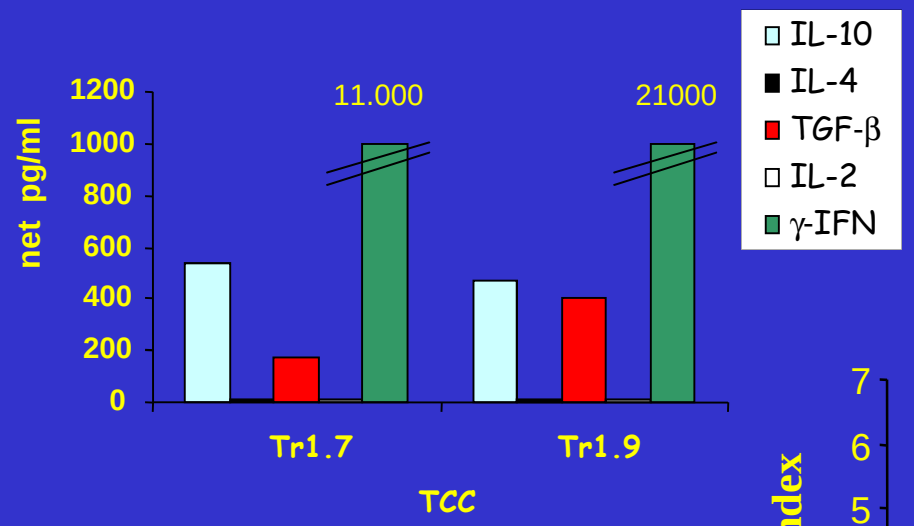
medium

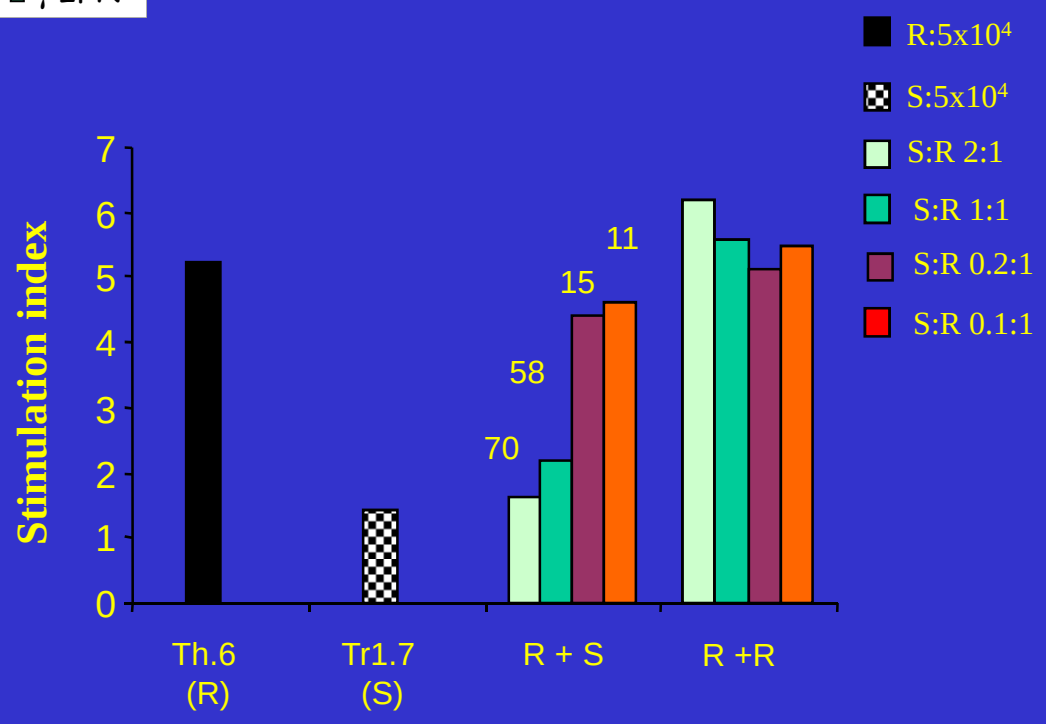


pA2

Mazzarella et al Gastroenterology 2008

Gliadin reactive Tr1 cells are present in celiac intestinal mucosa and are functional

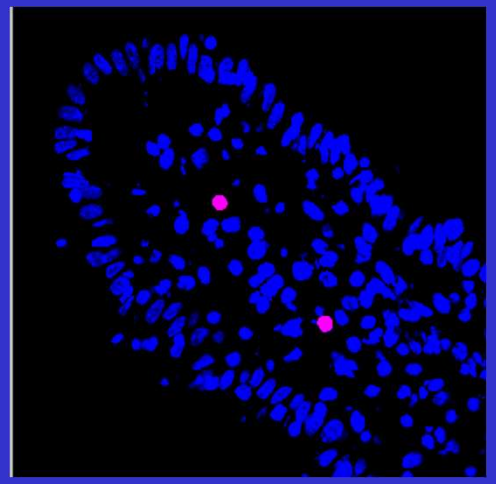




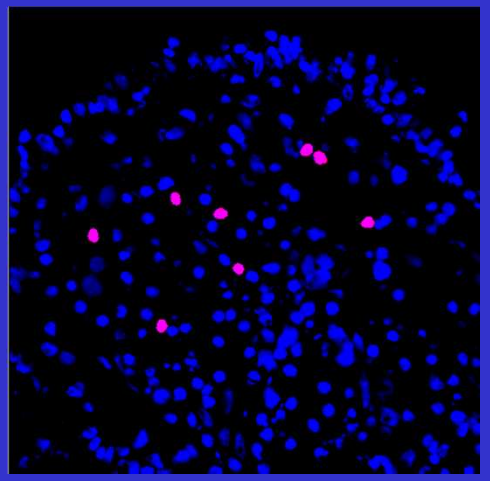
- ✓ **CD3+CD4+**
- ✓ **TCR α/β+**
- ✓ **B7 integrin+**
- ✓ **CD103^{neg}**
- ✓ **CD45RO+**

Gianfrani C. et al J Immunol 2006

Foxp3 expression in celiac mucosa:

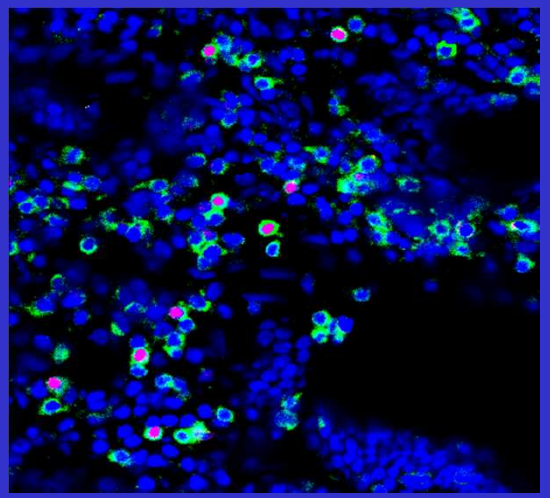


Control mucosa



Untreated mucosa

Foxp3+; CD4+



Mazzarella G. et al. submitted

T-regulatory cells in CD

No defect in the presence of T-cell regulatory cells in CD mucosa

- Tr1 cells are present in CD mucosa and not in controls and suppress the Th1 pathogenic T cells
- CD4+CD25+Foxp3+ are increased in CD mucosa might be to counteract the mucosal inflammation

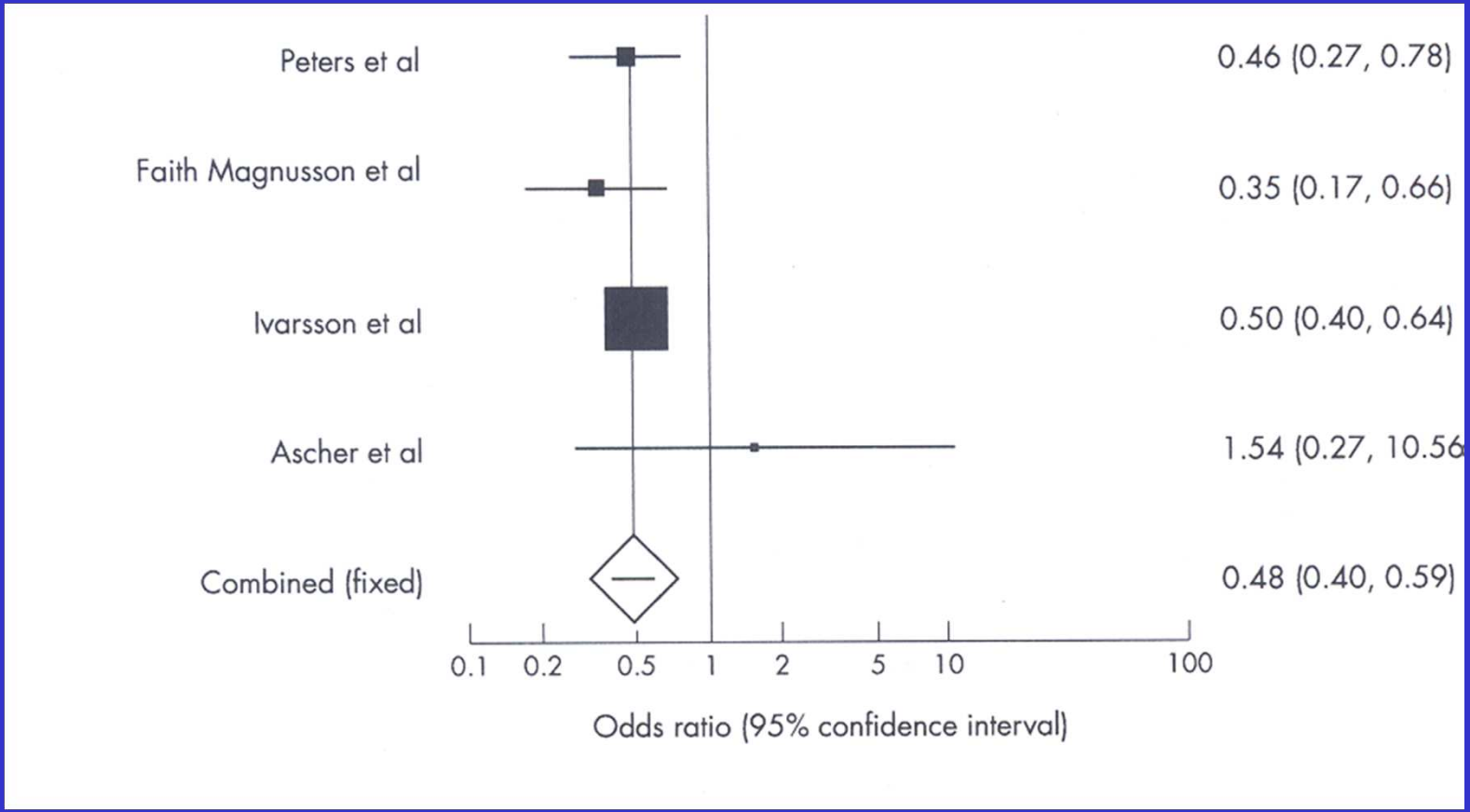
Genetics

Mechanisms

Prevention

Prevention is possible?

Breast feeding and coeliac disease
Metaanalysis of observational studies



Akobeng et al, Arch Dis Child 2006; 91:39-43

Early infant feeding practices and coeliac disease
Age at gluten introduction

In a population at risk for CD (HLA-DR3 positive), early exposition to gluten (before the 3rd month) or late exposition (after the 7th month) are associated with a significant increase in the production of antitissue transglutaminase antibodies (Norris, JAMA 2005)

Early infant feeding practices and coeliac disease Amount of gluten

- Sweden experienced a unique epidemic of celiac disease in children <2 years of age. The epidemic was partly explained by the increased proportion of infants introducing large amounts of gluten after breast feeding was ended (Olsson, Pediatrics 2008)

- The practice of introducing abruptly high amount of gluten, often without ongoing breast-feeding, might have contributed to the unexpectedly high prevalence of 3% recently found in this cohort (Myleus, JPGN 2009)

Recommendations

- Breast feeding for at least 6 months
- Gradual introduction of gluten in the diet, not before the 4th month of life, possibly when the child is still breast fed

And for those more at risk…

- ✓ Attribute the risk
- ✓ Active intervention?

Risk for a proband's brother according to DQ genotype of parents

	H1H1	H1H2	H1H3	H1H4	H1H5	H2H2	H2H3	H2H4	H2H5	H3H3	H3H4	H3H5	H4H4	H4H5	H5H5
H1H1			[8;29]	[8;29]	[8;29]		[8;29]	[8;29]	[8;29]						
H1H2		[7;29]	[8;29]	[7;29]	[1;29]	[7;29]	[7;29]	[7;29]	[1;29]	[8;24]	[7;24]	[1;24]			
H1H3			[1;29]	[1;29]	[1;29]		[1;29]	[1;29]	[1;29]						
H1H4				[7;29]	[1;29]	[7;29]	[1;29]	[7;2	[1;29]						
H1H5					[1;29]	[1;29]	[1;29]	[1;29]	[						
H2H2							[7;24]				[7;24]	[1;24]			
H2H3							[1;24]	[1;24]	[1;24]	[1;24]	[1;24]	[1;24]			
H2H4										[1;24]	[1;24]	[1;24]			
H2H5										[1;24]	[1;24]	[1;24]			
H3H3															
H3H4															
H3H5															
H4H4															
H4H5															
H5H5															

Risk evaluation based on parents' genotype

- Risk > 20%
- 15% < Risk < 20%
- 10% < Risk < 15%
- 1% < Risk < 10%
- Risk < 1%

Which possible intervention

- Delay the age at gluten introduction
- Give small amounts of gluten during breast feeding
- ?? Introduce gluten together with immunomodulatory molecules

Prevention of coeliac disease in at-risk babies
PREVENT-CD – 36383 – FP6

Protocol

ENROLLMENT
Families with at least one celiac member

BIRTH
HLA type in umbilical cord blood

Positive HLA DQ2/DQ8

- Breast feeding
- *Intervention* between 4th-6th month (100 mg gliadin/die)
- Gradual gluten introduction after 6th month
- Clinical and serologic controls every 3-6 months

Persistent positivity in serological tests

Clinical symptoms

INTESTINAL BIOPSY

Negative HLA DQ2/DQ8

Annual controls

Frontiers in Coeliac Disease

- Genetics: Clue to pathogenesis
- Pathogenesis: Gluten and activation innate immunity
- Clinical spectrum: CD and gluten sensitivity
- Diagnosis: New protocols
- Therapy: Alternative to GFD
- Prevention: Identification of at risk subjects and feeding pattern in the first year of life

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