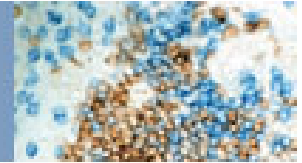


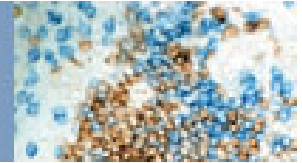
Autoimmunität

Overview

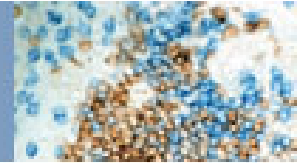


1. Introduction
2. Antigens, antibodies and B cells
3. Antigen presentation and T cells
4. Innate immunity
5. Adaptive immunity
6. Laboratory techniques
7. Tumor immunology
8. Hypersensitivity, Allergy – pathogenic immune reactions
9. **Autoimmune diseases**

Common autoimmune diseases



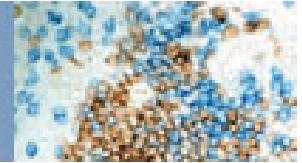
- Insulin-dependent diabetes mellitus (IDDM; Diabetes Typ 1)
- Multiple sclerosis (MS)
- Lupus erythematosus (Lupus erythematoses)
- Rheumatoid arthritis
- Ankylosing spondylitis (Morbus Bechterew)
- Grave's disease (Morbus Basedow)
- Myasthenia gravis



Definition

Diseases caused by failure of self-tolerance and subsequent immune reactions against self (autologous) antigens are called **autoimmune diseases**.

Autoimmune diseases can be caused by tolerance failure in **T cells** and/or in **B cells**.

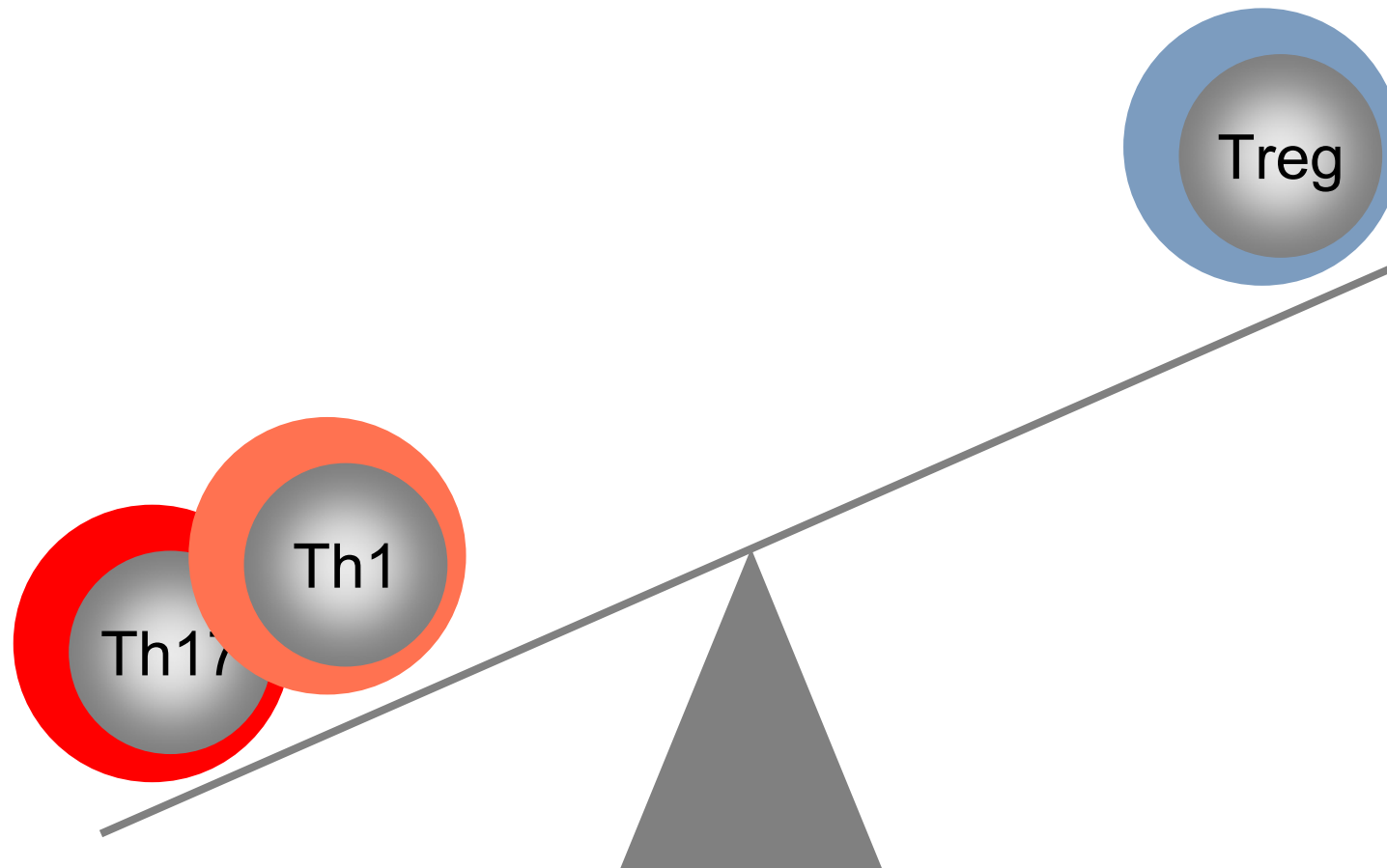
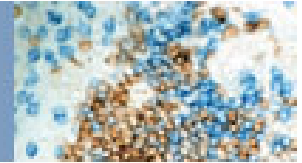


Definition

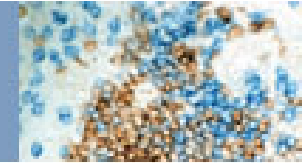
Tolerance is a physiologic state in which the immune system does not react **destructively** against the components of an organism that harbors it or against antigens that are induced to it.




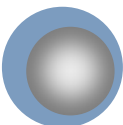

Destructive responses are prevented by a variety of mechanisms that operate during development of the immune system [**central tolerance**] *and* during the generation of each immune response [**peripheral tolerance**] .

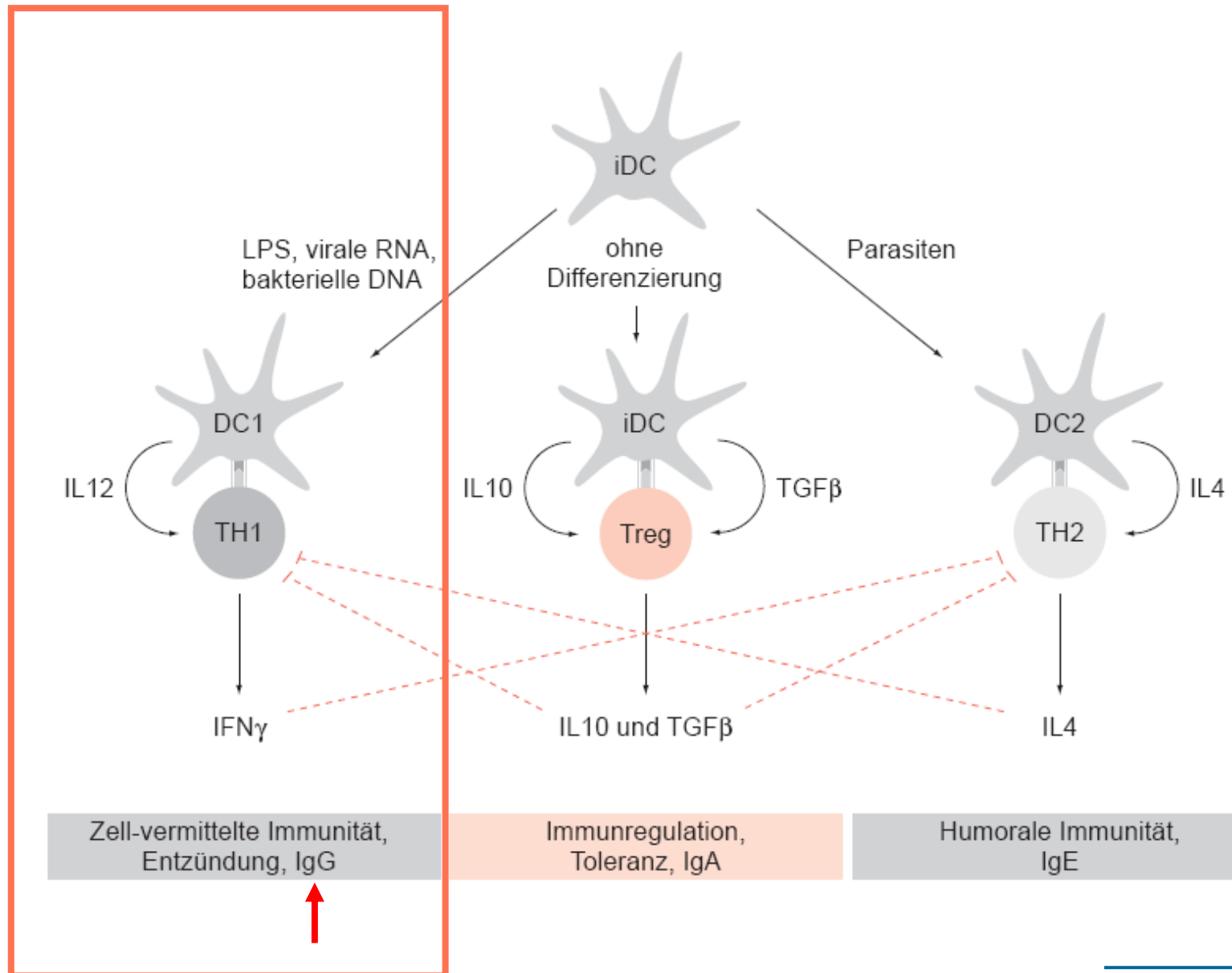
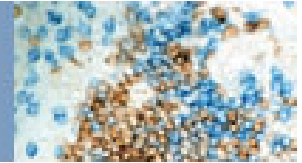
Dysbalance – Risk auf autoimmune disease



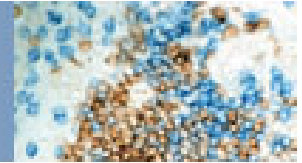
T cells come in different flavours



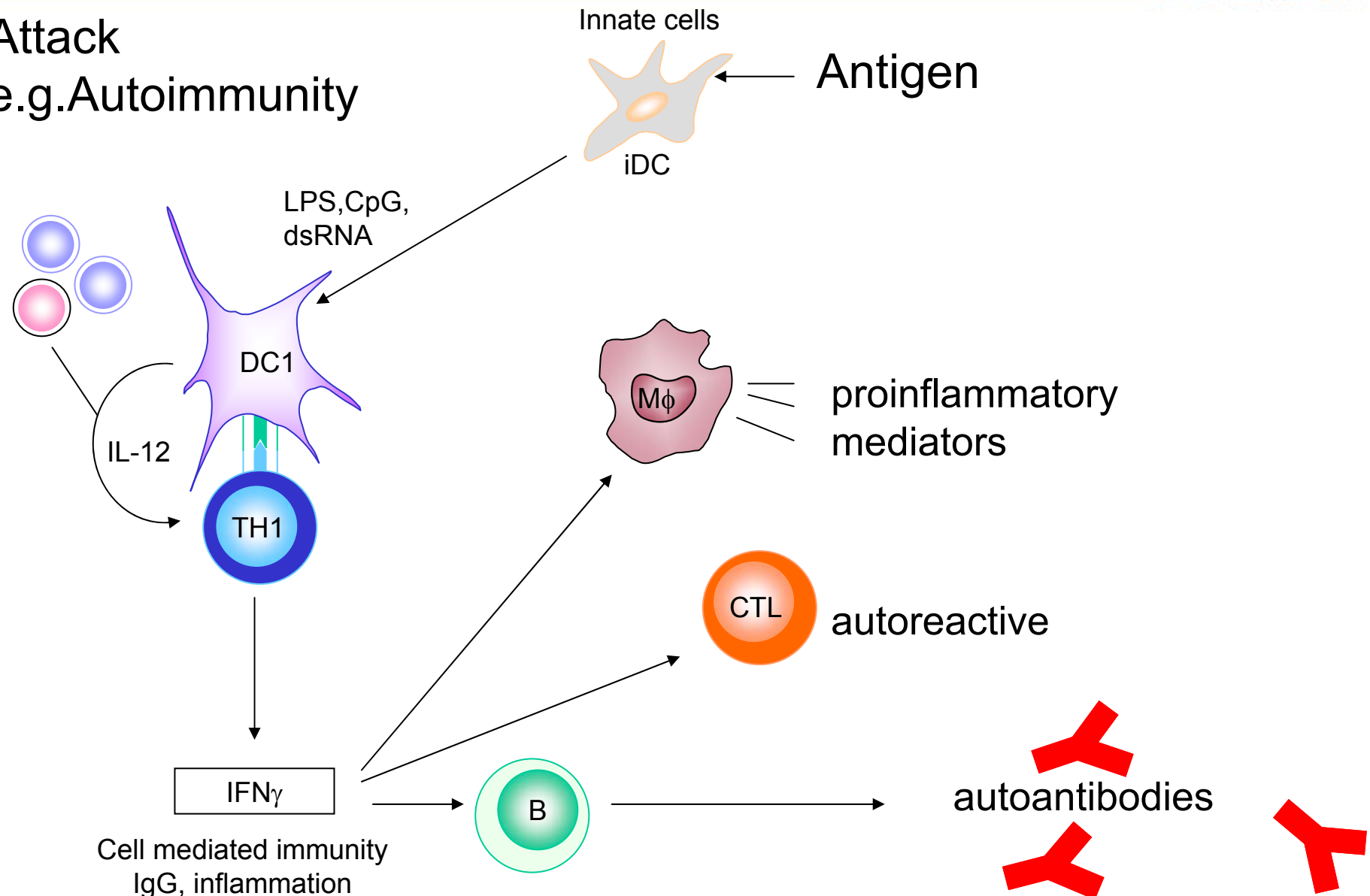
		Inducing cytokines	Lineage-specific transcription factor	Secreted cytokines
Th1		IL12 IFN γ	T-bet	IL2 IFN γ
Th2		IL4	GATA-3	IL4 IL5
Th17		TGF β + IL6	ROR γ T	IL17 IL6
Treg		TGF β	Foxp3	TGF β IL10
Tr1/Th3		IL10	?	IL10 TGF β



T helper cell differentiation



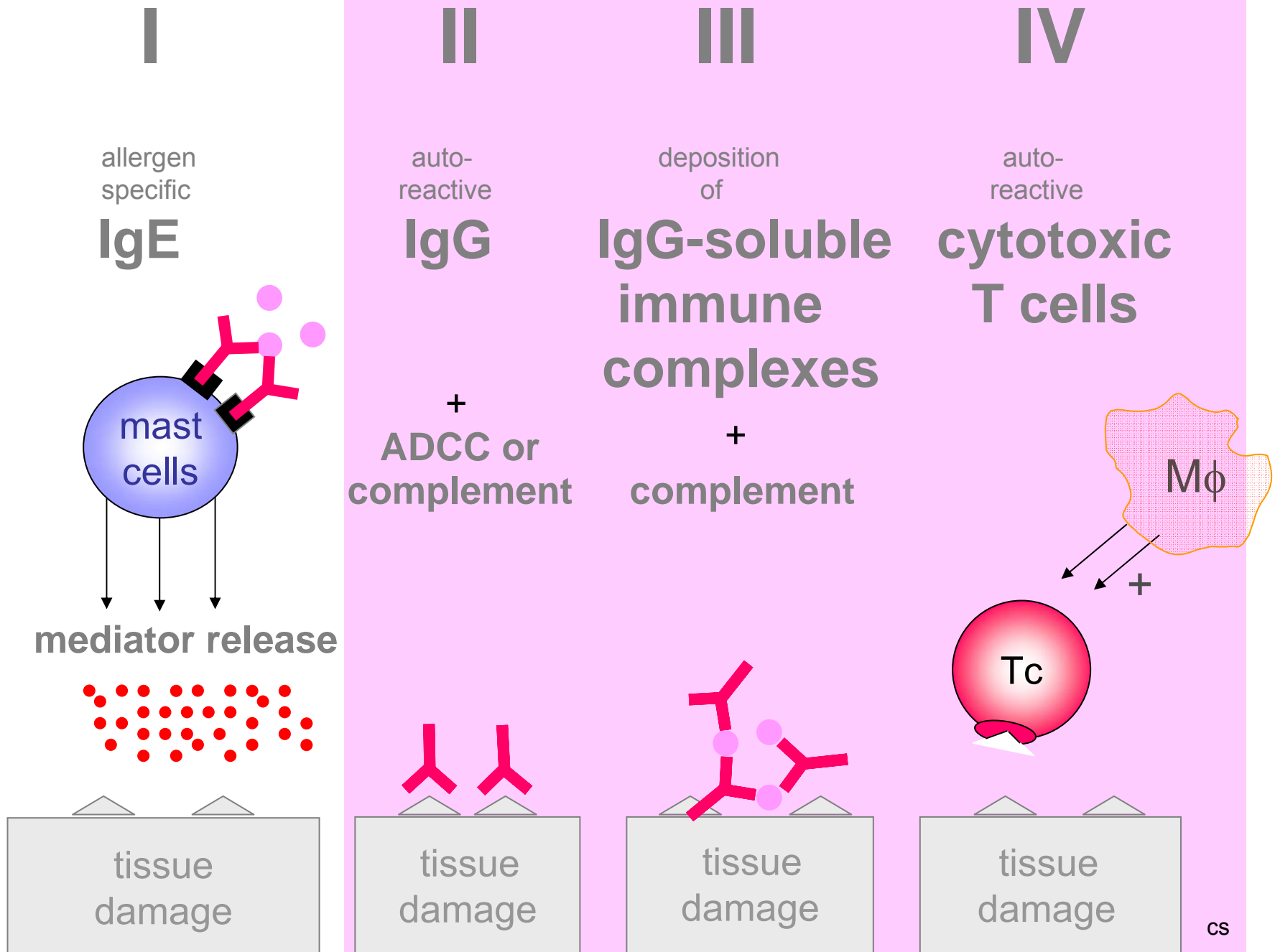
Attack
e.g. Autoimmunity



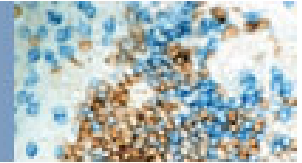
type		Antigen	Effector mechanism	Allergy	Graft rejection	Autoimmune disease
I	IgE	Soluble	Mast cell activation	Allergic rhinitis Allergic asthma Urticaria (Wheal and flare) Systemic anaphylaxis	None	None
II	IgG	Cell- or matrix-associated	Complement, FcR+ cells (phagocytes, NK cells)	Some drug allergies, where drug binds to cell surface, eg penicillin	Transfusion reaction (antibody to donor erythrocytes) Graft rejection (antibody to blood vessel wall)	Autoimmune hemolytic anemia Goodpasture's syndrome
	IgG	Cell surface receptor	Antibody alters cell signalling	None	None	Myasthenia gravis (Antagonist) Graves' disease (agonist)
III	Immune complexes	soluble	Complement, FcR+ cells (phagocytes, NK cells)	Serum sickness Farmer's lung	None	Systemic lupus erythematosus
IV	Th1	Soluble	Macrophage activation	Contact dermatitis Tuberculin reaction	Chronic graft rejection	Rheumatoid arthritis Multiple sclerosis
	Th2	Soluble	IgE; eosinophil activation, mastocytosis	Chronic asthma, Chronic allergic rhinitis	None	None
	CTL	Cell-associated	Cytotoxicity	None	Acute graft rejection	Diabetes mellitus

Summary: Pathogenic Immune Reactions Type I - IV

Autoimmune diseases



Breaking tolerance



Initially, tolerance for one or a few epitopes is lost

- spontaneously
- after infection
- after tissue destruction
- experimentally by immunization

The concept of **molecular mimikry**:

Infectious agents share epitopes with self antigens and activate autoreactive cells.

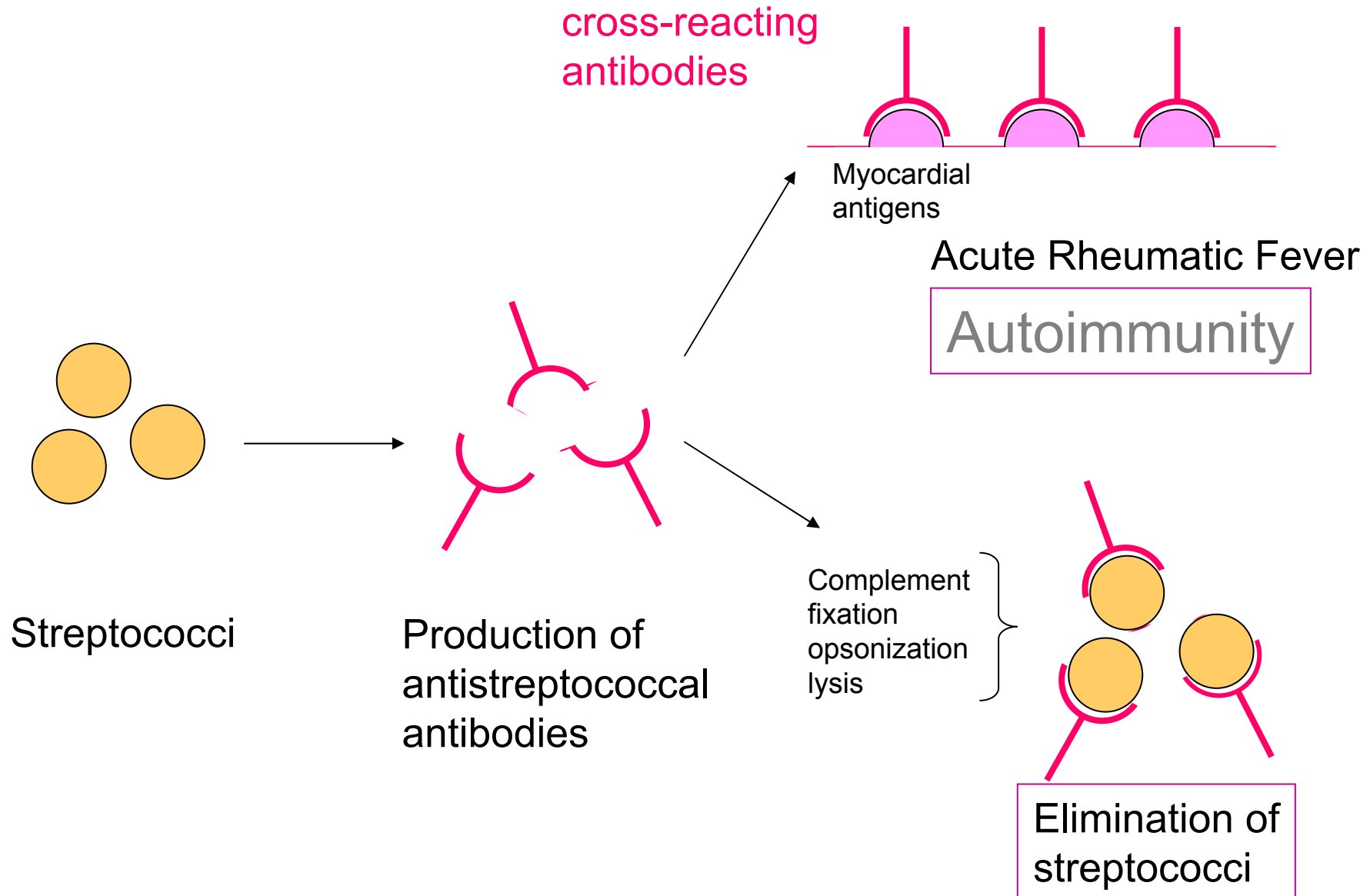
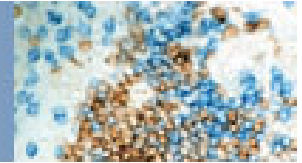
The concept of bystander-activation:

A local inflammation lowers the activation threshold of autoreactive T cells in a non-specific way.

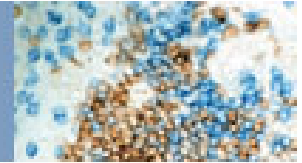
Loss of control mechanisms:

Any impairment of the downregulatory mechanisms of the immune system increases the risk of auto-immune disease.

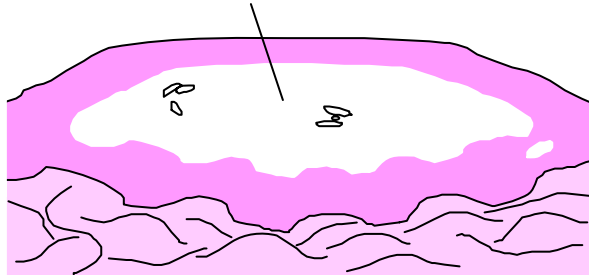
Cross-reacting antibodies



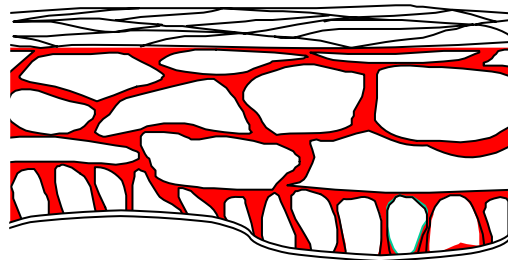
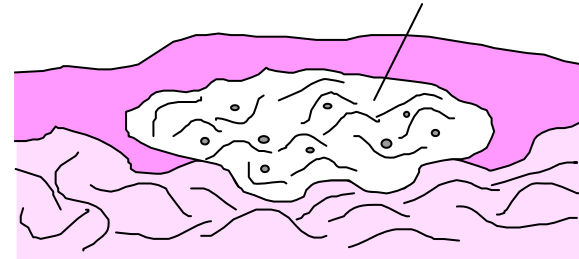
Blister formation



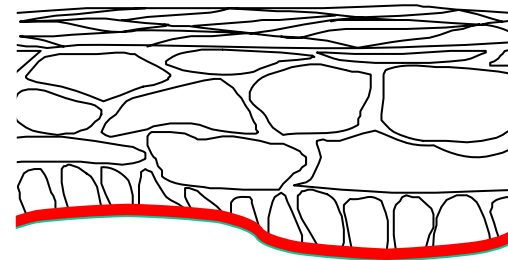
intraepidermal blister formation



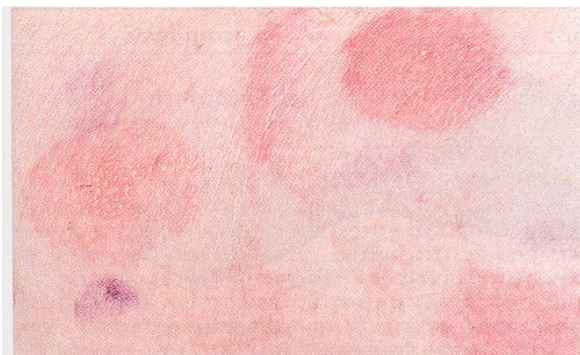
subepidermal blister formation



desmoglein - autoantibodies



BP - autoantibodies



Pemphigus vulgaris



Bullous Pemphigoid

Autoimmunity (Type II-IV)

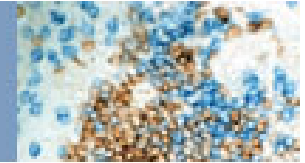
Some common autoimmune diseases classified by immunopathogenic mechanisms

Type II antibody to cell-surface or matrix antigens		
Syndrome	Autoantigen	Consequence
Autoimmune hemolytic anemia	Rh blood group antigens, I antigen	Destruction of red blood cells by complement and FcR ⁺ phagocytes, anemia
Autoimmune thrombocytopenic purpura	Platelet integrin GpIIb:IIIa	Abnormal bleeding
Goodpasture's syndrome	Noncollagenous domain of basement membrane collagen type IV	Glomerulonephritis pulmonary hemorrhage
Pemphigus vulgaris	Epidermal cadherin	Blistering of skin
Myasthenia gravis	Blocking autoantibodies against the acetylcholine - receptor	no muscle contraction

Type III immune-complex disease		
Syndrome	Autoantigen	Consequence
Mixed essential cryoglobulinemia	Rheumatoid factor IgG complexes (with or without hepatitis C antigens)	Systemic vasculitis
Systemic lupus erythematosus	DNA, histones, ribosomes, snRNP, scRNP	Glomerulonephritis, vasculitis, rash
Rheumatoid arthritis	Rheumatoid factor IgG complexes	Arthritis

Type IV T cell-mediated disease		
Syndrome	Autoantigen	Consequence
Insulin-dependent diabetes mellitus	Pancreatic β -cell antigen	β -cell destruction
Rheumatoid arthritis	Unknown synovial joint antigen	Joint inflammation and destruction
Experimental autoimmune encephalomyelitis (EAE), multiple sclerosis	Myelin basic protein, proteolipid protein, myelin oligodendrocyte glycoprotein	Brain invasion by CD4 T cells, weakness

Diagnosis of autoimmune diseases



- Medical history (Anamnese)
- Clinical examination
- Screening for autoantibodies
 - Binding to HEp2 cells (human epithelial cell line)
 - Binding to different tissue sections, such as
 - monkey muscle (anti-muscular autoantibodies)
 - monkey pancreas (anti-islet cell antibodies)
 - ELISA for binding to typical autoantigens, such as
 - phospholipids
 - citrullinated proteins

immune fluorescence

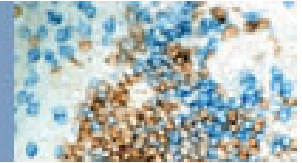


ELISA



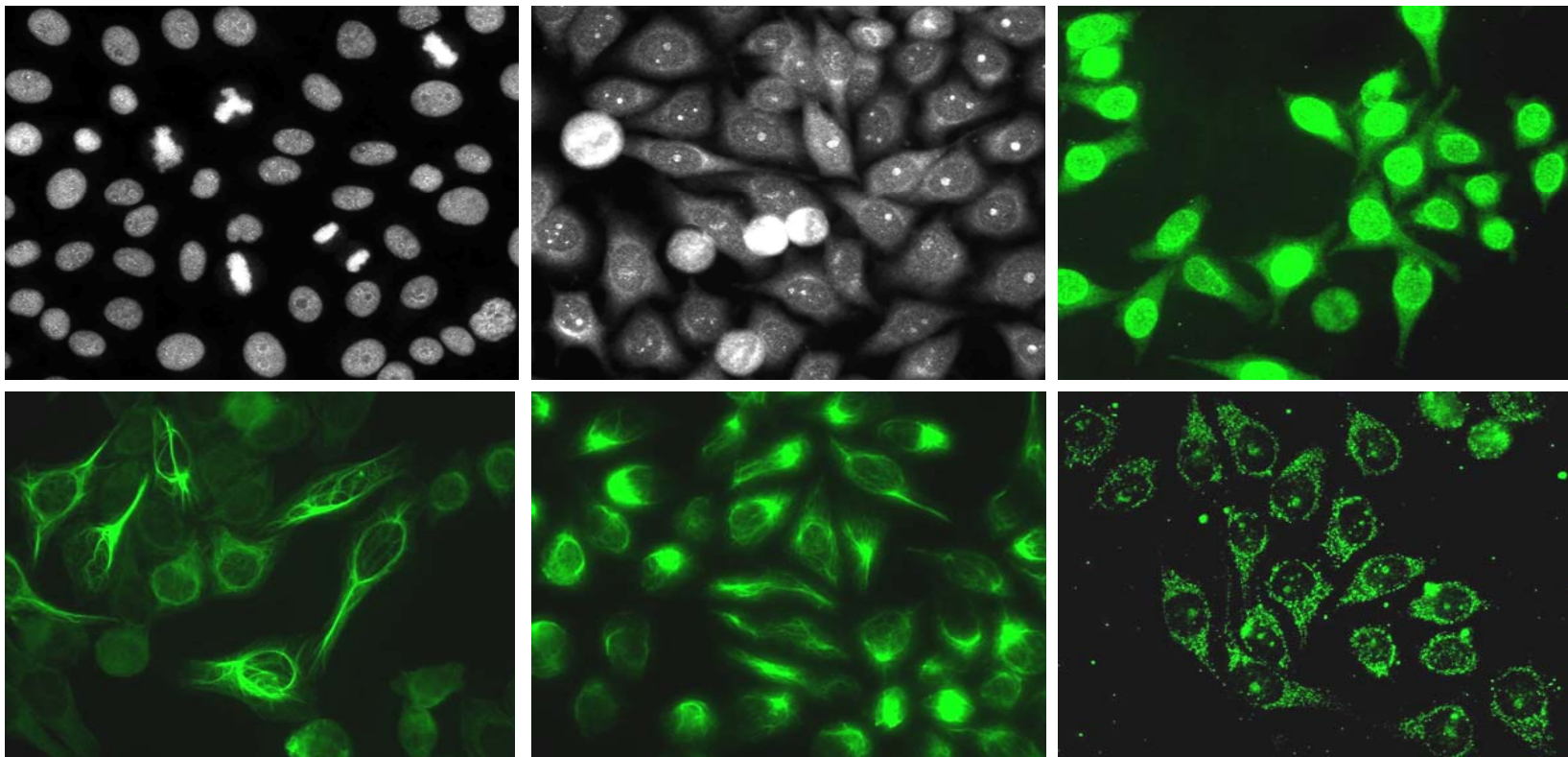
Praktikumsversuch

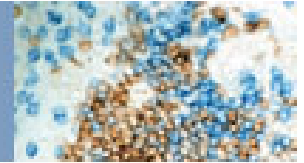
Test antibodies for autoreactivity



Binding to Hep-2 cells = screening test for autoreactive antibodies

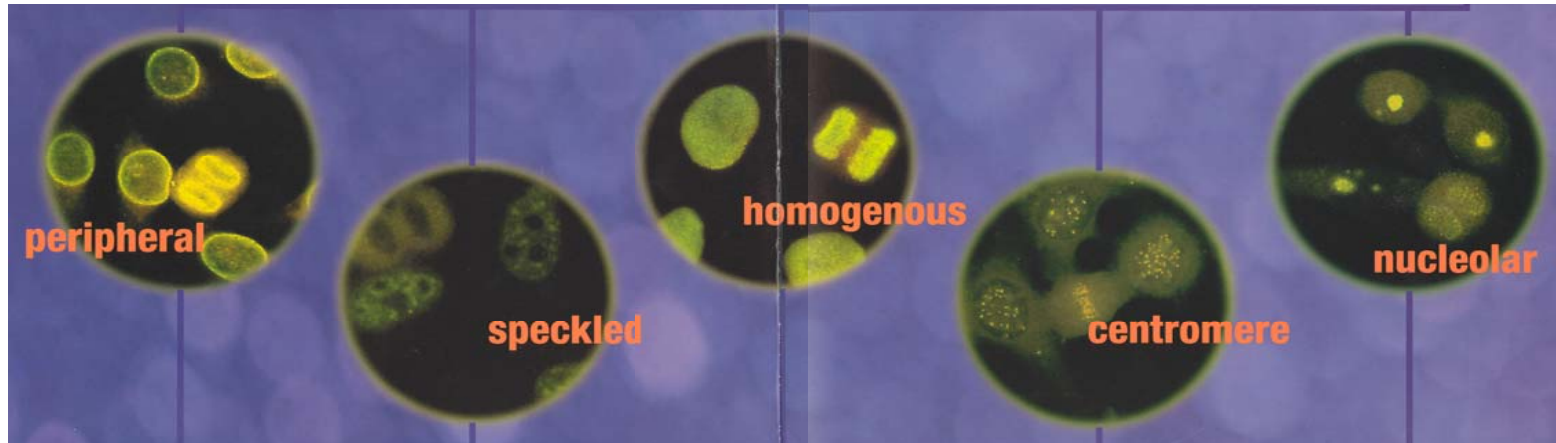
- e. g. nuclear antigens
- cytoskeleton
- cytoplasmic antigens ...



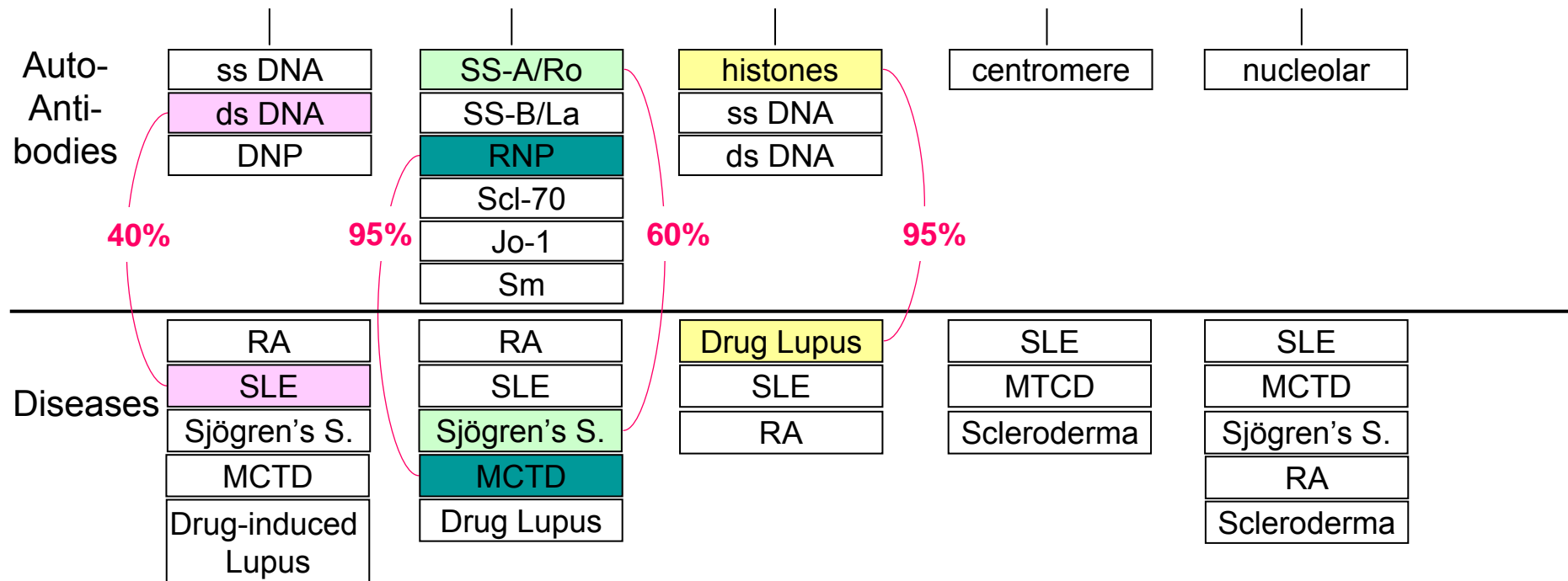


- Autoantibodies helpful for diagnosis
- Not always main cause of disease symptoms

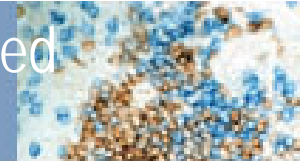
Measurement of autoAbs in certain diseases



Do these autoantibodies cause autoimmunity? No!



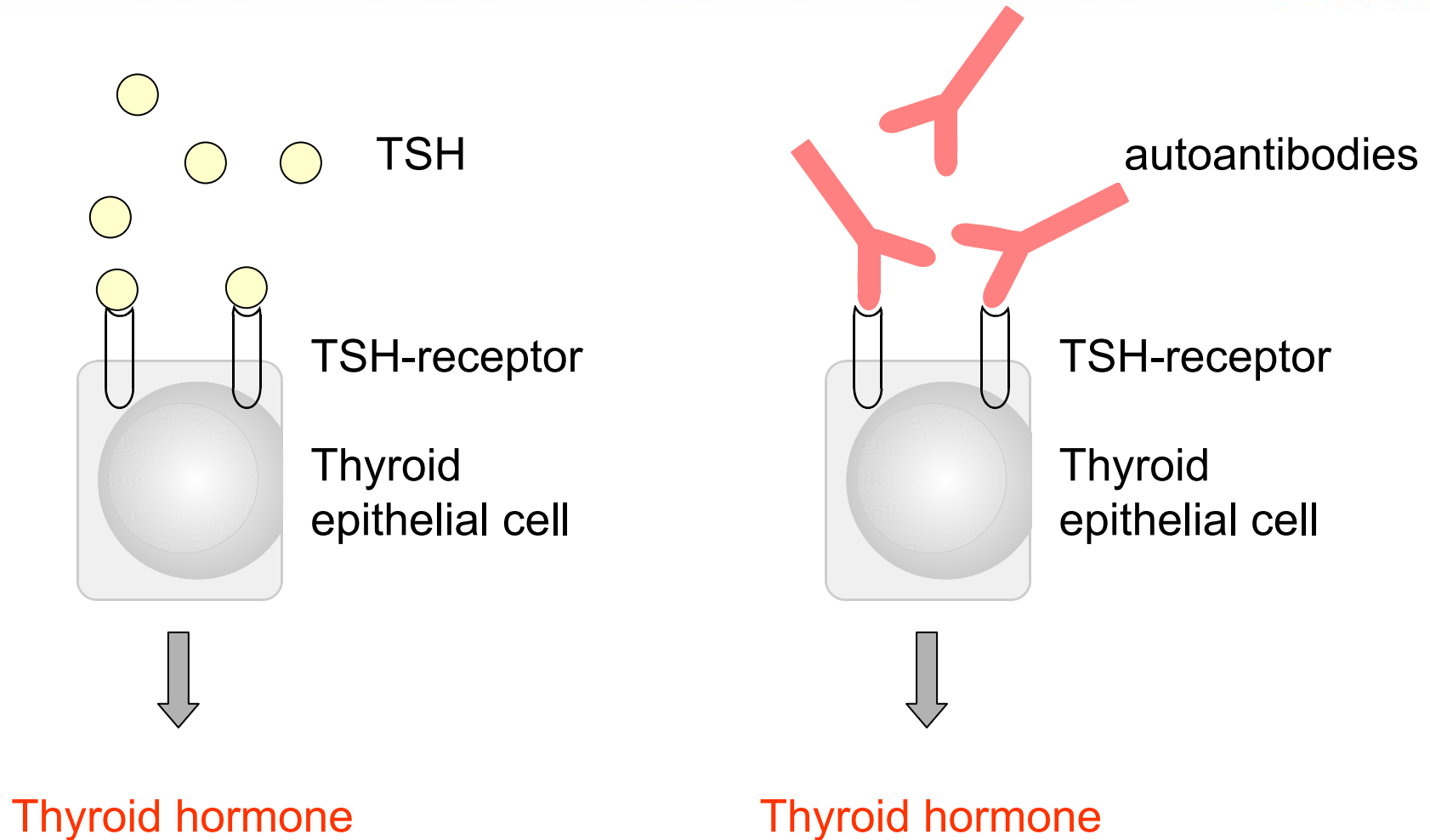
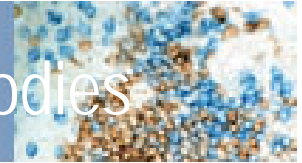
Prevalence (%) of autoantibodies in endocrine system-related autoimmune disorders



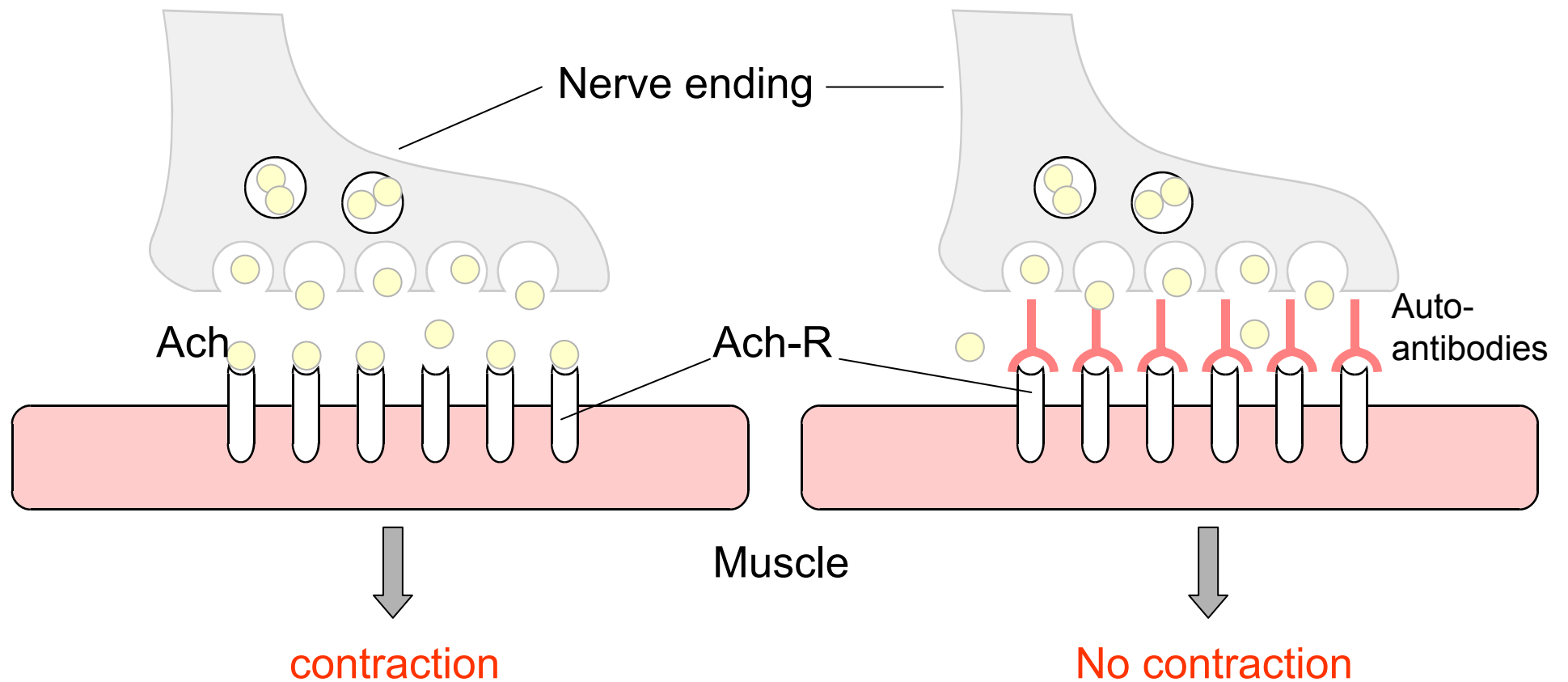
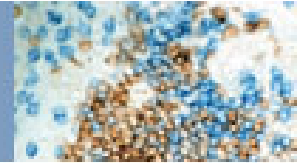
TSH receptor	95	-	?	-	-	-	-
tyroglobulin	80	95	95	-	10	30	9
thyroid cell microsomes	80	95	95	40	10	30	23
gastric parietal cells	50	50	50	33	10	90	21
pancreatic islet cells	5	5	5	5	70	-	-
adrenal cortical cells	5	5	5	65	-	5	4
gonadal cells	-	-	-	16	-	-	-
	Graves' disease	Hashimoto's thyroiditis	atrophic hypothyroidism	Addison's disease	IDDM	Pernicious anemia	Vitiligo

Do these autoantibodies cause autoimmunity?

Grave's disease (M. Basedow): Stimulating autoantibodies

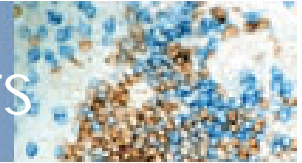


Myasthenia gravis: Blocking autoantibodies

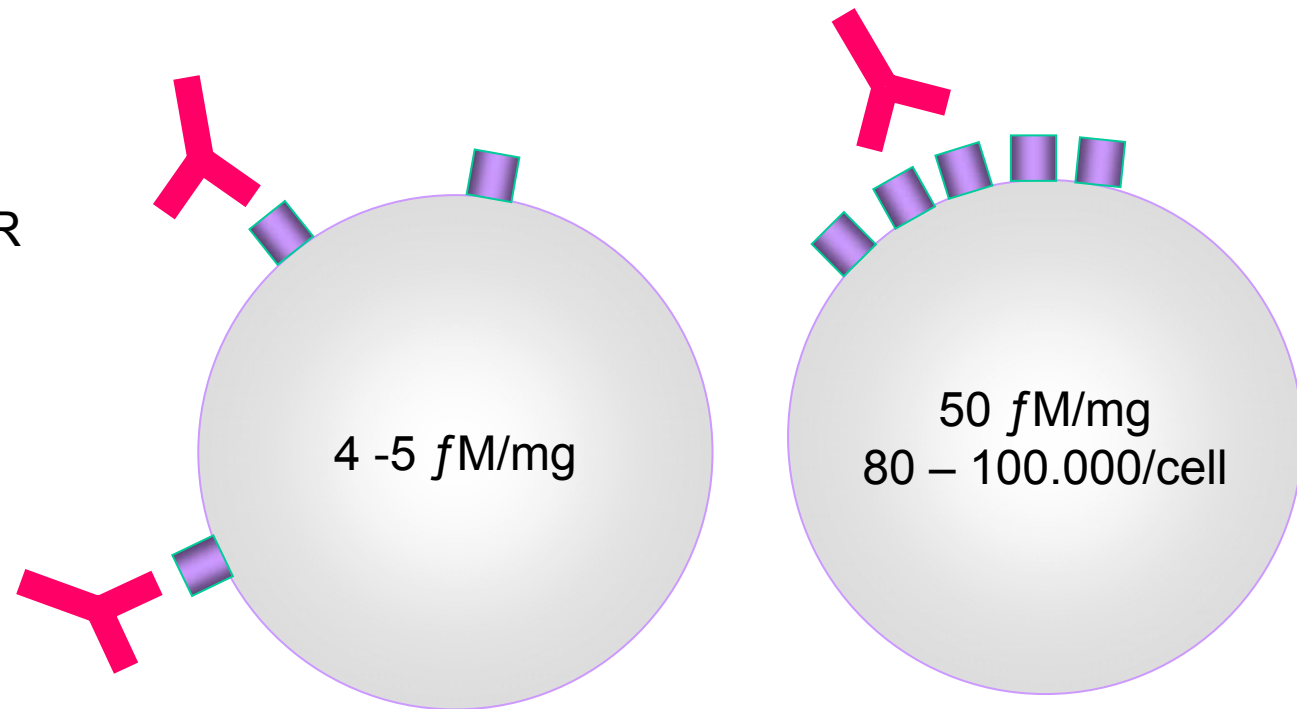


Ach: Acetyl choline; Ach-R: Acetyl choline receptor

Autoantibodies against G-protein coupled receptors



Serotonin (5HT₄)-R
Angiotensin (AT1)-R
Acetylcholin (Ach M2)-R
β₁-adrenoreceptor

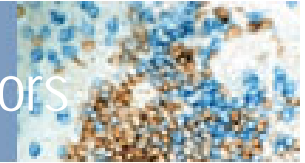


blocking antibody
(antagonist)

stimulating antibody
(agonist)

depending on the density of the receptor !

Agonistic autoantibodies against G-protein- coupled Receptors



→ β 1-adrenergic Receptors
(β 1R)

Chagas' Disease
Dilated Cardiomyopathy
Hypertension (therapy refractory)
Arrhythmia
Vascular diseases

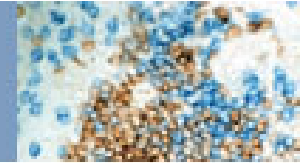
→ Angiotensin II Type 1 Receptors
(AT1R)

Preeclampsia
Acute vascular rejection in the
absence of immunological risk

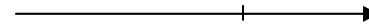
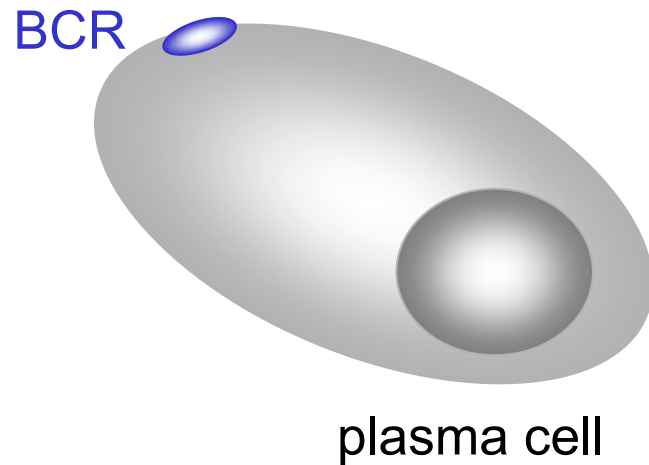
→ Protease activated Receptors
(PAR)

M. Raynaud
(Atopic dermatitis)

Therapy – Removal of all immunoglobulins?

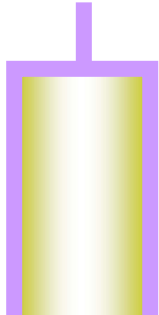
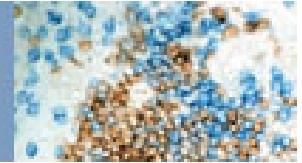


Plasma cells are highly resistant to immune suppressive and cytotoxic drugs

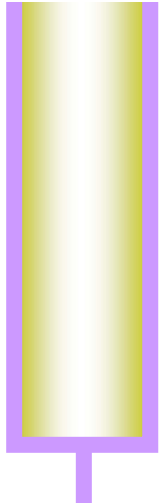


autoantibodies

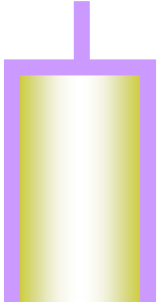
Antibody-coated extra-corporeal devices



Removal of continuously produced molecules?



Auto-antibodies

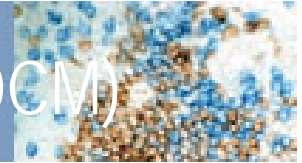


Removal of an acute excess of molecules



toxins, drugs

Agonistic autoantibodies in dilated cardiomyopathy (DCM)

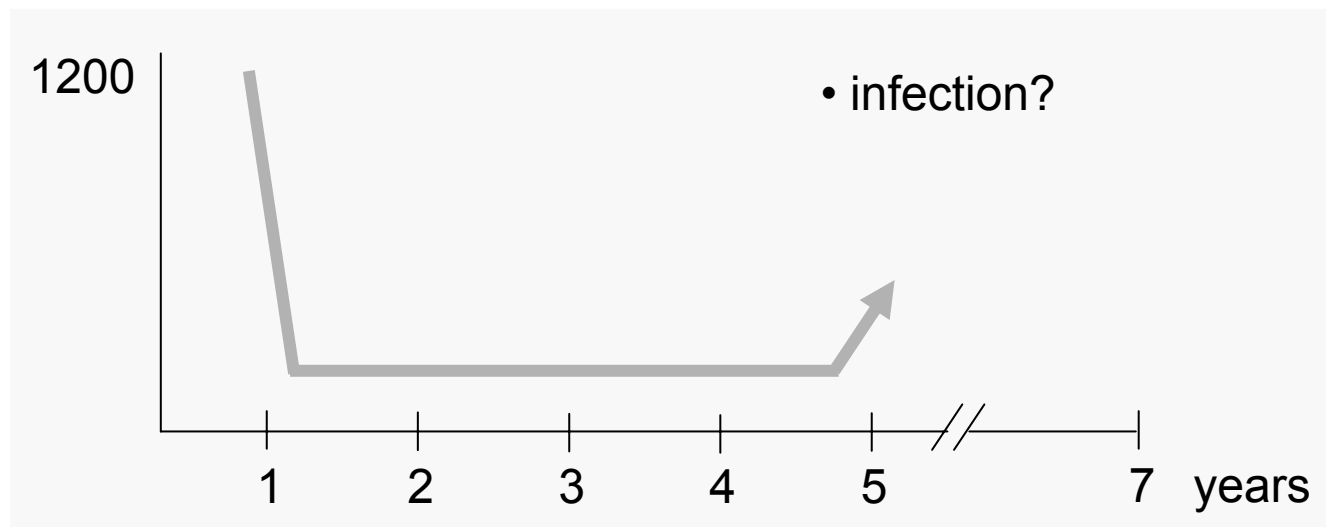


anti-muscarinic M2R Ab
anti- β_1 adrenoreceptor Ab

β -blocker prevent autoantibody effects!

Extracorporeal Immunoabsorption with anti-human IgG

1x/d*, 5d + Ig substitution (IVIg)

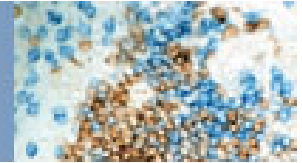


* each session removes
30-40% of total IgG

80% of DCM patients: autoantibodies ; survival rate ~ 50% !

7% of patients suffering from ischemic heart disease : autoantibody +

Breaking tolerance



Initially, tolerance for one or a few epitopes is lost

- spontaneously
- after infection
- after tissue destruction
- experimentally by immunization

The concept of molecular mimikry:

Infectious agents share epitopes with self antigens and activate autoreactive cells.

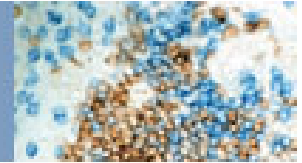
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Loss of control mechanisms:

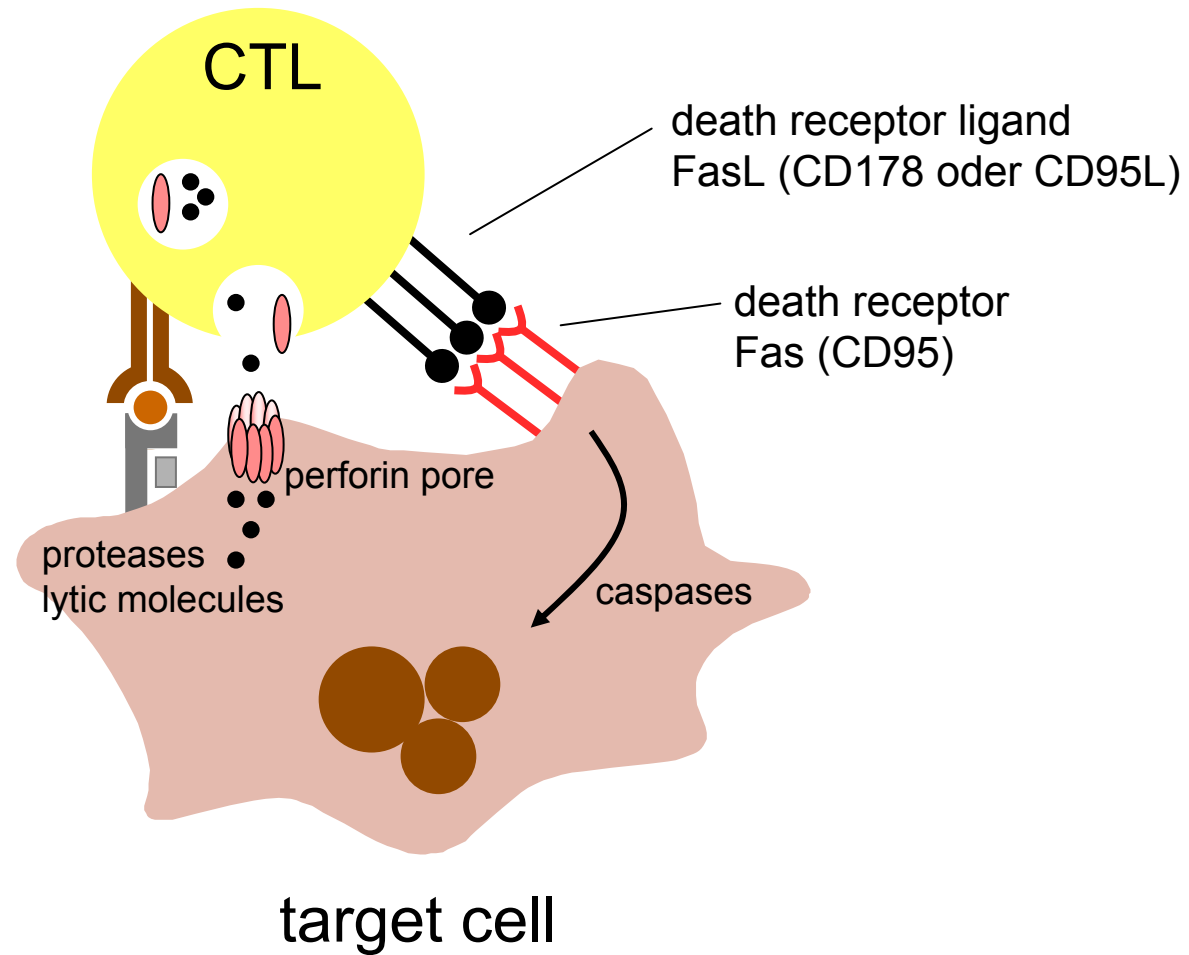
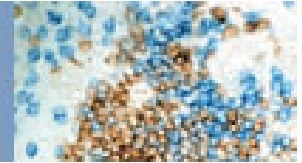
Any impairment of the downregulatory mechanisms of the immune system increases the risk of auto-immune disease.

Loss of control mechanisms

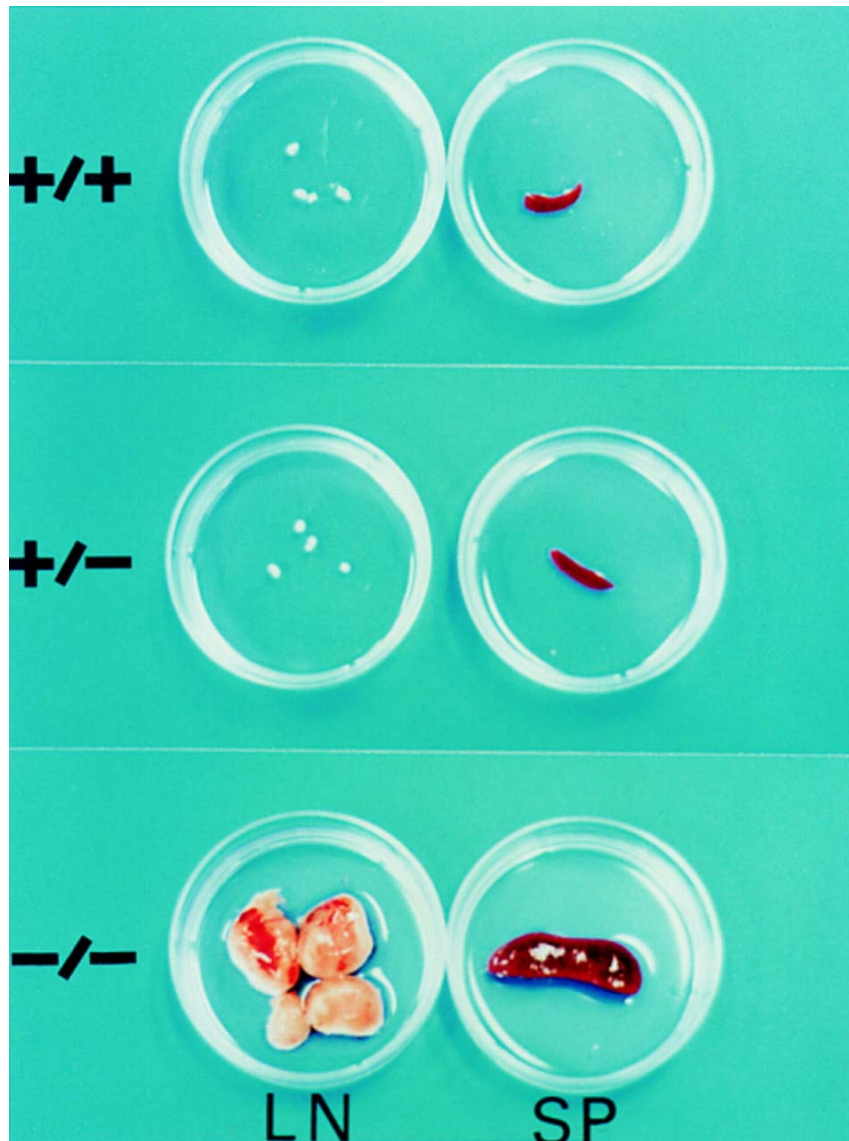
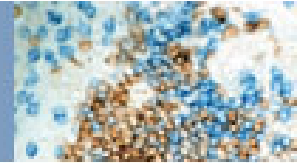


- All changes which impair the dampening mechanisms of the immune system confer an increase risk of autoimmune disease.
- Loss of function of the mechanisms of
 - Apoptosis
 - Anergy
 - Suppression

Death signals and perforin: The weapons of cytolytic lymphocytes



When apoptosis fails



+/+

Wild type (+/+)

+/-

heterozygous (+/-)

-/-

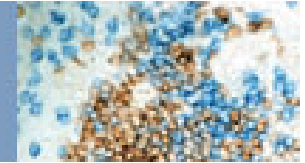
Fas-ko (-/-)

Lymph nodes and spleen from 16 week old mice

Nagata, Cell 88: 355-365 (1997)

B. Bröker, Uni Greifswald

Two extreme examples



ALPS – failure of cell death

Autoimmune lymphoproliferative syndrome

mutations in the **Fas**-gene.

- manifestation in childhood
- lymphnode swelling
- spleno- and hepatomegaly
- autoimmunity – hypergammaglobulinemia, autoimmune hemolytic anemia, thrombocytopenia, neutropenia
- 50fold increased risk of lymphoma

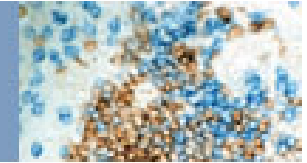
IPEX – failure of immune regulation

Immunodysregulation polyendocrinopathy enteropathy X-linked syndrome

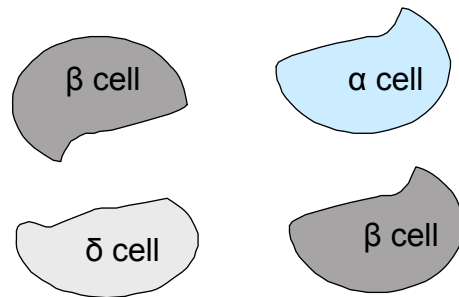
dysfunction of **Foxp3** → dysfunction of Treg

- manifestation early in childhood
- chronic diarrhea
- IDDM, autoimmune thyroiditis, autoimmune hemolytic anemia
- eczema, erythroderma, psoriasis, alopecia universalis
- sometimes atopia such as food allergy, increased IgE serum concentrations

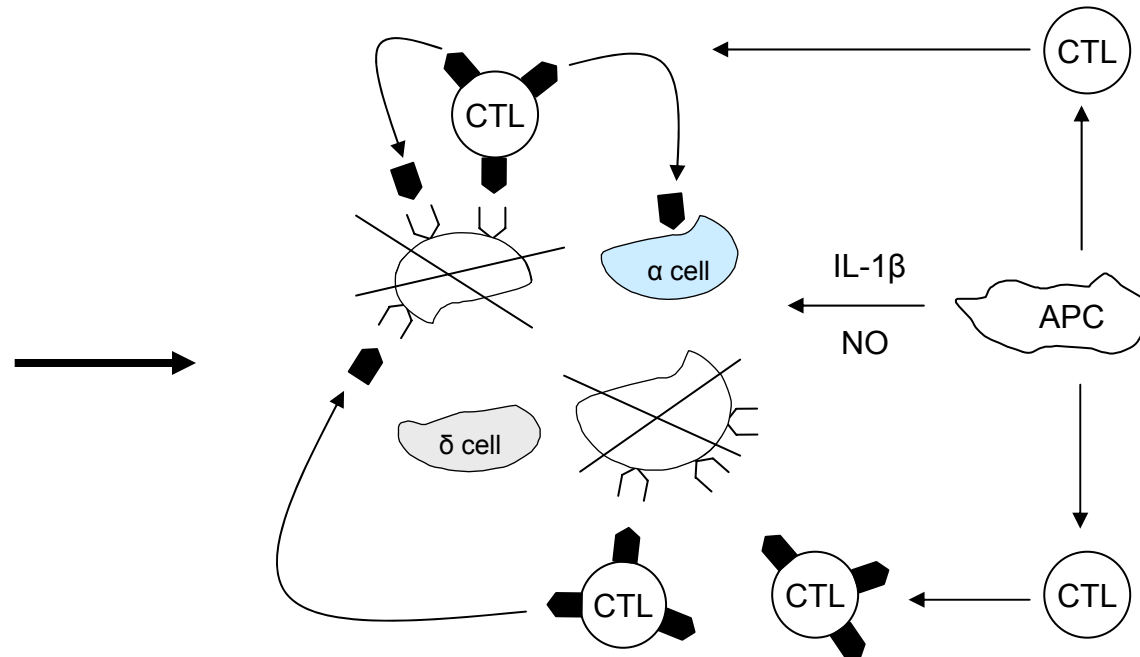
Insulin - dependent Diabetes mellitus (IDDM)



(a) Normal islet



(b) IDDM islet

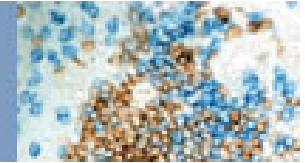


(a) Normal islet cells do not express Fas.

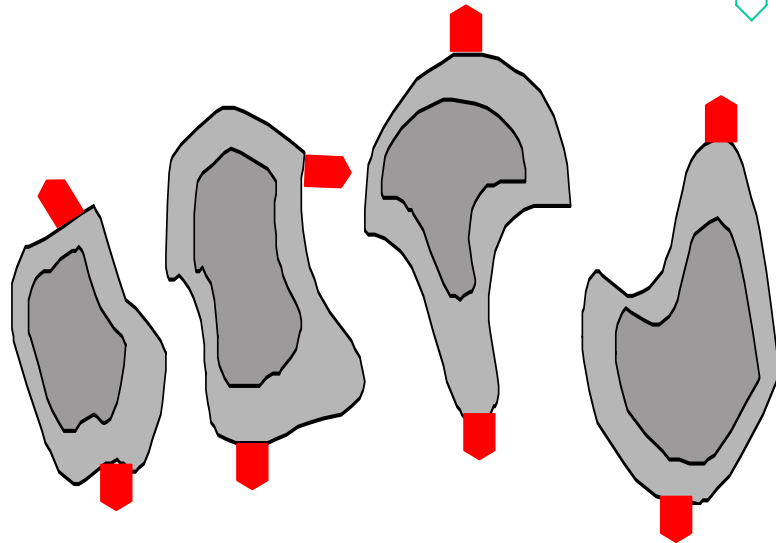


(b) Following an initial specific reaction causing limited damage to the β -cell mass, NO-induced Fas expression primes β cells for destruction through the secondary polyclonal T-cell response. CTLs are responsible for specific death of bystander Fas⁺ β cells by membrane-bound or released FasL, sparing Fas⁻ α and δ cells.

In contrast: Hashimoto's thyroiditis



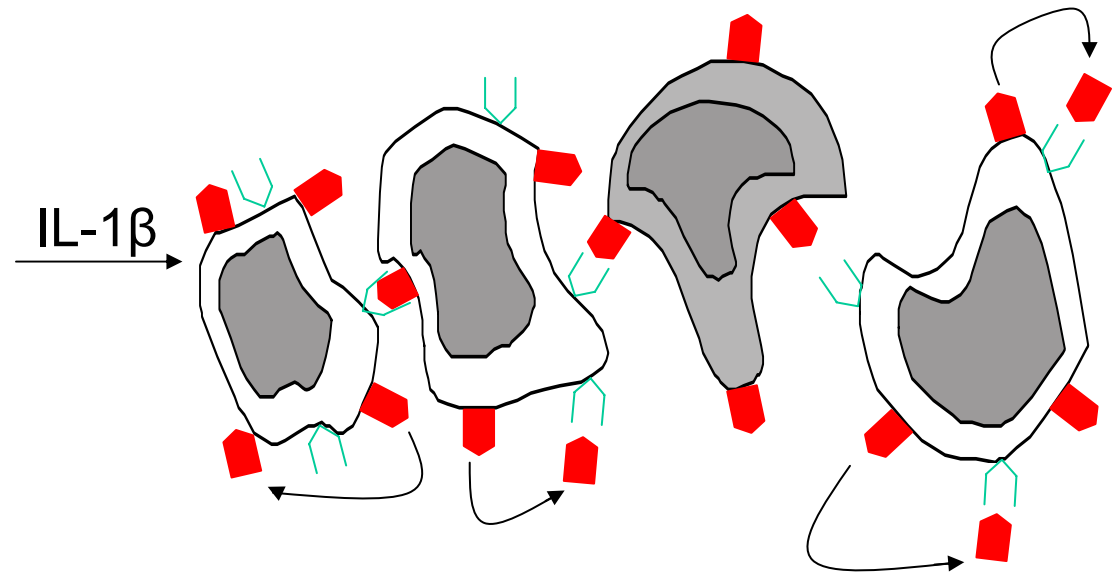
(a) Normal thyrocytes



Normal thyrocytes produce FasL but express negligible amounts of Fas (immune privileged site!).

■ FasL
∪ Fas

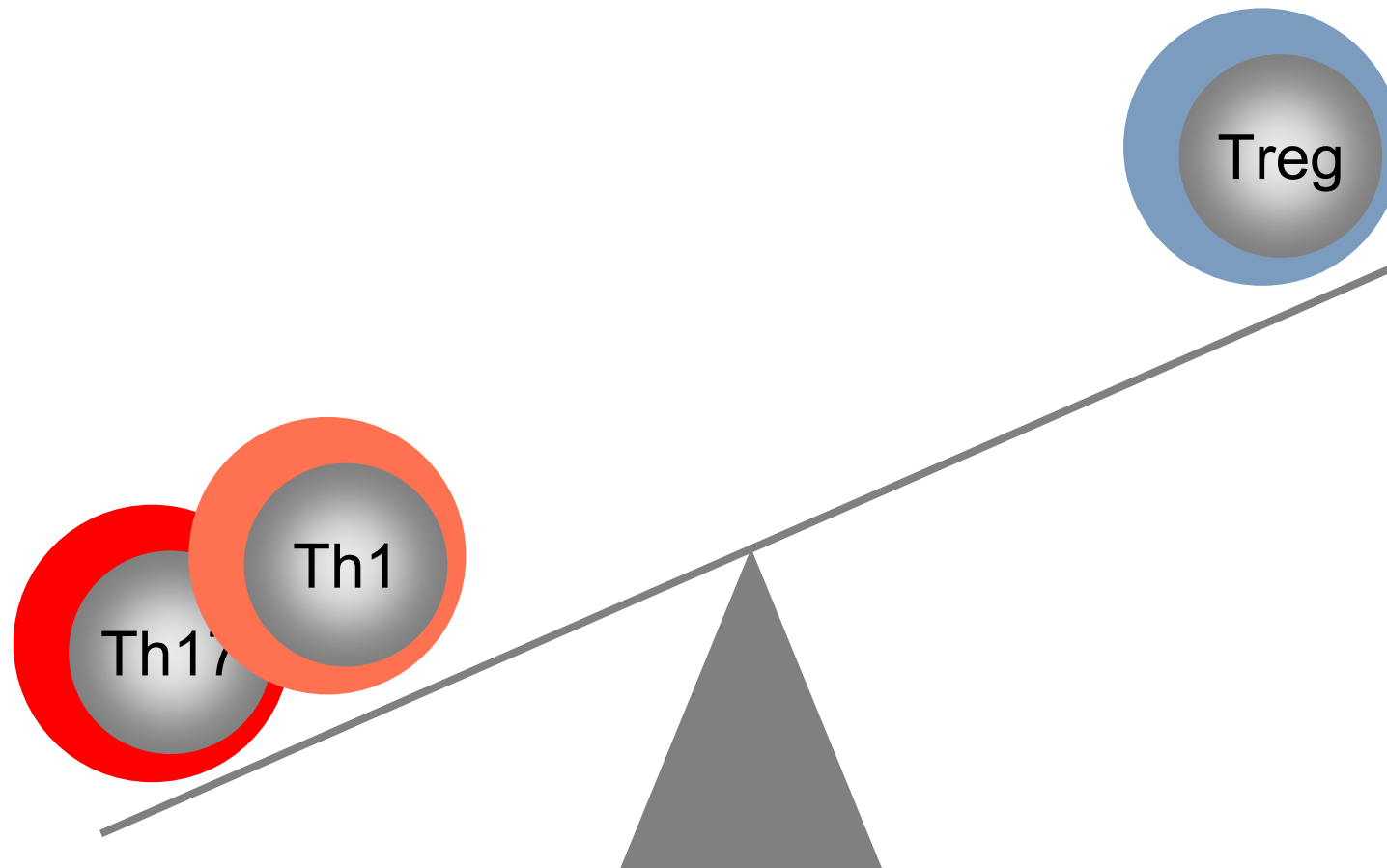
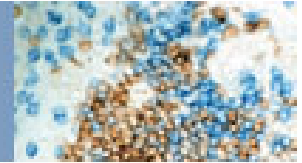
(b) Hashimoto's thyrocytes

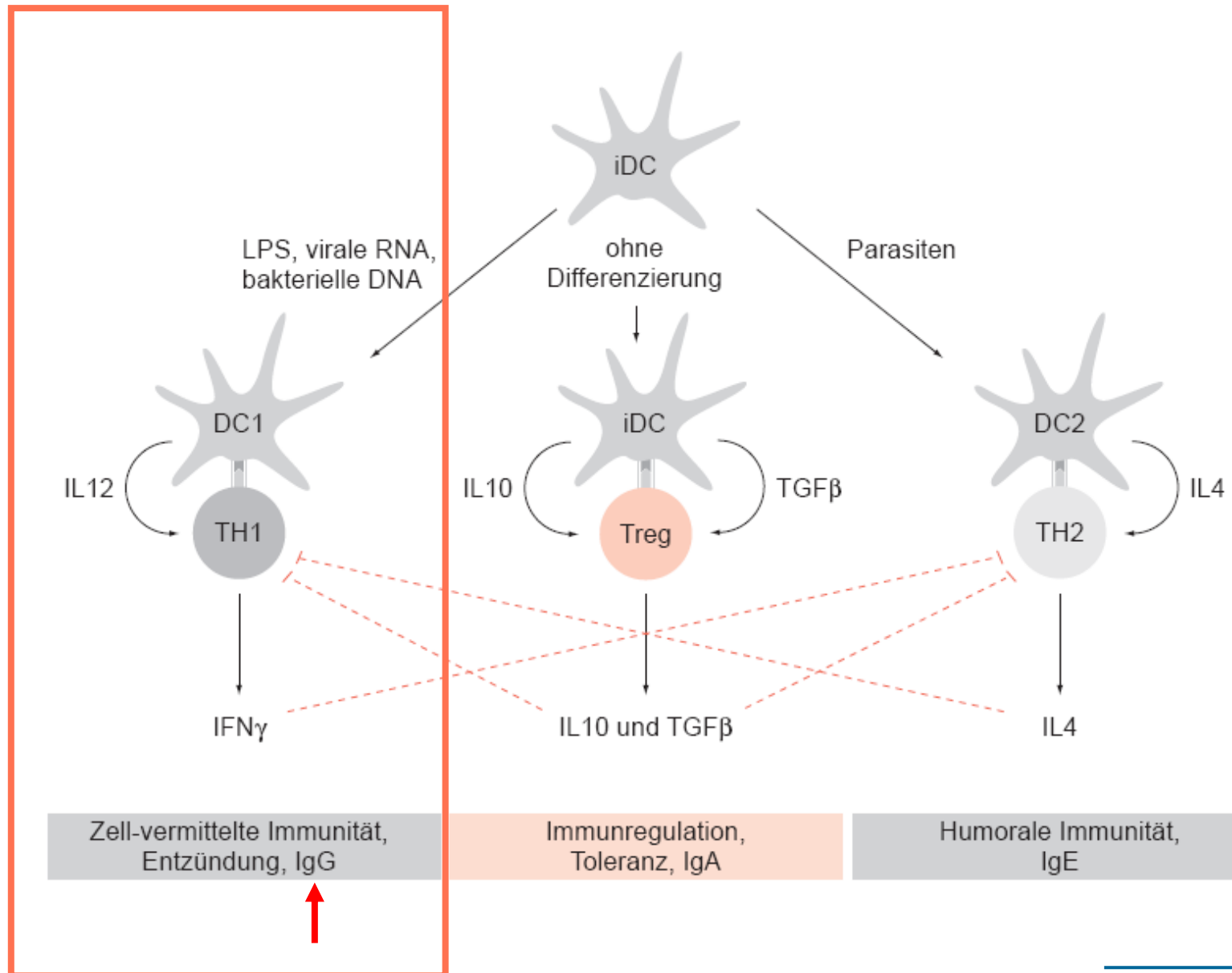
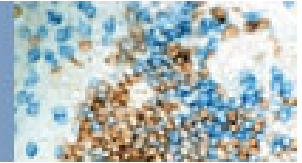


However, following inflammation, IL-1 β induces inappropriate Fas upregulation and consequent apoptosis through auto-crine or para-crine FasL production, leading to Hashimoto's thyroiditis. (no TSH receptor antibodies, but microsomal antibodies!)

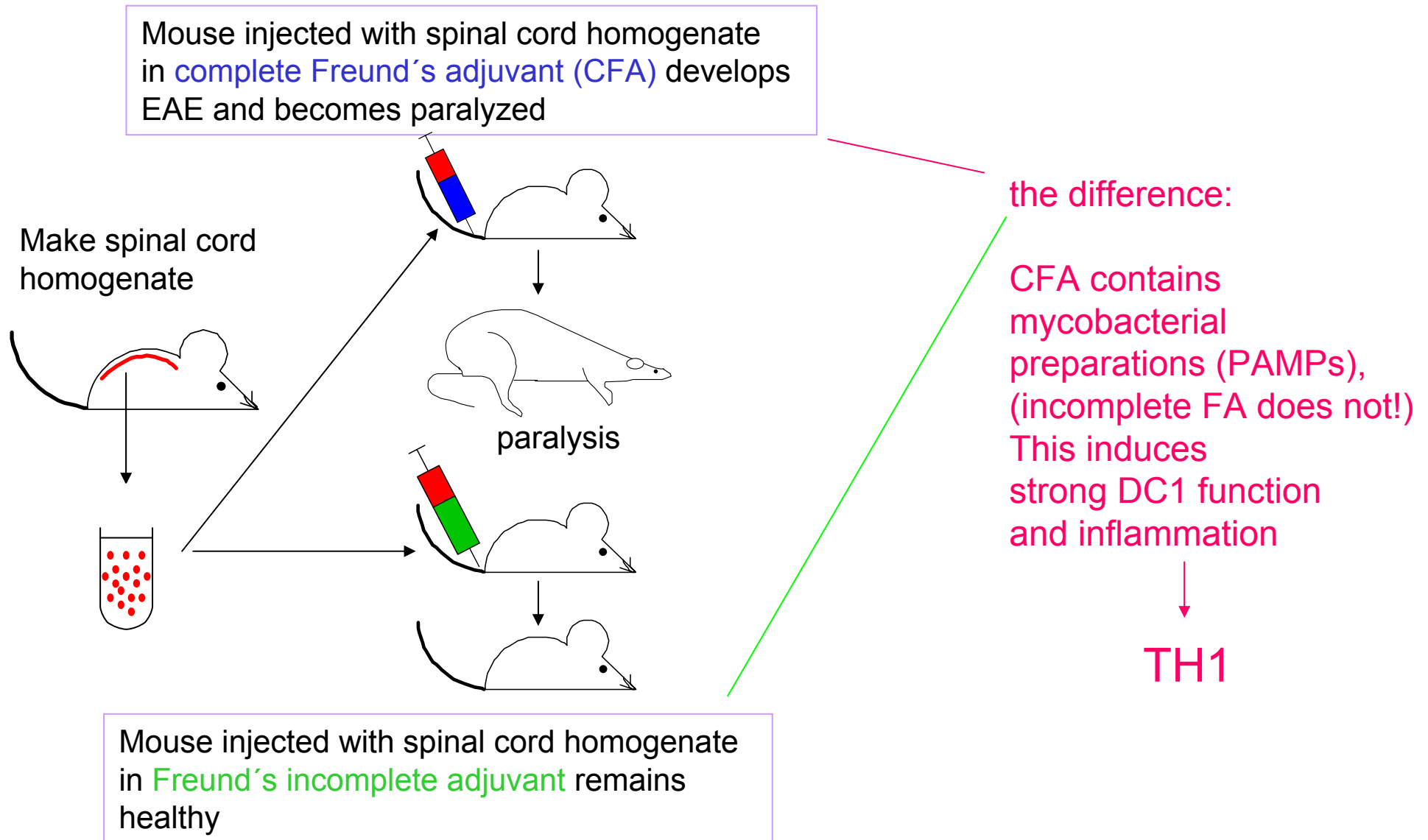
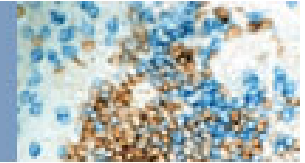
hier geht's weiter

Dysbalance – Risk auf autoimmune disease

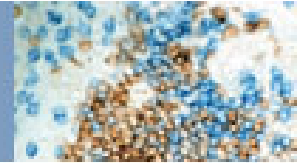




Breakdown of Peripheral Tolerance In Experimental Allergic Encephalomyelitis



Examples of TH 1-associated diseases



TH1 - associated diseases
in humans

Animal model with evidence for
anti-inflammatory effects of IL4
or TH2/Tr cells

Multiple sclerosis

EAE

Type I diabetes

Autoimmune diabetes

Acute uveitis

Experimental uveitis

Rheumatoid arthritis

Experimental arthritis

Contact hypersensitivity

Contact hypersensitivity

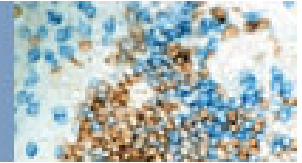
GVHD

GVHD

Transplantation

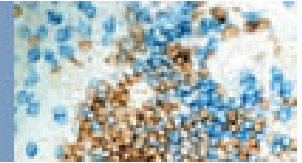
Alloreaction

Rheumatoid Arthritis



a patient with advanced RA
Gower Med Publ (5.8)

Classification of RA



American College of Rheumatology (1987)

Morning stiffness > 1h

Arthritis in three or more joints

Arthritis of the small joints of the hand and wrists

Symmetric localization of arthritis

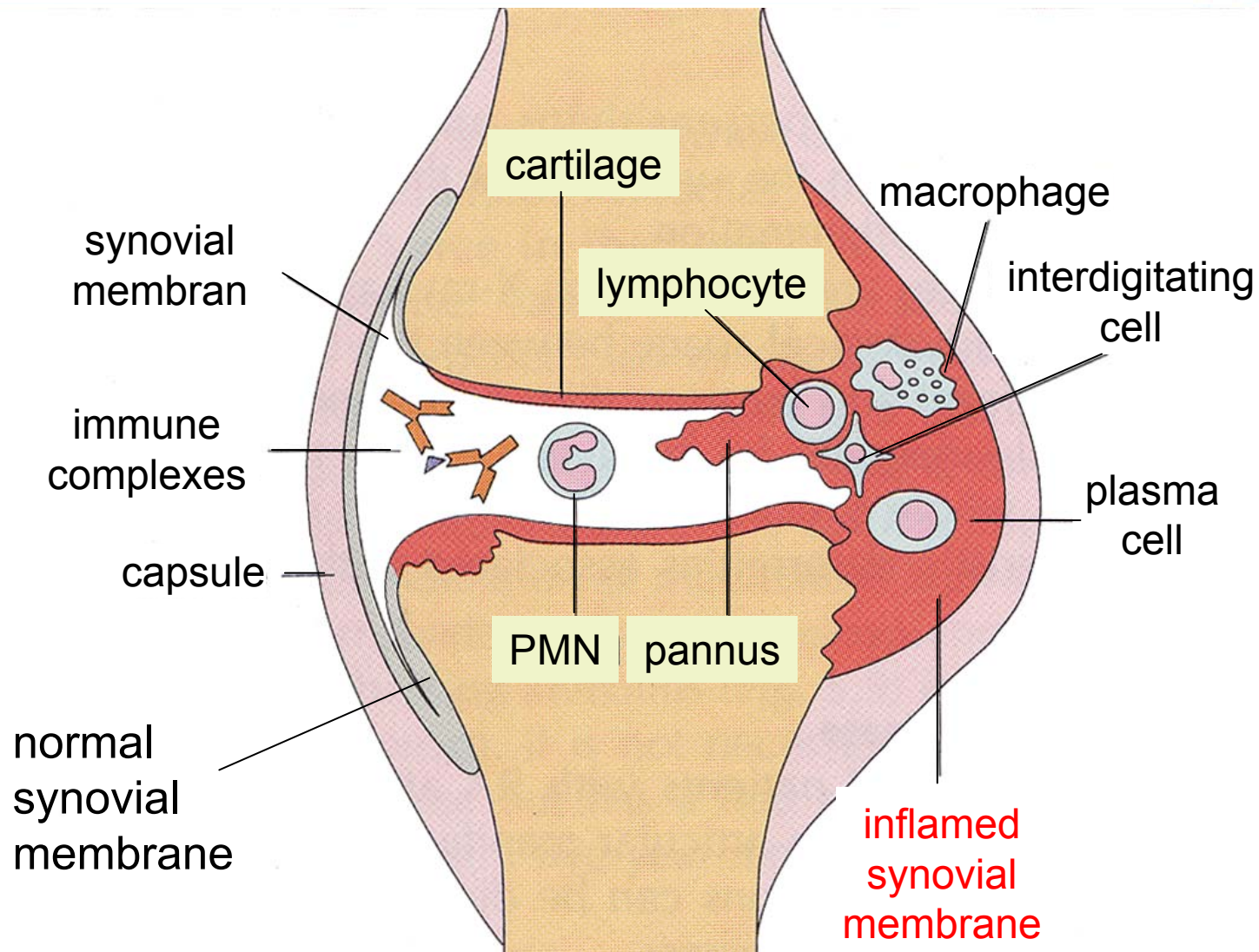
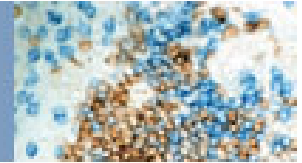
Rheumatoid nodules

Rheumatoid factor(s) positive

Radiographic changes in finger and carpal joints

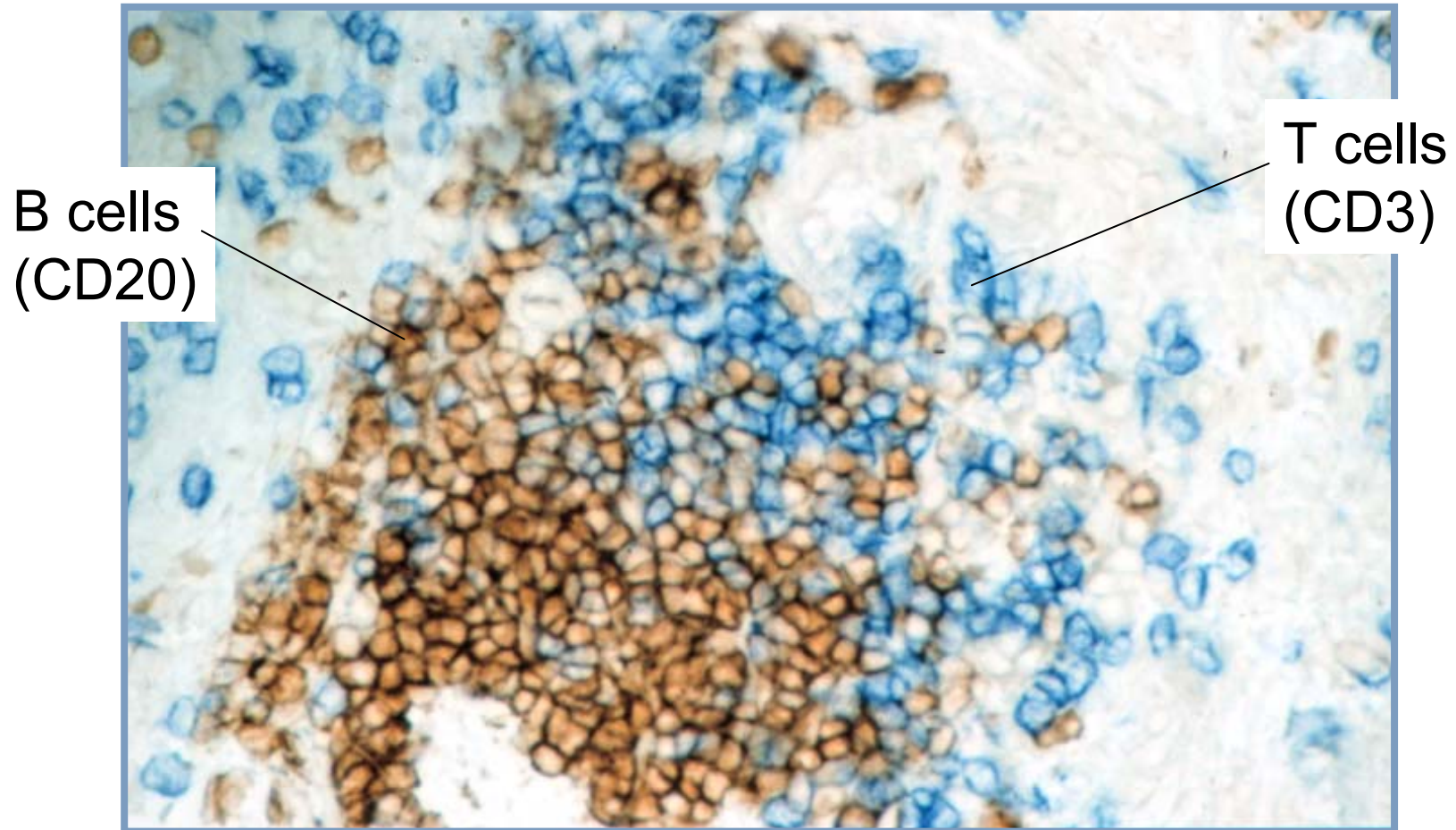
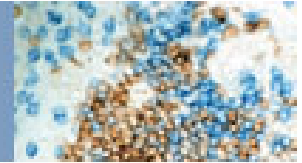
(erosions, decalcification)

Pathology of RA ?



synovial membrane hypertrophy and erosion of cartilage and bone

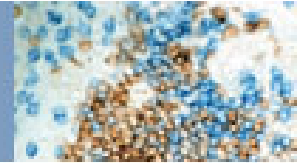
Ectopic lymphoid tissue in rheumatoid synovial tissue



kindly provided by B.Bröker

T- and B- cell specificities are unknown

Diagnostic specificity of auto-antibodies



high specificity

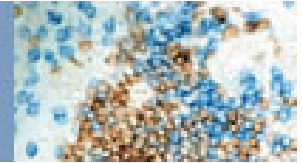
anti dsDNA	→	SLE
anti RNP	→	MCTD
anti Scl70	→	Progressive systemic sclerosis
anti PM-Scl	→	Polymyositis / Dermatomyositis
anti centromer protein	→	Skleroderma

low specificity

Rheumatoid factor (IgM)

Rheumatoid Arthritis

MCTD: mixed connective tissue disease



New: ● CCP - ELISA

Cyclic citrullinated peptides are targets of autoantibodies in Rheumatoid Arthritis

Proteolytic cleavage of profilaggrin during differentiation of epithelial cells
20% of arginine residues are enzymatically transformed into citrulline

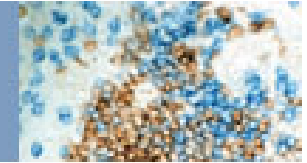
● Rheumatoid Factor (RF)

IgM anti-IgG antibodies

● HLA DR4 association

increased risk of RA

Anti-cyclic citrullinated peptide-antibodies

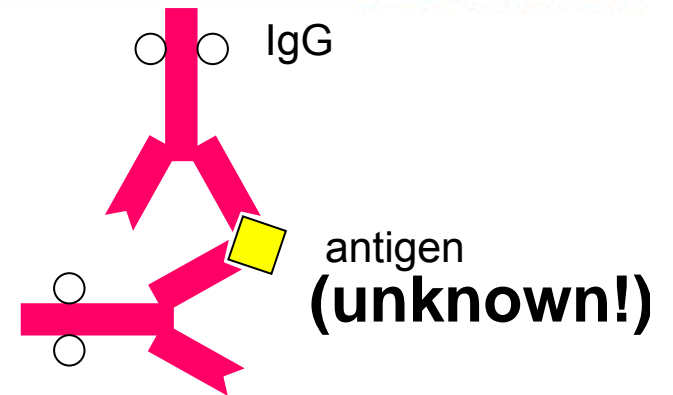
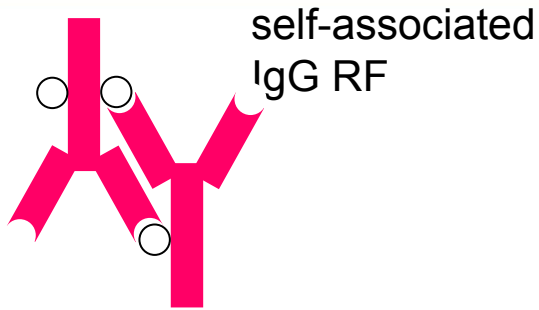
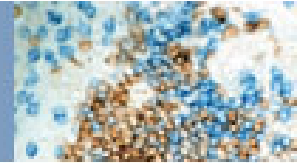


disease	n	anti CCP positive (%)
newly diagnosed RA	455	79
RA >2 years	696	80
blood donors	323	0
SLE	199	11
Sjögren Syndrome	86	1
Skleroderma	96	6
Polymyositis / Dermatomyositis	55	0
Osteoarthritis	66	11
Arthritis psoriatica	44	2
Autoimmune thyreoiditis	50	0
Borreliosis	45	2
Viremia	126	1

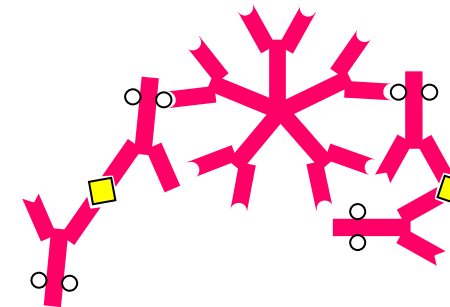
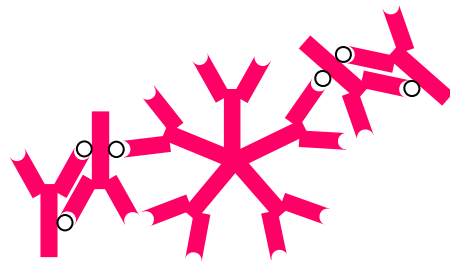
Sensitivity: anti-CCP 80%
RF 79%

Specificity: anti-CCP 96%
RF 64%

Immune complex formation in synovium in RA



+ IgM RF
"Rheumatoid factor"

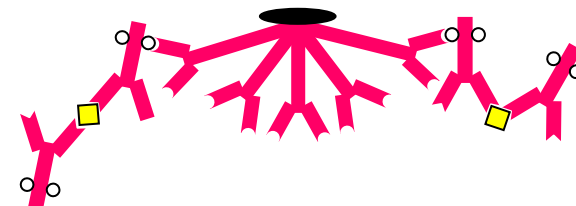


+ complement

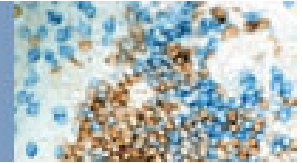
immune complex



immune complex

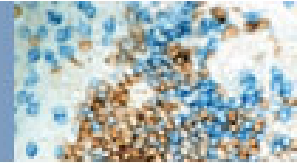


Therapeutic strategies in RA



- A nonsteroidal antiinflammatory drugs (NSAID)
- B glucocorticoids
- C Methylprednisolon bolus
- D Basic Drugs I Sulfasalazin, chloroquine
D-Penicillamin, Gold i.m.,
Methotrexate, Azathioprine
- E Basic Drugs II Cylosporin A, Cyclophosphamide
- F ...

RA therapy



1980 gold
low dose MTX

2000 **biologicals**

Anakinra IL1R antagonist

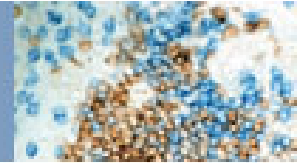
Celecoxib COX-2 inhibitors
Rofecoxib

Infliximab α TNF antibody ²

Etanercept TNFR fusion protein

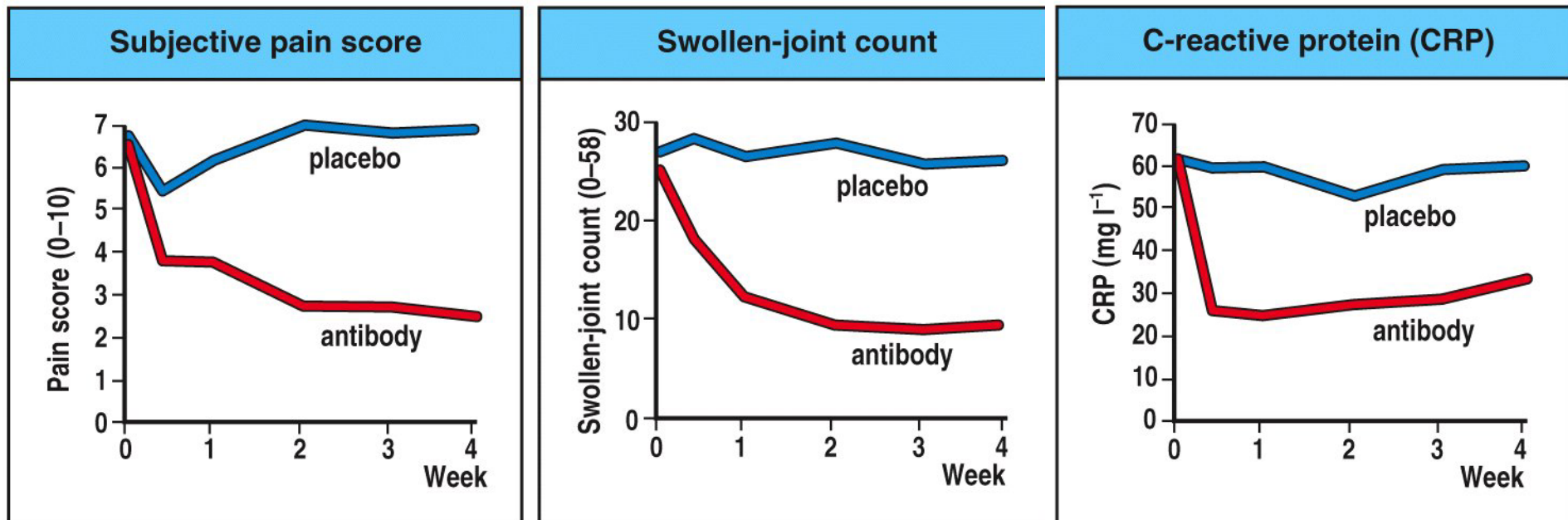
2005 antiCD20 mAb

Therapy of rheumatoid arthritis

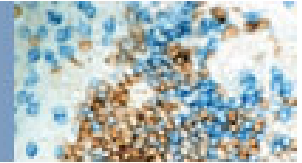


Anti-TNF-antibodies

- 10 mg/kg body weight
- for 4 weeks

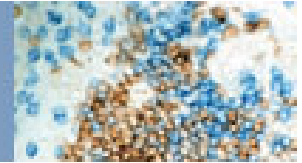


Outlook - a new immune system?



- Autologous bone marrow transplantation for life threatening autoimmune diseases (SLE)

Was ich vom Immunsystem wissen muss



- Autoimmunität wird als Zerstörung körpereigener Strukturen durch Immunzellen bzw. deren Produkte definiert. Dabei können Typ II-, III- und IV-Reaktionen ablaufen.
- Viele Autoimmunerkrankungen, wie IDDM, RA, MS werden als Typ IV-vermittelt definiert, ohne (!) dass die Pathophysiologie tatsächlich gesichert aufgeklärt wäre.
- Unspezifische Entzündungsmediatoren verstärken lokale Gewebeerstörung, weshalb neben Autoimmunität auch von Autoinflammation gesprochen wird. Deshalb werden z.B. Anti-TNF-Antikörper erfolgreich in der Therapie eingesetzt (Rheumatoide Arthritis, M. Crohn).
- Es werden viele neue *Biologicals* erprobt und eingesetzt, die an verschiedensten Stellen der unerwünschten unkontrollierten Immunantwort angreifen (Zellaktivierung, Adhäsion, Rezeptorblockade, Zellmigration, Blockade löslicher Mediatoren, Gabe antiinflammatorischer Proteine), die aber alle nur symptomatisch wirken.
- Ansonsten werden Autoimmunerkrankungen klassisch mit Glukokortikoiden bzw. Immunsuppressiva behandelt.
- Die therapeutische Induktion einer spezifischen Toleranz ist bislang in der Klinik nicht verfügbar.
- Wir unterscheiden organspezifische und nicht-organspezifische Autoimmunerkrankungen.
- Viele Autoantikörperbestimmungen geben mehr oder wenig sichere Hinweise auf Krankheitsentitäten.
- Sind Autoantikörper gegen intrazelluläre Strukturen gerichtet, haben sie keine ätiopathogenetische Bedeutung sondern sind Indikatoren von Gewebeuntergängen.
- Es gibt aber auch krankheitsverursachende Autoantikörper mit zytotoxischer Potenz.
- Viele Autoantikörper sind gegen Rezeptorstrukturen gerichtet und wirken blockierend (Myasthenia gravis) oder stimulierend (M. Basedow).
- In letzter Zeit sind vor allem Autoantikörper gegen G-Protein-gekoppelte Rezeptoren als Krankheitsursachen identifiziert worden, z.B. bei Dilatativer Kardiomyopathie (anti- β 1-adrenerge R), Präeklampsie (anti-AT1R), M. Raynaud (anti-PAR), hyperakuter steroidresistenter Nierentransplantatrejektion (anti-AT1R). Solche Antikörper wirken nur agonistisch, wenn die Zielzellen eine hohe Rezeptordichte aufweisen. Extrakorporale Immunadsorption ist hierbei eine neue Behandlungsstrategie.