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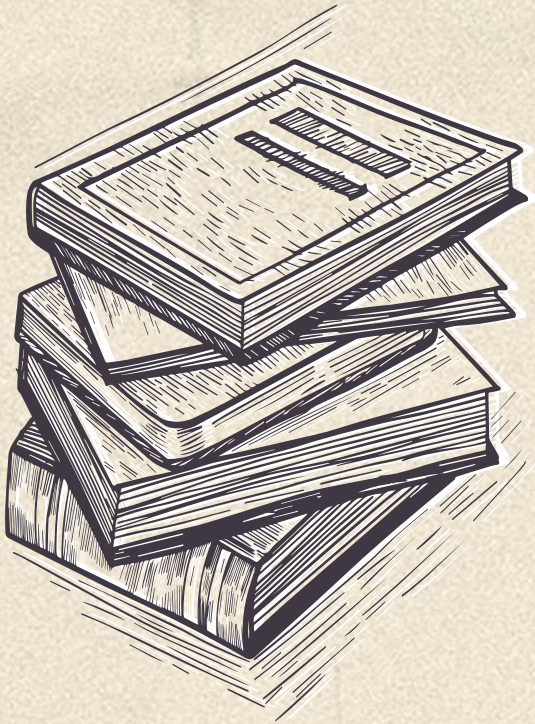

Immuno-Pharmacology

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Done by: Abdullah Hamdan

Quick Review

- Glucocorticoid (Glucocorticosteroid) has multiple side effects. However, we must use such drug. It is known also as the magical drug because there is no drug rather than Glucocorticosteroid can suppress pre- and post-organ transplantation rejection.

- Calcineurin inhibitors will inhibit eventually the production of IL-2 and autocrine activation for T-cell.

- These drugs are complex drugs, because we have a narrow therapeutic index. if you increase the drug concentration within the blood of your patient you will produce nephrotoxicity.

- Metabolized by the P450 3A (CYP 3A4, CYP 3A5), : CYP 3A4 and CYP 3A5 are polymorphic (has SNP single nucleotide polymorphism) which means that everyone has different activity of them.

Quick Review

- Any drug inhibits CYP 3A4 or CYP 3A5 will increase the concentration of Cyclosporine & Tacrolimus in the blood (drug-drug interaction).

- Cyclosporine & Tacrolimus are calcineurin inhibitors. However, one of them produce more toxicity than the other (Cyclosporine more toxic than Tacrolimus).

- Tacrolimus can cause diabetes mellitus and hyperglycemia, cyclosporin won't cause diabetes mellitus.

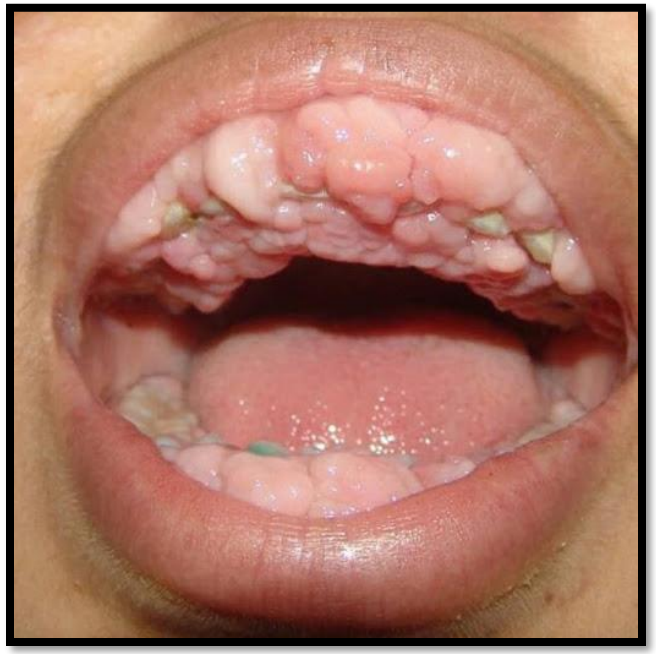
- Cyclosporine can cause Gingival Hyperplasia, we call it as trademark adverse effect, because there are only 3 or 4 drugs that can cause Gingival hyperplasia. The 2nd drug from these drugs is Calcium-channel blockers.

* الدكتور ركز عليها كثير, وحكى لازم تحفظوها زي اسمكم!

Quick Review

- Google: Gingival hyperplasia is an overgrowth of gum tissue around the teeth.

Just for clarification - Gingival hyperplasia



Sirolimus (RAPAMUNE)

- Closely related with Cyclosporine & Tacrolimus.

- Inhibits immune cell growth through inhibiting the kinase activity of mammalian target of rapamycin (mTOR) and decreasing IL-2 activities.

- Narrow therapeutic window
  - Levels too high: toxicities (i.e. mental confusion, nephrotoxicity)
  - Levels too low: transplant rejection

The target dose-range of these drugs will vary depending on clinical use.

Sirolimus (RAPAMUNE)

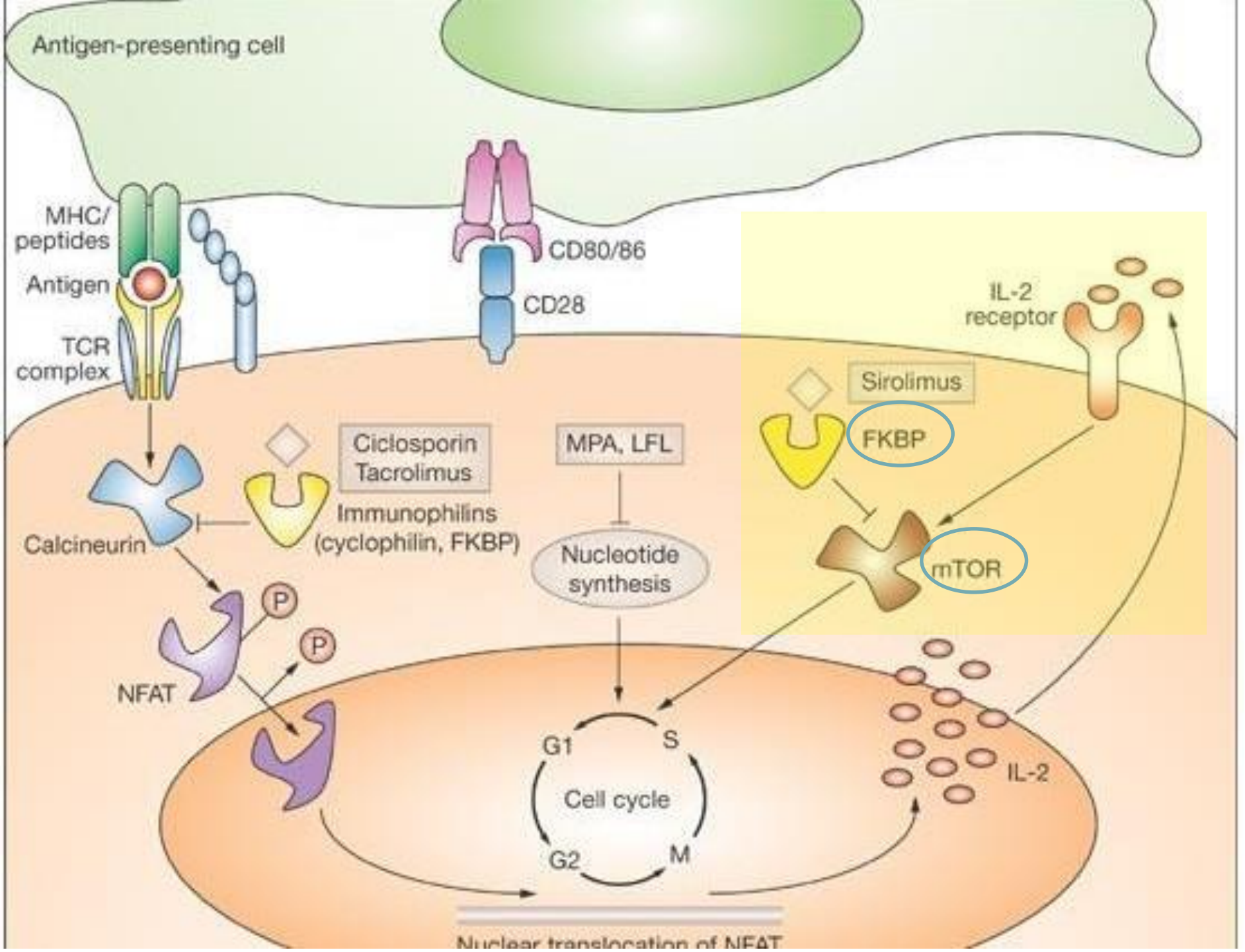
Mechanism of action:

- Sirolimus bind with FKBP, then FKBP will inhibit mTOR, the function of mTOR in normal statue as the following: It will be affected by IL-2, then it will enable the cell to enter the cell cycle and induce more proliferation of T-cells. So, Sirolimus will inhibit its action.

- Sirolimus and Calcineurin inhibitors are interchangeable which mean that we can't use them together, e.g. We either chose (Cyclosporin or Tacrolimus or Sirolimus).
  We can’t use cyclosporin with tacrolimus, or using sirolimus with tacrolimus, or using sirolimus with cyclosporin. We use only one of these 3 drugs to be self-acting alone.

- Although they have different mechanisms. however, there is a problem in its toxicity and there are some similarities in their mechanisms.

Sirolimus bind with FKBP, then FKBP will inhibit the mTOR, the function of mTOR in normal statue which will be affected by IL-2 and enable the cell to enter the cell cycle and produce more proliferation of T-cells.
So, Sirolimus will inhibit its action.



Anti-metabolites

Prevent the cell division in the S phase (DNA synthesis)

They affect the proliferation of both T cells and B cells.

In immune modulation cases we will use anti-metabolites and we will use it in cancer treatment to stop cancerous cell division completely.

So, in this case the dose will be a limiting factor. E.g. In cancer treatment we will give these anti-metabolites in very high doses than immunomodulation cases.

Thus, In immunotherapy, they are used in smaller doses than in the treatment of malignant diseases.

Anti-metabolites

There are 2 types of them:

- The 1st type which is responsible for inhibiting some enzymes involved in purine and pyrimidine synthesis. E.g. Methotrexate.
- The 2nd type is purine or pyrimidine analogs.

- We can use it in the treatment of cancer in acute lymphocytic leukemia in very high dose (5) more times than the dose that is used in treatment of Autoimmune disease.

- We use it in treatment of autoimmune disease including: psoriasis and inflammatory bowl disease but in lower doses which estimated to be 1/5 of the dose that we use to treat cancer.

Methotrexate

- Is a folic acid analogue. It binds dihydrofolate reductase and prevents synthesis of tetrahydrofolate.

- It is used in the treatment of autoimmune diseases (for example rheumatoid arthritis or Behcet's Disease or psoriasis) and in transplantations.

- The methotrexate is very important drug for rheumatoid arthritis. This is the drug of choice for rheumatoid arthritis and Behcet's Disease .

- We may use methotrexate in transplantations, but it’s not from the main stage drugs. The main stage drugs in treating transplantations are Azathioprine and mercaptopurine.

- Remember that Sulfonamides is the oldest anti-biotic and work by inhibiting dihydrofolate reductase in bacteria, so they work as anti-metabolites.

Azathioprine and mercaptopurine

- Azathioprine is the main immunosuppressive cytotoxic substance. هو الأساس بالحقيقة
- From its name it can indicate that it looks like purine.
- It is extensively used to control transplant rejection reactions.
- Azathioprine will be taken for long periods that will extend to more than 2 years.

MYCOPHENOLATE

- MPA (MYCOPHENOLATE) is a reversible inhibitor of the enzyme inosine monophosphate dehydrogenase (IMPDH).

- This leads to depletion of guanosine nucleotides.

- Depletion of guanosine nucleotides has antiproliferative effects on lymphocytes (Both T and B-cells).

MYCOPHENOLATE

- More effective than Azathioprine in preventing acute rejection.

- It is used in combination with cyclosporine and prednisolone (Glucocorticoids)

- Mycophenolate mofetil is used in solid organ transplant patients for refractory rejection:
  - يعني اذا صار في عندي رفض مناعي ببدل ال Azathioprine بال Mycophenolate

- In combination with prednisone, as an alternative to cyclosporine or tacrolimus in patients who do not tolerate those drugs.

- In renal transplants, it’s used with low-dose cyclosporine to reduce cyclosporine-induced nephrotoxicity.

س: ليش مش من البداية ما بنأخذ ال Mycophenolate؟

ج: نستخدمه بشكل أساسي كمنقذ. بحيث أذا صار عند المريض rejection حنستخدمه ... وإذا تم استعماله بالبداية قد يخفض المناعة بشكل كبير ويسبب سرطان!

- We can’t consider it to be in the main stage drug, because it decreases the immunity so much and we will be afraid to develop cancer in this case.

- Biologics they are the drugs that are targeted into biological component. E.g. Anti-CD3.

CD3 one of the signals that can be activated by antigen presenting cell, It's known as "Signal 1" and it constitutes T-cell triggering.

So, Scientists thought that they can inhibit this signal by Anti-CD3.

These signals are important because we will build our knowledge upon them.

The immune activation cascade can be described as a three-signal model.

Signal 1 constitutes T-cell triggering at the CD3 receptor complex by an antigen on the surface of an antigen-presenting cell (APC).

Signal 2 (costimulation) occurs when CD80 and CD86 on the surface of APCs engage CD28 on T cells.

Both Signals 1 and 2 activate several intracellular signal transduction pathways one of which is the calcium-calcineurin pathway.

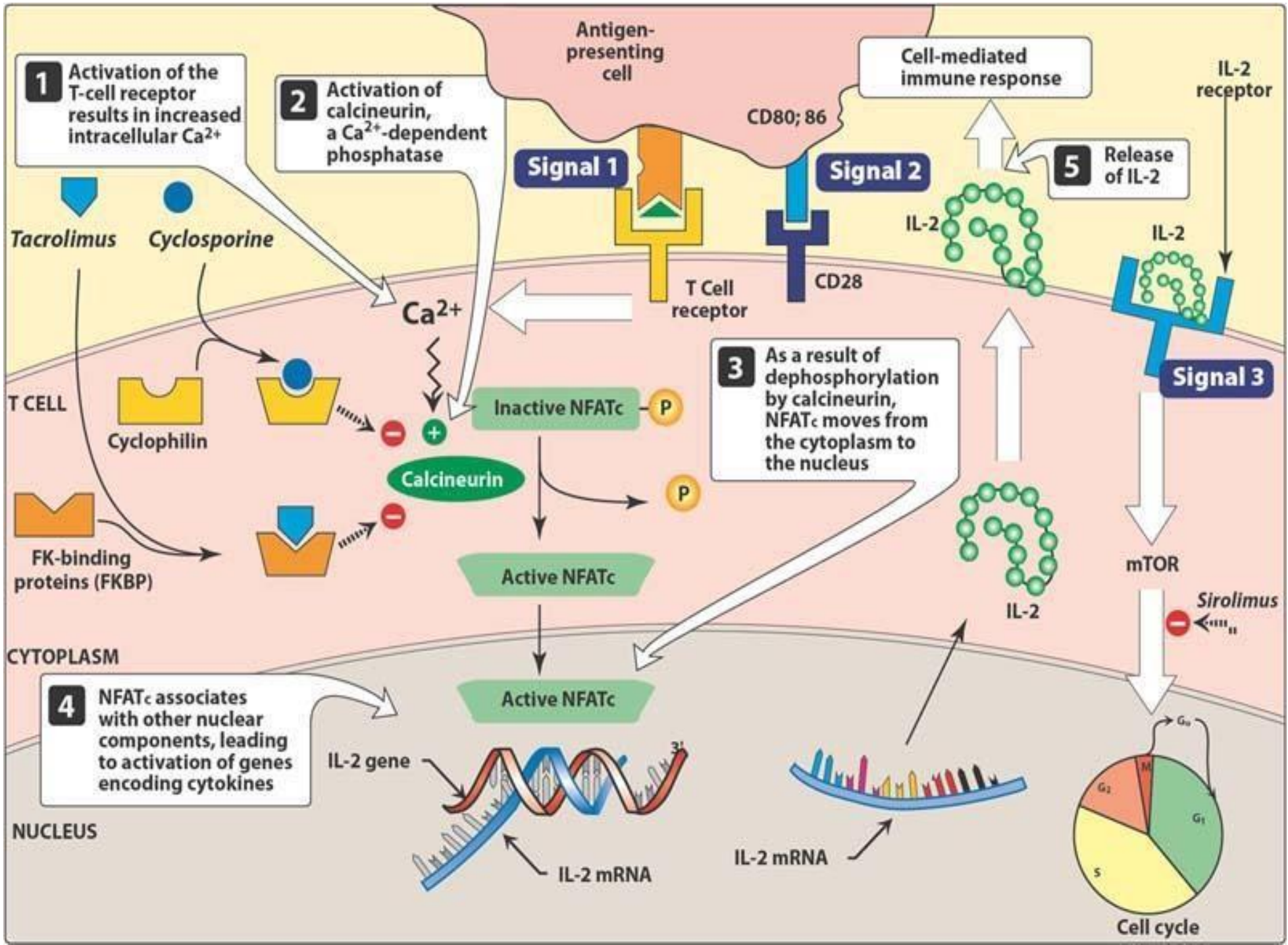
↓

Production of cytokines such as interleukin (IL)-2, IL-15, CD154, and CD25.

↓

IL-2 then binds to CD25 (IL-2 receptor) on the surface of other T cells to activate mammalian target of *rapamycin* (mTOR), providing Signal 3, the stimulus for T-cell proliferation.

Signal 3 (IL-2 will leave the cell and will then bind to IL-2 receptors and this will activate the mTOR and the cell will enter the cell cycle and will lead to more proliferation of T-cell. Thus, more induction of the immune system)



Immunosuppressive antibodies

We use Immunosuppressive antibodies :

- To suppress the activity of subpopulation of T-cells.
- To block co-stimulatory signals.

- Ab (Antibody) to the CD3 molecule of TCR (T cell receptor) complex results in a rapid depletion of mature T-cells from the circulation.

- It is used for treatment of acute rejection of renal allografts as well as for corticosteroid-resistant acute allograft rejection in cardiac and hepatic transplant patients.

- It is also used to deplete T cells from donor bone marrow prior to transplantation.

Anti CD3

Anti-CD3 is an injection, monoclonal antibody, antibody given in the blood, and this antibody will bind to CD3 and block it, e.g. if the patient had an acute rejection (rejection within the first 3 months) after the kidney transplantation, we would give him an anti-CD3, before the complete rejection takes place.

It is used for patients who has corticosteroid-resistant acute allograft rejection in cardiac and hepatic transplant patients. Corticosteroid is the primary drug, if the patient has corticosteroid-resistant we would then think to give him an anti-CDD3.

GvHD: We give the donor patient (who taking the bone marrow from him) an anti-CD3, he will have depletion of T-cells.

Anti CD3

Initial binding of muromomab-CD3 (اسم الدواء) to the antigen transiently activates the T cell and results in cytokine release (cytokine storm).

When I give the patient the muromomab-CD3 (IV) it will bind to CD3 and lead to activate it (This activation لحظي أو آني أو لوقت محدد) and it will result in cytokine release due to IL-2 production and autocrine activation. It depends on a main concept known as Occupation rather than inhibition. The initial binding starts the induction processes not the inhibition processes, and it can occupy the receptors for 3 weeks.

So to inhibit this cytokine storm, We shall give the patients some drugs earlier to Anti-CD3.

It is therefore customary to premedicate the patient with methylprednisolone (Glucocorticoid), diphenhydramine (Against Histamine) , and acetaminophen (إذا صار عند المريض حرارة) to alleviate the cytokine release syndrome.

IL-2-receptor antagonists

Targeting Signal 3 (IL-2 will leave the cell and will then bind to IL-2 receptors and this will activate the mTOR and the cell will enter the cell cycle and will lead to more proliferation of T-cell. Thus, more induction of the immune system)

Ab specific for the high-affinity IL-2 receptor is expressed only on activated T-cell, blocks proliferation of T-cells activated in response to the alloantigens of the graft.

Basiliximab is said to be “chimerized” because it consists of 25 percent murine and 75 percent human protein.

Daclizumab is 90 percent human protein and is designated “humanized.”
Both agents have been approved for prophylaxis of acute rejection in renal transplantation in combination with cyclosporine/tacrolimus and corticosteroids. To treat donor’s bone marrow before it is transplanted (Like Anti-CD3 but IL-2 Receptors antagonists are preferred to use more)

IL-2-receptor antagonists

-Both antibodies are given intravenously.

-The serum half-life of daclizumab is about 20 days, and the blockade of the receptor is 120 days.

- The serum half-life of basiliximab is about 7 days. Usually, two doses of this drug are administered—the first at 2 hours prior to transplantation, and the second at 4 days after the surgery.

- Well tolerated, Their major toxicity is gastrointestinal.

E.g Some patients who decide to undergo the kidney transplantation surgery, they sometimes buy this kidney from India, and the transplantation will take place in Jordan for example, the chance for rejection to occur will be high, because there is no relative donor to give him this organ, this is called as high risk of rejection patients, and here we can use anti IL-2 before the transplantation, two doses of this drug are administered—the first at 2 hours prior to transplantation, and the second at 4 days after the surgery.

Immunosuppression therapy in kidnay transplantation

- **Methyl Prednisolone (إبرة كورتيزون) 500 mg IV just prior to transplantation and again at 24 hours.**
- **إبرة 500 mg قبل وبعد واليوم الثالث:**

Tacrolimus led triple therapy. (مش للحفظ)

- Tacrolimus 0.1 mg/kg/day given as two doses at 10:00 and 22:00.
- Prednisolone 20 mg once daily at 08:00 PM
- Azathioprine 1-2 mg/kg (usually 75-100 mg) at 08:00 AM and Initially 1-2 mg/kg once daily. Maintenance 1 mg/kg once daily.

- It’s called as Regiments.

- **Just to understand that there is dozing timing, and the importance of drug half life. And we can’t neglect these guidelines because any change will lead to some problems.**

س: هل لازم أعرف كل دوا على أي ساعة بنعطيه أو شو معيار الجرعة المناسبة اله؟
ج: لأ

Prednisolone

Normally reduced according to the following schedule (Tipparing):

- 20 mg daily 1 month started on day 2
- 15 mg daily 1 month
- 10 mg daily 1 month
- 5 mg daily thereafter
- After that we must Decrease by 1 mg per month till 0mg.

This schedule may be altered if rejection occurs.

- All patients to receive Ranitidine (150 mgs od) along with Prednisolone.
- Steroid withdrawal should be discussed with the patient (نفهم المريض أنو ننزل الدواء تدريجياً) and they should be informed of the risk of rejection. The patient might have some rejection signs, and in this case we will increase the dose rather than lowering it.
- The steroids should be withdrawn according to the following schedule:

When we reach 5 mg in withdrawal program, we must then decrease by 1 mg per month till 0mg.

Tacrolimus

- Whole blood trough (concentration of the drug before the next dose) levels to be checked on Mondays, Wednesdays and Fridays.

- The target level for the first six months is 10 ng/ml – الدكتور ركز على الوحدة - (range 8-12 ng/ml) and 5-10 ng/ml after six months.

Patients who have an increased risk of rejection

- **Tacrolimus led triple therapy, but with MMF (Mycophenolate mofetil) substituted for Azathioprine (This can be applied to the example which was mentioned on slide 22).**
- Tacrolimus as per standard regime
- Prednisolone as per standard regime
- Mycophenolate Mofetil 2 grams/day given as two doses at 0800 and 2000 (note: not at the same time as Tacrolimus)

Basiliximab

- **Given to patients with expected delayed graft function** to allow reduced Tacrolimus dose (0.05mg/kg/day given as two doses), and sometimes to patients that are believed to be at increased risk of rejection.

Dose

- 20mg given 2 hours prior to transplantation
- 20mg given on day 4 post transplant

The first dose must not be administered until it is absolutely certain that the patient will receive the graft.

Autoimmune Disease

- An immune reaction against self
- Mechanism unknown, arises out of a failure in immune regulation
- Examples:
  - Rheumatoid arthritis
  - Systemic lupus erythematosus
  - Multiple sclerosis (MS)
  - Insulin-dependent diabetes mellitus
  - Many more

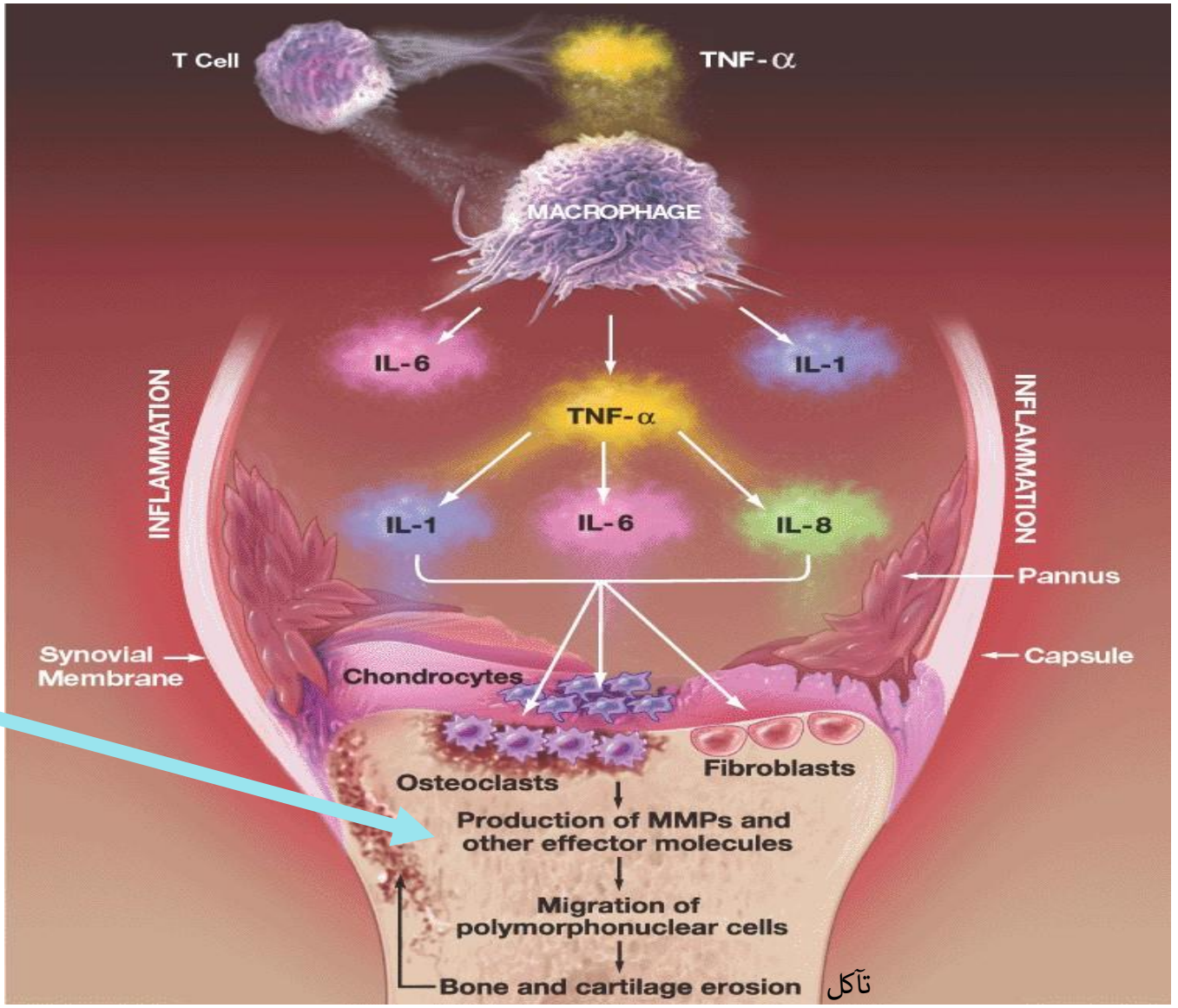
Rheumatoid arthritis

Macrophages will Secrete IL-1,IL-6 and TNF-α. Macrophage will go to the synovial fluid and can attack the chondrocytes and it will induce some problems on fibroblast and osteoclasts through TNF-α, which can stimulate IL-1, IL-6 and IL-8. Then it will lead to all of the following

And this can cause deformities.

We can give Anti-IL-1, Anti-IL-6, Anti-IL-8,…

But the question arise why we don't give the Anti TNF-α. And here is the Infliximab and Adalimumab.



Infliximab and Adalimumab

- Anti TNF-α
- Approved by the FDA in 1998

- Designated for use in patients who did not respond to methotrexate.

- Proven to slow the clinical progression of rheumatoid Arthritis.

Side Effects of TNF Inhibition

- **Infection**
  - Tuberculosis
  - Serious resulting in death
- **Neurologic**
  - Multiple Sclerosis, seizures, inflammation of the ocular nerve
- **Worsening of Congestive Heart Failure**

- Remember

STOP if develop a fever بسرعة, because the patient might have an infection,

Rituximab

- Anti-B cell (CD20) antibody
- 15-20% of rheumatoid arthritis cases there will be involvement of B-cells.
- First approved in 1997 for use in B-cell lymphoma
- Given in combination with Methotrexate
- Directed for patients who do not respond to Anti-TNF treatments
- Indicates the rheumatoid arthritis has a B cell component to its pathology.

Anti-IgE Antibodies Omalizumab

- Asthma has something to do with allergy, and this allergy related to IgE. (Meaning: If the patient had an elevated concentration of IgE, we would give him Omalizumab.

- Biologic antibody therapy (Omalizumab; Xolair) binds IgE in the circulation and prevents it from activating mast cells and basophils.

- Anti IgE therapy is recommended as an add-on to optimized standard therapy in asthmatics 12 years and older who need continuous or frequent treatment with oral corticosteroids.

- Elevated serum IgE.

Immunostimulants (زيادة المناعة)

- Increase the immune responsiveness of patients who have either selective or generalized immunodeficiency.

- Use for immunodeficiency disorders, chronic infectious diseases, cancer and HIV.

Cytokines Therapy

- Interferon (INF): INF-α,β,γ
  - Antiviral, anticancer, immunomodulating effects.
  - Antiviral effects : INF-α,β＞ INF-γ
  - immunomodulating effects: INF-γ
  - Adverse Effects: flu-like symptoms, fatigue, malaise
- Interleukin-2 (IL-2)
  - T cell proliferation, T_H, NK, LAK cell activation
  - Treatment of malignant melanoma, renal cell carcinoma, Hodgkin disease
  - Adverse Effects: fever, anorexia, etc .

Cancer Immunotherapy

- **Checkpoint inhibitor drugs that target PD-1 or PD-L1**

- PD-1 is a checkpoint protein on immune cells called T cells. It normally acts as a type of “off switch” that helps keep the T cells from attacking other cells in the body. It does this when it attaches to PD-L1, a protein on some normal (and cancer) cells. When PD-1 binds to PD-L1, it basically tells the T cell to leave the other cell alone. Some cancer cells have large amounts of PD-L1, which helps them hide from an immune attack.

- They include:

1- Nivolumab
2- Lambrolizumab
3- Pidilizumab

Cancer Immunotherapy

- Immune checkpoints refer to inhibitory pathways of the immune system that are crucial for maintaining self-tolerance and modulating the duration and amplitude of physiological immune responses in peripheral tissues in order to minimize collateral tissue damage.

- Tumors misuse immune-checkpoint to evade the immune system clearance, in particular to avoid tumor-antigen specific T-cell responses

T-cells activation can be inhibited, Some tumor cell can produce a relationship friendship like with T-cells through the production of modulators.