POST-TRANSPLANT MONITORING PROTOCOL

(JUNE 2021)

- **CHILDREN UNDER 4 YEARS OF AGE:**

  Methyl-PREDNISOLONA (Urbason® or Solumoderín®):

  20 mg/kg in reperfusion.

  10 mg/kg on day 1

POST-TRANSPLANT MONITORING PROTOCOL 1
(JUNE 2021) 1
- CHILDREN UNDER 4 YEARS OF AGE: 1
Methyl-PREDNISOLONA (Urbason® or Solumoderín®): 1
Tacrolimus (Prograf®): maintain levels of: 2
> 1 month: 15-20 ng/ml 2
BASILIXIMAB: days 0 and 4. 2
– IF POSITIVE CROSS-TEST: 2
- OVER 4 YEARS OLD: 3
– HIGH RISK (INTESTINAL ISOLATED T., RETRANSPLANT, ANTI-HLA ANTIBODIES +) 3
Methyl-PREDNISOLONA (Urbason® or Solumoderín®): 3
BASILIXIMAB: days 0 and 4. 5
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2 mg/kg from day 5 to 30.

1 mg/Kg 2nd month.

0.5 mg/kg 3 rd month

0.25 mg/kg 4th month.

0.25 mg/kg alternate days from year.

> plough withdrawn 6 months later in selected cases.

Tacrolimus (Prograf®): maintain levels of:

> 1 month: **15-20 ng/ml**

1-3 months: 10-15 ng/ml

3-12 months: 8-10 ng/ml

> 12 months: 5 ng/ml.

BASILIXIMAB: days **0 and 4.**

10 mg < 30 kg

20 mg > 30 kg

- **IF POSITIVE CROSS-TEST:**
  - Plasmapheresis: one session alternate days or every 2 days (depending on antibody levels)

- Repeat ↓-globulin (0.5 g/lg) alternating with plasmapheresis, up to 2 g/kg in total.
- Rituximab (antiCD20): 375 mg/m^2 weekly. Number of doses according to B-lymphocyte response.
- Review Bortezomib: 1.3 mg/m^2/72 hours

✇ **OVER 4 YEARS OLD:**

– **HIGH RISK (INTESTINAL ISOLATED T., RETRANSPLANT, ANTI-HLA ANTIBODIES +)**

- **PREMEDICATION:** By revascularising premedicate with:
  - Methylprednisolone (Urbason®): 10 mg/kg
  - Dexchlorpheniramine (Polaramine): 0.05 mg/kg iv

- **ALEMTUZUMAB (CAMPATH):** 0.5 mg/kg (max 30 mg), intratx **and day 4**

- **TACROLIMUS**: 0.05-0.10 mg/kg/dose every 12 hs (9 and 21 h) per NMS for levels of:
  - 10-15 ng/ml x 60-90 days
  - 5-10 ng/ml from 3 months.

- **STANDARD RISK**

Methyl-PREDNISOLONA (Urbason® or Solumoderín®):

20 mg/kg in reperfusion.

10 mg/kg on day 1

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2 mg/kg from day 5 to 30.

1 mg/Kg 2nd month.

0.5 mg/kg 3 rd month

0.25 mg/kg 4th month.

0.25 mg/kg alternate days from year.

Assess withdrawal 6 months later in selected cases.

Tacrolimus (Prograf®): maintain levels of:

< 1 month: **15-20 ng/ml**

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3-12 months: 8-10 ng/ml

> 12 months: 5 ng/ml.

BASILIXIMAB: days **0 and 4.**

10 mg < 30 kg

20 mg > 30 kg

EVOLUTIONARY IMMUNOSUPPRESSION

Assess SIROLIMUS in mixed therapy or monotherapy, in general, >6 months after Tx.

If:

- Kidney failure.
- EHD.
- AHAI
- SLPPT
- CMV infection
- Multiple allergies.

Start with:

- 1st day: 5 mg/m^2, maximum 6 mg/day.
- 2nd day: 2 mg/m^2
- Adjust to levels of 7 ng/ml approx. Mixed: sum between 7-10 ng/ml (sirolimus >5 ng/ml).

TREATMENT OF ACUTE CELL REJECTION

- **Increased dose of tacrolimus** (by oral or intravenous administration, if necessary) to achieve desired blood levels (around **15-20 ngr/ml***).*
- **Steroid bowling**: **Three days** of boluses of steroids at doses of **20 mgr/kg/day** at 3 doses with subsequent recycling for 5 days (10, 7, 5, 2, 1 mgr/kg/day) to reach the dose prior to the rejection episode. Dilute in glucose 5 % (maximum 4 mg/ml) and pass in 20-30 min.

- Establish CMV treatment

ACUTE CORTICO-RESISTANT REJECTION

- **RABBIT ANTITHYMOCYTIC GAMMAGLOBULIN**
  - TRADE NAME: THYMOGLOBULIN® (GEMZYME)
  - PRESENTATIONS: 25 mg/5 ml vials.
  - DOSE: **1.5-2 mg/kg/day** x 7-14 days. Adjust if leukopenia or thrombopenia.
  - RULES OF ADMINISTRATION: dilute to 0.5 mg/ml in SSF or SG 5 %. Go through central or large gauge with a 0.2 µ filter. In 6 hours the first dose and 4 the next.

- **INFLIXIMAB (ANTI-TNF):**
- TRADE NAME: REMICADE ® (SCHERING PL)
- PRESENTATION: 100 mg/20 ml vial
- DOSE: **5 mg/kg** one dose per week x 3-4 weeks.
- RULES OF ADMINISTRATION:
  - Dilute each 100 mg vial in 10 ml of sterile water for injection. DON'T SHAKE. Mix gently around.
  - Once diluted, let the vial stand for 5 minutes
  - Reconstitute the total dose by completing SSF up to 250 ml in glass container.
  - Purge the system with the SSF filter.
- Spend in 2 and a half hours through a filter of 1.2 microns or less.
- Purge again with SSF.
- Monitor blood pressure and HR every 15 minutes.

- **ALEMTUZUMAB (CAMPATH)**

***** Presentation: Blisters 30 g.

***** Dose: 0.5 mg/kg (maximum 30 mg) c/3 days. 4 doses

***** Premedicate with Urbason, Polaramine and Paracetamol.

- **MYCOPHENOLATE-MOFETIL**
- TRADE NAME: CELLCEPT® (ROCHE)
- FORMULATION: COMP 250 and *500* mg, susp 1 g/5 ml, vial iv 500 mg
- DOSE: **30 mgr/Kg/ day (or 600 mg/m^2)** divided into 2 oral doses, GNS or IV.
- RULES OF ADMINISTRATION (IV):
  - Due to its risk of producing pulmonary edema, evaluate prior chest Rx to associate diuretics in case of presence of intrapulmonary fluid. The number of lymphocytes should be greater than *3000* /mm^3.
  - Also assess intradermal test: intradermal arm administration of 0.1 cc diluted to 1 per *thousand*(0.1 cc ampoule diluted in 100 cc physiological saline). Observe within 1 hour with controls every 15 minutes of TA, FA, temperature, etc.
  - Rebuild each vial with 14 ml of SG 5 % and shake gently. Dilute the desired amount in SG 5 % so that the final concentration of the solution to be administered is 6 mg/ml.

- PERFORMANCE CONTROL: the CD3 lymphocyte rate is considered to be between 0-5 % (send blood samples to S. Immunology after the 2nd and 7th dose).

HUMOROUS REJECTION SURVEILLANCE

- Determination of anti-HLA antibodies, cross-match and donor typeage on the day of transplantation.
- Determination of anti-HLA Ac at 7 and 15 days postTx and then serialised every two weeks during the

  income (value frequency change according to risk)
- Determinations every 3-6 months after discharge, if not sensitised (individualise in sensitised)

TREATMENT OF HUMORAL REJECTION

- Plasmapheresis: alternate days or every 2 days (depending on antibody levels). 5 meetings.
- ↓-globulin (0.5 g/lg), up to 2 g/kg in total.
- Rituximab (antiCD20): 375 mg/m²/week, 4 doses.
- Value Eculizumab.

IMMUNOLOGICAL FOLLOW-UP

Complete TBNK:

- PreTx
- 7 days
- 15 days
- 1month
- 2months
- 3months
- 6months
- 9months
- 12 months
- 24 months

CHIMERISM CONTROL

- 7 days
- 15 days
- 1 month
- 30 days
- 2 months
- 3 months
- 6 months
- 12 months
- 24 months

TREATMENT GRAFT VERSUS ACUTE RECEPTOR DISEASE

1st Line:

- Optimise immunosuppression levels.
- Steroids: Two possible guidelines. Prednisone/Methylprednisolone
- 10-5 — 4-3 — 2 mg/kg/day. It does not appear to be higher than:

- 2 mg/kg/day

2nd Line:

- Photoapheresis.
- Ruxolitinib: 2.5 mg/12 hours <25 kg
  5 mg/12 hours >25 kg
  10 mg/12 hours in >12 years
  For 4-8 weeks.

3 rd Line:

- Thymoglobulin.
- Alemtuzumab
- Infliximab
- Rituximab
- Tocilizumab.
- Etc.

TREATMENT OF AUTOIMMUNE HEMOLYTIC ANEMIA

1st Line:

- Steroids: 5 mg/kg/day. Three days. Decrease 1 mg/kg/day to basal dose.
- Gammaglobilin: 0.5-1 g/kg/day. 5 doses.
- Rituximab: 375 mg/m^2, weekly, 4 doses

2nd Line:

Plasmapheresis.

Bortezomid: 1.3 mg/kg/72 hours.

Conversion to Sirolimus.

3 rd Line:

Alemtuzumab.

Splenectomy.

VIRAL INFECTION PROPHYLAXIS

GENERAL MEASURES

- It is recommended that the blood products used during the intervention and postoperative are CMV negative.
- Use of leukocyte filters during infusion of blood products.

Prophylaxis ANTIVIRAL

Two groups of patients are established in relation to the donor and recipient cytomegalovirus (CMV) pretransplantation.

- **HIGH-RISK PATIENTS (POSITIVE CMV DONOR/NEGATIVE CMV RECEPTOR):**

— GANCICLOVIR IV:

TRADE NAME: CYMEVENE

PRESENTATION: amp 500 mgrs. ADMINISTRATION: Dilute in physiological saline or glucose (maximum dilution 10 mgr/ml). Administration in 1 hour with 0.22 micron filter.

HOME: after transplantation as soon as permitted by neutrophils (>1000/mm^3*)* and platelets (>50.000/mm^3*).*

DOSE: **10 mgr/kg/day** divided into 2 doses for 2 weeks. The dose shall be adjusted for creatinine clearance:

- Greater than 50 ml/min/1.73 m^2: usual dose (5 mgr/kg/12 h).
- Between 25-50 ml/min/1, 73 m^2: 2,5 mgr/kg/12 h.
- Between 10-25 ml/min/1.73 m^2: 2.5 mgr/kg/24 h.
- Less than 10 ml/min/1, 73 m^2: 1.25 mgr/kg/24 h.

SUBSEQUENTLY: Valganciclovir (Valcyte®).

- DOSAGE: 14 mg/kg, 2 doses. Three months. 14 mg/kg/day. Three months.
- Suspension 1 cc = 50 mg. 400 mg tablets.

- **OTHER PATIENTS**

  - Ganciclovir IV during the two weeks post-transplantation at 10 mgr/kg/day divided into two doses.
  - Subsequently Valganciclovir (Valcyte®): 14 mg/kg/day. Three months.

SURVEILLANCE OF INFECTION VIRAL

- **SURVEILLANCE OF EPSTEIN-BARR VIRUS INFECTION (EBV)**

  - PRETRASPLANT SERVICES AND QUANTITATIVE PCR IN BLOOD OF VEB every 14 days post-transplantation during the period of hospital admission or whenever the clinical situation of the patient so requires. Then monthly until the 12nd month and every 2-3 months between 12 and 24 months post-transplantation. Samples: serum and total blood. (5 cc of blood in tube with heparin and 3 cc of serum).
  - Immunohistochemistry AND PCR for EBV in samples obtained from graft biopsies.

- **SURVEILLANCE OF CMV INFECTION**

  - PRETRASPLANT SERVICES AND QUANTITATIVE PCR OR Antigenemia IN CMV BLOOD every 14 days post-transplantation during the hospital admission period. After discharge: PCR every 3 months.
  - NMUNOHISTOQUOMIC AND PCR for CMV in graft biopsy samples.

TREATMENT OF CMV INFECTION

- Ganciclovir: 5 mg/kg/12 hours. 14 days. Followed by:
- Valganciclovir: 14 mg/kg/12 hours.
- Monitor viral load weekly.
- Maintain Valganciclovir for 3 months after undetectable viral load.
- If refractory:
  - Study of resistances
  - Foscarnet: 60 mg/kg/8 hours. 2-3 weeks. Adjust to renal function.

    Hyperhydrate.
  - Specific anti-CMV cytotoxic T-lymphocytes.
  - Cidofovir, Maribavir.
- Maintenance: Valganciclovir, Valaciclovir, letermovir.
- Value conversion to Sirolimus.

INFUSION PROTOCOLS

TACROLIMUS

—TRADE NAME: PROGRAF® (ASTELLAS)

—PRESENTATIONS:

Blisters for intravenous administration (1 ampoule = 1 ml = 5 mg).

0.5, 1 and 5 mgrs tablets.

— RULES OF ADMINISTRATION:

- Oral or NNS administration at an **initial dose of 0.1-0.2 mgr/kg/day** divided into 2 doses (9 and 21 hours).
  - Administration by SNG (not PVC): The diluted dose should be administered in water (10 cc) and then washed with a similar amount of water. The SNG shall be kept pressed for one hour after administration.
  - Oral administration: It should be given 1 hour before or 2 hours after eating.
  - If the desired blood levels have not been achieved at 48 hours of onset of the oral route or GNS for tacrolimus administration, it may be necessary to re-start concomitant intravenous use until these levels have been achieved.
  - Intravenous administration: if adequate levels are not achieved within 48 hours. Continuous infusion at **doses of 0.01-0.05 mgr/kg/day** with subsequent modification according to blood levels. It is appropriate to "mark" from the beginning the catheter by which it is to be administered to avoid contamination of the samples for determination of levels in subsequent days.
  - Diluents: physiological saline serum or 5 % glucose.
  - Concentration between the range 0.004 to 0.1 mgr/ml.
  - Total infusion volume in 24 hours: Between 20 and 250 ml.
  - Materials: syringes or bottles made of glass or polyethylene (never PVC). Stability of the infusion under these conditions: 24 hours after preparation.
- Frequency of samples: Daily during the first 15 days post-transplantation. Subsequently, according to clinical and analytical stability.

BASILIXIMAB (IL-2 antireceptor)

- TRADE NAME: SIMULECT® (NOVARTIS)
- PRESENTATION: vial+amp of 20 mg
- ADMINISTRATIVE RULES IV: after reconstituting the 20 mgr vial in 5 cc of sterile water dilute the solution in 50 cc give physiological saline or 5 % glucose and administer intravenously (central or peripheral) in 30 minutes. Once reconstituted the solution could be stored up to 24 hours between 2 and 8 degrees.

Alemtuzumab (anti CD52)

- TRADE NAME: CAMPATH® (BAYER)
- PRESENTATIONS: 30 mg/3 ml and 30 mg/1 ml vials

- ADMINISTRATIVE RULES IV:

  - DO NOT SHAKE THE VIAL.
  - Dilute the desired dose in 100 ml of SSF or 5 % glucose. Use within 8 hours after dilution. Invest to mix. Do not shake
  - Protect from light.
  - Manage in a few 2 hours
  - PREMEDICATION:
    - Urbason 1 mg/Kg i.v. (optional)
    - Acetaminophen 15 mg/Kg i.v.
    - Polaramine 0.05 mg/Kg i.v.
  - Maximum dose: 30 mg/dose i.v.

TIMOGLOBULLNA (human antithyocyte rabbit immunoglobulin)

- TRADE NAME: Thymoglobulin 5 mg/ml sun powder for infusion (Lab Gemzyme)
- COMMERCIAL PRESENTATION: 25 mgrs (5 ml) vials
- ADMINISTRATION: intravenous centrally.

  - Dosage: 2 single doses of **2-3 mgrs/kg/dose** (5 mgrs/kg total dose) administered the first doses in pretransplantation hours and second doses on the first post-transplant day. It is recommended to reduce doses by half in case of leucopenia (2000-3000 cels/mm^3) or thrombopenia (50,000-75,000 cels/mm^3). Below these figures assess discontinuation of treatment.

  - Form of administration: Remove 5 ml from the solvent vial and place it in the lyophilisate vial, stirring to a homogeneous solution (5 mgr/ml). Then extract the equivalent of the required dose and dilute in glucose 5 % or saline 0.9 % (recommended dilution 50 ml/vial). Infusion is recommended in 6 hours (4 h minimum). Use 0.22 µ filter. Once reconstituted use within 4-6 hours maximum.

  - Premedication:

- Methyl-prednisolone: 5 mgr/kg iv
- Nolotile: 0,1 cc/kg iv
- Polaramine: 0.05 mg/kg iv

- Monitoring of constants every half hour during infusion.

SIROLIMUS: as an alternative or combined with tacrolimus.

- TRADE NAME: RAPAMUNE® (WYETH).
- PRESENTATION: Oral solution 1 ml = 1 mgr and 1 and 2 mg tablets
- RULES OF ADMINISTRATION: oral or SNG. It can be diluted in water or orange juice. It is recommended to take it before meals, although it can be taken with or without food.
- DOSE: **2 mgr/m^2/day** in one dose.

  - Dose modifications: The dose of Sirolimus shall be modified based on clinical evidence of toxicity and efficacy, and to maintain blood **levels: 5-10 ngr/ml**
  - Levels: These are valley levels extracted before the morning dose. It is recommended not to remove them more than 2 times per week except situations that may affect their metabolism such as liver failure or association of inhibitors (fluconazole, ketoconazole, itraconazole, erythromycin, clarithromycin, metoclopramide, cimetidine etc) or activators (rifampicin, carbamazepine, phenobarbital or faith)
  - nitoin) of cytochrome P450.
  - Samples 3-5 mi of blood in tube with EDTA. If your processing is in 24-48 h store in a refrigerator at 2-8 degrees. If it will take longer to process the sample to freeze to -20°.

RITUXIMAB: (treatment of humoral rejection and AHAI))

- TRADE NAME: MABTHERA®
- PRESENTATION: 1 intravenous ampoule = 10 ml = 100 mg.
- RULES OF ADMINISTRATION: Dilute 200 mg in 200 ml of SSF or the proportional part corresponding to its dose (vials of 100 and 500 mg). Switch to 20 ml/hour on the 1st hour and 50 ml/hour the following. Total time: 4 and a half hours
- DOSE: 375 mg/m^2/week 4 doses.

— PREMEDICATION:

- Urbason 1 mg/Kg i.v.
- Acetaminophen 15 mg/Kg i.v.
- Polaramine 0.05 mg/Kg i.v.

INFLIXIMAB: treatment of cell rejection

TRADE NAME: REMICADE®

- PRESENTATION: 1 ampoule intravenous = 100 mg powder.
- RULES OF ADMINISTRATION: Dilute each 100 mg vial in 10 ml of sterile water for injection. **DON'T SHAKE**. Mix gently around. Once diluted, allow the vial to stand for 5 minutes. Reconstitute the total dose by completing SSF up to 250 ml in glass container. Purge the system with the SSF filter. Spend in 2 and a half hours through a filter of 1.2 microns or less. Purge again with SSF. Monitor blood pressure and HR every 15 minutes.
- DOSE: 5 mg/kg weekly dose.

PROSTAGLANDIN E1: if infusion iv of tacrolimus is needed.

- TRADE NAME: ALPROSTADIL® (PHARMAC SPAIN).
- PRESENTATION: 1 intravenous ampoule = 1 mi = 500 micrograms.
- RULES OF ADMINISTRATION: Dilute 1 ampoule (500 micrograms) in 25-100 cc of physiological saline or glucoseed serum. The solution is stable 24 hours once prepared.
- DOSE: home **0.03 micrograms/kg/minute** with gradual increase to **0.09 micrograms/Kg/minute** in the first 8-12 hours until intravenous infusion withdrawal of tacrolimus.

TOCILIZUMAB INFUSION (ROACTEMRA®)

- Presentation: Solution 20 mg/ml. Vials of 4 ml (80 mg), 10 ml (200 mg) and 20 ml (400 mg).
- Dosage: >30 kg: 8 mg/kg; ≤ 30 kg: 12 mg/kg. Every 2 weeks IV.
- Administer diluted in SSF. 100 ml in >30 kg; 50 ml in ≤ 30 kg.
- Remove from the SSF bag a volume equal to the volume of Tocilizumab required for the patient's dose. Replace it with the corresponding volume of the Tocilizumab solution. The final volume should be 50 or 100 ml, depending on the patient's weight. Mix the solution by inverting the bag.
- Rhythm: 10 ml/hour for the first 15 minutes. Rest at 65 ml/hour until the end. Wash with 10 ml of SSF at the

end.

- Monitor fever, skin reaction, rash, etc.
- Have 60 mg Urbason preparations, if needed.
- Constants at the beginning, 30 minutes and every hour until the end.

GAMMAGLOBULIN INFUSION

Presentation: 50 mg/ml 0.5-1 ml/kg/hour. For 30 minutes.

Then 5 ml/kg/hour until the end.