# SDM79 from PAA company

## Material :

* **ONLY 1** **bottle** of Medium from PAA : SDM79 – CGGGPPTA w/o glucosamine (Cat No : G3344,3005 Batch : #G311312-1054)
* 2 washed and autoclaved bottles (5 liters each).
* 5 Aguettant water bottles. (ref:3195085) (store room)
* 1 Nalgene 0.2 μm filter (500 mL).
* 10 bottles (500 mL) washed and autoclaved.
* 10 N NaOH in mQ water to adjust the pH.
* 500 mL of Foetal bovine serum (GE ref A15-101 Batch #A10111-0906).
* 25 mL of 2 mg/ml Hemin (Sigma H-5533) in 0,05N NaOH.
* A stirrer.
* Parafilm.
* Following Components :

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| **Components** | **Weight** | **Final concentrations** |
| NaHCO3 (Sigma #S-6014) | 11 g | 2,2 g/L |
| D-Glucose (Euromedex #UG3050) | 9,3 g | 10 mM |
| L-Glutamine (Sigma #G-8540) | 2,6 g | 3,5 mM |
| L-Proline (Sigma # P-5607) | 3,1 g | 5,3 mM |
| Sodium Pyruvate (Sigma #P-3662) (+4°C) | 0,45 g | 0,9 mM |
| L-Thréonine (Sigma #T-8441) | 2 g | 3,4 mM |
| Acide glutamique (Sigma #G-5889) | 0,11 g | 0,15 mM |
| Sodium Acetate (Euromedex #EU0310-B) | 0,051 g | 0,12 mM |
| D-glucosamine HCl (Sigma #G1514) | 0,25g | 0,23 mM |

## Procedure :

Mix all the powders in a 5L bottle with 4 L of Aguettant water.

When the powders are dissolved, adjust the pH to 7.4 with 10N NaOH (10-15 mL approx.).

Complete with the Aguettant water to 4.5 L.

Add 500 mL of Fetal Bovine Serum heated at 56 °C for 30 minutes

Add 25 mL of 2mg/ml Hemin (Sigma H-5533) in 0,05N NaOH freshly made

Under a flow hood , filter the media through a 0.2 μm Nalgene filter (500 mL) in 10 bottles of 500 mL.

Test for contamination 2 ml of each bottle into a 24 wells plates.

Store the media at +4°C.