

## ANNEXES

### 1. Recipes. Electrophoresis and Western Blot.

#### Polyacrylamide stacking gel (5%)

❖ H <sub>2</sub> O	3.22 mL
❖ Tris HCl 0.5 M pH 6.8	1.25 mL
❖ Acrylamide (37.5%)	0.53 mL
❖ SDS (10%)	50 µL
❖ APS	50 µL
❖ TEMED	5 µL

#### Polyacrylamide separating gel (10%)

❖ H <sub>2</sub> O	4.83 mL
❖ Tris HCl 1.5 M pH 7.5	2.50 mL
❖ Acrylamide (37.5%)	2.67 mL
❖ SDS (10%)	100 µL
❖ APS	100 µL
❖ TEMED	10 µL

#### Running buffer

❖ Glycine	144 g
❖ Tris base	30 g
❖ SDS	10 g
❖ H <sub>2</sub> O	1 L

#### Native running buffer

❖ Glycine	28 g
❖ Tris base	6 g
❖ H <sub>2</sub> O	2 L

Adjust to pH 7.5

### **SDS-PAGE sample loading buffer**

❖ Tris HCl 0.5 M pH 6.8	4 mL
❖ SDS	0.8 g
❖ Glycerol 100%	4 mL
❖ 0.5 M EDTA	1 mL
❖ Bromophenol Blue	8 mg
❖ H <sub>2</sub> O	5 mL

### **Native sample loading buffer**

❖ Tris HCl 0.5 M pH 6.8	2 mL
❖ Glycerol 100%	4 mL
❖ Ponceau	8 mg
❖ H <sub>2</sub> O	4 mL

### **Transfer buffer**

❖ Glycine	14.41 g
❖ Tris base	3.03 g
❖ Methanol/ethanol	200 mL
❖ H <sub>2</sub> O	to 800 mL

### **Tris HCl 0.5 M pH 6.8**

❖ Tris base	30.29 g
❖ Water	to 500 mL

Adjust pH to 6.8

### **Tris HCl 1.5 M pH 8.8**

❖ Tris base	90.8 g
❖ Water	to 500 mL

Adjust pH to 7.5

### **TTBS**

❖ 10x TBS	100 mL
❖ Tween 20	1 mL
❖ H <sub>2</sub> O	to 900 mL

### **Poinceau**

- ❖ Poinceau 100 mg
- ❖ Acetic acid 5 mL
- ❖ H<sub>2</sub>O mQ up to 100 mL

### **Blocking milk**

- ❖ Powder milk 1.25 g
- ❖ TTBS 25 mL

## **2. Recipes. Purification.**

### **NaCl 5 M**

- ❖ NaCl 58.44 g
- ❖ H<sub>2</sub>O 200 mL

### **Phosphate buffer pH 7.5**

- ❖ Na<sub>2</sub>HPO<sub>4</sub> 3.5 g
- ❖ NaCl 2.9 g
- ❖ H<sub>2</sub>O 500 mL

### **Imidazole 0.5 M (in phosphate buffer pH 7.5)**

- ❖ Na<sub>2</sub>HPO<sub>4</sub> 3.5 g
- ❖ NaCl 2.9 g
- ❖ Imidazole 17 g
- ❖ H<sub>2</sub>O 500 mL

### **Eluting solution (imidazole)**

- ❖ NaCl 5 M 2.5 mL
- ❖ Imidazol 5 mL
- ❖ Phosphate buffer 42.5 mL

### **NiCl<sub>2</sub> 0.1 M**

- ❖ NiCl<sub>2</sub> 1.3 g
- ❖ H<sub>2</sub>O 100 mL