Illustrated procedure for covalent immobilisation of dyes to foils, layers and textiles

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(Please also consult the original publications for the required amounts of materials and the procedures)

Chemicals and materials

HCl (37%; M = 36,46 g/mol)

H_2SO_4 (98%; M = 98,08 g/mol)

NaOH (pearls, MW = 40,00 g/mol)

Na_2CO_3 (MW = 105.99 g/mol)

pH indicator dye of choice (GJM- 492, GJM 503, GJM-534)

pH-insensitive dye Remazol Brillantblau R (RBBR, MW = 626.54, purity ~50%)

Cellulose material of choice (paper, foil, textile etc.)

Glassware and utensils

Glass dish or plastic tray (# is depending on the amount of different cellulose materials to be coloured separately)

Schott bottles 250 ml (4)

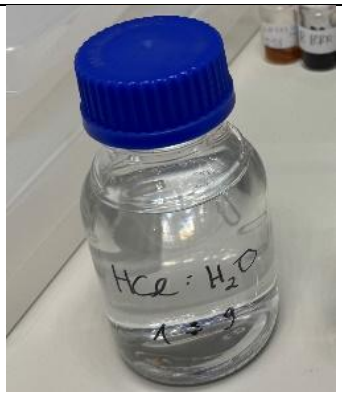
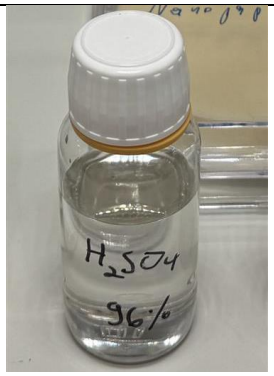
Mortar and pestle (1)

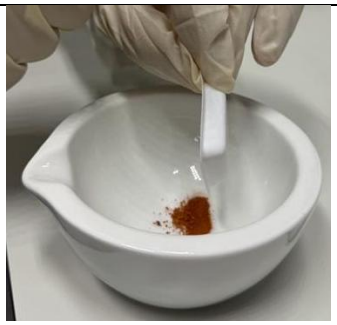
Graduated cylinder (10 ml, 100 ml, 250 ml)

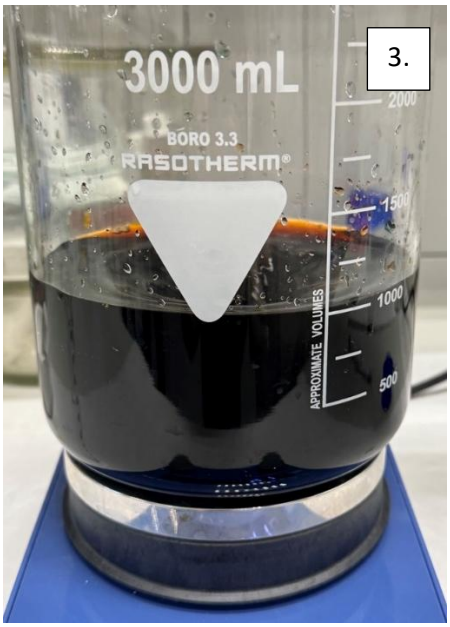
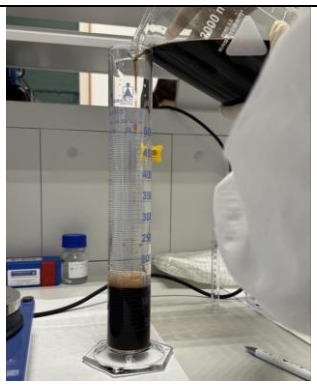
Beaker 1.5 L or bigger (1)

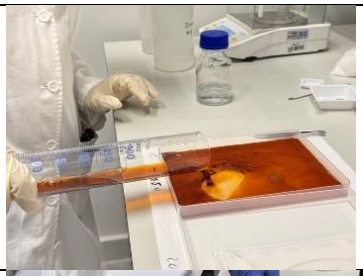
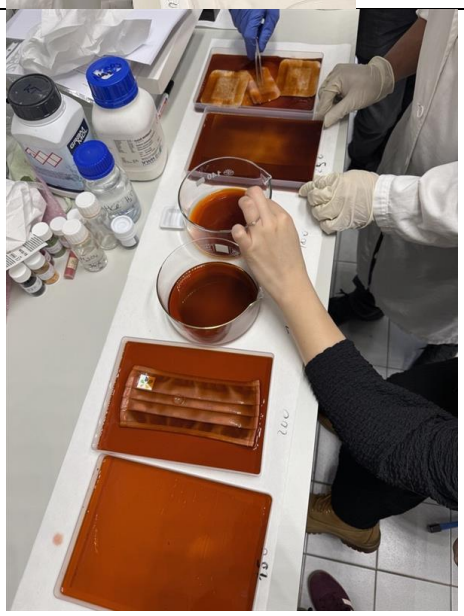
Magnetic stirrer, plus magnetic stirring bar, length > 5cm

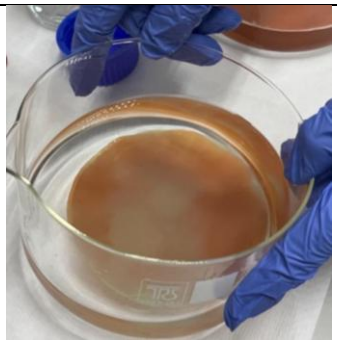
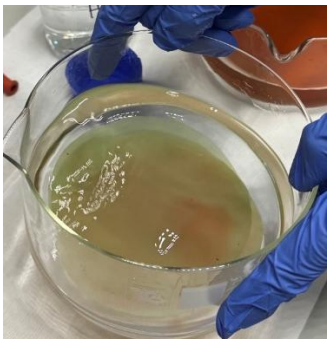
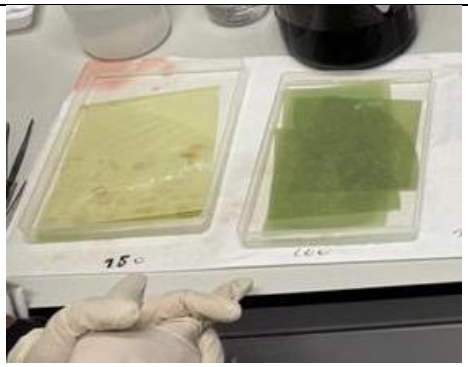
Glass/plastic pipettes

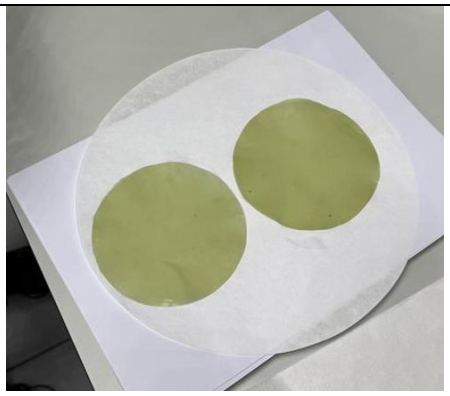
Step	Procedure description	Scheme
1.	Prepare diluted HCl solution; mix HCl:H_2O in 1:9 ratio (25 ml of HCl and 225 ml of distilled water) and put solution in Schott bottle.	
2.	In small air-tight container, put around 10 ml of 96% H_2SO_4 (directly from original bottle), to be used for esterification of the respective pH indicator dye.	
3.	Prepare 30% NaOH solution: Weigh 30 g of NaOH pearls and then put it in a Schott bottle in the fume hood; add 35 ml of distilled water in Schott bottle with NaOH, shake it carefully for approx. 2-3 minutes because of strong exothermic reaction. After that add remaining 35 ml of distilled water and shake it carefully again. Let the solution cool down.	
4.	Prepare Na_2CO_3 solution: Weigh 25 g of Na_2CO_3 and transfer it to the Schott bottle and fill it up with distilled water up to 200 ml mark.	

5.	Prepare the dye/acid mixture in a mortar: Weigh 100 mg of the selected pH indicator dye and carefully transfer it to the mortar. Then add 1 g of H_2SO_4 dropwise using a dropper or pipette. After adding the acid, gently move the pestle over the mixture until it becomes completely liquefied. Mix gently, as vigorous grinding may cause the dye to stick to the bottom of the pestle. Once the dye/acid mixture is properly homogenized, allow it to stand for 25 minutes. Then gently mix the dye/H_2SO_4 solution again for 5 minutes to complete the homogenization.	 
6.	After the 30-minute waiting period described in step 5, prepare the pH-insensitive RBBR dye solution (if required for dyeing) by weighing 80 mg of RBBR into a Schott bottle, adding 100 mL of distilled water, and gently shaking the closed bottle for approximately 5 minutes.	 
7.	Place the beaker containing the stir bar on the magnetic stirrer and add 600 mL of distilled water. Set the stirrer to mixing mode at 400 rpm.	

8.	**Add the following components to the beaker from step 7 in the order given below:** 1. Add the **viscous GJM dye/H_2SO_4 mixture** directly from the mortar. Then rinse the mortar with **100 mL of distilled water**, using the pestle to help remove any remaining residue. This will result in an **orange-colored solution** in the beaker. 2. Add **100 mL of the pH-insensitive dye RBBR solution**. When using the pH indicator dyes **GJM-492** or **GJM-503** together with RBBR, the solution changes color from **orange to dark green**. If no RBBR is used, as in the case of **GJM-534**, add **100 mL of distilled water** instead. 3. Add **200 mL of Na_2CO_3 solution**. When using **GJM-492** or **GJM-503** together with RBBR, the solution changes color from **dark green to dark orange**. 4. Add **7 mL of NaOH solution**. 5. Allow the solution to stir for **1 minute**. After this procedure, the solution is ready for **covalent immobilization**. The total volume of the dyeing solution should be **1000 mL**.	  
9.	Transfer the solution to a graduated cylinder, then distribute it into dishes or plastic trays if multiple different materials are to be colored.	

		
10.	Place the cellulose-based materials into dishes or plastic trays filled with the solution from step 9. Allow them to remain in contact with the solution for 30 minutes, stirring or gently moving them occasionally to ensure uniform dye immobilization.	
11.	Rinse the colored materials to remove unbound dye from the foils, layers, or textiles: Carefully decant the solution from the crystallizing dishes or plastic trays while gently holding the materials in place with a finger. Add distilled water to each dish or tray and allow the materials to soak for 3 minutes, gently stirring or moving them occasionally. Remove the water and repeat the rinsing procedure with fresh distilled water at least four additional times.	 

12.	Add HCl to neutralize the residual NaOH: After the fifth rinse with distilled water, add fresh distilled water to the dishes or plastic trays and add a few drops of HCl to each. Stir gently and observe the color change from orange/red to green. If the color does not change, add a few more drops of HCl until the change is visible.	   
13.	Once all cellulose-based materials have changed to the acidic form (green in this case), rinse them with distilled water at least five additional times to remove the acid completely. For textiles and thicker materials, more than five rinsing cycles may be necessary. Textiles should be wrung out several times during rinsing.	

		
14.	Place the cellulose materials between 3–4 layers of filter paper and cover them with a flat, heavy object (for example, a book) to ensure that they remain flat during drying. Take care to arrange all materials evenly, without shrinkage, wrinkles, or folds, between the filter paper layers. After drying, store the materials in the dark at room temperature until use.	 

If you have questions/comments, please write to:

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