

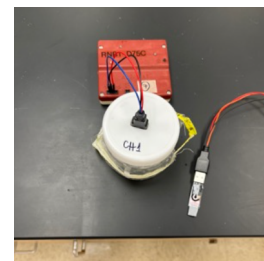
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Version 2

# Fabrication of laser inscribed graphene (LIG) 3-electrode plug-and-play chip V.2

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**We use this protocol and it's working**

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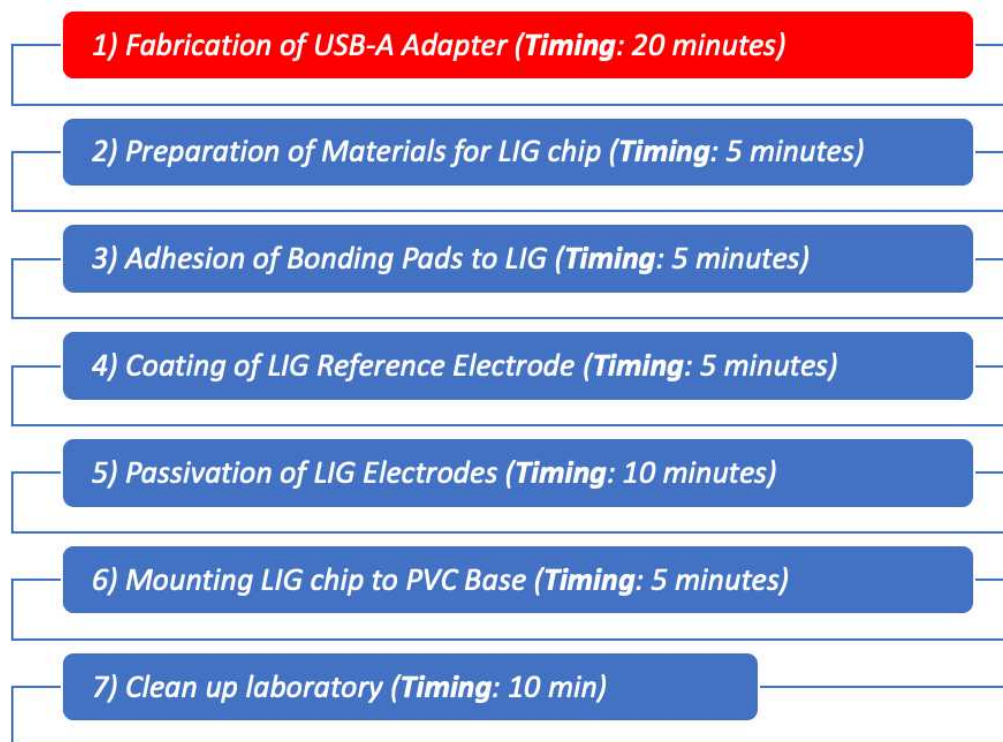
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## Abstract

Laser inscribed graphene (LIG) is a versatile material that is commonly used to prepare electrochemical sensors (Moreira et al 2023). This protocol describes the fabrication of a 3-electrode plug-and-play chip system based on LIG. The design is compatible with a female USB-A connector which can be modified for connection to any potentiostat using the protocol here. The complete process requires approximately 60 min to complete, including fabrication of USB-A connector (**Fig 1**).



**Figure 1.** Process flow for fabrication of laser inscribed graphene (LIG) chip compatible with USB-A. The process shown here is for a batch of 18 LIG chips, and requires approximately 60 min to complete. The USB-A adapter may be reused, and thus is shown as a separate section. The protocol depends on referenced protocols for fabrication of LIG (not shown here).

## Image Attribution

Plug and play system is compatible with any potentiostat and can be directly integrated with sample container(s)



## Materials

### *Hardware*

- Hardware required for fabrication of LIG using Universal Laser, see protocols here ([link](#))
- Soldering station ([link](#))

### *Software*

none

### *Chemicals, Reagents and Other Materials*

- Type HN Amber plain-back polyimide film (0.005" thick") ([link](#))
- 70% Isopropyl alcohol ([link](#))
- Low-Lint Cloth Wipes ([link](#))
- Masking Tape ([link](#))
- Conductive polyester metal tape (0.13 mm thick) ([link](#))
- Double-sided film tape ([link](#))
- Chemical-resistant PVC Sheets of 1/16" thick, 12X12 sheet ([Link](#))
- Certified eyewear ([Link](#))
- USB-A Female Port Socket with Plastic Cover ([link](#))
- 28AWG jumper wires, PVC insulation ([link](#))
- 395 nm UV flashlight ([link](#))
- Scissors (local store)
- Steel tweezers (local store)
- Fast Drying poly gel acrylic nail polish (UV curing) (local store)
- Timer (local store)



## Safety warnings

### ! Eye protection

- Laboratory eye protection is required when cutting PVC, as small shards may cause eye damage during the preparation of electrode base material process.

### Skin

- Avoid contact of acrylic nail polish with skin. Proper use of PPE should avoid any problems ([link to health hazard information](#))\

### Fumes/aerosols

- All use of the following chemicals must be conducted in a chemical hood or in a well-ventilated space
- Acrylic nail polish (or other types of nail polish ([link to health hazard information](#)))
- Isopropyl alcohol ([link to SDS](#))
- All soldering should be conducted in a chemical hood to avoid iron based solder fumes

### Heat and Flammable materials

- Avoid open flame or source of ignition when working with acrylic nail polish with skin ([link to health hazard information](#))

### Disposal

- Any excess polyamide, metal tape, or PVC sheet should be discarded in a labeled container on the counter.

## Ethics statement

N/A

## Before start

Ensure ample table space is available, and located in a well ventilated area.

### Safety information

- Wear eyewear protection at all times
- Wear gloves and lab coat at all times



## 1 *Step 1) Fabrication of USB-A Adapter*

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- Heat the soldering iron, and prepare a wet sponge and solder wick.
- See the following for an introduction to soldering techniques ([link](#))
- Solder one 28AWG jumper wire to each of the two outer contacts in the USB-A adapter.
- Create an electrical connection (i.e., “jump”) the two inner contacts in the USB-A adapter.
- Solder one 28AWG jumper wire to the “jumped” inner contact(s) in the USB-A adapter
- Place the plastic cover on the USB-A Female adapter
- Turn off soldering iron, and clean station when finished

## 2 *Step 2) Preparation of Materials for LIG chip*

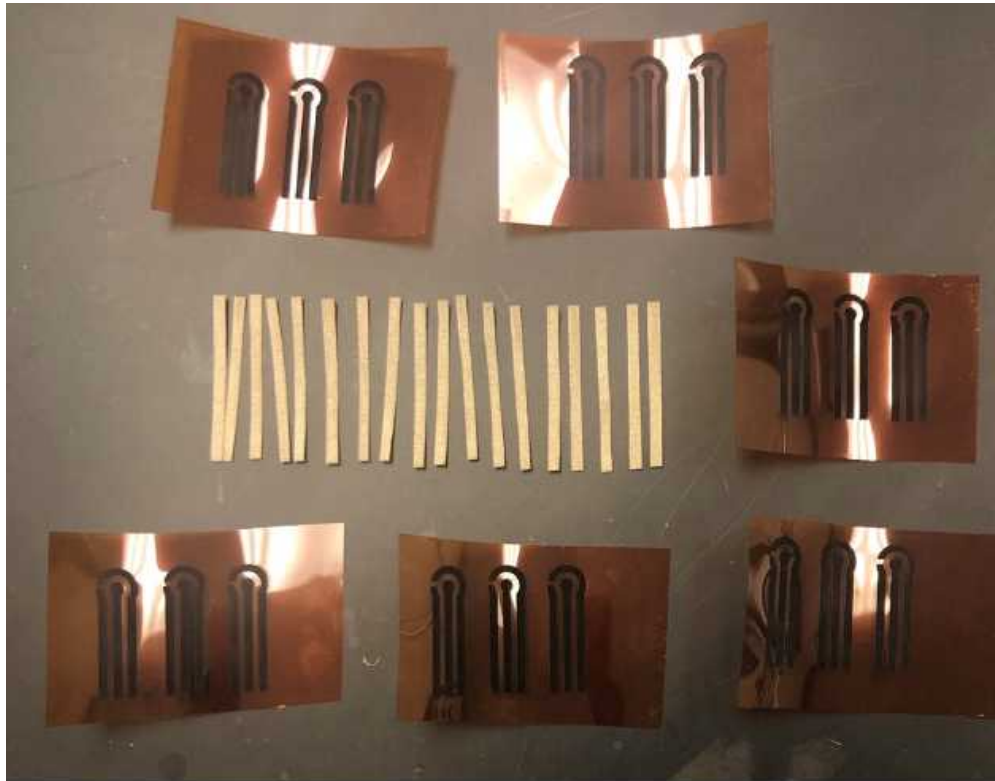
5m

- LIG should be prepared using the protocol developed for a 3-electrode sensor ([link](#))
- Using scissors, cut conductive polyester metal tape into 0.3 cm by 3.5 cm strips (one strip per LIG electrode)
- **Fig. 1** shows an example of a batch of 18 LIG electrodes with 18 metal tape strips
- For each LIG chip, cut a 1cm by 5 cm strip of PVC to act as the chip base
- Ample table space should be cleared to prepare the LIG chips

### Note

#### **Critical Step:**

Ensure that the PVC base is the correct width by inserting into the USB-A female adapter. The fit should not be too loose, but should be snug when inserted.



**Figure 1.** Photo of materials prepared for a batch of 18 LIG electrodes with 18 polyester metal tape strips (PVC base strips not shown in photo)

### 3 Step 3) Adhesion of Bonding Pads to LIG

5m

#### Note

**Critical step:**

Use caution and avoid touching the surface of the LIG material

- Using scissors, cut small 0.3 cm by 1.0 cm strips from the prepared polyester metal tape strips. One small subsection should be cut for each 3-electrode LIG sensor.
- Using steel tweezers, hold the metal tape and carefully remove the adhesive backing
- Carefully position the metal tape over one of the bonding pads on the printed LIG chip
- After coating the LIG reference electrode, the tweezers should be used to lightly squeeze the material and ensure good contact.
- Repeat the process for all three electrodes in the LIG chip (**Fig 2**)



**Figure 2.** Photo demonstrating adhesion of polyester metal tape strips to bonding pad of LIG design. Each bonding pad corresponds to the working, counter, or reference electrode.

#### Note

**Critical step:**

Care must be taken to ensure that the metal tape used to create bonding pads is not contacting other components of the chip, including other bonding pads.

#### 4 Step 4) Coating of LIG Reference Electrode

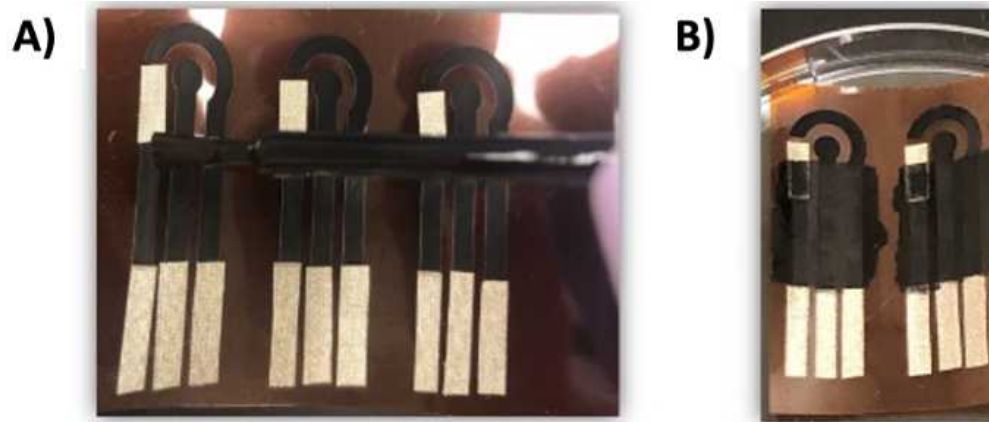
- The reference electrode may be coated with Ag/AgCl ink, or with a small 0.3 cm by 0.5 cm strip of polyester metal tape (as shown here)
- After coating the LIG reference electrode, the tweezers should be used to lightly squeeze the material and ensure good contact.
- If Ag/AgCl ink is used to coat the reference electrode, the material should be allowed to cure at room temperature for at least 15 min

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#### 5 Step 5) Passivation of LIG Electrodes

- Use poly gel acrylic nail polish to passivate the electrodes between the bonding pad and the functional electrode area (**Fig 3A**).
- The passivation may cover a small portion of the polyester metal tape if used as reference electrode coating material (**Fig 3B**)

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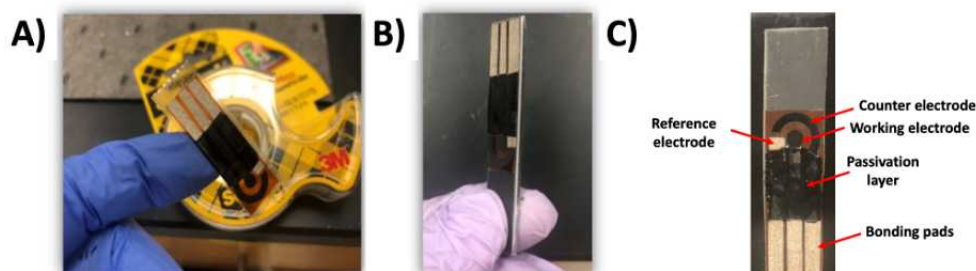


**Figure 3.** Passivation of LIG electrodes for creating functional electrode area and bonding pads.

## 6 Step 6) Mounting LIG chip to PVC Base

- Using scissors, cut a strip of double sided tape and adhere to the back of the LIG material (**Fig 4A**).
- Mount the LIG material to the PVC base and use scissors to trim any excess tape.
- The final chip should have clearance with exposed, unmodified PVC which may be handled without touching the LIG or other coating materials (**Fig 4B**)
- The three electrode system is composed of working, counter, and reference electrodes. The passivation area and bonding pad are easily discernable (**Fig 4C**)

5m



**Figure 4.** **A)** Cut a strip of double sided tape and adhere to back of LIG. **B)** LIG electrode mounted to PVC base. **C)** Example of completed LIG chip for plug-and-play with USB-A system. **C)** The final design is a plug-and-play 3 electrode LIG chip compatible with the USB-A adapter

## 7 Step 7) Clean up laboratory

- Any excess polyamide, metal tape, or PVC sheet should be discarded in a labeled container on the counter.

10m



- Turn off soldering iron, and clean station
- If storing electrodes, place in a sealed Petri dish with desiccant.

## Protocol references

Moreira, G. *et al.* A capacitive laser-induced graphene based aptasensor for SARS-CoV-2 detection in human saliva. *PLoS One* **18**, e0290256 (2023).