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CONDENSED MATTER

Determination of the Number of Graphene Layers on Different Substrates by Optical Microscopy Technique

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Abstract We present a method, based on transmission or reflection optical microscopy, to determine the number of graphene monolayers deposited on various substrates. To demonstrate the procedure, we synthesize graphene samples and deposit them on various substrates with the micromechanical cleavage technique. Our procedure initially relies on more classical approaches such as atomic force microscopy (AFM) and Raman to calibrate the equipment. After calibration, however, optical microscopy by itself is sufficient to characterize other samples, deposited on any substrate.

Keywords Graphene · Optical microscopy · AFM

1 Introduction

Graphene is a nanostructured material that has received much attention from the scientific community: a form of pure carbon constituted of flat, stacked layers of C atoms. Its bidimensional structure, in which the carbon atoms form a plane hexagonal network one atom thick, presents an enormous variety of peculiar properties, such as high electronic conductance [1], high mechanical resistance [2], unique optical properties [3], and numerous potential technological applications [4, 5].

The study of graphene starts with fabrication, usually via micromechanical cleavage of graphite [1], chemical vapor deposition (CVD) [6], or epitaxial growing [7, 8]. Next comes cataloging, which establishes the number of graphene layers in the samples. The sample must be subsequently characterized, after which nanodevice fabrication becomes possible.

To measure the number of number of grapheme monolayers, one usually resorts to relatively sophisticated techniques, such as atomic force microscopy (AFM) [1, 2] and Raman [9], which are restricted to small areas of the samples.

One of the most popular fabrication methods is the micromechanical cleavage technique, developed by Geim and Novoselov [1, 2], a technique that allows easy manipulation of the bidimensional system keeping unchanged its electronic, transport, mechanical and optical properties.

Historically, the optical properties of the new material were first studied with optical microscopy by Geim and Novoselov's group [3], who also used a spectrometer. Their work clearly showed that the number of graphene monolayers deposited on glasses substrate can be counted using optical microscopy transmittance, via measurement of the light percent transmittance or reflectance. Nonetheless, we are unaware of any subsequent attempts to determine the number of graphene monolayers on other substrates, such as silicon oxides or gold. Here, we demonstrate that the number of layers deposited on glass, silicon oxides, and gold substrates can be determined by optical microscopy, either by reflectance or transmittance measurements, depending on the substrate. This alternative to the already mentioned more sophisticated techniques is easily incorporated in any optical-microcopy system.

2 Experimental

Following the classical micromechanical cleavage synthesis technique, we used a Nitto blue tape to exfoliate natural graphite [1, 2]. We then deposited the graphene on different substrates, the choice being dictated by potential technological and scientific applications. Among the important substrates

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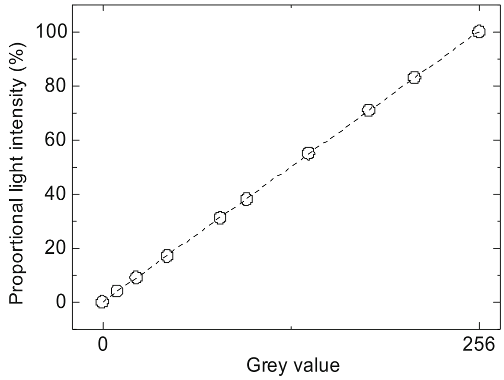

Fig. 1 Calibration function of the camera. The gray value of the 8-bit image must be a linear function of the proportional light intensity falling on the CMOS sensor

are Si wafers, used to make nanodevices, gold substrates, used in nanodevices and as substrates for FT-IR spectroscopy and AFM, and Corning glass 7059 and Corning glass 211 which are both very convenient for sample fabrication and optical study.

In order to establish a benchmark for our experiments, we used Raman-shift spectroscopy [9] to search for a sample with monolayer, bilayer, and trilayer regions in the same sample. Once one such sample was found, we went to the next step and calibrated our software and equipment to detect the different numbers of layers in this reference sample, which was deposited on a Corning glass 211 substrate.

Our measurements used a standard optical microscope from Carl Zeiss, with transmission- and reflection-imaging mechanisms. A CMOS monochromatic camera was attached to the microscope. This camera was chosen because it has a sensor that transfers digital data from the sample to the software under the conditions of our study. In a similar research, one that was nonetheless restricted to a single substrate, Nair et al. [10] monitored the transmission through monolayers of

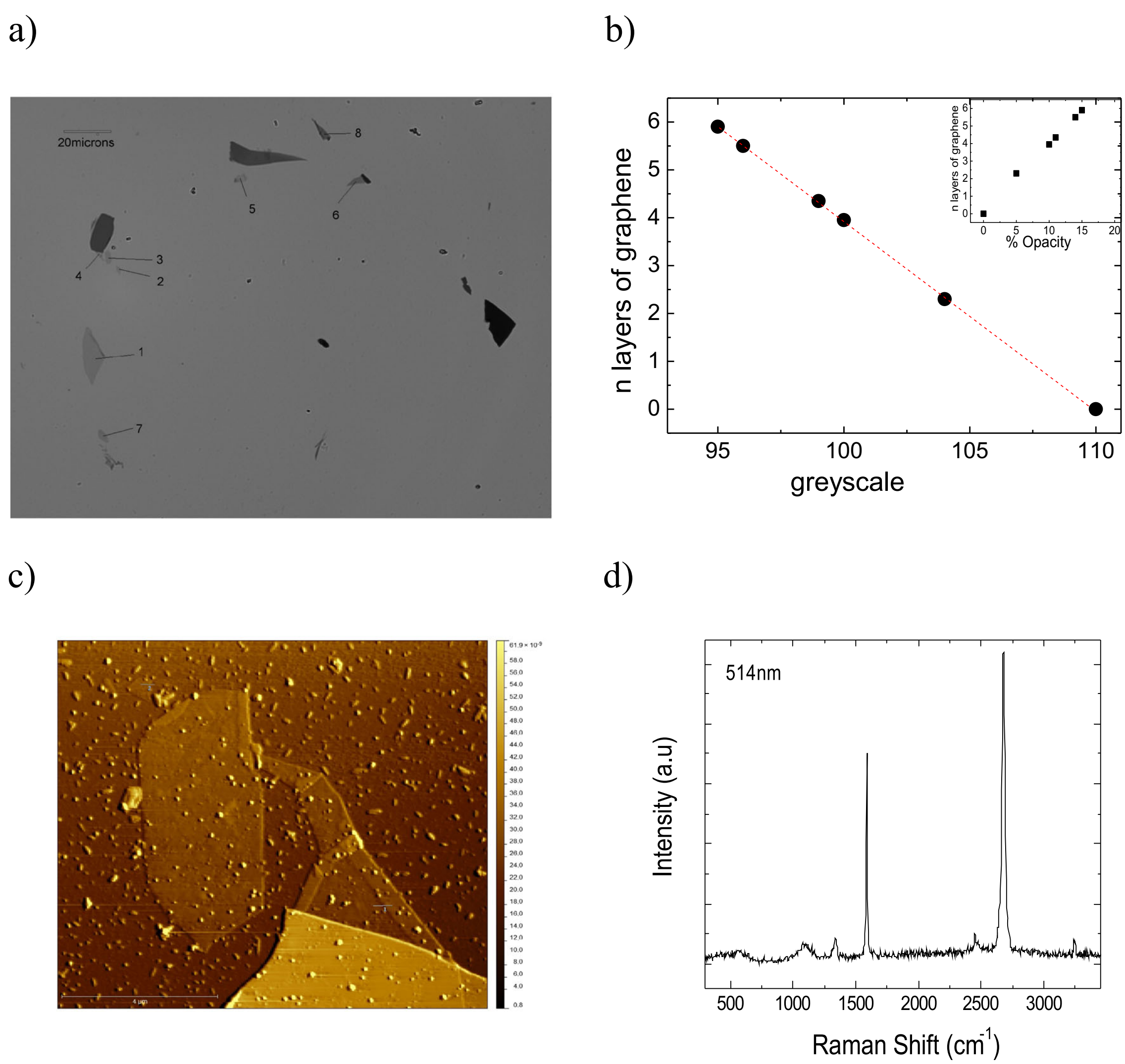

Fig. 2 **a** Optical microscopy of few layers of graphene deposited on Corning glass 7059, and **b** shows the number of monolayers (n) of graphene and the linear relation of the n with the opacity percent. **c** AFM and **d** Raman spectroscopy confirm monolayers of graphene in the sample deposited on Corning glass 7059

graphene under the light of a Xe lamp (250–1200 nm). In this and other studies, a relation between the opacity and the number of graphene layers was observed. We have taken advantage of this finding to study the transmission and reflection of graphene on different substrates under the white light of an optical microscope and try to relate it to the number of graphene layers. Mathematical treatments relating the optical constants to the contrast and wavelength of the incident radiation have been reported in the literature [11, 12]. It is not the purpose of our work to confirm such theoretical developments, which deal with phenomena that by now are amply understood. Our objective, instead, is to show that optical microscopy offers a simple, practical way of determining the number of graphene layers on a variety of substrates.

To this end, we have modified the camera to disable all automatic-contrast and gain configurations, so that the sensor detected the physical properties of the original image. While the software of certain cameras offers this reduction as an option, we had to physically modify our camera to bypass contrast enhancement and gain. Under these conditions, every pixel of the camera detects the light intensity falling upon it and transforms it to an 8-bit image datum that is transferred to the attached computer. The 8-bit image determines the graphene-sample opacity with 0.39 % resolution. Our camera is calibrated to linearly relate the light intensity to the resulting gray value, as shown by the agreement between the filled circles and the dashed line in Fig. 1; in practice, this is an important preliminary adjustment, which saves us the trouble of using a different equation to describe each substrate.

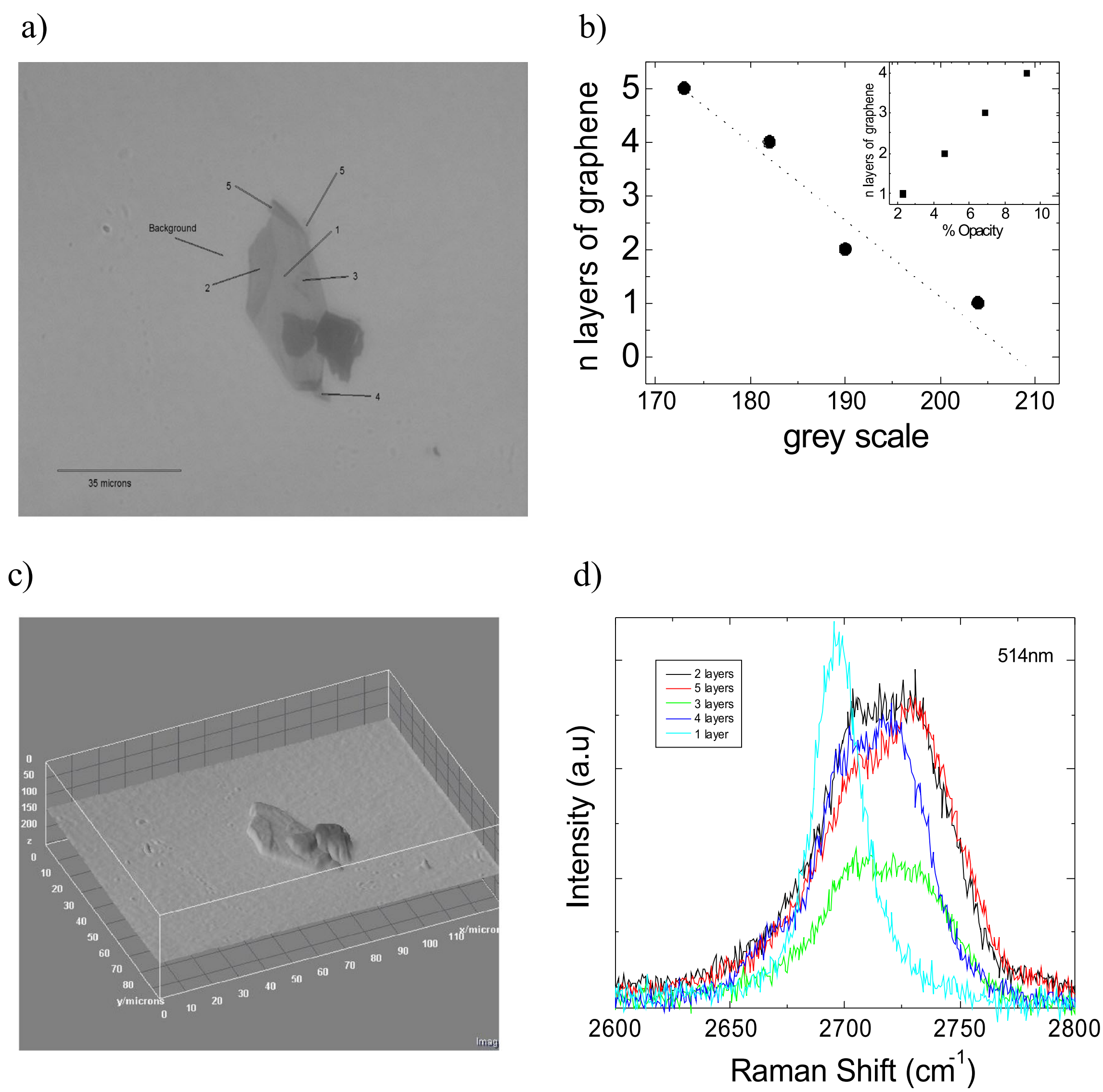

Fig. 3 **a** Optical microscopy of few layers of graphene deposited on Corning glass 211, and **b** shows the number of monolayers (n) of graphene and the linear relation of the n with the opacity percent. **c** 3D optical picture and **d** Raman spectroscopy confirm monolayers of graphene in the sample deposited on Corning glass 211

We used the ImageJ software to process the sample images captured by the sensors. To calibrate the software, we first imaged the reference sample. Since the number of layers in each region of this sample was known, we were able to construct a matrix relating the gray-scale readout to the number of layers in each region. The elements of this (diagonal) matrix are factors correcting for the distinct opacities detected by the different CMOS sensors. In other words, the calibration matrix yielded a linear relation between the gray-scale readout and the number of graphene layers in the reference sample. To analyze other samples, we had to calibrate the software to the distinct transparencies of the substrates, a relatively simple task: we contrasted images from a (black) region containing graphite and from the (near white) blank substrate and associated these extremes to 0 and 100 % transparency, respectively. For each region of graphene layers, we then applied a simple Thales theorem reasoning to find the opacity. Accordingly, given the correction factors and the opacity indicated by the software, we determined the number of layers from the following equality:

$$n\text{layers} = \frac{\text{Gray value background} - \text{grey value sample}}{\text{grey value background} \times 2.3} \times \text{correction factor} \quad (1)$$

From the monochromatic image generated by the camera, we analyzed, with a modified image-analysis program, the proportion of gray in each region of the image, following which simple manipulation of the scales yielded a 3D model of the sample. For a given sample, one such 3D model is expected to help the design of experiments to probe the physical properties of the sample.

3 Results and Discussion

Figure 2a shows the optical-microscope picture from a reference sample with few layers of graphene on a Corning glass 7059 substrate. To initially calibrate the equipment, we obtain the AFM image in Fig. 2c and Raman spectra, such as the one shown in Fig. 2d. Analysis of the Raman spectrum in each region determines the number of layers in each of the various islands in the optical image. Comparison between this information and the opacity data collected by the optical microscope then yields the calibration curve depicted by the main plot in Fig. 2b, which is a graphical representation of Eq. (1).

Once the camera and the analysis software are calibrated, we no longer needed Raman spectroscopy and can determine the number of layers directly from Eq. (1).

Brief discussion of this procedure seems warranted. Equation (1) is akin to the definition $\otimes C=(C_{\max}-C_{\min})C$, of the contrast on an 8-bit image, where $C_{\max}$, $C_{\min}$, and C are the maximum, minimum, and average gray values among the pixels composing the image. To insure uniformity between measurements in samples on different subtracts, for each substrate we scale the right-hand side of Eq. (1) so that the opacity, i.e., the first factor on the right-hand side, is 100 % for graphite and 0 % for the blank substrate. Multiplication of the opacity by the appropriate, substrate-independent correction factor then yields the number of graphene layers in each region of the sample.

As an illustration, Fig. 3 shows the procedure applied to the sample on a Corning glass 211 substrate. Figure 3a displays the microscope image, the inset of Fig. 3b shows the opacities of different regions of the samples, and the main plot, the number of layers as a function of the gray scale. For completeness, we

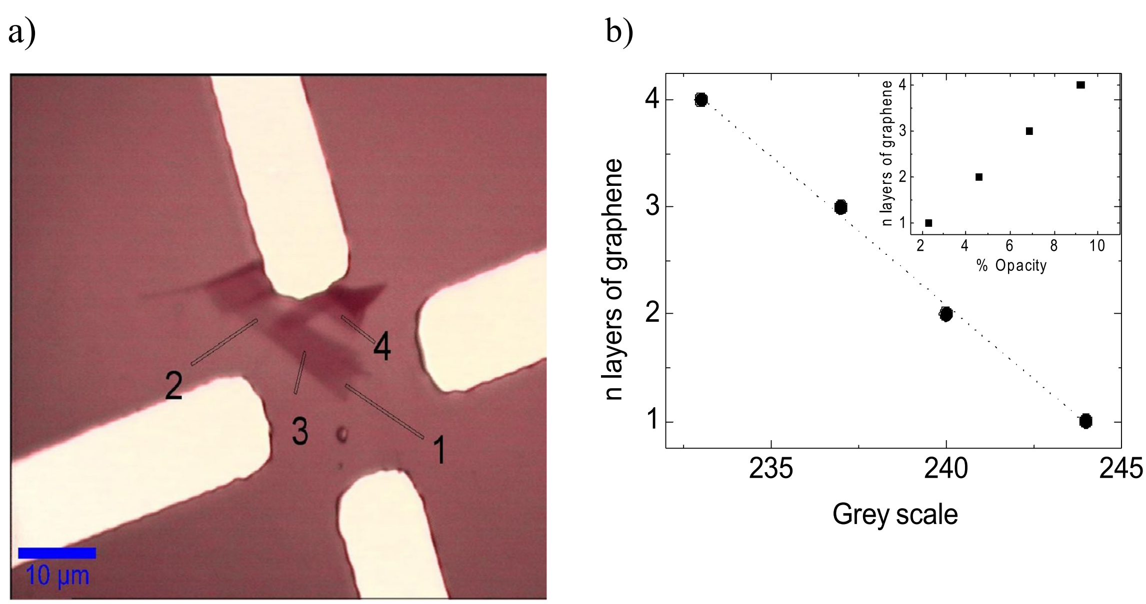

Fig. 4 **a** Optical microscopy of few layers of graphene deposited on SiO2, and **b** shows the number of monolayers (n) of graphene and the linear relation of the n with the opacity percent

also show the AFM image and the Raman spectra of the various regions in Fig. 3c, d, respectively.

For another example, Fig. 4a, b shows the optical image and the resulting plots for the sample on a Si wafer. The substrate now being opaque, one might argue that the light back reflected by the substrate through the graphene might modify our correction factors. As the inset of Fig. 4b shows, however, this is not the case, because the adjustment of the opacity to zero for the blank substrate accounts for the back reflection.

As a final consideration, we note that the correction factors are not universal, since they depend on the characteristics of the optical microscopes. For a given experimental setup, the correction factors can be determined as explained above, via comparison with the Raman spectra for a reference sample. Alternatively, one may resort to comparison between the measured opacity and the theoretical expression for the opacity as a function of the number of layers.

4 Conclusion

Starting with the monochromatic image generated by a camera, we have shown that a simple modification of the ImageJ image-analysis package to analyze the proportions gray-scale intensity in each region of the image yields a 3D model of a sample comprising few layers of graphene deposited on any substrate. We expect models of this kind to be instrumental in the planning stage of experiments focused on graphite samples. We have shown that our technique yields reliable measurement of the number of monolayer number in different regions of samples deposited on various substrates.

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