Cu_2O ordered nanoarrays for non-enzymatic glucose detection

Chenchen Liu[1], Kaifeng Xue[2], Aixin Sun[1], Dexiang Chen[1], Pinhua Zhang[1*], and Guangliang Cui[1**]

[1] School of Physics and Electrical Engineering, Linyi University, Shandong 276005, China
[2] School of Mechanical Vehicle Engineering, Linyi University, Shandong 276005, China
[*] Corresponding author. E-mail: zhangpinhua@lyu.edu.cn
[**] Corresponding author. E-mail: cuiguangliang@lyu.edu.cn

Received: Mar. 10, 2021; Accepted: Apr. 25, 2021

Non-enzymatic glucose sensors have been proposed and widely investigated due to their better stability and higher durability against external environmental conditions compared with enzymatic sensors. In this work, the Cu_2O long-range ordered nanoarrays with periodic bamboo-like nanostructure were in situ assembled by 2D electrochemistry method for non-enzymatic glucose detection. Combined with ion sputtering, we developed a portable non-enzymatic glucose sensor based on Cu_2O ordered nanoarrays. The sensor exhibits excellent sensitivity and specificity toward glucose and a linear relationship with glucose concentrations ranging from 0.05 to 15 mM. In short, the Cu_2O ordered nanoarrays developed with special periodic and long-range order nanostructure are expected to be used for the application of non-enzymatic glucose sensors.

Keywords: Cu_2O, Nanoarrays, 2D electrochemistry, Non-enzymatic, Glucose

http://dx.doi.org/10.6180/jase.202112_24(6).0002

1. Introduction

Diabetes mellitus is a worldwide public health problem. This metabolic disorder results from insulin deficiency are reflected by blood glucose concentration higher or lower than the normal range of 80-120 mg/dL (4.4-6.6 mM) [1]. For diabetics, the frequent test of physiological glucose levels is of great significance to avoid emergencies (e.g., hyperglycemia and hypoglycemia) and to assess whether a diabetes treatment protocol is efficient or not. This huge requirement has made glucose sensors dominate approximately 85% of the entire biosensor industry [2]. Due to the extremely large financial burden caused by diabetes and its serious complications, glucose detection is becoming paramount in battling diabetes and reducing financial loss [3].

The rising demand for glucose sensors with high sensitivity, good reliability, long-term durability, and excellent selectivity has impelled researchers to make tremendous efforts for decades [4]. Up to now, a variety of glucose detection methods have been developed such as electrochemical, optical, thermometric, and fluorescent sensors [5]. Compared with the above methods, the amperometric method exhibits high sensitivity, reliable low detection limit, which becomes one of the important methods to detect glucose accurately [6–8]. Among these different types of sensors, enzymatic amperometric glucose sensors have a broad application prospect due to the convenient operation, based on which good selectivity and high sensitivity have been achieved for glucose detection [5]. However, owing to the intrinsic not only expensive but also vulnerable of enzymes (enzymes can easily be affected by temperature, pH, humidity and toxic chemicals), enzymatic sensors maybe not the best strategy for glucose detection [9]. The non-enzymatic glucose sensor with the same detection performance and more stability becomes the first choice for glucose detection [10].

Cu_2O has attracted intensive attention because of its

great potential for catalytic oxidation of glucose ascribed to its good redox potential and higher stability [11–13], and it has been reported in many studies as the sensing material for non-enzymatic glucose detection [14–16]. In addition, many kinds of sensors were prepared by modifying electrodes with Cu_2O nanoparticles, such as Cu_2O-straight multiwalled carbon nanotube hybrid nanostructures and Cu_2O nanoparticles deposited on polypyrrole nanowires [17]. Furthermore, in some studies, many metal oxides modified with Cu_2O were fabricated for non-enzymatic glucose detection [18, 19]. It is acknowledged that the electrocatalytic performance is directly related to the specific surface area and morphology of Cu_2O nanostructures [20].

In order to improve the glucose detection performance, Cu_2O nanostructures need to be designed specially with larger specific surface area and more distinctive morphology. In our work, we propose a Cu_2O ordered nanoarrays prepared by in situ electrodeposition and used directly for glucose detection. To achieve a stronger sensitivity of glucose, the nanoarrays were designed to possess a larger specific surface area and a more organized structure than that we have prepared by the same method before [21–23]. The sensor based on the Cu_2O ordered nanoarrays has good linearity (0.05-15 mM), low detection limit (0.05 mM), high sensitivity, and good stability towards glucose test. Hence, the Cu_2O ordered nanoarrays prepared by this method to meet the requirements of the enzyme-free glucose sensor. The ordered and controllable assembly strategy has an important reference for the design of new nano-microstructure biosensor materials.

2. Material and Methods

2.1. Materials

Experimental reagents include $Cu(NO)_2$, HNO_3, glucose, KOH, uric acid (UA), ascorbic acid (AA), NaCl, L-cysteine (L-Cys) and Deionized water were used to prepare the electrolyte and test solutions. All reagents were directly used without treatment. The copper foils (30 μm thick, 1 mm wide, 2 mm long) were used as electrodes, and cover glasses were used as substrate. Petrolatum was used as an insulation waterproof layer to prepare detection elements.

2.2. Preparation of Cu_2O ordered nanoarrays

The detailed description of the experiment has been given in our previous reports [21–23]. Briefly, the ultra-thin sample growth space is constructed by freezing the electrolyte (50 mM $Cu(NO)_2$ solution) at -1.7 °C. Then a DC voltage of 0.5 V was applied to the electrodes for the beginning of the deposition. When the sample grew to 0.5-1 mm, the deposition voltage was changed to a 0.5 Hz semi-sine wave potential with an amplitude varied from 0.3 V to 0.7 V. After the deposition, the sample was rinsed 3 to 5 times with deionized water and dried at ambient environment.

2.3. Characterization of structural and glucose detection

The morphology and nanostructure of Cu_2O ordered nanoarrays were characterized using scanning electron microscopy (SEM, ZEISS EVO 18) and transmission electron microscopy (TEM, JEOL JEM-2010F). The surface elemental composition was measured with X-ray photoelectron spectroscopy (XPS, ESCALAB MKII, VG). The electrochemical characterization of the sensor involved cyclic voltammetry (CV) and chronoamperometry experiments were carried out using a custom-made four-probe tester (including a four-electrode probe, a Keithley 2400 source meter and a computer). The CV experiments were performed between 0 and 1.8 V using a scan rate of 0.01 V/s, while a DC bias voltage of 1.6 V was utilized in the chronoamperometry experiments.

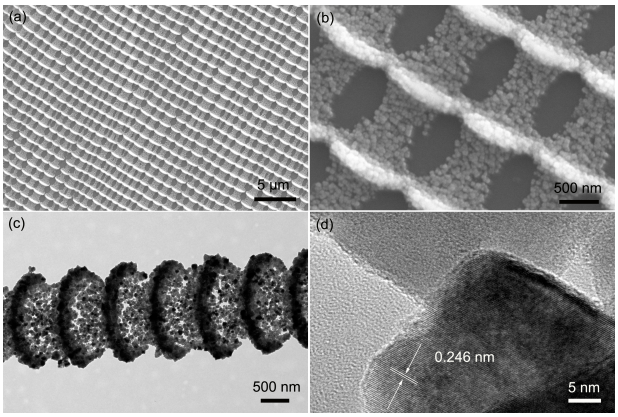

Fig. 1. SEM images of Cu_2O ordered nanoarrays with high magnification (a) and low magnification (b). (c) and (d) are TEM and HRTEM images of Cu_2O ordered nanoarrays.

3. Results and Discussion

Fig. 1a and b present the morphology, size, and microstructure of Cu_2O ordered nanoarrays by SEM at different magnification. We can clearly see in Fig. 1a, Cu_2O ordered nanoarrays are bamboo-like nano-microstructures with uniform cycle length and long-range order. Fig. 1b proves that the bamboo-like pattern is composed of periodically accumulated nanoparticles. The nanoparticles accumulate together compactly at the bamboo joints, while the accumulation is insufficient between the bamboo joints. The noncompact distribution of Cu_2O nanoparticles leads to

large specific surface area and more available catalytic active sites. The TEM image of the sample in Fig. 1c is a further indication of the periodic variation of accumulation. According to the HRTEM image of the nanoparticle shown in Fig. 1d, the Cu_2O nanoparticles are similar to cube structure, and the lattice fringes with d-spacings of 0.246 nm correspond to (111) facets of Cu_2O.

XPS spectrum was further employed to investigate the compositions and element chemical states of the samples. Fig. 2a indicates that the high distinguishability of the Cu 2p XPS spectrum has two strong main peaks at the binding energy of 952.2 and 932.4 eV, which are attributed to the $2p_{1/2}$ and $2p_{3/2}$ spin-orbital components of Cu^{1+}. In addition, two weak binding energy peaks appear at near 954.4 and 934.6 eV, which are attributed to the $2p_{1/2}$ and $2p_{3/2}$ spin-orbital components of Cu^{2+}. There is the observation of other satellite peaks on the high binding energy side of the main peak of Cu^{2+}, which are absent in the spectra for Cu_2O [24]. The appearance of Cu^{2+} may be due to the slight surface oxidation of Cu_2O in the air [11]. Fig. 2b displays that O 1s has a strong peak at 531.4 eV, indicating the O element exists only in the form of O^{2-}.

The Cu_2O ordered nanoarrays were explored as a non-enzymatic biosensor for glucose detection, and its electrocatalytic activity was assessed by cyclic voltammetry (CV) measurement. The flow chart of constructing glucose sensor based on Cu_2O ordered nanoarrays is shown in Fig. 3a. After the sample (about 5 mm long) was dried naturally, the top of the sample was covered with a narrow strip mask 1 with a width of 1 mm. And another 3 mm wide strip mask 2 was fixed to the top of the mask 1 subsequently. Then an Au film was deposited on the whole upper surface of the substrate by vacuum ion sputtering. After that, mask 2 was removed leading to the exposure of mask 1 and part of the sample. The vaseline was applied as an insulating layer to cover the exposed sample and avoid direct contact of this side Au film with the test solution. At last, we get a non-enzymatic glucose sensor after removing the mask 1. The glucose solution was dripped onto the sample and directly contacted with the corresponding side of the gold film.

The kinetically controlled reactions of Cu_2O ordered nanoarrays on glucose oxidation were verified and analyzed by CV measurements. 0.1 M KOH was selected as the electrolyte for all the chronoamperometry experiments. In this typical experiment, 20 μL 0.2 M KOH aqueous solution and 20 μL glucose solution were dripped onto the sensor simultaneously. The CV curves of 5, 8 and 10 mM glucose solutions at a scan rate of 0.01 V/s were recorded as shown in Fig. 3b. All the CV curves of 5, 8 and 10 mM glucose solutions exhibit two significant redox peaks. They have the same redox peak at ~0.85 V even in case of different glucose concentrations, indicating that the peak is independent of glucose. According to the previous reports, this peak is attributed to the oxidation of Cu^{1+} to Cu^{2+} and Cu^{3+} [11, 25]. The second peak due to the oxidation of glucose because of the shift from ~1.38 V for 5 mM glucose solution to 1.42 V for 8 mM and 1.48 V for 10 mM glucose solution. The current signal is significantly higher than the peak current of Cu^{1+} oxidation, and the oxidation voltage value increases with the increasing concentration, while the oxidation peak becomes more obvious at higher concentration conditions. The optimized potential should not only provide sufficient current response but also tremendously avoids the oxidation of any other interference molecules or substances [26]. Hence, the bias voltage was set to 1.6 V in our chronoamperometry test to satisfy the redox potential of the glucose with higher concentration.

In the typical test process, 20 μL 0.2 M KOH solution was first dripped onto the sensor with a bias voltage of 1.6 V, and then 20 μL glucose solution was dripped into the KOH solution. As we can see in Fig. 4a, the plot of I-t image of detection of 10 mM glucose solution based on Cu_2O ordered nanoarrays, the current increased immediately accompanied by an obvious fluctuation when the 20 mM glucose solution was dripped into the KOH solution. The current reached a maximum value in 12 s and then decreased due to the reduction of glucose concentration. As shown in Fig. 4a, the R-value calculated according to the formula (R = (Imax-IKOH)/IKOH) is used as the response of glucose catalyzed by electrooxidation to express the concentration of glucose. The chronoamperometric current response of glucose with concentrations ranging from 0.05 to 20 mM was recorded to evaluate the linear relationship of response and glucose concentration. As Fig. 4b shown, the result proves the linear relationship of response and glucose concentration in 0.05-15 mM. The fitting equation is R = 0.17204c + 0.05657 (R^2 = 0.99962), which can cover a broad dynamic range required for human glucose detection. In addition, the detection limit of Cu_2O ordered nanoarrays is 0.05 mM, far lower than the normal blood glucose level, which can be applied to the measurement standard of glucopenia.

There are lots of co-existing interfering species with glucose in the human blood serum, such as AA, UA, NaCl, L-Cys, lactose, and soluble starch. Although at low concentrations relative to glucose, they are known to sometimes have high electron transfer rates [27]. For example, AA and UA could be easily oxidized at a relatively positive potential and accordingly obtained overlapped responses

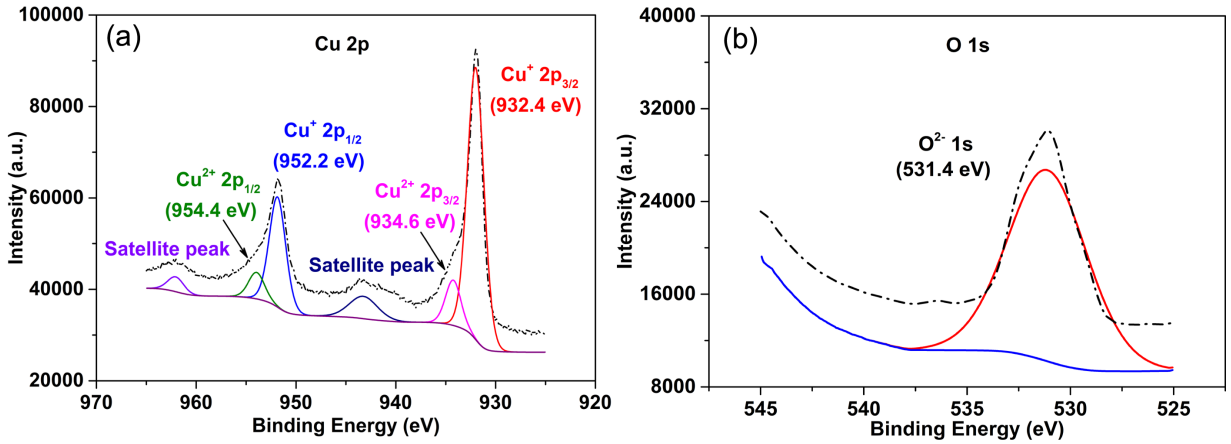

Fig. 2. XPS spectra of the (a) Cu 2p and (b) O 1s core level of Cu_2O ordered nanoarrays.

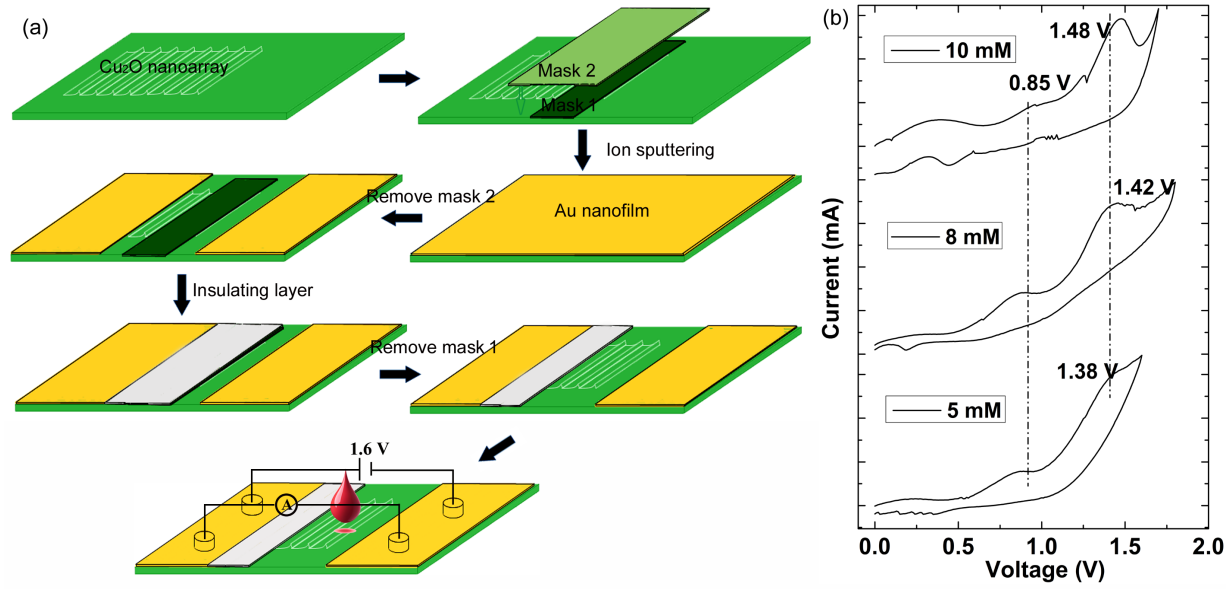

Fig. 3. (a) Schematic illustration of construction and application of non-enzymatic glucose sensor. (b) The CVs of 5, 8 and 10 mM glucose solutions based on this sensor at a scan rate of 0.01 V/s.

to interfere with the detection of glucose [9]. Amperometric response of upon interfering species were carried out by adding 40 mM NaCl, 40 mM UA, 40 mM L-Cys and 40 mM AA to 0.2 M KOH solution separately at 1.6 V. As Fig. 4c shown, for all the interfering species, the sensors showed no any significant responses, indicating that the Cu_2O ordered nanoarrays had good specificity of glucose detection even in the presence of interfering species. Furthermore, to determine the storage stability of the sensors, the prepared Cu_2O ordered nanoarrays were stored in air for certain lengths of time, and at the end of these times their detection performances were tested. As can be seen from Fig. 4d, the response maintained the initial strength even after 30 days of storage. Meanwhile, the effect of temperature on response value was also investigated and shown in Fig. 4e and f. It can be seen that the response value of the sensor to 10 mM glucose in the temperature ranges of 0-60 °C changed slightly in the extent of -20%-20%, which indicates the relative outstanding resist ability to temperature interference. The accuracy of the sensor is in line with the Chinese national standard (GB/T19634-2005, $\leq \pm$ 20% when the concentration of glucose > 4.2 mM).

To make a contrast, the performances of other non-enzymatic glucose sensors that based on the Cu_2O micro or nanostructures are summarized in the Table 1. Compared with the previous reports, the Cu_2O ordered nanoarrays also demonstrates a wider detection range with a higher detection accuracy, which would be much more suitable

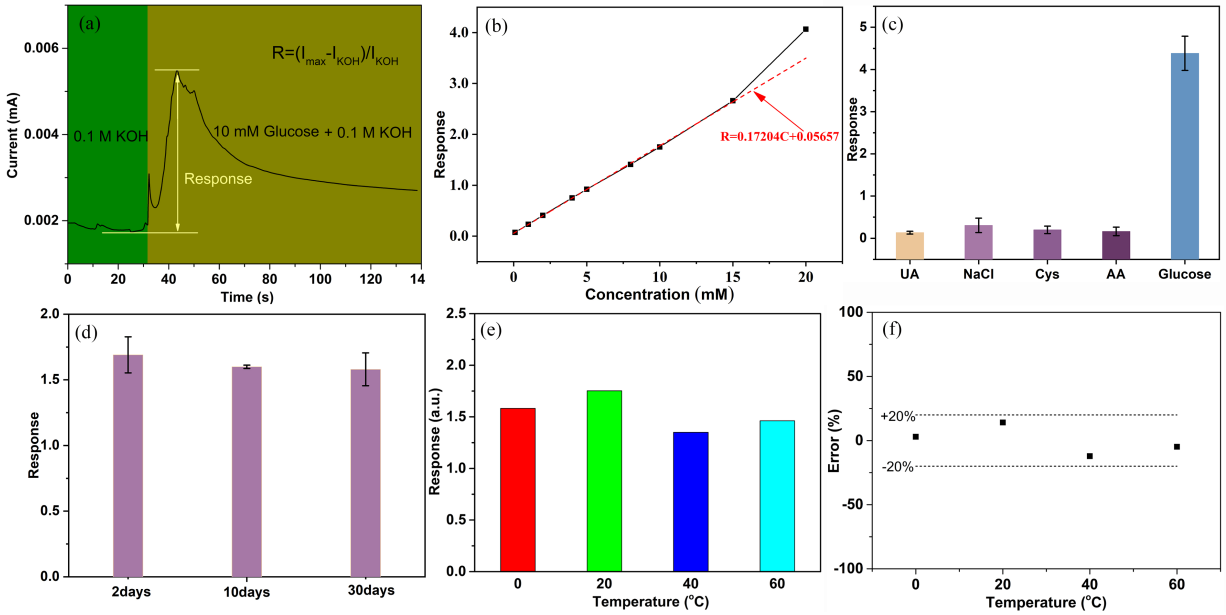

Fig. 4. (a) The chronoamperometric current response of the sensor to 10 mM glucose. (b) Calibration curve versus glucose concentration. (c) Interference test of the sensor with the same bias voltage of 1.6 V. (d) Stability of the sensor stored at ambient conditions over a month. (e) The temperature dependence of the sensor to 10 mM glucose. (f) Response error calculated from the data in (e).

Table 1. Comparison of the fabricated Cu_2O non-enzymatic glucose sensor performance in our work with other reported works.

Materials	Linear range (mM)	Detection Limit (mM)	Reference
Octahedral Cu_2O particles	1-14	0.51	[20]
Hexapod Cu_2O particles	1-14	0.6	
Cu_2O micronanoparticles	0.5-10	0.1	[28]
Cu_2O ordered nanoarrays	0.05-15	0.05	Our work

for the detection of human blood glucose.

The large specific surface area and ordered nanostructure of Cu_2O ordered nanoarrays are the fundamental reasons for the excellent performance. Furthermore, the nanoarrays in situ assembled by 2D electrochemistry method have a relatively firm adhesion on the substrate, which is a benefit for the preparation of the detection sensor. Above all, the sensor based on Cu_2O ordered nanoarrays exhibits high sensitivity, large linear range, specificity and stability for glucose detection, it proves our work has reference value in the research and commercial application of non-enzyme glucose detection.

4. Conclusions

In summary, Cu_2O ordered nanoarrays were prepared by 2D electrochemistry method for non-enzymatic glucose detection. The nanoarrays are bamboo-like nanomicrostructure composed of periodically accumulated nanoparticles with uniform cycle length and long-range order. We fabricated a sensor for fast and efficient glucose monitoring with a 0.05 mM detection limit and a linear relationship in a range of 0.05-15 mM, which covers a broad dynamic range required for human glucose detection. Cu_2O ordered nanoarrays show a wide linear range, high sensitivity, good stability and fast response for glucose detection, these excellent detection performances meet the urgent need of glucose sensor. This study reveals a promising strategy for the design of nano-microstructure materials for facile non-enzymatic glucose detection.

5. Acknowledgements

This research was funded by the National Natural Science Foundation of China (NO. 11704168, 11404158), Natural Science Foundation of Shandong Province (NO.

ZR2018JL003).

References

[1] Joseph Wang. Electrochemical glucose biosensors, 2008.

[2] Na Lu, Changlu Shao, Xinghua Li, Teng Shen, Mingyi Zhang, Fujun Miao, Peng Zhang, Xin Zhang, Kexin Wang, Yang Zhang, and Yichun Liu. CuO/Cu2O nanofibers as electrode materials for non-enzymatic glucose sensors with improved sensitivity. *RSC Advances*, 4(59):31056–31061, 2014.

[3] Yuchan Zhang, Yixin Liu, Liang Su, Zhonghua Zhang, Danqun Huo, Changjun Hou, and Yu Lei. CuO nanowires based sensitive and selective non-enzymatic glucose detection. *Sensors and Actuators, B: Chemical*, 191:86–93, 2014.

[4] M. U. Anu Prathap, Balwinder Kaur, and Rajendra Srivastava. Hydrothermal synthesis of CuO micro-/nanostructures and their applications in the oxidative degradation of methylene blue and non-enzymatic sensing of glucose/H 2O 2. *Journal of Colloid and Interface Science*, 370(1):144–154, 2012.

[5] Ding Jiang, Qian Liu, Kun Wang, Jing Qian, Xiaoya Dong, Zhenting Yang, Xiaojiao Du, and Baijing Qiu. Enhanced non-enzymatic glucose sensing based on copper nanoparticles decorated nitrogen-doped graphene. *Biosensors and Bioelectronics*, 54:273–278, 2014.

[6] Jian Lv, Chuncai Kong, Yun Xu, Zhimao Yang, Xiaojing Zhang, Shengchun Yang, Ge Meng, Jinglei Bi, Jianhui Li, and Sen Yang. Facile synthesis of novel CuO/Cu2O nanosheets on copper foil for high sensitive nonenzymatic glucose biosensor. *Sensors and Actuators, B: Chemical*, 248:630–638, 2017.

[7] Marc Delvaux and Sophie Demoustier-Champagne. Immobilisation of glucose oxidase within metallic nanotubes arrays for application to enzyme biosensors. *Biosensors and Bioelectronics*, 18(7):943–951, 2003.

[8] F. Valentini, L. Galache Fernàndez, E. Tamburri, and G. Palleschi. Single Walled Carbon Nanotubes/polypyrrole-GOx composite films to modify gold microelectrodes for glucose biosensors: Study of the extended linearity. *Biosensors and Bioelectronics*, 43(1):75–78, 2013.

[9] Susu Zhang, Guangli Zhang, Ping He, Wen Lei, Faqin Dong, Dingming Yang, and Zhirong Suo. Novel one-pot hydrothermal fabrication of cuprous oxide-attapulgite/graphene for non-enzyme glucose sensing. *Analytical Methods*, 7(6):2747–2753, 2015.

[10] Huang Chen, Guokang Fan, Jie Zhao, Meijia Qiu, Peng Sun, Yifeng Fu, Dongxue Han, and Guofeng Cui. A portable micro glucose sensor based on copper-based nanocomposite structure. *New Journal of Chemistry*, 43(20):7806–7813, 2019.

[11] Yuqin Wang, Zhenyuan Ji, Xiaoping Shen, Guoxing Zhu, Jiheng Wang, and Xiaoyang Yue. Facile growth of Cu2O hollow cubes on reduced graphene oxide with remarkable electrocatalytic performance for non-enzymatic glucose detection. *New Journal of Chemistry*, 41(17):9223–9229, 2017.

[12] Yuxin Zhao, Yanpeng Li, Zhaoyang He, and Zifeng Yan. Facile preparation of Cu-Cu2O nanoporous nanoparticles as a potential catalyst for non-enzymatic glucose sensing. *RSC Advances*, 3(7):2178–2181, 2013.

[13] Sakthi Velmurgan, Rajkumar Devasenathipathy, Shen Ming Chen, and Sea Fue Wang. A Facile Chemical Synthesis of Cu2O Nanocubes Covered with Co3O4 Nanohexagons for the Sensitive Detection of Glucose. *Electroanalysis*, 28(7):1547–1552, 2016.

[14] K. M. El Khatib and R. M. Abdel Hameed. Development of Cu2O/Carbon Vulcan XC-72 as non-enzymatic sensor for glucose determination. *Biosensors and Bioelectronics*, 26(8):3542–3548, 2011.

[15] Zhenjing Zhuang, Xiaodong Su, Hongyan Yuan, Qun Sun, Dan Xiao, and Martin M.F. Choi. An improved sensitivity non-enzymatic glucose sensor based on a CuO nanowire modified Cu electrode. *Analyst*, 133(1):126–132, 2008.

[16] Xinyi Zhang, Dan Li, Laure Bourgeois, Huanting Wang, and Paul A Webley. Direct electrodeposition of porous gold nanowire arrays for biosensing applications. *ChemPhysChem*, 10(2):436–441, 2009.

[17] Qian Yang, Mei Long, Lin Tan, Yi Zhang, Jin Ouyang, Ping Liu, and Aidong Tang. Helical TiO2 Nanotube Arrays Modified by Cu-Cu2O with Ultrahigh Sensitivity for the Nonenzymatic Electro-oxidation of Glucose. *ACS Applied Materials and Interfaces*, 7(23):12719–12730, 2015.

[18] Mei Long, Lin Tan, Hongtao Liu, Zhen He, and Aidong Tang. Novel helical TiO2 nanotube arrays modified by Cu2O for enzyme-free glucose oxidation. *Biosensors and Bioelectronics*, 59:243–250, 2014.

[19] Xuemei Zhou, Huagui Nie, Zhen Yao, Youqing Dong, Zhi Yang, and Shaoming Huang. Facile synthesis of nanospindle-like Cu 2O/straight multi-walled carbon nanotube hybrid nanostructures and their application in enzyme-free glucose sensing. *Sensors and Actuators, B: Chemical*, 168:1–7, 2012.

[20] Yu Ho Won and Lia A. Stanciu. Cu2O and Au/Cu2O particles: Surface properties and applications in glucose sensing. *Sensors (Switzerland)*, 12(10):13019–13033, 2012.

[21] Guangliang Cui, Liang Gao, Binbin Yao, Shuang-

ming Wang, Pinhua Zhang, and Mingzhe Zhang. Electrochemistry of CuO/In2O3 p-n heterojunction nano/microstructure array with sensitivity to H2 at and below room-temperature. *Electrochemistry Communications*, 30(3):42–45, 2013.

[22] Guangliang Cui, Mingzhe Zhang, and Guangtian Zou. Resonant tunneling modulation in quasi-2D Cu2O/SnO2 p-n horizontal-multi-layer heterostructure for room temperature H2S sensor application. *Scientific Reports*, 3:1–8, 2013.

[23] Guangliang Cui, Pinhua Zhang, Li Chen, Xiaoli Wang, Jianfu Li, Changmin Shi, and Dongchao Wang. Highly sensitive H2S sensors based on Cu2O/Co3O4 nano/microstructure heteroarrays at and below room temperature. *Scientific Reports*, 7(November 2016):1–10, 2017.

[24] Jorge Ramírez-Ortiz, Tetsuya Ogura, Jorge Medina-Valtierra, Sofía E. Acosta-Ortiz, Pedro Bosch, J. Antonio De Los Reyes, and Victor H. Lara. A catalytic application of Cu 2 O and CuO films deposited over fiberglass. *Applied Surface Science*, 174(3-4):177–184, 2001.

[25] Jingwei Huang, Zhengping Dong, Yingdong Li, Jing Li, Jia Wang, Haidong Yang, Shuwen Li, Shujing Guo, Jun Jin, and Rong Li. High performance non-enzymatic glucose biosensor based on copper nanowires-carbon nanotubes hybrid for intracellular glucose study. *Sensors and Actuators, B: Chemical*, 182:618–624, 2013.

[26] Xiangjian Liu, Ling Long, Wenxiu Yang, Lulu Chen, and Jianbo Jia. Facilely electrodeposited coral-like copper micro-/nano-structure arrays with excellent performance in glucose sensing. *Sensors and Actuators, B: Chemical*, 266:853–860, 2018.

[27] Ying Mu, Dongling Jia, Yayun He, Yuqing Miao, and Hai Long Wu. Nano nickel oxide modified non-enzymatic glucose sensors with enhanced sensitivity through an electrochemical process strategy at high potential. *Biosensors and Bioelectronics*, 26(6):2948–2952, 2011.

[28] Petra Majzlikova, Jan Prasek, Jana Chomoucka, Jana Drbohlavova, Jan Pekarek, Radim Hrdy, Jaromir Hubalek, and Libuse Trnkova. Cu2O based electrochemical sensor for direct glucose detection. In *Proceedings of IEEE Sensors*, 2013.