

Blood Cholesterol Monitoring With Smartphone as Miniaturized Electrochemical Analyzer for Cardiovascular Disease Prevention

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Abstract—Currently, cardiovascular diseases become one of the major threat to human's life. The early prevention of cardiovascular diseases plays a critical role in the healthcare engineering. Point of care monitoring the blood lipid level is capable of making the positive contribution to the prevention of cardiovascular disease. Ubiquitous smartphones paved the way as the flexible and widespread platform for the interaction of various health information. In this manuscript, we report the world's first medical smartphone as an electrochemical analyzer for blood lipid monitoring. Integrating an electrochemical analyzer into a smartphone allows us to measure the current generated by the enzymatic reaction with the total cholesterol test strip. The disposable test strip is used to convert the biochemical signal to electrical signal through the electrochemical reaction. The proposed medical smartphone can provide accurate evaluation of patient's blood lipid level as compared to the clinical biochemical analyzer. The proposed medical smartphone system is a promising platform as a point-of-care device for blood total cholesterol (TC) monitoring, which can be applied for long-term prevention of cardiovascular disease due to its portability, reliability, lower cost, convenience, and internet-based medical data interaction.

Index Terms—Cardiovascular disease prevention, electrochemical biosensor, medical smartphone, total cholesterol monitoring.

I. INTRODUCTION

BASED on the statistical analysis by the World Health Organization (WHO), cardiovascular diseases (CVDs) have become one of the major threats to cause human's direct or indirect death in the world. In 2015, the statistical result indicated that 31% of the reported global deaths are resulted from the CVDs, which reveals a terrible truth that about 17.7 million people's lives were taken by CVDs [1]. According to a related investigation, it is estimated that 7.4 million of the patients' deaths are caused by coronary heart disease. More than 75% of CVD deaths happened in medical resource-poor countries [1]. Dyslipidemia is one of the most important factors to cause atherosclerosis (also known as arteriosclerotic vascular disease or ASVD). ASVD includes ischemic heart disease (IHD), stroke

and peripheral artery disease (PAD) [2], [3]. During the last 30 years or so, the popularity of dyslipidemia among Chinese citizens is becoming worse because of the changes in people's diet, lifestyle, and environmental pollution. The statistical result of NHFPC (national health and family planning commission of the people's republic of China) in 2004 shows that the dyslipidemia ratio is as high as 18.6% among all Chinese. This means that the number of people suffering from dyslipidemia is as many as 230 million. Dyslipidemia has become a major hidden danger to Chinese. To further improve the prevention and treatment of dyslipidemia, experts on Chinese Society of Cardiovascular Diseases of Chinese Medical Association cooperate with those of Chinese Medical Association Diabetes Society etc. and promulgated the "Guidelines for prevention and treatment of dyslipidemia in Chinese adults" in 2007. The guidelines indicated that the developed products should have not only the accuracy to meet the requirements of *in vitro* diagnosis but also simple, rapid and safe in order to improve quality of dyslipidemia self-management. The products also should be applied for non-professionals' self-monitoring of blood total cholesterol (TC) level and estimating the effect of lipid-lowering drugs [4]–[6].

The mIoTs is believed to be a promising solution for flexible and point of care interaction with human beings, family doctors, hospital, medical device etc [7]–[10]. The mIoTs is aiming at optimizing the healthcare management. In the last twenty years, the communication techniques have experienced a prosperous development which paved the way for the emerging IoTs-based healthcare and chronic disease prevention engineering [11]–[19]. With the popularization of 4G smartphone and pad, professional medical resource such as the remote family doctor service can be precisely and rapidly delivered to the healthcare consumers with rigid demand. The flexible medical devices enable the medical data accessing to the mobile internet to upload the health consumer's biomedical information into her personalized health center and obtain the timely professional suggestion from her remote doctor. It provides an efficient solution of long-term chronic disease prevention and management. With the emerging popularity of the next-generation communication technique, 5G technology is capable of allowing the patients to transmit and upload more flexible biomedical information such as the digital NMR and CT image information through 5G platform. Smartphone as the ubiquitous device is the most suitable medical analyzer since it has the IoT-scale users.

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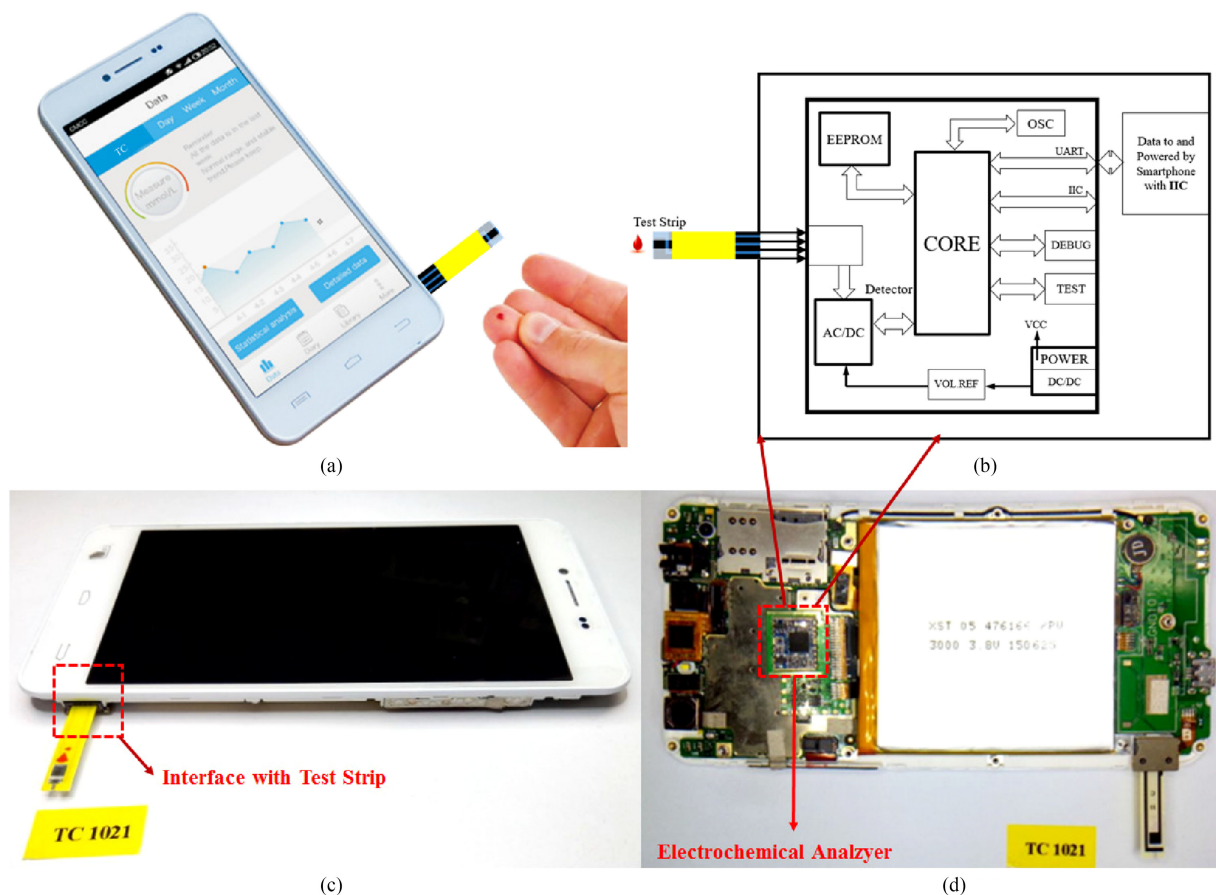
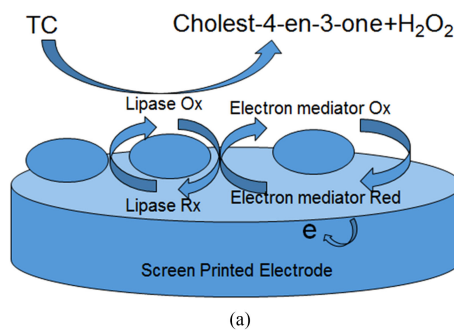


Fig. 1. (a) the schematic view of the proposed medical smartphone in which a miniaturized electrochemical analyzer has been integrated (b) the working diagram of the integrated electrochemical module (c) the side view of the presented medical smartphone: the specific slot working as the interface for connecting TC test strip (d) the practical photograph from the top view of the main printed circuit board layout of the medical smartphone with the insertion of TC strip. The electrochemical analyzer is labelled with red dash line.

The biomedical microdevices have been reported to emerge as a powerful analytical platform for biological sample characterization [20]–[25]. Smartphone-based microdevices have been utilized in the biomedical field with the application extending from the biochemical diagnosis, immunoassay to molecular diagnosis. For examples, a smartphone with medical accessory dongle for blood ketone evaluation was demonstrated [26]. Smartphone performing as a compact electrochemical reader has been presented for the point of care estimation of UA (Uric Acid) level [27], [28] and other biomedical application [29]–[33]. The working principle of the abovementioned research works is based on the electrochemical method to measure the targets molecules level, which indicated that the miniaturized system is compact, cost-effective, rapid response, accurate.

In this paper, we first implement the integration of the compact electrochemical modules in the smartphone for rapid, quantitative evaluation of blood TC with one 1 μ L finger pricked whole blood drop. The electrochemical module is composed of an embedded circuit for signal processing and communication with smartphone. It bonds together with the main printed circuit board in smartphone, and the disposable test strip for blood TC sensing. At the first step, the operator inserts the test strip into the smartphone through the designed slot to make it



1. Cholesterol ester + Cholesterol esterase + $H_2O \longrightarrow$ Cholesterol + Fatty Acid
2. Cholesterol + ChOx + $H_2O + O_2 \longrightarrow$ 4-Cholesteryl-3-Ketone + H_2O_2
3. $H_2O_2 \rightarrow 2H^+ + O_2$ and $2e^-$

Fig. 2. (a) the schematic view of the electrochemical reaction of TC sensor (b) the working principle of the proposed TC sensor by cascading enzyme reaction.

contact with the electronic interface in electrochemical modules, then blood drop was loaded into the test strip by capillary flow. After electrochemical reaction under the specialized enzyme, the concentration of TC was sensed by the printed carbon electrode. The generated electrochemical current was acquired

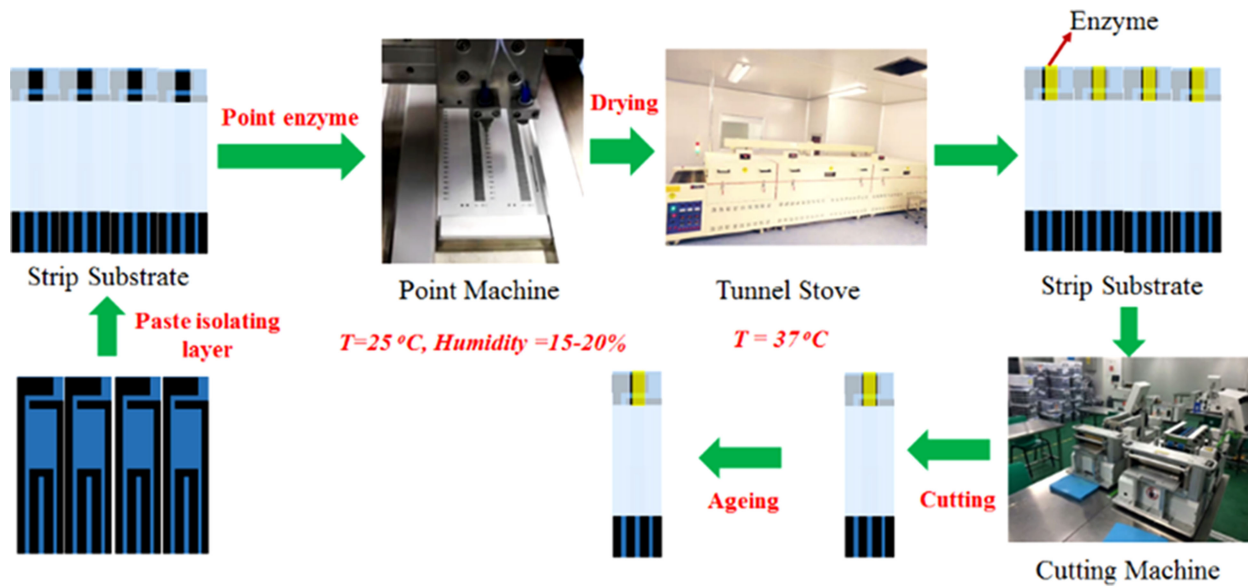


Fig. 3. the practical fabrication process of the proposed total cholesterol test strip.

by the ADC (Analog to Digital converter) and analyzed by the MCU (micro-controller unit) of the module. The pre-buried algorithm can map the current into the measured TC concentration by the calibration line. The module was powered with 3 V power source obtained from the smartphone. The data was communicated through the IIC (one typical communication standard). The measured data was saved and uploaded to personal health management center through the internet. Since it is equipped with a medical level reader, this smartphone provides a very powerful and portable platform as a point-of-care diagnostic platform, especially in smart management of personal health. Fig. 1 shows the schematic view of the medical smartphone integrated with the electrochemical reader. The overall cost of the proposed medical smartphone is less than 50 USD.

II. MATERIALS AND METHODS

In the process of the enzyme catalysing reaction, TC oxidase catalyze the TC into Cholest-4-en-3-one with concomitant reduction of electron mediator Fe(III) to Fe(II) [9]; The Fe(II)(CN)_6^{4-} ions generated were detected by the amperometric method. Fig. 2(a) and (b) illustrate the working principle of enzymatic catalysis of TC. Ferricyanide plays a role as the electron mediator to catalyze the reduction of Fe(III) to Fe(II); the Fe(II)(CN)_6^{4-} ions were evaluated by detecting the current change between WE and CE through integrated electrometer in the medical smartphone. The buried electrometer is capable of translating the current signal into the concentration of TC, which can be easily recorded by the smartphone and upload through the 4G mobile internet.

The TC test strips were fabricated with polyethylene terephthalate (PET) substrate, screen printed electrodes, isolating layer, covering layer. The complete fabrication process of the proposed TC strip was illustrated in Fig. 3. Initially, the PET is chosen as the test strip substrate. Carbon ink was screen printed above the PET and works as the working and counter

electrode for electrically sensing the transferred electrons generated within the electrochemical reaction under the catalysis of TC oxidase. The isolating layer was pasted on the PET with electrodes to form the reaction channel full of blood sample. As the following step, the paramount TC oxidase solution was mixed with the electrical mediator to be loaded within the narrow reaction channel by the microliter point machine. Then the strip with wet enzyme solution experienced the stove tunnel with temperature at 38 °C for 35 min in order to immobilize the enzyme solution on the electrodes within the reaction channel. Once the enzyme was dried, the covering hydrophilic slip was laminated on the top of the semi-finished test strip to form the final product. The overall cost of the TC strip was estimated less than 0.3 USD, while the existing optic-based TC strips is more than 0.6 USD.

The proposed medical smartphone with the function as the amperometric analyzer was used to resolve the electrochemical current induced by the enzymatic reaction. The resolved current was mapped into the blood TC concentration by the calibration comparison with clinical biochemical analyzer. The measured biochemical parameter was stored into the smartphone and be the updated with the users' personalized health data center.

III. RESULT AND DISCUSSION

The presented medical smartphone was utilized as a portable and compact electrochemical analyser to provide the potential with applying the amplitude at 350 mV on the proposed TC test strip. Chronoamperometric characterization has emerged as a cost-effective, rapid response, accurate and sensitive electrochemical method in point of care test. By resolving the electrochemical current induced by the enzymatic reaction on the test strip, the target molecules can be precisely estimated. The pricked finger whole blood from the healthy target is utilized to test the stability of the proposed system for three times. The typical chronoamperometric curves captured by medical

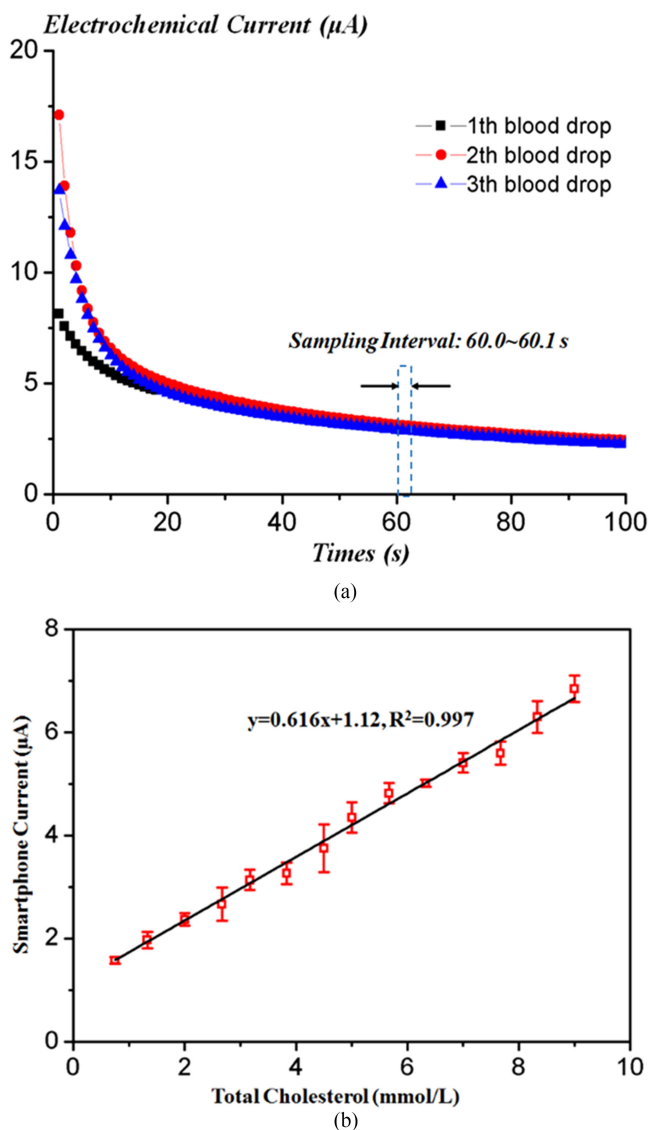


Fig. 4. (a) the three measured chronoamperometric profiles by the proposed TC test strips and medical smartphone by applying the practical finger blood sample from the healthy human with blood TC level at the 3.23 mmol/L; (b) the linearly fitted relationship profile between the measured steady-state electrochemical current and total cholesterol concentration, the error bar indicated the standard error of the mean and was calculated d by analysing 5 test results at each TC concentration.

smartphone as a result of applying the real finger blood sample are obtained in Fig. 4(a). The blood TC were verified by the clinical laboratory biochemical analyser with the TC concentration 3.23 mmol/L. The three tested results of chronoamperometric curves with the response to the real blood sample from the same target health human are very close which demonstrated that the proposed TC test strip was relatively stable and has good reproducibility. The typical $I-t$ profile described an interesting and complete electrochemical sensing process. During the early state, the transferred electrons in unit time increase significantly since the enzymatic paramount catalysis under the externally electrical potential excitation. After a transient time, the electrochemical current undergoes a significant decay due to the fast enzymatic oxidation of the target molecules around the working

TABLE I
THREE DIFFERENT CONCENTRATIONS OF BLOOD SAMPLES
ARE USED FOR THE TEST

TC level	Test strip #1		Test strip #2		Test strip #3	
	Test Result (mmol/L)	CV (%)	Test Result (mmol/L)	CV (%)	Test Result (mmol/L)	CV (%)
Low	2.53 ± 0.10	3.95	2.62 ± 0.11	4.20	2.56 ± 0.09	3.52
Mid	4.38 ± 0.12	2.94	4.51 ± 0.08	1.78	4.41 ± 0.13	2.95
High	7.47 ± 0.17	2.27	7.60 ± 0.19	2.50	7.52 ± 0.18	2.39

Each blood sample three batches of TC dipstick test was repeated 10 times in three multi-function testers respectively. Samples are randomly selected in three blood TC concentrations, namely, low concentration range (< 3 mmol/L), median range (3.0 mmol/L \sim 6.0 mmol/L), and high range (> 6.0 mmol/L).

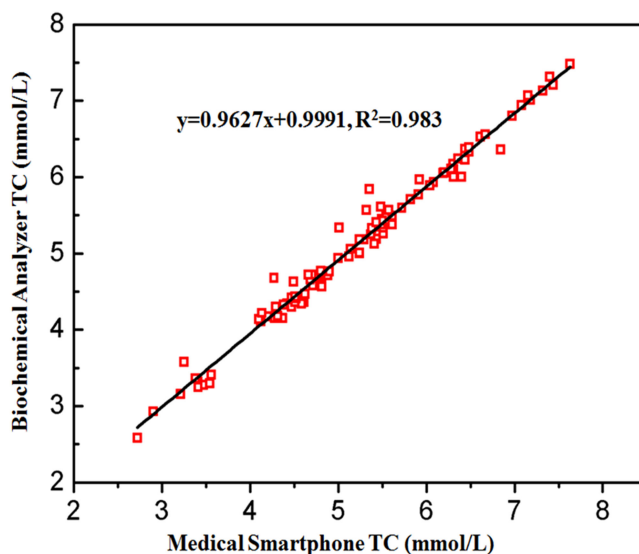


Fig. 5. the measured biochemical parameters of TC comparison between the proposed medical dongle and clinical biochemical analyser.

electrode surface. As the following step, the current decreased very slowly resulting from the principle that the electrochemical reaction was mainly dominated by the mass transfer of TC molecules from the bulky blood solution to the surface of the working electrode. In the choice of the sampling interval, the beginning time is critical important. The slope of the curve become smaller as the function of time. The ideal sampling point with its slope is close to zero, however, the time consuming will be quite longer. The blood clot happened if the sampling beginning time is set with longer time. Therefore, the balanced choice is to choose the sampling point with relative flat tangent line. The sampling interval set between the 50 and 65 s is preferable. In the proposed design, the sampling time begins at 60th s. According to the chronoamperometric curve in Fig. 4(a), the amplitude of the electrochemical current keep a relative steady value around the 60th second. Therefore, the electrochemical current was set to be captured by the ADC in medical smartphone between 60th s to 60.1th s with 20 sampling points. The

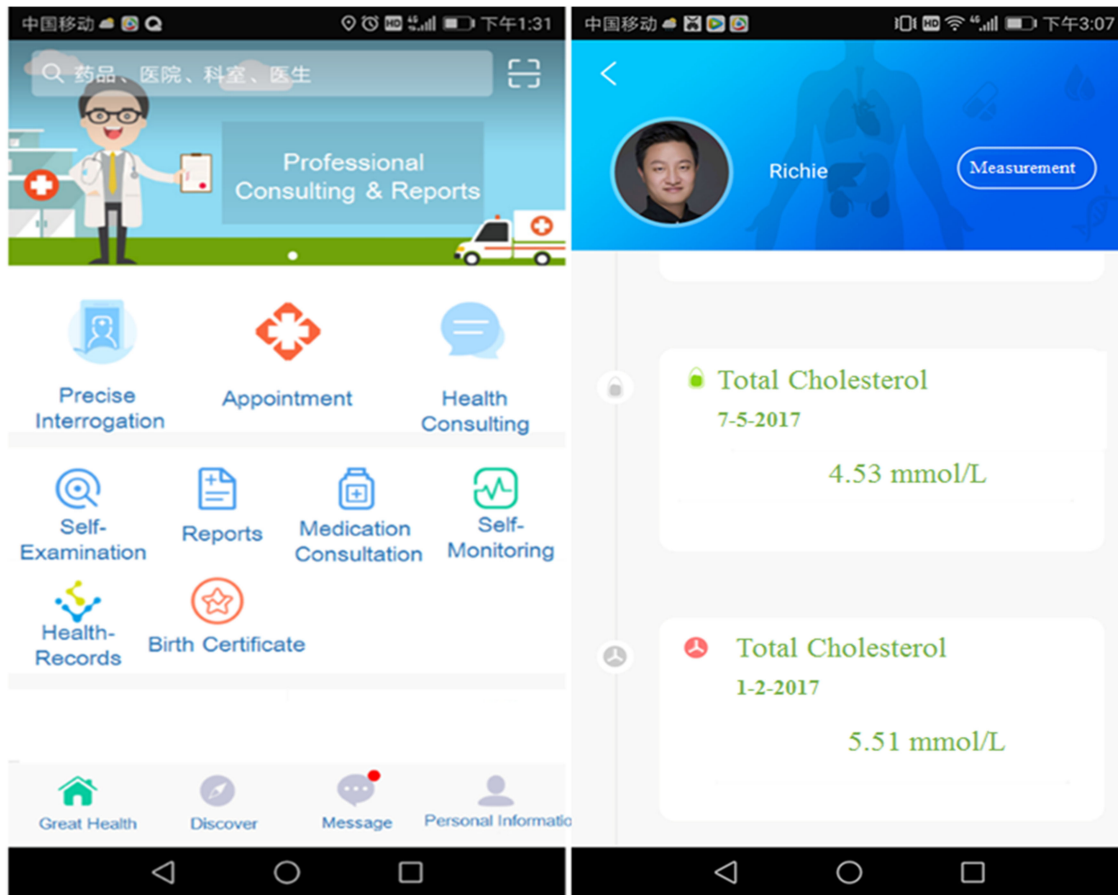


Fig. 6. (a) The front mobile page of the health management app, which includes the online medical interrogation, appointment with doctor, health consulting, self-examination, medical reports, medication consultation, self-blood test, health parameter records etc. (b). the personal health page for health management user, which recorded the medical biophysical or biochemical parameters such as the total cholesterol, which enable the user can continuously monitor the related biomedical parameters.

captured average current was mapped into the biochemical concentration by the calibration comparison with the biochemical analyser. Fig. 4(b) shows the linear relationship between the blood TC concentration and the induced electrochemical current with the linear regression coefficient $R^2 = 0.997$.

To evaluate the coefficient of variation (CV) of the TC test strip, the pricked fingertip whole blood samples with different blood TC level are tested. Three different concentrations (lower, middle and higher) of blood samples are used for the test compared with the biochemical analyser. Blood sample was tested by three groups of TC test strips and each group was repeated 10 times in three smartphone readers, respectively, and then 90 test results were generated. The manufacture differences of TC test strips are statistically evaluated respectively. Samples are randomly selected in three blood TC concentrations, namely, low concentration range (<3.0 mmol/L), medium range (3.0 mmol/L ~ 6.0 mmol/L), and high range (>6 mmol/L). The CV of the proposed TC strip with response to the three groups TC blood was summarized in Table I. The 126 blood sample tests were approved by IRB.

Besides, in order to evaluate the precision of the TC test strip compared to the laboratory biochemical analyser, the scattering points between the TC in human serum from venous blood by

biochemical analyser and the whole blood droplet with finger prick blood drop by the proposed TC test strip incorporating with medical smartphone were plotted in Fig. 5. 126 blood samples taken from 126 targeting patients with different blood TC were performed for the comparative test. The pricked finger blood drop of each target patient was loaded to the TC test strip with the signal reader of the medical smartphone and the human serum obtained from the venous blood from the same target patient were compared to the bulky biochemical analyser. Fig. 5 showed the blood TC by medical smartphone VS the clinical biochemical analyser. The compared results demonstrated that the medical smartphone with the proposed TC test strips was relatively accurate as compared to the clinical laboratory equipment. There still exists obvious difference between the TC test strip and the bulky biochemical analyser. The accuracy of the electrochemical current was affected by the red blood cells, which may retard the UA molecules transferring from the bulky solution to the electrode surface. In the conventional biochemical laboratory, the test blood sample was blood serum, which gets rid of the effect by the blood cells. It's the limitation of the proposed TC test strips as compared to the optical test strip. However, the blood cell effect can be compensated by the impedance measurement of the blood.

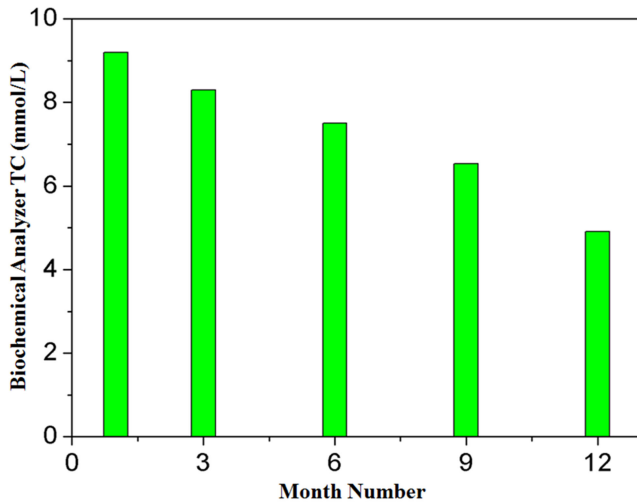


Fig. 7. the measured blood total cholesterol of hyperlipidemia patient for 12 months by using the proposed medical system to manage the chronic disease under the professional suggestion of the remote doctor.

The on-line medical consulting network service was integrated with the smartphone to store the patients' personalized biomedical parameters and provide the medical service such as on-line doctor inquiry. The on-line doctor is capable of directly access the patient's health information through the system. The Great-in-Health APP can be equipped from the apple or android application store. The home page of the Great-in-Health APP is shown in Fig. 6. Fig. 6(a) is the first APP page when accessing the system which integrated many professional medical services. The Fig. 6(b) demonstrated the page for the patients or consumer. It allows the users to track their history test result easily.

A typical example of one Hyperlipidemia patient was given to show the positive contribution of the proposed system in chronic disease management. The patient was asked to register in the Great-in-Health APP system to measure and monitor his TC one time every three months within one year. The professional advice from his on-line doctor was given and the patient followed. It included the continuous sports and suitable diet, such as preferring the fruit and vegetables rather than the greasy meat. Fig. 7 shows the one year TC through the whole year. It demonstrated the professional suggestion take the positive effect on lowering the blood lipid. Therefore, the proposed system was believed to be a promising technology to prevent the cardiovascular diseases.

IV. CONCLUSION

Cardiovascular diseases have become the top threat to humans' lives with the improved life of human beings. The early prevention of cardiovascular diseases plays a critical role in the healthcare and biomedical engineering. The mIoTs is believed to tackle the serious situation for cardiovascular disease prevention. In the presented work, we demonstrated the world's first medical smartphone as an electrochemical analyzer for blood lipid monitoring, which is implemented by integrating an electrochemical analyzer with smartphone. This enables it to work

as an electrochemical reader to resolve the current generated by the enzymatic reaction within the total cholesterol (TC) test strip. The disposable test strip is used to convert the biochemical signal to electrical signal by the enzymatic reaction. Blood TC test strips were used to demonstrate that the proposed medical smartphone is capable of providing clinical level information of patient's blood lipid level by comparing the test results of the medical smartphone with the results from the clinical biochemical analyzer. In the emerging situation of mIoTs, the presented system is capable of providing a very feasible solution for the huge commercial need of cardiovascular disease prevention since it connects the medical sensors, mobile communication, big data information processing and on-line doctor services.

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