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A low cost, simplified and battery-powered mobile electrophoresis system suitable as edutainment tool.

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ABSTRACT: Electrophoresis originated since 1807 and has been a staple tool used by biologist and chemist over the centuries since its inception. From paper electrophoresis system to today's modern automated electrophoresis system, the development of electrophoresis systems have been driven by the advancement of technology and also by the requirement of better and faster resolution of results. This paper reviews the progress of electrophoresis over the decades and into possible future development of electrophoresis.
Keywords: Battery-powered; Edutainment; Electrophoresis; Mobile; Simplified.

INTRODUCTION

Electrophoresis is the process used by researchers to separate a chemical entity based on its charge. An example of the use of electrophoresis is in the separation of DNA, RNA, and protein by taking advantage of their negatively charge nature [1]. Most of the current electrophoresis system uses the electrical power from a wall socket to do this. However, there is very limited development for a battery powered electrophoresis system that is both portable and mobile.

The inability to perform analysis instantly and directly on site has often caused many issues when real time analysis is required for fast decision to be made. One of the main challenges in field studies is the degradation of samples sensitive to the environment conditions which eventually affects the final product or result [2]. This risk may increase due to poor handling of materials when the analysis of the samples is delayed, especially when the samples are collected from remote areas. The sample would have started to deteriorate without any proper storage facilities. Although this problem can be tackled through the use of sophisticated and expensive equipment, such as a mobile refrigeration system, they are often expensive and bulky [3]. Other non-cryogenic methods, which are less effective and less commonly used to preserve samples collected from the field, are the use of silica gel to dry the collected samples for storage [4], or addition of saturated brine containing cetyltrimethylammonium bromide detergent/disinfectant (NaCl-CTAB) for preservation [5]. As analysis cannot be carried out at the site, samples are often collected and accumulated before they are sent to the laboratories. Due to large number of samples, they may be sent to different laboratories which use equipment with varying sensitivity or accuracy [6]. In this case, variation in results may be produced.

In this paper, a novel electrophoresis system has been developed for field usage. The system consists of a miniaturized and battery powered portable electrophoresis system. The system was evaluated for its ability on resolution and efficiency, comparing it to a commercial electrophoresis system.

MATERIALS AND METHODS

Chemicals and reagents

The performance of the developed system and the commercial electrophoresis unit was evaluated by analysing the migration of 1 kb and 100 bp DNA ladder (Fermentas, Canada) on 1% agarose gel (Vivantis, USA) in TBE buffer [7]. The agarose gels were stained with RedSafe according to the protocol recommended by the manufacturer (INtRON Biotechnology, Korea), before they were visualized by using a Gel Documentation System (Biovision, Model: 1000/26MX X PRESS, San Francisco).

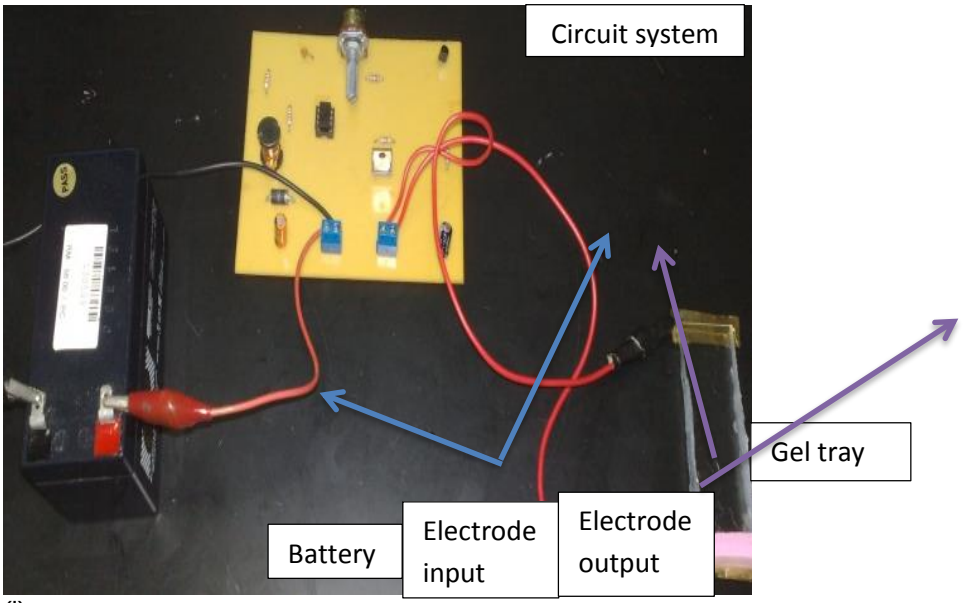
Commercial Electrophoresis System

The commercial electrophoresis unit which was used in this study for comparison with the performance of the prototypes was from Cleaver Scientific, UK (Model: MSMINI, Unit dimension: 21 X 9 X 9 cm)[8].This unit was connected to a power pack (Consort, Model: E835, Belgium) and the gels were analysed at 100V. Generally, 50 ml of agarose was required to cast the gel and 200 ml of TBE buffer was used for each electrophoresis [9].

RESULTS AND DISCUSSION

Prototype system architecture

One of critical part for the development of a miniaturized battery powered gel electrophoresis system was the need of a suitable gel tray [10]. Two glass slides with a dimension of 7.5cm x 2.5cm was used to build the gel casting tray for the developed system with the electrodes located at both ends of the glass slide. A space is included between the two glass slides to allow the filling of agarose gel matrix and a gel comb is used to form the wells. 200 µl of TBE buffer is required to fill the wells. The design of the prototype system consist of a single layer direct current-direct current (DC-DC) step up converter circuit board hardware used to up-regulate and maintain the battery power input at 45v for up to an hour to the gel tray (Figure 1). The circuit board works by up-regulating the battery input via the boost converter circuit. The output will then be controlled by the feedback system, in which if the volt increases to beyond the set volume, the switch control circuit will activate to reduce it to maintain the desired output [11]. This also works vice versa in the event the output is below the required setting.



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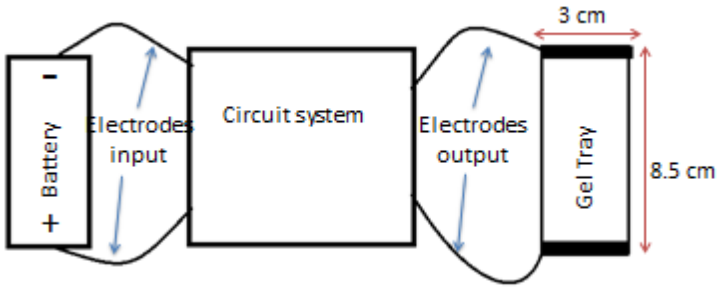


(ii)

Figure 1: (i) Set up and (ii) sketch diagram showing the set-up of Prototype seven (circuit board coupled).

Data analysis

Data evaluation was determined by measuring the distance travelled by the sample, the volt loss per cm travelled by the sample (v/cm), power efficiency of the battery (%), resolution and resolving ability of each prototypes. For this parameter, the 100 bp band in the 1 kbp DNA ladder was use as the standard reference to calculate the distance travelled by the sample, on both the prototypes and the conventional electrophoresis system using the same parameter of 45v for 1 hour in a 1% agarose gel [12]. The 100 bp DNA ladder was only

used as an indicator on the ability of the prototype systems to separate small size DNA molecule. The resolution and resolving ability was measured on the ability of the prototype systems to separate random samples. The power efficiency of the batter was calculated by using the following equation;

Efficiency= $(V^2_f/V^2_o)*100$

Where Vf=Initial voltage of battery and V0= Voltage of battery after use [13].

0.2 µg/µl of DNA or RNA ladder and 0.5 µg/µl of sample, both DNA and RNA samples, from the different sample test was used per well

Figure 2 shows a representative result from using the prototype system using both 1 kbp DNA ladder and 100 bp DNA ladder. Copper plate was used as the electrode in the gel tray of the prototype system, achieving an average distance travelled of 2.05 cm for the 1 Kbp DNA ladder.

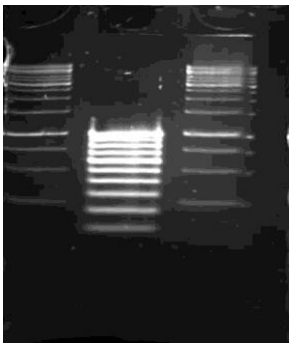


Figure 2: Representative of a result from using the prototype system with a copper plate as electrodes. (Lane 1,3=1 kbp DNA ladder, Lane 2=100 bp DNA ladder)

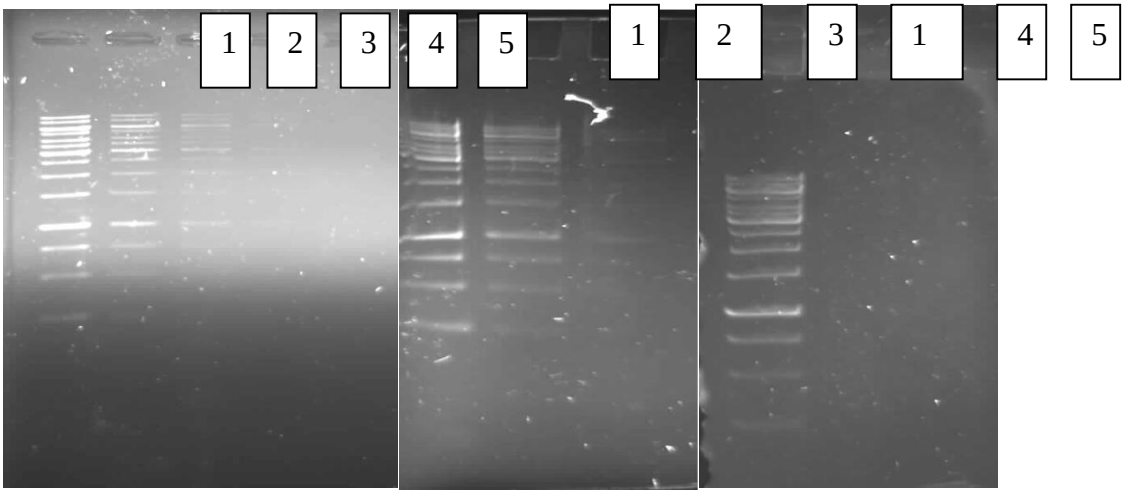
The prototype system has a power efficiency of 99.6% in its battery power consumption per electrophoresis process. The efficiency of the boost converter could be just calculated by using the formula:

Efficiency, $\eta = \frac{P_{out}}{P_{in}}$

Where P_{in} is the amount of battery power input and P_{out} is the amount of battery power output.This shows that although the prototype system conducts an electrophoresis using an independent external power source, it has little waste of its energy expenditure, allowing it to perform multiple electrophoresis process without worry of lack of energy [14].

Sensitivity test

To determine the resolving and sensitivity the prototype system, a simple test was used. Serial dilution of the 1 kbp DNA ladder was done, forming a concentration gradient of 0.1 µg/µl, 0.033 µg/µl, 0.011 µg/µl, $3.67 x10^{-3}$ µg/µl and $1.22x10^{-3}$ µg/µl. Figure 3 showed the comparison of the sensitivity for the prototype system and the conventional electrophoresis system [15]. In the prototype system it was capable of resolving up to 0.011 µg/µl while the conventional electrophoresis system was able to resolve up to $3.67x10^{-3}$ µg/µl.

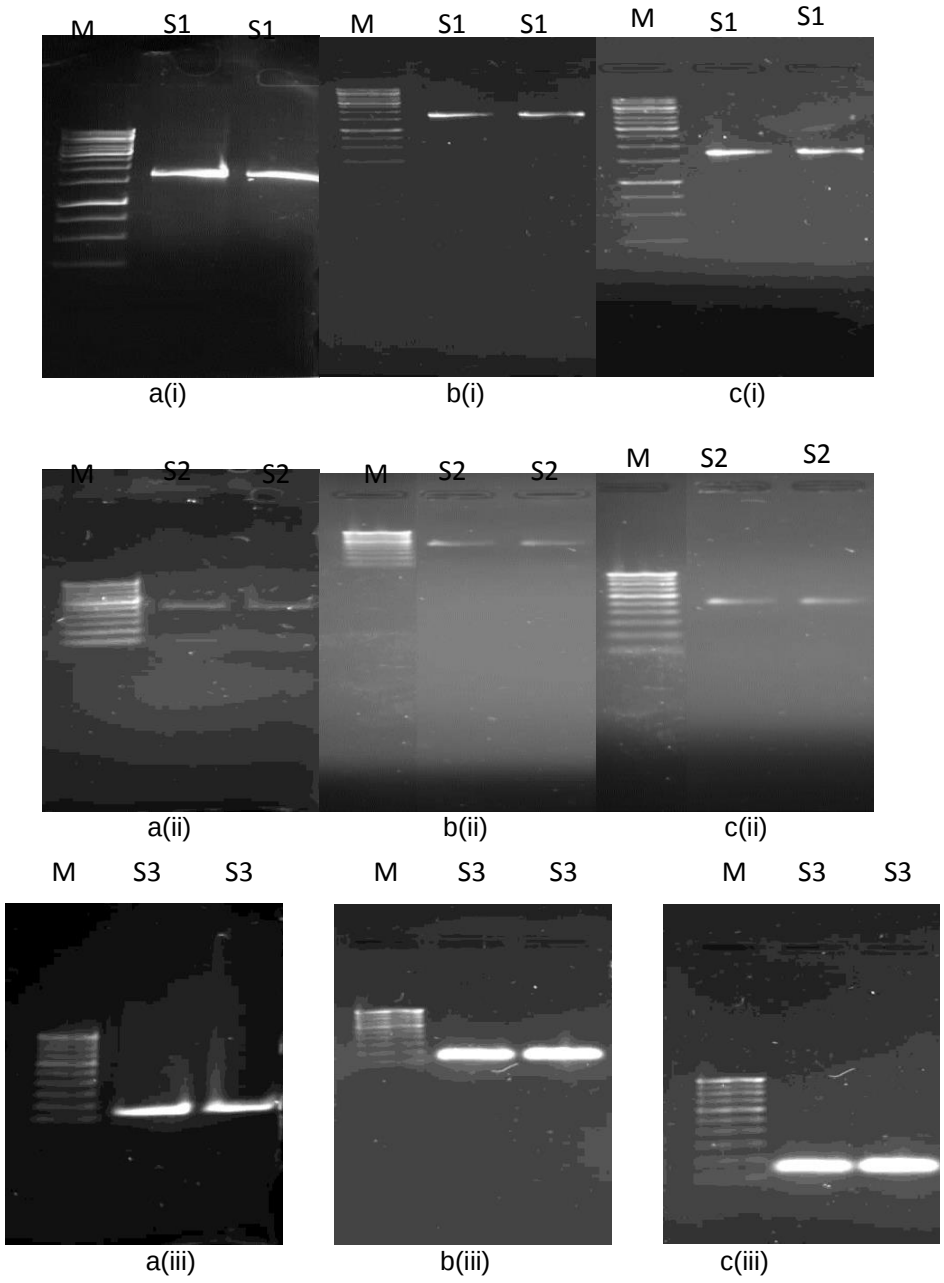


(i) (ii)

Figure 3: Sensitivity and resolving ability test for (i) Conventional electrophoresis system (ii) Prototype seven (Circuit board) (Lane 1=0.1 µg/µl, Lane 2=0.033 µg/µl, Lane 3=0.011µg/µl, Lane $4=3.67x10^{-3}$ µg/µl, Lane $5=1.22x10^{-3}$ µg/µl)

Different Samples test using Prototype seven (circuit board coupled) against Conventional Electrophoresis System

The prototype system was next used to determine the efficiency of the prototype's resolution ability for various samples of different sizes and types. Three PCR products of various size: 1700 bp, 500 bp and 152 bp (PCR product for pcDNA DSN-1, Toxoplasma gondii and peroxisome proliferator-activated receptors (PPARγ) respectively) was used to determine the resolution ability of the prototypes [16]. Genomic DNA of Toxoplasma gondii was used as the next type of DNA sample. RNA of TW01 (Nasopharyngeal Carcinoma (NPC) cell line) was the third type of sample used for this test. The samples were analysed using the same parameter of 45 v for 1 hour in both the prototype system and conventional electrophoresis system, except for the RNA sample which was run at 100 v for 1 hour with the conventional electrophoresis system and 45 v for 1 hour using the prototype system [17, 18]. However, analysis for the conventional system was continued for another hour after observation at the first hour of electrophoresis except for the RNA test due to the instability of the RNA sample. The migration of the bands is as shown in Figure 4 while Table 1 showed the results from the comparison of the distance travelled by the sample between the two systems.



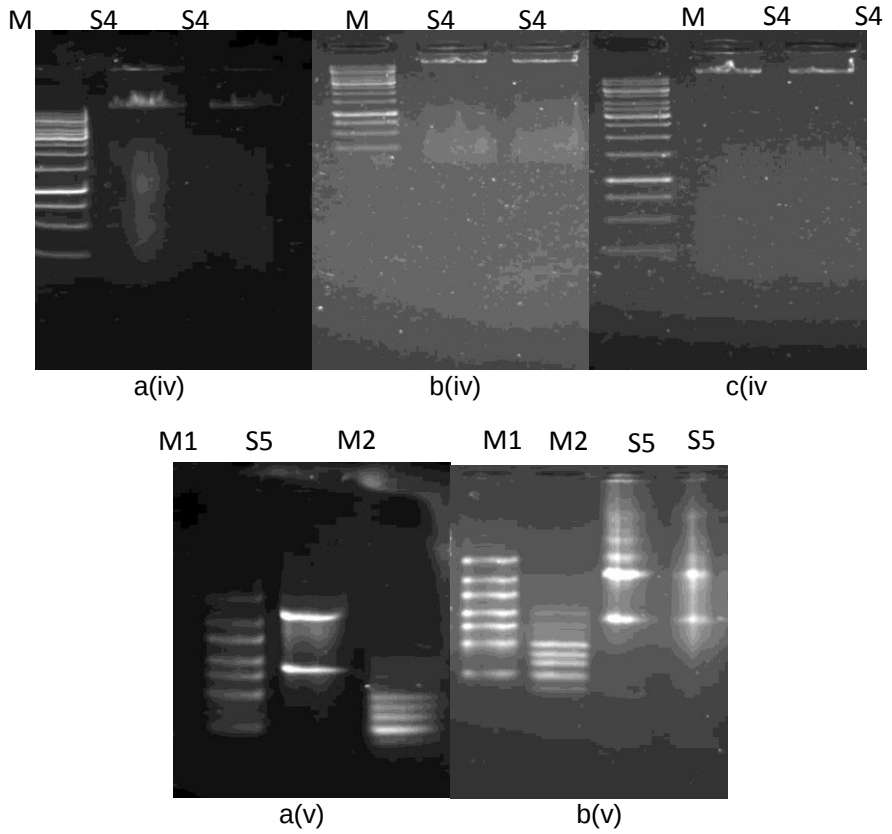


Figure 4: The migration of 5µl of sample DNA bands of different sizes (i) 1700 bp (pcDNA DSN-1)(Lane S1), (ii) 500 bp (Toxoplasma gondii) (Lane S2), (iii) 150 bp (PPARγ) (Lane S3), (iv) 17414 bp (Genomic) DNA of Toxoplasma gondii (Lane S4) and (v) RNA of TW01 NPC cell line (Lane S5) in (a) Prototype seven (circuit board coupled) and (b) conventional unit for 1 hour and in (c) conventional unit for 2 hour (M= DNA marker, M1= High range RNA marker, M2= Low range RNA marker)

Table 1: Distance travelled by the various samples using either the Conventional or Prototype seven (circuit board coupled) systems.

	Sample size (bp)	Distance travelled by sample (cm) Conventional	Prototype	Duration
S1 pcDNA DSN-1	1700	0.8	1.3	1 hour
		1.5	Not available	2 hour
S2 Toxoplasma gondii	500	1.1	2.4	1 hour
		2.2	Not available	2 hour
S3 PPARγ	152	1.8	3.1	1 hour
		3.6	Not available	2 hour
S4 Toxoplasma gondii	17414 (Genomic)	0.2	0.7	1 hour
		0.4	Not available	2 hour
S5 TW01 NPC cell line	1.9 kbp (18s) 5 kbp (28s) (RNA)	2.8	3	1 hour

Improvements

One of the major advantages of the prototype system is the reduction in the use of both gel materials and overall weight of the system. In the prototype system (inclusive of a mobile battery power pack), it only weights a total of 0.2 kg while the conventional system weights 4.5 kg (inclusive of power pack connected to wall power output). For the gel material, only 6 ml of gel matrix was used, compared to the 50-100 ml in the conventional system. This reduction in weight is a major advantage when using the prototype system out in the field.

The portable system is slightly similar in design to currently available portable system, in terms of size and design [19,20]. However, it has an advantage over these systems that the developed prototype system is more robust, and has a longer battery lifespan compared to the current available portable systems [21].

Concluding remarks

One important point for the development of this prototype system was for the on the spot analysis of the sample. The prototype system tested for its ability for reproducing results, and also in its battery power consumption efficiency allowed for it to be used for outdoor field experiments would be beneficial in the long

run. It will also enable a faster and accurate as possible diagnostic or result for on the spot decision making for samples collected.

The performance of the prototype system has also been proven. Although the design of the system is still in its infancy, it was capable of performing as reliable as a conventional electrophoresis system.

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