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EFFECTS OF STATIC MAGNETIC FIELD ON *Escherichia coli* MUTATION

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ABSTRACT

The effects of a dipolar static magnetic field of strength 400, 800, 1200 and 1600 Gauss were prepared locally, on the ultra-structure of *E. coli* type 1 bacterium cells have been studied. Equal volumes of liquid culture media were exposed to the magnetic field for different periods, the three most effective periods, namely: 24 hrs, 48 hrs and 72 hrs were chosen for all our experimental studies. After API kit test of treated *E. coli* culture media as control group. Results indicated that exposure of the microorganisms to the demonstrated magnetic field caused pronounced changes in the Arginine dihydrolase (ADH), Citrate utilization (CIT) and Gelatinase (GEL) were observed on the cell growth. Besides, changes in the morphology of the *E. coli* colonies were observed after exposure period and subculture on MacConkey agar. Furthermore, the bacterial growth subculture tested for morphological and biological activity, the results suggested that a mutation occurred in bacterial cells.

Keywords: Magnetic field; optical density; *E coli*; mutation.

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INTRODUCTION

A magnetic field is the area of influence exerted by a magnetic force. This field is normally focused along two poles. These poles are usually designated as north and south. However these directions are not the only two that a magnetic field can have, most magnetic objects are composed of many small fields called domains. The search for a biological effect due to magnetic fields has a long history dating back a hundred years. The literature on biomagnetic effects on the growth and development of various organisms has been quite extensive showing both positive and negative findings. Among the positive findings attributed to strong magnetic fields are: altered growth rate, enzyme activities, cellular metabolism, DNA synthesis and animal orientation (Gremion *et al*, 2009).

A wide variety of methods have been reported in the literatures which are directed to the use of magnetic energy as a diagnostic technique and also for the treatment of diseases in warm blooded animals including humans. For example, magnetic energy has been utilized quite successfully over the past several years to promote the formation of osteoblasts in conjunction with the healing of bone fractures. In many instances markedly improved results in healing times have been achieved by the application of magnetic energy to the site of bone fractures and other injuries. Magnetic enhancement or retardation of bacterial or cellular growth rates has been reported in the literature. For example, Davis and Rawls presented numerous examples of enhancement to seeds and various types of cellular growth (Albert *et al*, 1996). Magnetic inhibition of bacterial growth in wounds healing reported by (Henry. *et al*, 2008). Investigations sponsored by Bio-Magnetics Systems, Inc. have shown that unidirectional magnetic fields inhibited or increased the growth of cancer cells, depending on the field polarity (Trappier *et al.* 1990).

A mutation is a permanent change in the DNA sequence of a gene. Mutations in a gene's DNA sequence can alter the amino acid sequence of the protein encoded by the gene (Rosche *et al*. 2000).

Mutations can be acquired. This happens when environmental agents like magnetic field forces damage DNA, or when mistakes occur when a cell copies its DNA prior to cell division (Ji *et al*, 2009).

MATERIALS AND METHODS

Bacterial suspension of *E coli* isolated from human urine sample and cultured on MacConkey agar will be inoculated into five groups of tubes containing nutrient broth media and exposed four tubes to one of dipolar magnetic field which were prepared locally with different forces including 400, 800, 1200, 1600 G and measured by Teslometer in Physical Department /College of Science. The tube number five regard as a control and not subjected to magnetic power, all of these tubes incubated separately for 24, 48 and 72 hours at 37ºC. The inoculation of API kit (BioMerieux Company) with bacteria from each groups were performed separately to identify the enteric bacteria type (Appelbaum *et al*, 1980 , A plastic strip holding twenty mini-test tubes inoculated with a saline suspension of a pure culture. This process also rehydrates the desiccated medium in each tube. A few tubes are completely filled (CIT, VP and GEL, and some tubes overlaid with mineral oil such that anaerobic reactions can be carried out (ADH, LDC, ODC, H2S and URE) (Lindquist *et al*., 2001).

After incubation in a humidity chamber for 18-24 hours at 37°C, the color reactions were read. Note especially the color reactions for amino acid decarboxylations (ADH through ODC) and carbohydrate fermentations (GLU through ARA). The amino acids tested are (in order) arginine, lysine and ornithine. Decarboxylation is shown by an alkaline reaction (red color of the particular pH indicator used). The carbohydrates tested are glucose, mannitol, inositol, sorbitol, rhamnose, sucrose, melibiose, amygdalin and arabinose. Fermentation is shown by an acid reaction (yellow color of indicator). Hydrogen sulfide production (H_2S) and gelatin hydrolysis (GEL) result in a black color throughout the tube. A positive reaction for tryptophan deaminase (TDA) gives a deep brown color with the addition of ferric chloride (Lindquist *et al* 2001).The enzymes ADH, CIT and GEL of API test from all groups are affected by magnetic field, one drop of this bacterial suspension and was inoculated added to 5-ml nutrient broth media, incubated for 24hrs without magnetic field at 37ºC, performing API test, subculture on MacConkey agar and measured the OD again to determine the mutation or inhabitation of bacterial sample.

RESULTS AND DISCUSSION

Changes in the structure and characteristic behavior of bacteria resulting from the exposure to the static magnetic field have been reported. These results may be of a great importance for evaluating the benefits and hazards of exposure to the static magnetic field. Also the importance of this work lays in the fact that *E. coli* as a microorganism is a unit cell behaving as a complete living biological system. (Table 1 and Fig 1); shows the change in the *E.coli* type 1 enzymes such as ADH, CIT and GEL. It is clear from this table and figure that the24 hrs exposure period to different forces of SMF (400, 800, 1200 and 1600) G can inhibit or promote enzyme activity according to API test, these results are in a good agreement with (Gremion *et al*, 2009). The literature on biomagnetic effects on the growth and development of various organisms has been quite extensive showing both positive and negative findings. Among the positive findings attributed to strong magnetic fields are: altered growth rate, enzyme activities, cellular metabolism, DNA synthesis and animal orientation. Also (Saunders *et al*, 1991) reported that the biological effects of magnetic fields may critically depend on the physical characteristics of the magnetic signal, in particular the wave forces. Many studies indicated that magnetic field has a biological effect on living things (Gremion *et al*, 2009). .

Table (1): The API test for *E. coli* samples ± magnetic forces at 24 hours

ONDG	ADH	LDC	ODC	Cit	H2S	URE	TDA	IND	VP	GEL	GLU	Man	INO	SOR	RHA	SAC	MEL	AMY	ARA	24 hours
+	+	+	+	-	-	-	+	+	-	-	+	+	-	+	+	+	+	-	+	Control
+	-	+	+	+	-	-	+	+	-	-	+	+	-	+	+	+	+	-	+	400 Gauss
+	-	+	+	+	-	-	+	+	-	+	+	+	-	+	+	+	+	-	+	800 Gauss
+	-	+	+	+	-	-	+	+	-	-	+	+	-	+	+	+	+	-	+	1200 Gauss
+	-	+	+	+	-	-	+	+	-	-	+	+	-	+	+	+	+	-	+	1600 Gauss

Table (2): The API test for *E. coli* at 48 and 72 hours

ONDG	ADH	LDC	ODC	Cit	H2S	URE	TDA	IND	VP	GEL	GLU	Man	INO	SOR	RHA	SAC	MEL	AMY	ARA	48 & 72 hours
+	+	+	+	-	-	-	+	+	-	-	+	+	-	+	+	+	+	-	+	Control
+	-	+	+	+	-	-	+	+	-	-	+	+	-	+	+	+	+	-	+	400 Gauss
+	-	+	+	+	-	-	+	+	-	-	+	+	-	+	+	+	+	-	+	800 Gauss
+	-	+	+	+	-	-	+	+	-	-	+	+	-	+	+	+	+	-	+	1200 Gauss
+	-	+	+	+	-	-	+	+	-	-	+	+	-	+	+	+	+	-	+	1600 Gauss

Table 3 shows the growth rate of *E. coli* for each groups incubated in magnetic determined by spectrophotometer. There is a decrease in the growth rate of the exposed *E. coli* cells to different magnetic field forces. Kohno *et al.* (2000) reported ferrite magnet caused strength dependent decrease in the growth rate and maximum number of bacteria for *S. mutans, S. aureus* when cultured under anaerobic conditions but their growth was not inhibited under aerobic conditions. However no growth effects were detected in *E. coli* culture which was contrary to our result, the static magnetic field affected the growth rate of *E. coli*. . Table(4) and figure 3 (A and B). Shows that the growth rate of *E. coli* for each groups incubated without magnetic fields are determined by spectrophotometer. The 800G bacterial suspension incubated again without magnetic field at 24hrs, resulted in the mutation of *E. coli* growth, because the enzyme gelatinase can be affected again by magnetic field, and the colony size of bacteria on media agar was changed and the growth rate also decreases. We found obvious cell surface damage when the cells were exposed to SMFs. . Mutations can be caused by mistakes in DNA replication or by damage to DNA Portier *et al*,1998).We reported that MF exposures did not increase mutation rates in any tester strain although other investigators have reported that power frequency MFs can alter fidelity of replication via a defect on DNA repair systems. For instance (Henry L *et. al*, 2004) reported that 60Hz, 0.1–0.5mT magnetic fields inhibit DNA repair in rat cells in vivo. Gui *et al.* (2001) reported that the higher density 50Hz MF (400mT) affects DNA repair in a human cell line, and that the mutation frequency varies with induced current.

Table (3): The growth rate of *E.coli* for each groups incubated in magnetic field are determined by spectrophotometer

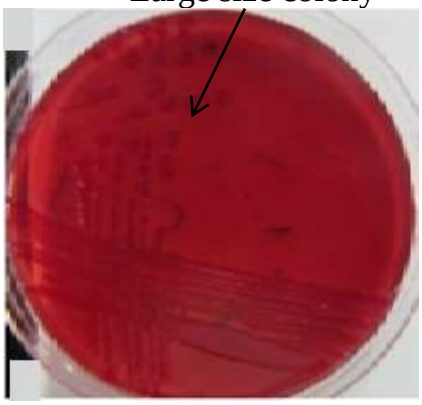
Samples incubation in different MF forces	OD 600nm at 24 hours	Bacterial cell count X 10^6
control	1.192	715.2
400G	1.403	841.8
800G	1.120	672
1200G	1.080	648
1600G	1.112	667.2

Table (4): The growth rate of *E.coli* for each groups incubated without magnetic fields are determined by spectrophotometer

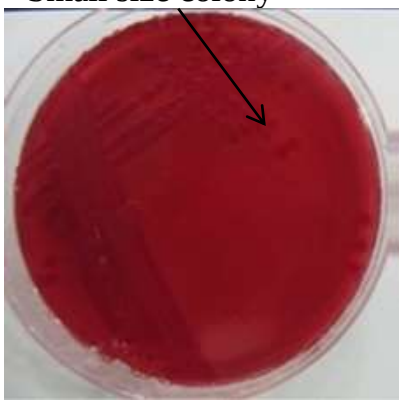
Samples incubation	OD 600nm at 24 hours	Bacterial cell count X 10^6
control without magnetic field	1.192	715.2
400G	1.200	720
800G	0.110	66
1200G	0.010	60
1600G	0.123	73.8

Fig (3): A. Treated *E.coli* culture with 800 G magnetic field.
B. *E.coli* culture without magnetic field (mutation occur from A large to B small colony size).

A
Large size colony



B
Small size colony



The different forces of static magnetic field affect the growth rate, and enzyme activity of *E. coli*. 800G static magnetic field was mutagenic to *E. coli* type 1.

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تأثير المجال المغناطيسي الثابت على طفرات البكتريا الايشيريشيا القولونيه

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الخلاصة

درست تأثير المجال المغناطيسي ثنائي القطب وبقوى 1600،1200،800،400 كاوس المحضرة محليا على التركيب البنائي لبكتريا *E.coli* type 1. استعملت احجام متساوية من مرق الوسط الزرعي السائل للبكتريا وعرضت للمجالات المغناطيسية المختلفة ولفترات زمنية متفاوتة والمتضمن 24، 48 و 72 ساعة كل على انفراد مقارنة بنموذج السيطرة السالبة للمزروع البكتيري غير المعرضة للمجال المغناطيسي. اختبر مزروع البكتريا المعوية المعاملة باختبار API kit، اشارة النتائج حدوث تغيير في نتائج الاختبارات Arginine dihydrolase (ADH), Citrate utilization (CIT Gelatinase (GEL)). كما لوحظ تغيير في شكل مستعمرات البكتريا المعاملة على الوسط MacConkey agar. استنتج من نتائج دراسة شكل المستعمرات وفعاليتها الحياتية حدوث الطفرة لخلايا البكتريا المعرضة للمجال المغناطيسي.

الكلمات الدالة:

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