Iraqi National Journal of Chemistry

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Improve Biosafety Level in Advance Molecular Laboratory at College of Science

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Abstract

Many progressing biological researches have been done in advance molecular laboratory at college of science Al-Mustansiriyah university. Therefore many consequences were appeared like contamination area with ethidium bromide, infectious pathogens and chemical toxic materials so many practical steps have been done to over takes these consequences such as replacement the conventional PCR with Real-Time PCR, replacement gel electrophoresis with automated gel electrophoresis, replacement ethidium bromide with other safe stains also used PCR stations and biosafety cabinet, to improve biosafety level in molecular laboratory.

Keywords: Biosafety, Molecular Laboratory, ethidium bromide**,** Real-Time PCR**,** gel electrophoresis**.**

Introduction

Biosafety is extensive loss prohibition of biological integrity, focusing both on ecology and human health. So these prohibition mechanisms include conduction of methodical reviews of biosafety in laboratory environment, furthermore hard guidelines to follow. When we entered in the new millennium, the application of DNA technologies have been occupied wide range in filed molecular genetics great payoff in the service ofhumanity, especially in pathogens detection, mutation detection, virus detection and sequencing, gene therapy additional to important role in detection the SNP that play role in genetic diseases and cancers furthermore in forensic DNA. So these technologies were applied in many laboratories in (researches lab,

university lab and heath lab).Despite of the great benefit, these technology have many consequences it important to highlight these consequences

Furthermore each year new chemical enter market and generate an increasing volume of residue raising health and environment concern although these chemicals describe as toxic, have a proportion hazard that not classified can do harmful biological effect. So the evaluation of biohazards periodically and doing assessment to biorisk which ensure the biosafety in lab and health workers.Especially the Biosafety is used to protect from harmful incidenceadditional to high security facilities are demanded when dealing with synthetic biology belong to the possibilities of bioterrorism acts or release of hazardous chemicals or organisms into environment.

Objective and Goals: evaluate the biohazardous in molecular lab and estimate the assessment of biosafety levels to improve biosafety and good working practice in lab. So the complete comprehensive of experimental risks that associated with synthetic biology is assisting to enforce the knowledge and performance of biosafety.

Material and methods

Conduct a laboratory risk assessment with assistance of biosafety committee and specialized persons was done to describe the research activities in the lab, identify the organisms and biological materials was used in lab, describe the risk associated with laboratory procedures and appropriate controls and/ or practices to minimize those risk. As it appear in diagram below.

Results and discussion

Polymerase Chain Reaction(PCR)

In conventional PCR used many chemical reagents like mastermix this contain some toxic dyes to person and environment ,furthermore the PCR product should be electrophoresis in gel electrophoresis by using agarose and ethidium bromide dye (3,8-dimmino-5-ethyl-6-phenylphenanthridinium bromide), which interacting agent used in molecular genetics and structures studies of DNA and chromatin, EB showed the ability to inhibit cell growth in tissue culture even in low concentration additional to mutagenic capacity. EB was used in electrophoresis to sized, and detected PCR product under UV trans-illiumiator.

UV trans-illiuminator consider carcinogenetic to skin and destroyed collagen layer.PCR techniques required a suitable area one of the most important criteria that demanded wide area from the bench lab, as shown in figure (1).

Figure (1) bench lab crowded with equipment

from these three steps of PCR techniques the electrophoresis representative the hazardous stepto person and environment especially when we dealing with viruses because the possibility of infection by virus while loading the sample or laboratory contamination of the environment also the agarose and buffer contaminated with virus and ethidium bromide dye, it very carcinogenetic additional to contaminate all the equipment and working area with it furthermore contaminate the swage.

In this case we work hardly to overtake this problems in our lab by using:

Firstly : Real-time PCR technique is more accurate and sensitive in molecular genetics also the result appear after one step, detection by fluorescent dye inside the instrument and the product can directly disposal, so this technique devoid us the toxic dye, UV, contamination and takes short time as shown in figure (2)

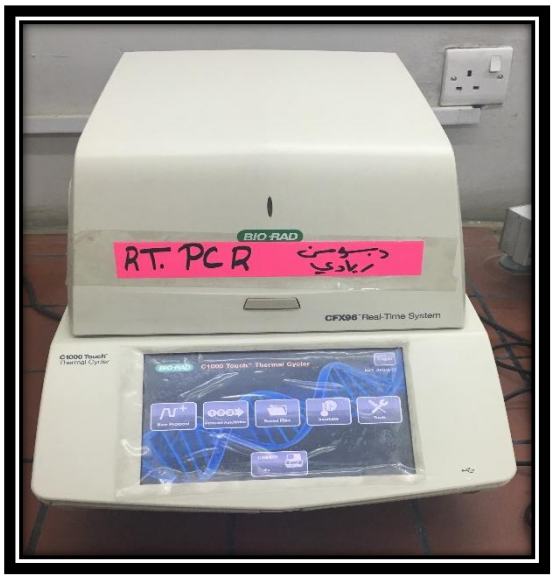

Figure (2) Real-PCR from Bio-Rad

Secondly: using PCR station and biosafety cabinet to avoid lab contamination and provide limit working area with power full of sterilization by UV as shown in figure (3)

Figure (3) PCR station and biosafety cabinet

Thirdly: replacement the ethidium bromide dye with diamond dye it is more safe and has high efficiency to bind with nucleic acid as shown in figure (4)

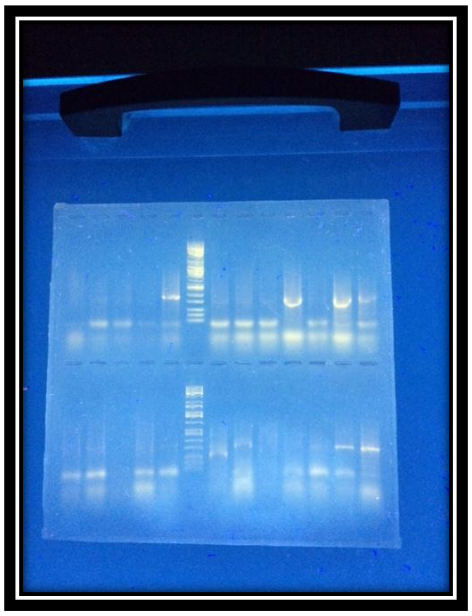

Figure (4) agarose gel stained with diamond dye

Fourthly: experion electronic gel electrophoresis technique is very accurate and sensitive to nucleic acid band (can separate two band have differences up to 5bp), protein and RNA integrity so this technique devoid us using agarose and toxic dyes also the result appear only after 20 mins.figure (5) show the hazardous area while figure (6) show the experion electronic gel electrophoresis system space

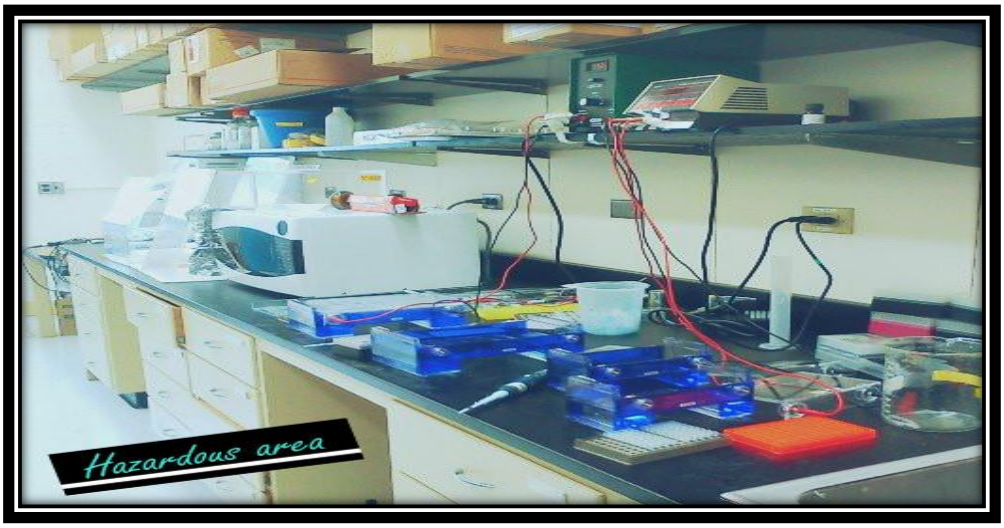

Figure (5): crowded area with hazardous

Figure (6) experion electronic gel electrophoresis system

Conclusion

As an college of Science related to ministry of higher education and scientific research, this project will improve the safe work practices in molecular laboratory that deal with synthetic biology to ensure maintenance a safe working environment in all research and teaching laboratories where the biological material are used additional to establish biorisk management system.

Recommendation

1- Increase personal understanding to the risks connected with work in laboratoryby offering appropriate training to mitigate those risks and follow the required safety practices in their work.
2- Determining the roles, responsibilities and procedures of the labs and changing according the needs.
3- Provide the emergency plans for spills and/ or personal exposure as set by safety committee.
4- Ensure the performance of all safety equipment used in the laboratory including personal protective equipment, biosafety cabinet,aerosol proof centrifuges, etc.
5- Reported any significant problems belong to research accidents and illness to the workers in the laboratory.
6- Implementing and following the legal instructions

Acknowledgement: we would express our great thankful to ministry of higher education and scientific research specially research development department to their financial support to develop infrastructure of our laboratory and department of united states for grand to attended training courses with CRDF

Global and Sandia international laboratory to developing our skills in biorisk management.

تحسين مستوى السلامة الأحيائية في مختبر علم الاحياء الجزيئي المتقدم في كلية العلوم الجامعة المستنصرية

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الخلاصة:

العديد من البحوث تتم في مختبر علم الاحياء الجزيئي المتقدم في كلية العلوم الجامعة المستنصرية. ولذلك ظهرت العديد من المشاكل مثل منطقة التلوث مع بروميد إيثيديوم ومسببات الأمراض المعدية والمواد السامة الكيميائية. الكثير من الخطوات العملية أجريت لتقليل او ازالة هذه النتائج المعتمدة على الوقت. استبدال بروميد إيثيديوم مع صبغة آمنة أخرى تستخدم أيضا محطات ، لتحسين مستوى السلامة الحيوية في المختبرات الجزيئي.

References:

1- Ouchi, R. Y.; Manzato, A.J.; Ceron, C. R. and G. O. Bonilla-rodriguez.(2007). Evaluation of the effect of single exposure to ethidium bromide in Drosophila melanogaster (Diptera-Drosophilidae). J. Bull Environ. Contam Toxical.78:489-493.
2- War, K. K.(2011).Advanced Clinical Applications of PCR Technology in the Molecular Genetics & Cancer. J.Syrian clinical laboratory association. Vol:6 no. 2.
3- von Wurmb-Schwark, N.; Cavelier, L. and G.A. Cortopassi .(2006). A low dose of ethidium bromide leads to an increase of total mitochondrial DNA while higher concentrations induce the mtDNA 4997 deletion in a human neuronal cell line. Mutat Res. **596** (1-2): 57–63.
4- Ohta, T.; Tokishita, S. and H. Yamagata H. (2001). Ethidium bromide and SYBR Green I enhance the genotoxicity of UV-irradiation and chemical mutagens in E. coli.. *Mutat Res.* **492** (1-2): 91–7.
5- www.bio-rad.com
6- Laboratory manual safety Michigan technological university. https://www.mtu.edu/research/administration/integrity-compliance/pdf/biosafety-manual.pdf
7- Wilson, D.E. and Chosewood, L.C., (eds.) 2009., Biosafety in Microbiological and Biomedical Laboratories 5th ed., Centers for Disease Control and Prevention, and National Institutes of Health., Washington, D.C., US Government Printing Office.