MEETING OF THE Minds

Undergraduate Research Symposium

APRIL 30 | 2019

Carnegie Mellon University Qatar

CARNEGIE MELL
PITTSBURGH PEN

Meeting of the Minds is an annual symposium at Carnegie Mellon University that gives students an opportunity to present their research and project work to an audience of faculty, fellow students, family members, industry representatives and the larger community. Students use posters, videos and other visual aids to present their work in a manner that can be easily understood by both experts and non experts. Through this experience, students learn how to bridge the gap between conducting research and presenting it to a wider audience. A review committee consisting of industry experts and faculty members will review the presentations and choose the best projects and posters. Awards and certificates are presented to the winners.

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From the Dean

The Meeting of the Minds student research symposium is a celebration of ingenuity, hard work, scientific exploration and intellectual curiosity. It is a highlight of the academic year, and we are exceptionally proud of the fine body of work produced by our students.

Research is an essential element of the undergraduate experience. For some students, this is the beginning of a career in scientific exploration, experimentation and analysis. For others, the intellectual rigor of research is invaluable experience in problem solving, which develops critical skills they will use throughout their professional careers.

At its heart, scientific research brings together creativity and reason. The projects at Meeting of the Minds 2019 are a showcase of this process. I encourage you to explore the projects, ask questions and learn about the unique perspectives that our students bring to scientific questions.

Michael Trick
Dean
Carnegie Mellon University in Qatar

Undergraduate research at CMU-Q

A research institute like no other, Carnegie Mellon is home to the world's leading experts in a range of fields. In this tradition, Carnegie Mellon University in Qatar nurtures and develops opportunities for faculty members and students to build regionally relevant research programs in their areas of expertise.

Faculty members contribute to the CMU-Q body of work through studies funded by Qatar National Research Fund (QNRF) and internal seed funds. These projects often provide a framework for undergraduates to learn about the research process and contribute to a larger project.

Students also undertake senior thesis projects, pursue independent studies guided by faculty mentors, initiate their own projects, and partake in summer research programs within Carnegle Mellon University and Education City. Meeting of the Minds is a showcase of these projects.

Acknowledgements

Special Awards

Carnegie Mellon University in Qatar acknowledges and thanks the Ministry of Development Planning and Statistics and Qatar National Research Fund for recognizing students and researchers with special awards.

Judges

Carnegie Mellon University in Qatar would like to express deep appreciation to the judges, who offer their time, expertise and feedback to make this research symposium a success. Thank you.

- **Dr. Essam Abdelalim,**
  Hamad Bin Khalifa University
- **Nesrine Affara,**
  Carnegie Mellon University in Qatar
- **Dr. Ali Alaboudy,**
  Qatar National Research Fund
- **Law Alsobrook,**
  Virginia Commonwealth University School of the Arts in Qatar
- **Houda Bouamor,**
  Carnegie Mellon University in Qatar
- **Salim Bougarn,**
  Sidra Medicine
- **Jennifer Bruder,**
  Carnegie Mellon University in Qatar
- **Lauren Burakowski,**
  Carnegie Mellon University in Qatar
- **Julie Decock,**
  Hamad Bin Khalifa University
- **Mohammed Dehbi,**
  Hamad Bin Khalifa University
- **Hasan Demirkoparan,**
  Carnegie Mellon University in Qatar
- **Muhammad Elnaggar,**
  Sidra Medicine
- **Jason Ford,**
  Sidra Medicine
- **John Gasper,**
  Carnegie Mellon University in Qatar
- **Mohammad Hammoud,**
  Carnegie Mellon University in Qatar
- **Henning Horn,**
  Hamad Bin Khalifa University

- **Karl Richard Alexander Knuth,**
  National Center for Cancer Care and Research,
  Hamad Medical Corporation
- **Ramesh Krishnamurti,**
  Carnegie Mellon University in Qatar
- **Rafah Mackeh,**
  Sidra Medicine
- **Nayef Mazloum,**
  Weill Cornell Medicine-Qatar
- **Enas Mohammed,**
  Qatar National Research Fund
- **Mohamed Mokbel,**
  Qatar Computing Research Institute
- **Preslav Nakov,**
  Qatar Computing Research Institute
- **Basem Shomar,**
  Qatar Environment and Energy Research Institute
- **Munir Tag,**
  Qatar National Research Fund
- **Kin-Ming Tsui,**
  Sidra Medicine
- **Stephan Vogel,**
  Qatar Computing Research Institute
- **Ingmar Weber,**
  Qatar Computing Research Institute
- **Barak Yehya,**
  Planning and Statistics Authority

Carnegie Mellon University in Qatar awards

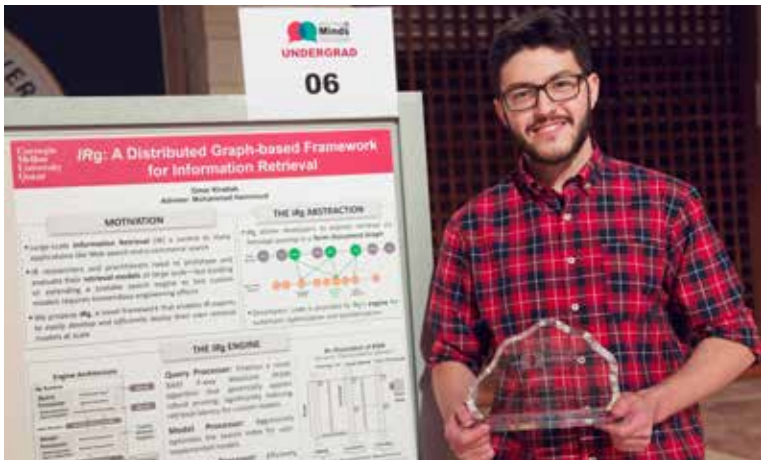
There were 26 poster presentations in the undergraduate category at Meeting of the Minds 2019, representing each of the five programs of study at CMU-Q: biological sciences, business administration, computational biology, computer science and information systems. CMU-Q awards the top three poster presentations and recognizes one poster for best design.

Best Project

First place

Omar Khattab, IRg: A distributed graph-based framework for information retrieval

Advisor: **Mohammad Hammoud**



Omar Khattab, who graduated with a degree in computer science, received the Best Project Award and an award from Qatar National Research Fund at Meeting of the Minds 2019. This project was Khattab's senior honor thesis, which was later recognized by Carnegie Mellon University's School of Computer Science with the Alumni Award for Undergraduate Excellence in Computer Science.

See page 58 for the poster and abstract.

Best Project

Second place

Beom Jin Jayden Park and Hawra Al-Saygh, Effect of aspartame on kinetics of calf intestinal alkaline phosphatase

Advisor: **Annette Vincent**

See page 42 for the poster and abstract.

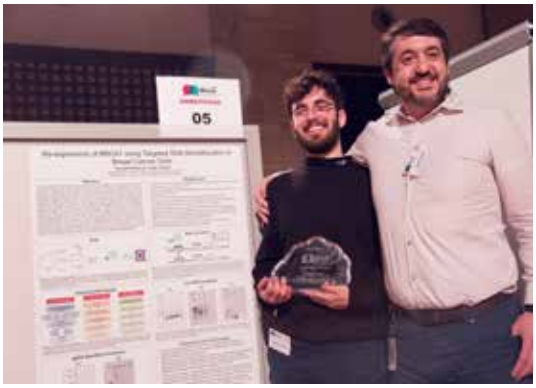
Best Project

Third place

Youssef Kanbour, Re-expression of BRCA1 using targeted DNA demethylation in breast cancer cells

Advisor: **Ihab Younis**

See page 54 for the poster and abstract.



Best Poster Design

Al-Dana Al-Mohannadi, Educating girls in Qatar: Toward enhancing technology use in public schools

Advisor: **Susan Hagan**

See page 60 for the poster and abstract.

Special awards: Qatar National Research Fund

Qatar National Research Fund and CMU-Q have a long history of partnership and collaboration, and the fruits of this partnership are on display at Meeting of the Minds. Many of the student projects are off-shoots of larger, faculty-led projects that have been generously funded by QNRF.

QNRF has offered special awards at Meeting of the Minds for many years. This year's QNRF awards were presented by senior program manager for ICT at QNRF, **Dr. Munir Tag**.

QNRF awards

Albandari Al-Khater, Modulating PARP1 splicing in breast cancer as potential therapeutic approach,

Advisor: **Ihab Younis**

Abstract and poster: page 22

Omar Khattab, IRg: A distributed graph-based framework for information retrieval

Advisor: **Mohammad Hammoud**

Abstract and poster: page 58

Anis Charfi, Syed Mehdi and Esraa Mohamad, ARAP – Author profiling and its application for market segmentation

Abstract and poster: page 72

Special awards: Planning and Statistics Authority

For many years, the Planning and Statistics Authority (formerly known as the Ministry of Development Planning and Statistics), has supported undergraduate research with special awards that are relevant to Qatar.

The awards from the Planning and Statistics Authority were announced by **Dr. Barak Yehya**, a longtime friend and supporter of CMU-Q.

Aisha Fakhroo, Boshra Al-Sulaiti and Reem Elasad, PTEN gene encodes a ncRNA that acts as a potent tumor suppressor in breast cancer

Advisor: **Ihab Younis**

Abstract and poster: page 34

Kawthar Al-Sadat, Molecular tools for microbial viability assessment in environmental samples: Case study of ballast water

Advisors: **Basem Shomar**, Qatar Environment and Energy Research Institute and **Annette Vincent**

Abstract and poster: page 26

Al-Dana Al-Mohannadi, Educating girls in Qatar: Toward enhancing technology use in public schools

Advisor: **Susan Hagan**

Abstract and poster: page 60

Faiq Defiandry, What does the eye say?

Advisor: **Jennifer Bruder**

Abstract and poster: page 62

Anis Charfi, Esraa Mohamad and Syed Mehdi, Deception detection in Arabic text

Abstract and poster: page 74

Class projects

Carnegie Mellon University follows a distinct approach to undergraduate education that combines professional training with a firm grounding in the arts and sciences. This approach teaches students to draw connections between disciplines and work effectively outside their focused area of study.

This year at Meeting of the Minds, select course projects were showcased to demonstrate the breadth of thought, research and exploration at Carnegie Mellon.




Course 82-286, Understanding Cultural Complexities: The French in the Middle East
Taught by **Bonnie Youngs**, Teaching Professor of French and Francophone Studies

Throughout the semester, students explored how acceptance and rejection interfere with our ability to communicate effectively across cultures. Students were asked to write a short essay and create a poster shaped around how the course affected their personal cultural views.

- This I believe... Religion doesn't define you, **Dina Abelazeem**
- This I believe... I should not have to explain myself... anymore, **Amna Ali**
- This I believe... I believe the past should stay in the past, **Khalid Al-Naemi**
- This I believe... My Palestinian identity, **Noora Al-Shurafa**
- This I believe... I should explain myself to avoid any misunderstandings, **AlDana Al-Sulaiti**
- This I believe... Cultural education could save humanity, **Naram Hajjar**
- This I believe... Learning to accept, **Sharoq Hassan**
- This I believe... Owning my mental hurdles, **Fahim Mahdi**
- This I believe... Fear of the unknown, **Faiha Sahirah**
- This I believe... Unite two countries and two religions, **Mariam Syed**
- This I believe... No home, **Moussa Zekak**

Course 62-238, Looking at Shapes
Taught by **Ramesh Krishnamurti**, Professor of Architecture

- What am I looking at? Using machine learning to resolve typology, **Hasan Nadeem, Muhammad Ibrahim Ghous**

67-475 Innovation in Information Systems

- Breeze, **Maryam Al-Maadeed, Aisha Al-Misnad, Maha AlMarri, Dana Al-Sheeb**
- CliQue, **AlDana Al-Sulaiti, Al-Danan Al-Mohannadi, Amna Ali, Rachel Marella**
- Scene, **Abdulaziz Al Haddad, Hassan Marafih, Ibrahim Ghous, Mohammed Al Khuzaei**

Undergraduate Posters

Effects of pH and temperature on the activity of alkaline phosphatase from sheep's brain

Authors
Sara AlDarwish, Maha AlTamimi

Advisors
Annette Vincent

Category
Biological Sciences

Abstract

The goal of this experiment is to find the optimum pH and temperature for alkaline phosphatase activity extracted from sheep brain. We anticipate that the optimum pH and temperature would be different from the literature value published from a research paper as it used purified AP from sheep brain and not a crude extract from sheep brain like what was done in this experiment. The research paper was published in 1977 by Bachhawa and Dorai that stated that the optimum pH is 9.0 and optimum temperature is at 37ºC. Therefore, it is hypothesized that the Km and Vmax would vary when comparing a crude extract to a purified form of AP from sheep brain and thus we can study the effects of pH and temperature on substrate binding affinity (Km). The optimum temperature was the same as the literature value of 37ºC, but the pH was 11.0 which is higher than the literature pH of 9.0. From which the Km was calculated to be 0.036 mM and Vmax was 0.0025 min-1.

Effects of pH and Temperature on the Activity of Alkaline Phosphatase from Sheep's Brain.

Sara AlDarwish and Maha AlTamimi Advisor: Annette Vincent
Biological Sciences Program, Carnegie Mellon University in Qatar

Abstract

The goal of this experiment is to find the optimum pH and temperature for Alkaline Phosphatase activity extracted from sheep brain. We anticipate that the optimum pH and temperature would be different from the literature value published from a research paper as it used purified AP from sheep brain and not a crude extract from sheep brain like what was done in this experiment. The research paper was published in 1977 by Bachhawa and Dorai that stated that the optimum pH is 9.0 and optimum temperature is at 37°C. Therefore, it is hypothesize that the Km and Vmax would vary when comparing a crude extract to a purified form of AP from sheep brain and thus we can study the effects of pH and temperature on substrate binding affinity (Km). The optimum temperature was the same as the literature value of 37°C, but the pH was 11.0 which is higher than the literature pH of 9.0. From which the Km was calculated to be 0.036mM and Vmax was 0.0025s^{-1}.

Introduction

Alkaline phosphatase (AP) is a **homodimeric** protein enzyme that is found in nearly all living organisms, with the exception of some plants. It is found in the periplasmic space of *E. coli* bacteria. In humans, it is found in many forms depending on its origin within the body, it plays an integral role in metabolism within the liver and development within the skeleton. Alkaline phosphatase can break down a substrate called p-nitrophenylphosphate (NPP) which gets dephosphorylated to produce yellow colored p-nitrophenol. This reaction is vital in helping the detection of AP through the color change which is measurable at wavelength of 410nm, the reaction is shown below:

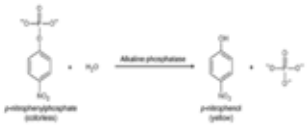
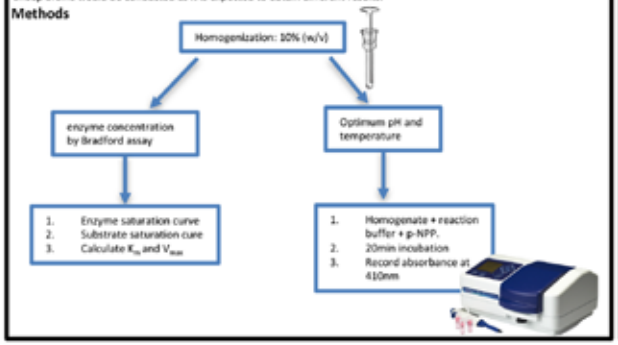


Figure 1[12]: Dephosphorylation of p-nitrophenylphosphate by AP

Many published research papers address the characteristics of AP that is present in E. Coli, some plants, and different organs in animals. One of the focused organs to study AP is from the brain. This raised an interest in studying the AP extracted from a Qatari Sheep brain as it has never been done before, and a comparison between the AP from crude extract of a Qatari sheep brain and the published literature values (pH and temperature) of purified AP from other sheep brains would be conducted as it is expected to obtain different results.

Methods



Results

Figure2: Lineweaver Burke plot of alkaline phosphatase from sheep brain

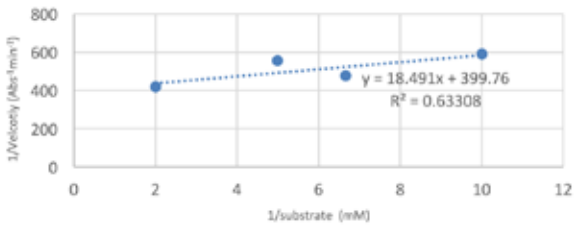


Figure3: Substrate saturation curve of 0.19 mg/ml Alkaline phosphatase from sheep brain

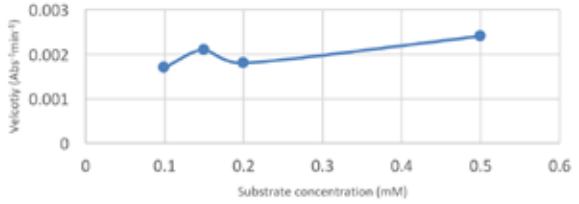


Figure4: Effects of pH on the activity of Alkaline phosphatase from sheep brain

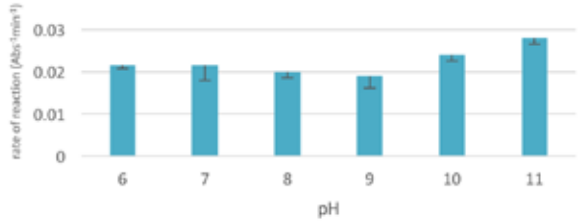
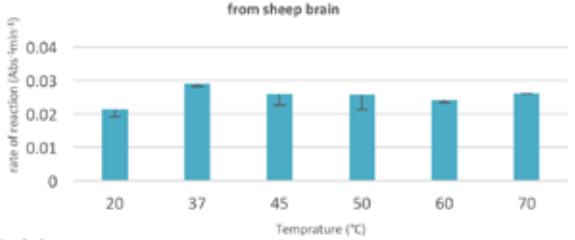


Figure5: Effects of temperature on the activity of Alkaline phosphatase from sheep brain



Analysis

The inverse was taken to plot Lineweaver Burke Plot (figure 3) from which we obtain Km and Vmax as the y-intercept is 1/Vmax, the x-intercept is -1/Km, and the slope is Km/Vmax. Since the x-intercept is not shown in figure 3, the Km is calculated from the slope and y-intercept to be 0.036mM and the Vmax is 0.0025s^{-1}. The literature value of Km for AP in sheep brain is 0.1mM. The Km obtained is 0.036mM which is lower than the published value which indicates that the substrate binding affinity is higher for the AP extracted in this experiment. The accuracy of the Km obtained is related to the r-squared value which is of 0.63, this suggests that the Km calculated is not reliable and hence not accurate due to low R^2. To explain the difference in the Km values, we have to check for the optimum pH and temperature values obtained in this experiment. The optimum temperature was 37C which is the same as the literature value. However, for the pH it was quite different as the literature value was 9.0 but the obtained value was 11.0. This suggests that the change in optimum pH had caused the change in Km value, and thus the temperature does not seem to have a vital role in substrate affinity. Therefore, the increase in pH from 9.0 to 11.0 led to affecting the ionic charges in AP's active site which increased the substrate's affinity and this is shown in the large decrease of Km value (lower Km value, higher substrate affinity).

Conclusion

With regards to the hypothesis, it was partially correct as the optimum pH of AP obtained from the crude extract differed from the literature value of purified AP, however, the optimum temperature remained the same. This shows that the change in Km is due to pH change in which at pH 11.0 (higher than 9.0) the substrate's affinity is higher (lower Km).

Acknowledgment

Special thanks to Dr. Annette Vincent, Maya Kemaldean, and Maria Bernales for help and guidance throughout the research

Reference:

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Effect of high temperatures on alkaline phosphatase isolated from Escherichia coli

Authors
Khulood Al-Haroon, Noora Al-Shukri

Advisor
Annette Vincent

Category
Biological Sciences

Abstract

The goal of this project is to examine the effect of high temperature on the activity and kinetic parameters of alkaline phosphatase isolated from Escherichia coli. The enzyme alkaline phosphatase is known to be highly thermostable and functions properly at elevated temperatures. According to Irenus, A. et al., 2015, we hypothesize that the optimum temperature of E.coli alkaline phosphatase is 80°C at a substrate concentration lower than the Km value (0.0256 mM). This is because the bonds that maintain the secondary structure of the enzyme are buried and not affected by elevated temperatures. The activity of alkaline phosphatase was assayed spectrophotometrically using the substrate conversion to nitrophenol and the absorbance was measured at 410 nm. From our data results, the Km value for the substrate pNPP was the highest at 70°C then decreased at 90°C. While the Km value was the lowest (0.005 mM) at 80°C suggesting that this is the optimum temperature for the enzyme to optimally dephosphorylate pNPP.

Effect of High Temperatures on Alkaline Phosphatase Isolated from *Escherichia coli*

Khulood Al-Haroon and Noora Al-Shukri Supervisor Dr. Annette Vincent Biological Sciences Program CMU Qatar

Abstract

The goal of this project is to examine the effect of high temperature on the activity and kinetic parameters of Alkaline phosphatase isolated from *Escherichia coli*.

The enzyme Alkaline phosphatase is known to be highly thermostable and functions properly at elevated temperatures. According to Irenus, A. et al., 2015, we hypothesize that the optimum temperature of *E.coli* Alkaline phosphatase is 80°C at a substrate concentration lower than the Km value (0.0256mM). This is because the bonds that maintain the secondary structure of the enzyme are buried and not affected by elevated temperatures. The activity of alkaline phosphatase was assayed spectrophotometrically using the substrate conversion to nitrophenol and the absorbance was measured at 410nm. From our data results, the Km value for the substrate pNPP was the highest at 70°C then decreased at 90°C. While the Km value was the lowest (0.005 mM) at 80°C suggesting that this is the optimum temperature for the enzyme to optimally dephosphorylate pNPP.

Introduction

One has to study the kinetic parameters of an enzyme to understand how it functions in a chemical reaction. The enzyme efficiency depends on specific parameters such as temperature, substrate concentration, pH, and metabolic effectors. Alkaline phosphatase is one enzyme that is found in mammalian cells, and its use is widely spread as it plays an important role in dephosphorylation processes inside the human body.[1]

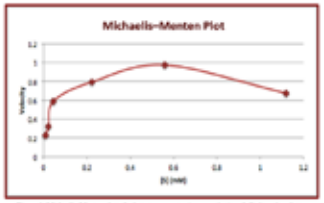
In this project, we are determining the efficiency of alkaline phosphatase isolated from *E. coli* by measuring the kinetic parameters under varying temperatures which are 37°C to 70°C, 80°C, and 90°C. The efficiency of the enzyme is measured by determining the velocity of the enzyme-substrate reaction. V_{max} and K_m, are two main parameters that correspond to the enzyme efficiency and binding affinity respectively. They are obtained by plotting a Lineweaver Burke plot where the substrate concentration reciprocal is plotted against the velocity reciprocal. Alkaline phosphatase dephosphorylates p-nitrophenolphosphate (pNPP) into p-nitrophenol (pNP) and Pi. The accumulation of p-nitrophenol results in a yellow color that can be measured using UV-Vis spectrophotometer as it absorbs at 410 nm. The absorbance value of this yellow product is directly proportional to the enzyme activity. Literature have shown that *E. Coli* alkaline phosphatase has maximum activity at 80°C.

Methods

The activity of alkaline phosphatase was assayed using UV-Vis spectrophotometer. The standard assay for *E. coli* alkaline phosphatase was performed at 37°C for 3 minutes using 11.2 mM (pNPP) while varying the enzyme concentration. The rate of dephosphorylation of pNPP was continuously measured spectrophotometrically at 410nm over the linear period. The plot of the initial velocity versus different enzyme concentrations was generated in order to determine the proper enzyme concentration for further kinetic determination.

The V_{max} and K_m was also determined under constant enzyme concentration (0.05 U) and varied substrate concentration.

In addition, the effect of high temperatures on the activity of *E. coli* alkaline phosphatase was studied. The optimum temperature was determined by the standard assay method under high temperatures in 1.0 M Diethanolamine buffer with 0.05 mM $MgCl_2$ (pH 9.8). The plot of initial velocity versus varying concentrations of substrate was generated for all enzyme assays to determine V_{max} and K_m, along with the Line-weaver Burke plot.



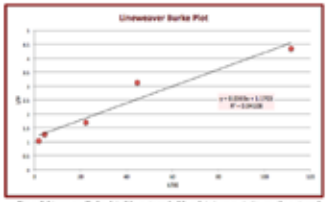


Figure 3. Effect of different temperatures on alkaline phosphatase activity

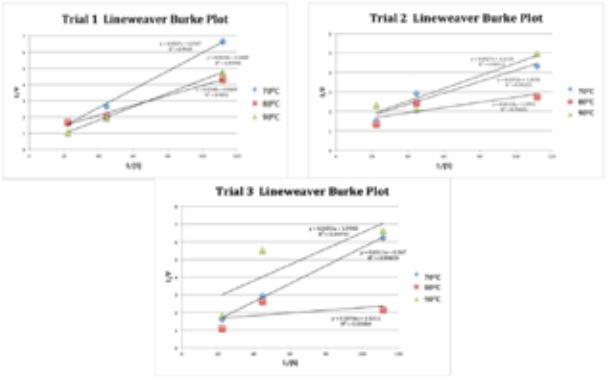
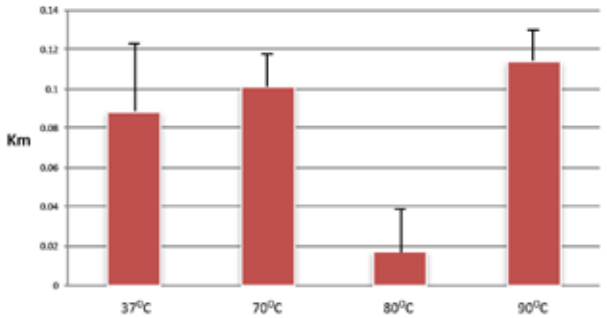


Figure 4. Effect of different temperatures on K_m of alkaline phosphatase



Discussion

The chosen enzyme concentration was 0.05 Units. The Line-weaver Burke plot was generated as shown in figure 2.

Since the y-intercept reciprocal is equal to V_{max}, and the slope of the line is equal to K_m/V_{max}, by using calculations, we get V_{max} value of 0.854 and K_m as 0.0256 mM. The K_m value obtained for *E. coli* alkaline phosphatase was 0.0256 mM at 37°C, while the K_m value provided by the instructor is 0.013 mM, which is lower than what we have obtained. If the R^2 value is closer to 1, the K_m value becomes more accurate.

According to Irenus et al.[2], we hypothesized that the optimum temperature of *E. coli* alkaline phosphatase is 80°C at a substrate concentration lower than the K_m value. Our data showed that the K_m for the substrate pNPP was the lowest at 80°C in all trials as shown in figure, indicating that this is the optimum temperature for the enzyme.

- When the enzyme activity was assayed at 37°C which is the positive control for this experiment, the K_m value in 37°C is higher than 80°C. Meaning that the enzyme functions more efficiently in 80°C than in 37°C.
- K_m value was higher at 70°C, which indicates that this temperature gives low activity of *E. coli* alkaline phosphatase.
- K_m value was the highest at 90°C, indicating that the enzyme is inactive and might have denatured due to extreme high temperature.

Conclusion

We conclude that the optimum temperature of *E. coli* alkaline Phosphatase is 80°C with the lowest K_m values in all three trials compared with the other temperatures 70°C and 80°C. This proves our hypothesis in which the optimum temperature of *E.coli* Alkaline phosphatase is 80°C according to Irenus et al.[2]. The alkaline phosphatase is a highly thermostable enzyme that can catalyze the formation of products efficiently at high temperatures.

References:

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2 Tazisong, N. Senwo, & Ile (2015). Phosphatase Hydrolysis of Organic Phosphorus Compounds. *Advances in Enzyme Research*. Retrieved 2015, from https://file.scirp.org/pdf/AER_2015062411112845.pdf

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Kinetic study on effects of the inhibitor L-Phenylalanine on calf intestinal alkaline phosphatases

Authors
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Advisor
Annette Vincent

Category
Biological Sciences

Abstract

Mammalian Alkaline Phosphatase and their allosteric properties make them susceptible to inhibition by L-amino acids, such as L-Phenylalanine. Since the inhibition mechanism of L-phenylalanine remains largely unknown, we examined the effect of L-Phenylalanine on Calf Intestinal Alkaline Phosphatase (CIAP) using a different concentration of L- Phenylalanine inhibitor. The inhibition constant (Ki) of L-Phenylalalnine was found to be 1.1mmol (René, B., 2009). Without inhibitor, Calf Intestinal Alkaline Phosphatase showed the following kinetic characteristics with pNPP in 0.5 M Tris-HCl buffer (pH 10.4) containing 5 mM MgCl2·6H20 at 37°C: Vmax is 6.34 µmoles min-1 unit-1 and the Km= 1.51 mM, respectively. Vmax is higher than that of published value, this shows that the our CIAP acts faster than the published one. While the published value for Km is smaller than that found by our result. This shows that our CIAP has less affinity to p-NPP. According to Figure 4, when adding 0.8 mM of L-Phy the Vmax is -3.106 µmoles min-1 unit-1 and the Km is -0.99 mM, while when adding 1.1 mM of L-Phy the Vmax is -2.156 µmoles min-1 unit-1 and the Km is -0.73 mM, when 1.5 mM is added, the Vmax is -7.8 µmoles min-1 unit-1 and the Km is -1.65 mM. The values do not have a specific trend since for the Vmax and Km it increased and then decreased again. This can be due to experimental errors, use of different enzyme stocks and low R2 values that gave negative values which is not possible for the Km to be negative since it is the concentration of substrate when the reaction reaches half the maximum velocity.

Kinetic study on effects of the inhibitor L-Phenylalanine on calf intestinal alkaline phosphatases

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Abstract

Mammalian Alkaline Phosphatase and their allosteric properties make them susceptible to inhibition by L-amino acids, such as L-Phenylalanine. Since the inhibition mechanism of L-phenylalanine remains largely unknown, we examined the effect of L-Phenylalalnine on Calf Intestinal Alkaline Phosphatase (CIAP) using different concentration of L-Phenylalanine inhibitor. The Inhibition constant (Ki) of L-Phenylalalnine was found to be 1.1mmol (René, B., 2009). Without inhibitor, Calf Intestinal Alkaline Phosphatase showed the following kinetic characteristics with pNPP in 0.5 M Tris-HCl buffer (pH 10.4) containing 5 mM MgCl2·6H20 at 37 °C: Vmax is 6.34 µmoles min^{-1} unit^{-1} and the Km= 1.51 mM, respectively. Vmax is higher than that of published value, this shows that the our CIAP acts faster than the published one. While the published value for Km is smaller than that found by our result. This shows that our CIAP has less afinity to p-NPP. According to Figure 4, when adding 0.8mM of L-Phy the Vmax is -3.106 µmoles min^{-1} unit^{-1} and the Km is -0.99mM, while when adding 1.1mM of L-Phy the Vmax is -2.156µmoles min^{-1} unit^{-1} and the Km is -0.73 mM, when 1.5mM is added, the Vmax is -7.8µmoles min^{-1} unit^{-1} and the Km is -1.65mM. The values does not have a specific trend since for the Vmax and Km it increased and then decreased again. This can be due to experimental errors, use of different enzyme stocks and low R^2 values that gave negative values which is not possible for the Km to be negative since it is the concentration of substrate when the reaction reaches half the maximum velocity.

Introduction

Alkaline phosphatase is a homodimeric protein with a molecular weight of 86 kDa. It is found in many eukaryotic and prokaryotic cells to catalyze the hydrolysis and transphosphorylation of diverse phosphate monoesters. Example on substrates are phenylphsphate & para-Nitrophenylphosphate known as p-NPP. The enzyme contains two zinc atoms and a magnesium ion. Monomers of the enzyme alone do not function, both subunits are required. The optimum pH of the enzyme is at 8.0 and it is stable in hot and hostile non-reducing environments outside the cells.(1)

Mammalian Alkaline Phosphatase and their allosteric properties make them susceptibile to inhibition by L-amino acids, such as L-Phenylalanine. L-Phenylalalnine is found to be an aminoacid that acts like a uncompetitive inhibitor of Alkaline Phosphatase (2). This type of inhibitor does not resemble the substrate and doesn't bind to the active site, but rather to a separate site on the enzyme (1). Since, there is not enough research papers who proved L-Phenylalalnine effect on CIAP, we will examine the effect of L-Phenylalalnine on Calf Intestinal Alkaline Phosphatase (CIAP) using different concentration of inhibitor. CIAP is a 68kDa enzyme that has an optimum pH of 10.4.The Ki was found to be 1.1mmol.

Methods

First assay tested enzyme concentration versus reaction velocity, to identify whether our CIAP enzyme amount is rate limiting or not. The substrate used para-Nitrophenylphosphate (pNPP), with a constant concentration of 11.2?mM (5µl). Enzyme concentrations tested was 0.05,0.1,0.2,0.3,0.4,0.5,1 (U). Secondly, Michaelis Menton plot (Vo vs [S] vs Velocity) was perfomed, this is to find the linear part of the curve and the Km & Vmax value. Enzyme concentration used was 0.5U (5µl). pNPP concentration tested was 0.2,0.3,0.5,1,1.5,2,3.32,3.35,10mM. Thirdly, Lineweaver burke plot was made for 3 substrate concentrations 0.2,0.3,0.5mM, each with the 3 inhibitor concentrations testes, 0.8mmol, 1.1mmol,1.5mmol. 20mM of L-Phenylalanine was made from 20mM stock prepared from 200mM stock..
Total volume in cuvettee in all assays was 300µl. Buffer used in all assays is 0.5 M Tris-HCl buffer (pH 10.4) containing 5 mM MgCl2·6H20 was incubated at 37 °C water bath, it's amount varied according to amount of enzyme and substrate concentration added. All assays were measured using spectrophotometer at 410nm for 2-3 minutes

Data

Figure 1: Enzyme concentration versus Rate of reaction of Calf Intestinal Alkaline Phosphatase at room temperature using para-Nitrophenylphosphate substrate

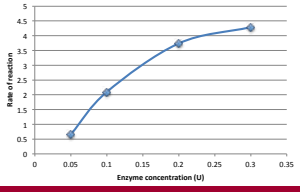


Figure 2: Michaelis Menton plot of Calf Intestinal Alkaline Phosphatase at room temperature using para-Nitrophenylphosphate substrate

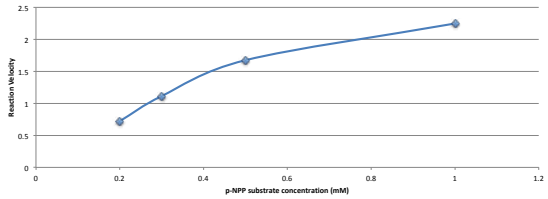


Figure 3: Reciporocal of p-NPP substrate concentration versus Reaction velocity of of Calf Intestinal Alkaline Phosphatase at room temperature

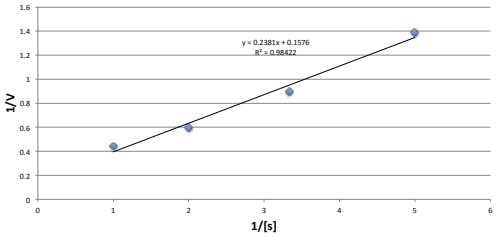
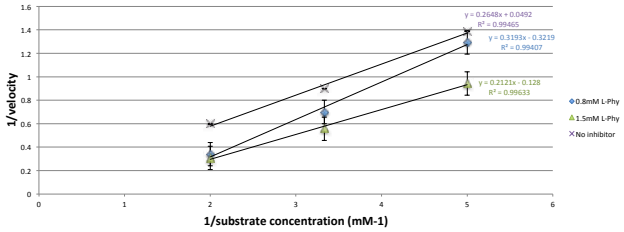


Figure 4: Lineweaver plot of of Calf Intestinal Alkaline Phosphatase at room temperature with L-Phenylalanine inhibitor



Analysis of Results

In figure one, 0.5U of enzyme was chosen as the best concentration used for following assays because it lies in the most saturated point in the curve, which shows the highest speed of reaction the enzyme can go through. In figure 2, the first 3 substrate concentration lied in the linear part of the graph, which exhibits first order of reaction. In figure 3, the Vmax is the inverse of the y intercept, where the Km is the negative inverse of the x intercept value, thus, the Vmax is 1/0.1576= 6.34 µmoles min^{-1} unit^{-1} and the Km= 1.51 mM. The published value for Vmax Km in Tris-HCl buffer at pH 11 is 3.12 µmoles min^{-1} unit^{-1}, which is lower than that found, this shows that the our CIAP acts faster than the published one. While the published value for Km in Tris-HCl buffer at pH 11 is 7.6×10^{-4} M, which is smaller than that found by our result. This shows that our CIAP has less afinity to p-NPP. According to Figure 4, when adding 0.8mM of L-Phy the Vmax is -3.106 µmoles min^{-1} unit^{-1} and the Km is -0.99mM, while when adding 1.1mM of L-Phy the Vmax is -2.156µmoles min^{-1} unit^{-1} and the Km is -0.73 mM, when 1.5mM is added, the Vmax is -7.8µmoles min^{-1} unit^{-1} and the Km is -1.65mM. The values does not have a specific trend since for the Vmax and Km it increased and then decreased again. This can be due to experimental errors and low R^2 values that gave negative values which is not possible for the Km to be negative since it is the concentration of substrate when the reaction reaches half the maximum velocity.

Conclusion:

We expected with the increasing concentration of inhibitor added, the inhibition would be higher, however, looking at this graph we observed that the higher amounts added of inhibitor added, the less inhibtion takes place. This might be because of using different enzyme stocks, the results appear to be inconsistent. In Future work, we will make sure that we have more replicates in order to have significant values.

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Comparing thermostability and enzyme kinetics of bacterial alkaline phosphatase and calf-intestinal alkaline phosphatase at high temperatures

Authors
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Category
Biological Sciences

Abstract
Thermostability property of enzymes are necessary in the industrialized economy. Some of the enzymes that are involved in the industrial processes are: oxidoreductases, transferases, hydrolases, lyases, isomerases and ligases (Ibrahim & Ma, 2017). The type of enzyme in focus of the lab would be alkaline phosphatase. The purpose of the lab is to determine and compare the changes and significance of high temperatures (60°C) in calf-intestine and bacterial alkaline phosphatase kinetic activity. The hypothesis is that calf-intestinal alkaline phosphatase (CIAP) would be more thermostable and have higher Vmax value at 60°C than bacterial alkaline phosphatase (ECAP) because the optimum temperature for CIAP is higher than ECAP (respectively 45°C and 37°C). Using literature values of Km and enzyme kinetics at optimum temperature for each enzyme, the amount of substrate to add were recorded. The substrate used in both enzymes was pNPP (para-Nitrophenylphosphate). The presence of NPP (nitrophenol) can be detected using UV-spectroscopy at 410 nm wavelength. Lineweaver plot was plotted to calculate and compare the Vmax. The results were that the Vmax and enzyme activity increased for CIAP at 60°C than in 45°C.

Comparing Thermostability and Enzyme Kinetics of Bacterial Alkaline Phosphatase and Calf-intestinal Alkaline Phosphatase at High Temperatures

Reem Al-Karbi, Sondoss Hassan **Professor Annette Vincent**

Biological Sciences Program, Carnegie Mellon University Qatar

Abstract

Thermostability property of enzymes are necessary in the industrialized economy. Some of the enzymes that are involved in the industrial processes are: oxidoreductases, transferases, hydrolases, lyases, isomerases and ligases (Ibrahim & Ma, 2017). The type of enzyme in focus of the lab would be alkaline phosphatase. The purpose of the lab is to determine and compare the changes and significance of high temperatures (60℃) in calf-intestine and bacterial alkaline phosphatase kinetic activity. The hypothesis is that calf-intestinal alkaline phosphatase (CIAP) would be more thermostable and have higher Vmax value at 60°C than bacterial alkaline phosphatase (ECAP) because the optimum temperature for CIAP is higher than ECAP (respectively 45°C and 37°C). Using literature values of Km and enzyme kinetics at optimum temperature for each enzyme, the amount of substrate to add were recorded. The substrate used in both enzymes was pNPP (para-Nitrophenylphosphate). The presence of NPP- (nitrophenol)can be detected using UV-spectroscopy at 410 nm wavelength. Lineweaver plot was plotted to calculate and compare the Vmax. The results were that the Vmax and enzyme activity increased for CIAP at 60°C than in 45°C.

Introduction

Alkaline phosphatase is an enzyme that hydrolyses a phosphate to a free inorganic phosphate group that can be transferred to phosphorylate other proteins, DNA strand and other organic compounds. The industrial application of alkaline phosphatase are bioremediation, cloning and more. The bioremediation process uses purified calf-intestine alkaline phosphatase to separate metal ions and uranium contaminants from the protein samples isolated. This is used by chromium-plating and leather manufacturers (Venu-Babu,.. 2018). The other application of calf-intestine alkaline phosphatase was that it helps in DNA cloning as the 5' and 3' ends of the DNA are dephosphorylated so avoids self-ligation of the DNA (Biolabs, n.d.).

Methods

The Enzyme activity for both enzymes was measured at optimum temperature (45°C for calf intestine alkaline phosphatase and 37°C for E.coli alkaline phosphatase). It was measured by adding different concentrations of substrate pNPP and hydrolyzed to NPP- that absorb at 410 nm. The enzyme saturation curve was done first to know the units of enzyme needed to reach plateau, that indicates the maximum number of enzyme that can hydrolyze the substrate pNPP. This was done to know the Km, and Vmax values at the enzyme's optimum temperatures (Alkaline Phosphatase Kinetics. (n.d.)).

0.67M pNPP + Enzyme (0.1U of ECAP and 1.0 U of CIAP in different tubes) + 1.0M Diethanolamine Buffer, 0.50mM Magnesium Chloride. pH 9.8

Product in NPP- absorb at 410 nm. For 3min, at 1s interval

Data

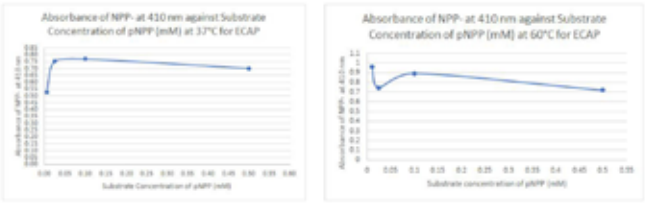

Figure 1: The absorbance of NPP- at 410 nm against the concentration of pNPP (mM) at optimum temperature (37℃), *on the left,* and high temperature (60℃), *on the right,* for ECAP.

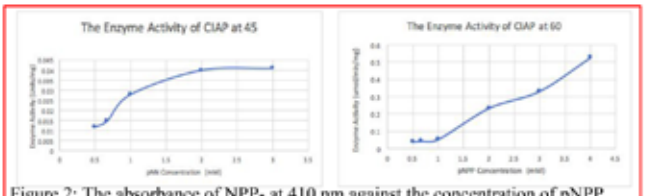

Figure 2: The absorbance of NPP- at 410 nm against the concentration of pNPP (mM) at optimum temperature (37℃), *on the left,* and high temperature (60℃), *on the right,* for ECAP.

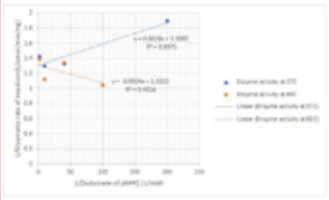

Figure 3: Lineweaver plot of ECAP enzyme activity at 37℃ and 60℃.

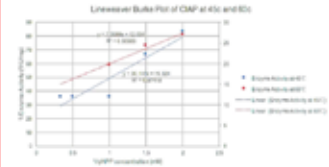

Figure 4: Lineweaver plot of CIAP enzyme activity at 45℃ and 60℃.

Analysis

In figure 1.0, the R^2 value for ECAP enzyme activity at 37°C, was 0.8975, and at 60°C was 0.4016, so since both are less than 0.95, there are some variability in our data. In figure 2.0, the R^2 value for CIAP enzyme activity for 37°C was 0.9913, but at 60°C it was 0.8874, which means there are some variability in this data. According to Figure 2.0, the calculated Km value for CIAP at 45℃ was 2.466 M and at 60℃ it was calculated to be -18.12 M.

Table 1.0 Kinetics of ECAP and CIAP at optimum temperatures and 60℃.

	Kinetics at optimum temperatures		**Kinetics at 60℃**	
Sample	**Calculated Km value (mM)**	**Calculated Vmax value (Units/mg)**	**Calculated Km value (mM)**	**Calculated Vmax value (Units/mg)**
ECAP	2.14×10^{-3}	7.65×10^{-1}	-1.82×10^{-3}	7.40×10^{-1}
CIAP	2.466	8.6×10^{-2}	6.0×10^{-1}	8.0×10^{-2}

Conclusion

In conclusion, the enzyme activity for both ECAP and CIAP decreased after incubation at 60℃. The Vmax decrease for ECAP was 0.025 umol/min/mg but for CIAP the Vmax decrease was 1.255 umol/min/mg. Therefore, the more thermostable enzyme was ECAP due to smaller decrease in Vmax comparing to CIAP.

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MEETING OF THE Minds Undergraduate Research Symposium

Carnegie Mellon University Qatar

Modulating PARP1 splicing in breast cancer as potential therapeutic approach

Author
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Category
Biological Sciences

Abstract

The DNA damage repair pathway is highly enriched in genes that contain minor introns. Minor introns are typically removed by splicing. Specifically, PARP1 is a protein that initiates DNA damage repair in cells and contains a minor intron. The primary objective of this project was to study the effect of DNA damage on the splicing of PARP1 in breast cancer cells. We used PARP1 antisense morpholino oligonucleotide that targets and inhibits its minor intron splicing, followed by PCR and qPCR analysis of splicing as well as western blotting to check for the protein level. We also analyzed cell viability over three days. We found splicing alterations upon DNA damage. In addition, upon inhibiting PARP1 splicing, we show decreased cell survival. Interestingly, treating cells with a DNA damaging agent along with inhibiting PARP1 minor intron splicing caused significant cell death, suggesting that a combination therapy would be possible.

Modulating PARP1 Splicing in Breast Cancer as Potential Therapeutic Approach

Albandari Al-Khater, Prof. Ihab Younis
Biological Sciences Program, Carnegie Mellon University in Qatar

Abstract

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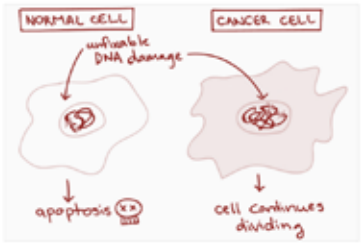
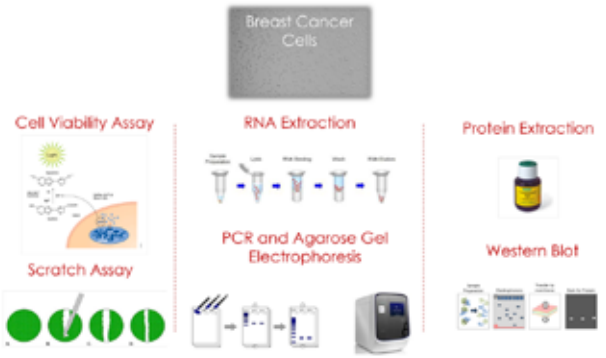
Background

- DNA damage repair pathway highly enriched with minor introns
- PARP1 functions in initiating DNA damage repair and additional transcription factor functions through DNA binding domain
- **Hypothesis:** Breast cancer cells regulate minor intron splicing of genes in the DNA damage repair pathway for their genomic instability



Methods



Results

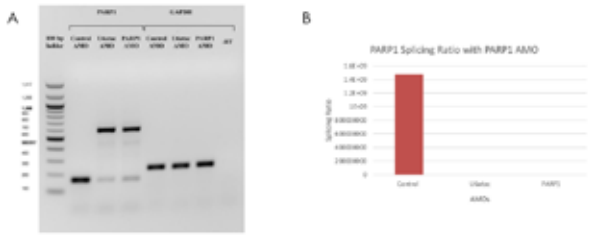


Figure 1: PARP1 AMO Efficiently Inhibits Splicing of PARP1 Minor Intron in Breast Cancer Cells. A) MDA-MB-231 cells were treated with control, Ubatac and PARP1 AMOs. Followed by a PCR amplification of spliced and unspliced PARP1, and run on 1.5% agarose gel in 1X TAE buffer. Loading control for the reactions was GAPDH. B) Splicing Ratio based on ImageJ Quantification of PARP1 Bands.

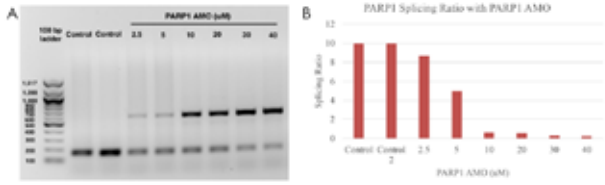


Figure 2: PARP1 Minor Intron Splicing shows Dose Dependency on PARP1 AMO Concentration in Breast Cancer Cells. A) MDA-MB-231 cells were treated with control and PARP1 AMOs. Followed by PCR amplification of spliced and unspliced PARP1, and run on 1.5% agarose gel in 1X TAE buffer. Loading control for the reactions was GAPDH. B) Splicing Ratio based on ImageJ Quantification of PARP1 Bands.

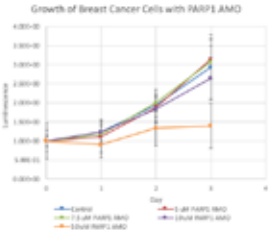


Figure 3: Growth of Breast Cancer Cells was Inhibited upon treatment with 50 nM PARP1 AMO. MB-231 cells were treated with 5, 7.5, 10 and 50 nM PARP1 AMO and followed for 3 days to assess the viability of the cells with cell titer glo. Each well had 2000 cells and each condition and time point had triplicates.

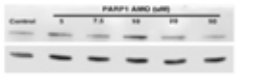


Figure 4: Slight Decrease in PARP1 Protein Upon PARP1 AMO Treatment. Protein was extracted from control MDA-MB-231 cells and AMO treated cells. Followed by SDS-PAGE, loading control was GAPDH. The gel was run at 100 volts for 1 hour and 15 minutes in 1X TGS Buffer. The gel was then blocked onto a nitrocellulose membrane that was then stained with anti-PARP1 and anti-GAPDH antibodies.

Figure 5: Death of Breast Cancer Cells upon treatment with 50 nM PARP1 and 0.5 nM Doxorubicin. MB-231 cells were treated with Control and 50 nM PARP1 AMO and 0.1 nM Doxorubicin. Cells were followed for 3 days to assess the viability with cell titer glo. Each well had 2000 cells and each condition and time point had triplicates.

Conclusion and Significance

Overall, the results indicate that a combined treatment of an AMO that inhibits PARP1 minor intron splicing with induction of DNA damage is efficient in killing breast cancer cells through synthetic lethality. So far, targeting PARP1 with inhibitor has been approved for patients with BRCA1 mutations. Our results show that this combinatorial treatment could be a novel therapeutic method to treat breast cancer and yield better prognosis regardless of BRCA1 status.

Acknowledgements

Nourhan Elkhateb, Professor Mohammed Bouaouina, Maya Kemaldean, Maria Bernales. This project was funded by Carnegie Mellon University Qatar Seed Grant for Professor Ihab Younis.

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MEETING OF THE Minds Undergraduate Research Symposium
Carnegie Mellon University Qatar

Integrin-mediated signaling in breast cancer cells

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Category
Biological Sciences

Abstract

Integrin are essential transmembrane proteins that function as cell adhesion receptors, there are various types of integrins due to the receptor being a heterodimeric complex. In addition to adherence, integrins have a signaling role that transduces cellular signaling to facilitate various cellular process. In breast cancer cells, changes in integrins levels and types provide cancer cells with the ability for metastasis through adhesion to various Extra Cellular Matrix (ECM) proteins and more aggressive phenotypes. We hypothesized that integrin-mediated cell adhesion to various ECM proteins could change cellular signaling in breast cancer cell lines (MDA-MB 231 & MDA-MB 468).

Initially, we determined integrin expression profile of both cell lines in addition to MCF-7 noninvasive cells. We observed the expression of integrin αvβ6 in MDA-MB 468 and MCF-7 cells and β2 integrin in MDA-MB 231. Then, we assessed whether cell detachment or matrix-specific cell adhesion such as adhesion to collagen, fibronectin and fibrinogen could change the phosphorylation state of key signaling effectors, namely AKT, Erk and p38 MAP kinases. The data shows an effect of detachment on the over-phosphorylation of both Erk and Akt in MDA-MB 468 while it causes down regulation of phosphorylation of both proteins in MDA-MB 231 compared to adhesion to serum. PMA addition shows a varying effect on the phosphorylation state in which it up-regulates Erk phosphorylation and downregulation of Akt phosphorylation. The aim of the research is to determine which effects matrices could have on cell signaling that could alter cell behavior such as cell division, adhesion, migration, etc. Following determination of adhesion effect on cell signaling, the next process that will be investigated is cell migration. The differential phosphorylation of various signaling molecules within the cells could bring about an effect on cell migration and metastasis.

Integrin-Mediated Cell Signaling in Breast Cancer Cells

Khalid Al-Naemi Dr. Mohamed Bouaouina
Biological Sciences Program CMU Qatar

Background

Integrin are essential transmembrane proteins that function as cell adhesion receptors, there are various types of integrins due to the receptor being a heterodimeric complex. In addition to adherence, integrins have a signaling role that transduces cellular signaling to facilitate various cellular processes. In breast cancer cells, the upregulation/downregulation of integrins provides the ability for cancer cells for easier metastasis through adhesion to various ECM proteins. Thus the differential expression of integrins in cancer could enable varying effect of aggressive phenotypes for the cancer cells. Our previous work has determined the integrin expression profile in MDA-MB 231.

Objective

Matrix-Specific integrin-mediated signaling in breast cancer cells is not fully understood. Our hypothesis is that we predicted that adherence to various ECM proteins will have an effect on the cellular signaling within the three breast cancer cell lines (MDA-MB 231, MDA-MB 468 & MCF-7). Adhesion-induced phosphorylation of various signaling proteins will be investigated.

Methods

Cell Culture:

Cells were cultured In Dulbecco's Modified Eagle's Medium (DMEM), supplemented with Fetal Bovine Serum (FBS), Penicillin-Streptomycin, Sodium Pyruvate and Non-essential amino acids. Incubated at 37C° Dry incubator with 5% CO_2 flow. The cells were monitored on a daily basis to sustain cell culture propagation.

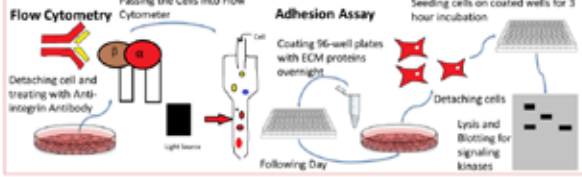
Flow Cytometry:

Cells are counted to be $5x10^5$ and are transferred into FACS tubes, and subjected to 3 conditions: Cells only, Cells + Secondary Ab, Cells + Anti-Integrin Primary Ab + Secondary Ab . The incubation time for each antibody staining is 30min at 4C°, while the staining with Secondary Ab was done in low light condition alongside previous set conditions. The cells are processed and analyzed using BD LSR Fortessa Flow Cytometer analyzer. Collected data was analyzed using Flow Jo.

Adhesion Assay:

96-well plates are coated with ECM protein overnight. Following washing with PBS, $1x10^5$ cells are seeded in each well, fed with serum-lacking medium and incubated in 37°C. A negative control is set with cell maintained at 37C° in suspension. After 3 hours, cells are washed and lysed. Cell lysate is ran in SDS gel, and western blot is carried with specific antibodies against Phospho and total: AKT, P38 and Erk MAP Kinases.

Scheme



Results

Flow Cytometry

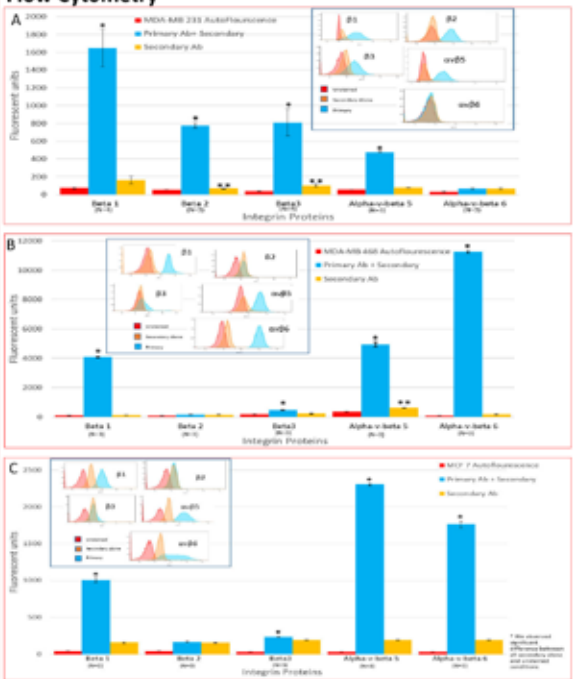


Figure 1: Analyzed fluorescence data representing specific integrins expression in A) MDA-MB 231, B) MDA-MB 468 and C) MCF7 with a panel data obtained from Cytometer as a histogram plots.
(*): Statistically significant difference between the primary staining and secondary staining (p-value <0.05)
(**): Statistically significant difference between secondary staining and MDA-MB 231 Autofluorescence (p-value <0.05)

Adhesion Assay

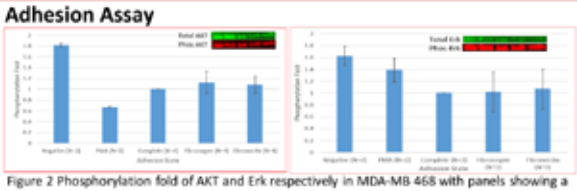


Figure 2 Phosphorylation fold of AKT and Erk respectively in MDA-MB 468 with panels showing a representative bands

Adhesion Assay

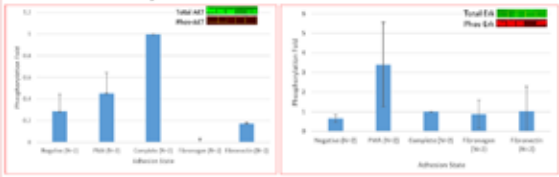


Figure 3: Phosphorylation fold of AKT and Erk respectively in MDA-MB 231 with panels showing a representative bands

Analysis

- Each Cell line has a distinct integrin expression profile, in terms of type and quantity
- Detaching cells has varying effect on the phosphorylation state of proteins
- Treating with Phorbol 12-myristate 13-acetate (PMA) as positive control for P38 showed upregulation of Phosphorylation for Erk and downregulation of phosphorylation for AKT while P38 phosphorylation could not be detected
- AKT phosphorylation due to adhesion to fibronectin is reduced to 20% compared to adhesion to serum.
- MDA-MB 468 cells express 6x times of αvβ6 compared to MCF7, which is known to be expressed in specific physiological conditions.
- MDA-MB 231 solely expresses Integrin-β2, which is leukocyte-specific integrin

Conclusion & Future Research

We were able to determine the integrin expression profile for the 3 breast cancer cell lines. In addition, we obtained a preliminary data that suggest possible effect of adherence states (adhering, non-adhering, adhering to various ECMs) on the phosphorylation state of AKT and Erk.
Subsequent experiments will look into links between specific integrins and these key signaling kinases and troubleshoot for P38 by seeding higher number of cell for the assay.

Acknowledgements

I would like to thank to Kareem Hassan and Balasubramanian Moovarkumudalvan for providing guidance in lab and Professor Ihab Younis for providing some insight on P38 activation. Similar thanks extend to Maya Kemaldean, Maria Navarro and Maria Bernales for providing resources for the lab

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MEETING OF THE Minds Undergraduate Research Symposium

Carnegie Mellon University Qatar

Molecular tools for microbial viability assessment in environmental samples: Case study of ballast water

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Advisor
Basem Shomar, Qatar Environment and Energy Research Institute
Annette Vincent

Category
Biological Sciences

Abstract
16S ribosomal RNA gene sequences are used in the study of bacterial phylogeny and taxonomy and is found in almost all bacterial species. It is specifically used as housekeeping genetic marker or as loading control for the following reasons: (I) the presence of 16S rRNA sequence in most of bacterial species as multigene family or as operons (II) overtime and despite of evolution, the function of 16S rRNA has been conserved, therefore, random changes in 16S rRNA sequence can be used as an accurate measurement of evolution (III) the size of 16S rRNA gene sequence, which is 1500 basepairs, is large enough to be used for informatics and sequences purposes [2]. 16S rRNA gene sequence is made up of nine different variable and hypervariable regions in addition to multiple conserved regions [1]. The variable and hypervariable regions express considerable sequence diversity among different bacteria, thus, act as finger prints for different bacterial species [1]. Therefore, species-specific sequences within a given hypervariable region constitute useful targets for diagnostic assays and other scientific investigations [1].

Molecular Tools for Microbial Viability Assessment in Environmental Samples: Case Study of Ballast Water

Kawthar Al-Sadat[1] , Basem Shomar [2] and Annette Vincent[1]

[1]Biological Sciences Program, Carnegie Mellon University Qatar; [2] Qatar Energy and Environment Institute (QEERI)

Introduction

16S ribosomal RNA gene sequences are used in the study of bacterial phylogeny and taxonomy and is found in almost all bacterial species. It is specifically used as housekeeping genetic marker or as loading control for the following reasons: (I) the presence of 16S rRNA sequence in most of bacterial species as multigene family or as operons (II) overtime and despite of evolution, the function of 16S rRNA has been conserved, therefore, random changes in 16S rRNA sequence can be used as an accurate measurement of evolution (III) the size of 16S rRNA gene sequence, which is 1500 basepairs, is large enough to be used for informatics and sequences purposes [2]. 16S rRNA gene sequence is made up of nine different variable and hypervariable regions in addition to multiple conserved regions [1]. The variable and hypervariable regions express considerable sequence diversity among different bacteria, thus, act as finger prints for different bacterial species [1]. Therefore, species-specific sequences within a given hypervariable region constitute useful targets for diagnostic assays and other scientific investigations [1].

Gap

Lack of studies on the bacterial viability in specific water samples and the effect of the use of RNA as the source of information on the accuracy of the results.

Aim

Study bacterial populations and their viability in ballast water samples and assess the efficiency of results obtained from RNA analysis as a different information source.

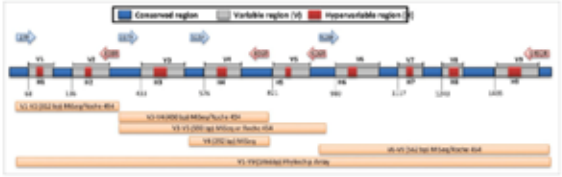

Figure 1: A diagram of the 16S rRNA sequence showing the conserved, variable, and hypervariable regions of the sequence [1].

Method

Four main techniques were used in this experiment: Live/Dead assay, RNA extraction, reverse transcription, and real time Polymerase Chain Reaction (PCR). Prior to performing RNA extraction, water samples were spiked with samples of *Escherichia coli* C600 at four different dilutions 25, 50, 100, and 200-fold and were filtered using a 0.22 μm sterile membrane. The filter papers were stored at -80ºC. Similarly for ballast water samples.

Live/Dead assay was used to estimate the number of live and dead cells in the sample. RNA extraction was performed using the RNeasy PowerWater Kit (Qiagen). The extracted RNA was used to perform reverse transcription using the ProtoScript® II Reverse Transcriptase kit, (NEB). This resulted in the production of cDNA from the extracted RNA. qPCR was used to plot the standard curve and determine the efficiency of the designed primers used through the melting curve analysis. This was applied to ballast water samples.

Results

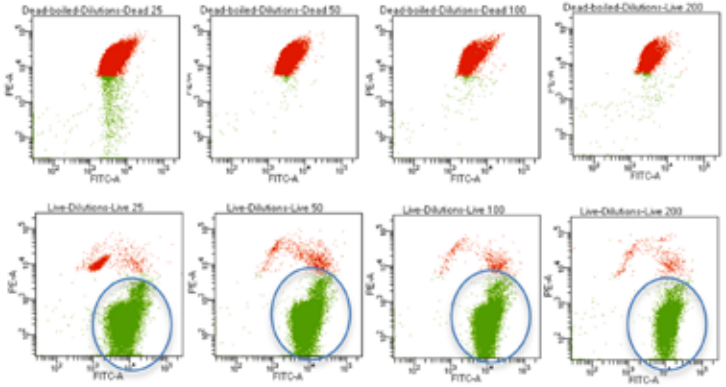

Figure 2: Live/Dead BacLight™ Bacterial Viability assay. The assay distinguishes and quantitate live and dead bacteria with the aid of a flow cytometer. A mixture of two nucleic acid stains: green-fluorescent SYTO™ 9 dye and red-fluorescent propidium iodide are used for viability determination. Dead cells show increased PI staining as seen in the increased red population in boiled samples; viable cells stain green and reduced green population as samples are more diluted (circle).

References

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[2] Patel, J. B. (2001). 16S rRNA gene sequencing for bacterial pathogen identification in the clinical laboratory. *Molecular Diagnosis*, *6*(4), 313-321.

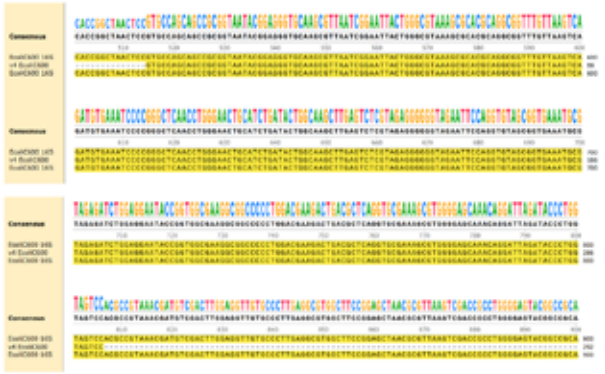


Figure 2: Alignment using MUSCLE. RNA extracted from spiked water samples were reverse transcribed to cDNA and the v4 region of the 16S was amplified using the 515F and 806R primers. Amplicons were sequenced using the MiSeq Illumna 150. Sequences were aligned to *Escherichia coli* C600 16S rDNA (NCBI) using Muscle. 100% identity to the v4 region was obtained.

Primer3 Output

```
No mispriming library specified
Using 1-based sequence positions
OLIGO            start  len      tm    gc%   any   3' seq
LEFT PRIMER          6   18   62.25   61.11  6.00  3.00 AGCAGCCGCGGTAATACG
RIGHT PRIMER        58   20   59.43   45.00  6.00  3.00 TACGGCCAGTAATTCCGATT
SEQUENCE SIZE: 61
INCLUDED REGION SIZE: 61

PRODUCT SIZE: 53, PAIR ANY COMPL: 5.00, PAIR 3' COMPL: 3.00

     1 GTGCCAGCAGCCGCGGTAATACGCAGGGTGCAAGCGTTAATCGGAATTACTGGGCGTAAA
       ||||||||||||||||||||                  ||||||||||||||||||||

  61 G
```

Figure 3: Primer designed for qPCR which binds to conserved region outside the v4 hypervariable region

Conclusion

RNA is a better measure of viable microrganisms in water samples both for accurate identification and quantitation.

Acknowledgements

I would like to show my gratitude to Professor Annette Vincent for her guidance throughout this research project. I would also like to thank Professor Ihab Younis, Professor Mohammad Bouaouina, Maria Ali, Maya Kemaldean, and Bernadette Bernales for their assistance

Metagenomic analysis of DNA and RNA profiles in ballast water

Author
Najlaa Al-Thani

Advisors
Annette Vincent

Category
Biological Sciences

Abstract

Ballast water is fresh or salt water, sometimes containing sediments, held in tanks and cargo holds of ships to increase stability and maneuverability during transit. Cargo ships then proceed with the discharge of ballast water stored from their initial port into the waters of the final or destination port. This ballast water contains a variety of bacterial species, enriching the local bacterial profile within the surrounding aquatic environment with possible foreign and harmful bacteria. Bacterial species can be identified based on the analysis of variable regions within the conserved 16S rRNA sequences found in all bacterial species.

In this research project, bacterial species within ballast water samples are analyzed based on the study of variable regions within the 16S rRNA sequences within bacterial DNA and RNA profiles. Initially, bacterial DNA and RNA from ballast water samples collected by Qatar Petroleum are extracted. Following extraction of bacterial DNA and RNA, reverse transcription will be used to convert the extracted RNA to cDNA and along which will be used for sequencing and. Using PCR, extracted DNA and RNA will be used to create 16S rRNA libraries by amplifying the variable regions within the 16S rRNA sequences. The results will be used to identify bacterial populations and correlate the evidence from both nucleic acid libraries available on water samples to compare between DNA and RNA profiles within the ballast water. It is crucial to study the viability of bacterial species in ballast water samples since it may alter the quality of water in the environment. Furthermore, bacterial species can affect the viability of other species in water and equipment installed underwater such as underwater oil pipelines through biocorrosion. Finally, it is important to be able to recognize the bacterial species that are being relocated from one region to another through tanks or cargo holds of ships.

Metagenomic Analysis of DNA and RNA profiles in Ballast Water

Najlaa Al-Thani
Biological Sciences Department, Carnegie Mellon University Qatar

MEETING of the Minds Undergraduate Research Symposium

Carnegie Mellon University Qatar

Abstract

Ballast water is fresh or salt water, sometimes containing sediments, held in tanks and cargo holds of ships to increase stability and maneuverability during transit. Cargo ships then proceed with the discharge of ballast water stored from their initial port into the waters of the final or destination port. This ballast water contains a variety of bacterial species, enriching the local bacterial profile within the surrounding aquatic environment with possible foreign and harmful bacteria. Bacterial species can be identified based on the analysis of variable regions within the conserved 16S rRNA sequences found in all bacterial species. In this research project, bacterial species within ballast water samples are analyzed based on the study of variable regions within the 16S rRNA sequences within bacterial DNA and RNA profiles. Initially, bacterial DNA and RNA from ballast water samples collected by Qatar Petroleum are extracted. Following extraction of bacterial DNA and RNA, reverse transcription will be used to convert the extracted RNA to cDNA and along which will be used for sequencing and. Using PCR, extracted DNA and RNA will be used to create 16S rRNA libraries by amplifying the variable regions within the 16S rRNA sequences. The results will be used to identify bacterial populations and correlate the evidence from both nucleic acid libraries available on water samples to compare between DNA and RNA profiles within the ballast water. It is crucial to study the viability of bacterial species in ballast water samples since it may alter the quality of water in the environment. Furthermore, bacterial species can affect the viability of other species in water and equipment installed underwater such as underwater oil pipelines through biocorrosion. Finally, it is important to be able to recognize the bacterial species that are being relocated from one region to another through tanks or cargo holds of ships.

Introduction

Ballast water is fresh or salt water, sometimes containing sediments, held in tanks and cargo holds of ships to increase stability and maneuverability during transit. Cargo ships then proceed with the discharge of ballast water containing a variety of bacterial species into the ocean, enriching the local bacterial profile within the surrounding aquatic environment. In general, bacterial genetic profiles contain several conserved regions, such as the 16s ribosomal RNA, that allows for the the study of bacterial phylogeny and taxonomy as the 16S ribosomal RNA is usually found in almost all bacterial species. 16S rRNA sequences are specifically used as housekeeping genetic markers or as loading control. As for the V4 region within the 16S ribosomal RNA, this region is a semi-conserved hypervariable region that allows for the identification of specific bacterial species that contain their own unique v4 sequence. By targeting these two regions within bacterial specifies found in Ballast water, an overview of the proliferation and transcription is obtained based on the data obtained from targeting the 16S ribosomal region whereas an indication of the type of bacterial species present in the sample is obtained through the data obtained from targeting the V4 region.

Methods

Sample Collection

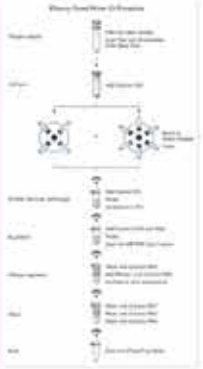
Ballast Water samples were collected by Maersk Oil Company from a variety of vessels. Prior to extractions, samples were filtered.

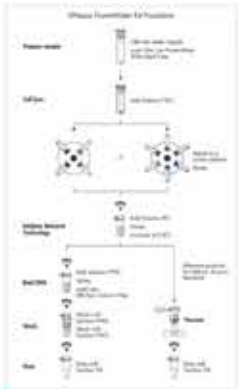
Extraction

Followed DNeasy® and RNeasy® PowerWater® Kit User Guides:

RNeasy® PowerWater® Kit (Cat No./ID: 14700-50-NF)

DNeasy® PowerWater® Kit (Cat No./ID: 14900-100-NF)





Conversion to cDNA (only for RNA extracts)

Followed Protoscript® II RT-PCR Kit User guide :
Protoscript® II RT-PCR Kit (#E6400S)

Metagenomic Analysis

Ion 16S™ Metagenomics Kit User Guide:
Ion 16S™ Metagenomics Kit (Cat. No. A26216)
Ion Plus Fragment Library Kit (Cat No. 4471252)

Results

UAE Analysis

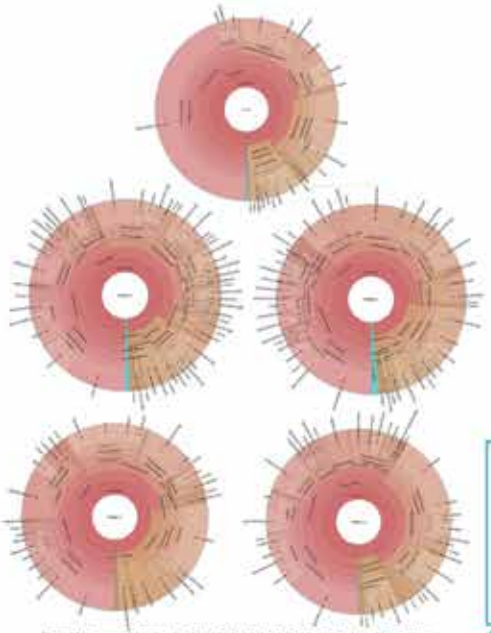


Figure (1): Sequencing DNA (S1 v1) and RNA (P42 v1, P46 v1, N7 v1, P48 v1) analysis of Ballast Water samples originating from UAE ports showcasing common species of samples.

Arabian Sea Analysis

Figure (3): Sequencing DNA (S1_v1) and RNA (P45_v1, P44_v1) analysis of Ballast Water samples originating from ports located in the Arabian Sea showcasing common species of samples.

Japan Analysis

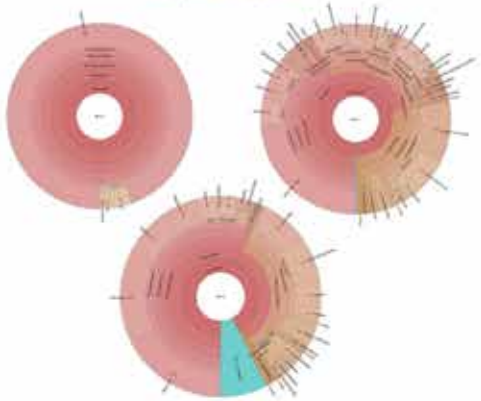


Figure (2): Sequencing DNA (T8 v1, S3 v1, S4 v1) analysis of Ballast Water samples originating from UAE ports showcasing common species of samples.

Discussion

Based on Figure (1), most bacterial species populating UAE waters that are at risk of infecting the Qatar oceanic environment are proteobacteria. Based on Figure (2), similar bacterial profiles can be seen for Arabian Sea bacterial species as proteobacteria also seems to populate this region which is expected due to the close regions. Based on Figure (3), an unmapped bacterial species can be seen in sample 78 from the Japan ballast waters suggesting a possible harmful bacterial species.

Acknowledgements

I would like to show my gratitude to Professor Annette Vincent for her guidance throughout this research project. I would also like to thank Maya Kemaldeen, Maria Ali and Bernadette Bemales for their assistance during lab work. Finally, I am grateful for Carnegie Mellon University Qatar for providing me with the faculties required to carry out my research.

References

[1] Chakravorty, S., Helb, D., Burday, M., Connell, N., & Alland, D. (2007). A detailed analysis of 16S ribosomal RNA gene segments for the diagnosis of pathogenic bacteria. Journal of microbiological methods, 69(2), 330-339.
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Role of kindlin-2 in breast cancer cell adhesion and migration

Author
Sayeda Sakina Amir

Advisor
Mohamed Bouaoina

Category
Biological Sciences

Abstract

Integrins are a family of receptors that mediate cell adhesion and migration and kindlin-2 is a ubiquitous regulator of integrin expression and activation in cells. When activated by kindlin-2, integrins can be used by the cell to adhere and migrate. Clinical research has shown that there is a positive correlation between the amount of kindlin-2 in breast cancer cells and cancer aggressiveness and lethality. Previous studies have also linked kindlin-2 to cancer cells invasion and metastasis, both of which are integrin-mediated processes. Therefore, the role that kindlin-2 plays in integrin-mediated MDAMB468 cancer cells adhesion and migration was investigated. To do this, we determined which integrins breast cancer cells use during adhesion and migration, and what effects acute depletion of kindlin-2 would have in these integrin function and breast cancer cells survival. Recent studies have also linked kindlin-2 to the process of EMT (Epithelial-to-Mesenchymal) such that reducing kindlin-2 levels inhibits the expression of key markers of the EMT process. Thus, we assessed whether kindlin-2 upregulation is sufficient for the expression of EMT markers and the increase in cancer cell invasiveness.

We also investigated whether kindlin-2 involvement in EMT is depending on its ability to bind and therefore activate integrins. We found that MDAMB-468 cells mainly rely on αvβ3 and αvβ5 integrins to adhere in-vitro. We confirmed that MDAMB468 cell migration is inhibited when kindlin-2 is knocked down. Moreover, we show that kindlin-2 is sufficient to increase cancer cells invasiveness and for their upregulation of αvβ6 integrin and the down-regulation of E-cadherin, both of which are key EMT markers. Moreover, we demonstrate that kindlin-2 effects on EMT markers and cell motility require kindlin binding to integrin. Our data clearly establishes that kindlin-2 is a driver of EMT in breast cancer and that kindlin-2 uses integrins.

ROLE OF KINDLIN-2 IN BREAST CANCER CELL ADHESION AND MIGRATION

SAYEDA SAKINA AMIR, DR. MOHAMED BOUAOUINA
BIOLOGICAL SCIENCES PROGRAM, CARNEGIE MELLON UNIVERSITY QATAR

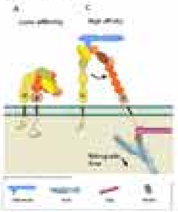
ABSTRACT

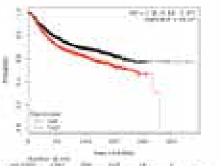
Integrins are a family of receptors that mediate cell adhesion and migration and Kindlin-2 is a regulator of integrin expression and activation in cells. When activated by kindlin-2, integrins can be used by the cell to adhere and migrate. Clinical research has shown that there is a positive correlation between the amount of kindlin-2 in breast cancer cells and cancer aggressiveness and lethality. Previous studies have also linked kindlin-2 to cancer cells invasion and metastasis, both of which are integrin-mediated processes. Therefore, the role that kindlin-2 plays in integrin-mediated cancer cells adhesion and migration was investigated. To do this, we determined which integrins breast cancer cells use during adhesion and migration, and what effects acute depletion of kindlin-2 would have in these integrin function and breast cancer cells survival. Recent studies have also linked kindlin-2 to the process of EMT (Epithelial-to-Mesenchymal) such that reducing kindlin-2 levels inhibits the expression of key markers of the EMT process. Thus, we assessed whether Kindlin2 upregulation is sufficient for the expression of EMT markers by performing Flow Cytometry and Western Blots. This protein is important to study because, if fully understood, kindlin-2 could be used as a potential target for breast cancer therapy.

We found that MDAMB-468 cells use integrins αvβ3 and αvβ5 to adhere. Cell migration is negatively affected when kindlin-2 is knocked down. Finally, kindlin-2 overexpression leads to upregulation of avb6 and down regulation of e-cadherin which corresponds to what happens during EMT.

INTRODUCTION

Integrins are a large family of αβ heterodimeric transmembrane receptors. They bind to the glycoproteins found in the extracellular matrix such as collagen, fibronectin or transmembrane proteins expressed by other cells and mediate adhesion and migration of the cells as well as cell survival, motility, and proliferation. Kindlins are adaptor proteins that bind to integrins. They bind to the C terminal region of ß integrin cytoplasmic tails and activate them[1]. Research has shown that cells lacking kindlin-2 were unable to activate their integrins[2]. Additionally, Kindlin-2 is shown to play a key role in integrin signaling that helps the cell regulate adhesion and spreading[2]. Thus, it is evident that Kindlin-2 is a critical component involved in integrin signaling and activation.





Interestingly, recent clinical studies have shown that there is a positive correlation between the amount of kindlin-2 and the aggressiveness and lethality of breast cancer cells[4]. Studies have also linked kindlin-2 to breast cancer metastasis[3] and claimed that the acute reduction of kindlin-2 in cells reduces their adhesion and disturbs their focal adhesion sites[2]. Inarguably, we can say that kindlin2 plays an important role in breast cancer cells adhesion and migration. To date, the specific integrin receptors involved in cancer cells adhesion and spreading are unknown.

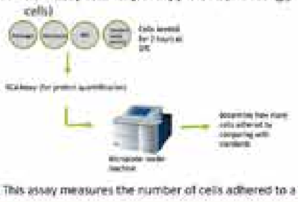
Previous research also states that EMT (Endothelial-to-Mesenchymal, a process in which cells lose their polarity and adhesion and become motile mesenchymal cells with more aggressive phenotype) correlates with more aggressive cancer. Recent studies also state that breast cancer cells lacking kindlin-2 showed a reduced expression of EMT markers.

Therefore, we were set which integrins cancer cells use to adhere and migrate and which role K2 plays in regulating these integrins. Then, we wanted to determine whether an overexpression of kindlin-2 in these cells will be sufficient to drive the expression of EMT markers like avb6 integrin and E-cadherins. This is critical because understanding the function of kindlin-2 will open doors for the discovery of alternate methods for breast cancer therapy.

METHODS

CELL ADHESION ASSAY

- Prepared four wells coated with Fibrinogen, Fibronectin, BSA, and Serum
- 30,000 cells seeded in plain culture media for 3 hours
- Cilengitide, a specific inhibitor of αvβ3 and αvβ5 integrins, was added to the wells in increasing concentrations
- BCA Assay used to quantify proteins (indicating presence of bound cells)

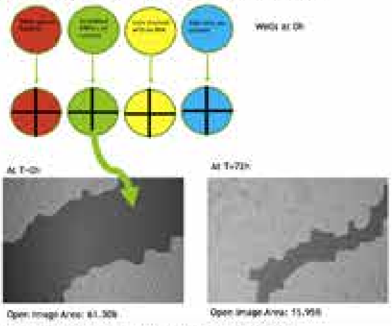


This assay measures the number of cells adhered to a surface.

METHODS

SCRATCH ASSAY

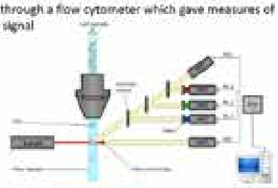
- Seeded cells that were electroshocked with short interfering RNAs Well surface was scratched 24 hours later
- Took images at each scratch region for the next 72 hours



This assay measures the rate of migration of the cells.

FLOW CYTOMETRY

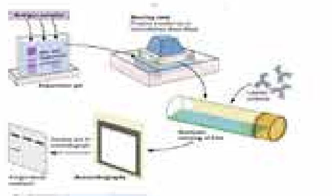
- Cells were stained with anti-avb6 and anti-e-catherin primary antibodies and Alexa 647 mouse secondary antibody
- Samples run through a flow cytometer which gave measures of fluorescence signal



This assay gives information regarding the properties of cells as well as the fluorescence emitted due to the anti-avb6/anti-e cadherin antibodies

WESTERN BLOTS

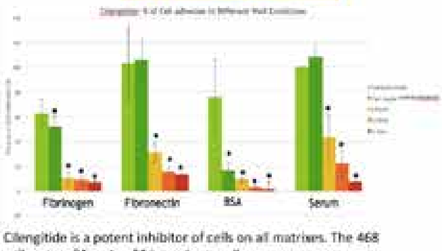
- Cells were lysed and the lysate was run on a SDS gel
- The gel was transferred onto a membrane which was incubated with a primary and secondary antibody and then imaged



Western Blots give information regarding the quantity of proteins

RESULTS

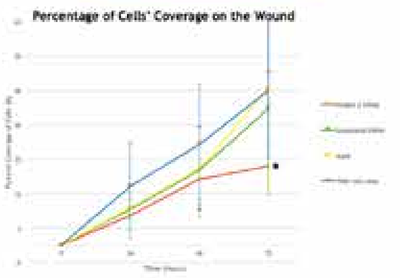
CELL ADHESION ASSAY



Cilengitide is a potent inhibitor of cells on all matrixes. The 468 cells use αvβ3 and αvβ5 integrins to adhere.

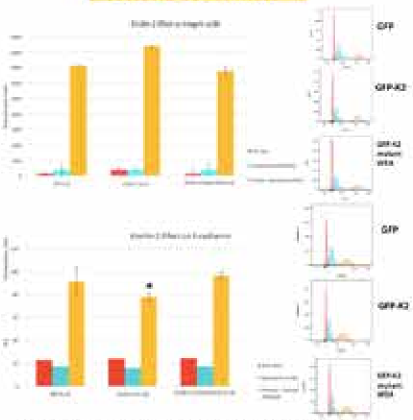
RESULTS

SCRATCH ASSAY



There is around 50% less migration in cells lacking 80% of their Kindlin-2 compared to cells that were transfected with the negative control Scrambled

FLOW CYTOMETRY



There seems to be an increase in the intergrin avb6 and a significant ($p<0.05$) decrease in E-cadherins in cells that have 20% more kindlin-2 than normal.

CONCLUSION

MDAMB-468 cells use integrins αvβ3 and αvβ5 to adhere. When kindlin-2 is knocked down from these cells, the cells migrate around 50% less. This implies that Kindlin-2 is essential for 468 cells' migration. Our data shows that kindlin-2 overexpression leads to upregulation of avb6 and down regulation of e-cadherin which corresponds to what happens during EMT. This proves that Kindlin-2 is sufficient to drive EMT in these MDAMB 468 cells and plays a huge role in breast cancer cells adhesion and migration.

ACKNOWLEDGEMENTS

I would like to thank Professor Mohamed Bouaouina for letting me work his lab. I would also like to thank my collegues Balasubramanian Moovarkumuldaivan and Kareem Hassan. I would also like to thank Maya Kemaldean, Maria Bernadette Bernales, and Maria Victoria for all their help on my project.

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MEETING of the Minds Undergraduate Research Symposium
Carnegie Mellon University Qatar

Role of P21 in the regulation of apoptosis in breast cancer tumor formation

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Category
Biological Sciences

Abstract

P21 is a cyclin-dependent kinase inhibitor (cdkn) gene which promotes cell cycle arrest in response to certain stimuli. This gene inhibits the cyclin-cyclin dependent kinase 2 & 4 and acts as a regulator for the G1 check point in the cell cycle. Previous research shows that the upregulation of CDKN1A in breast cancer tumors has caused an inhibitory effect on cell apoptosis. However, not much has been found to comprehensively explain this gene's function in tumors.

In this project, we studied the function of P21 in terms of its different splice forms, gene & protein structure, and homologs. We then looked into two of CDKN1A's protein partners (EGFR and ERBB2) which help in disrupting the cell cycle arrest function in breast cancer cells, along with CDKN1A. The data was obtained from public databases: NCBI/GenBank, mutation analysis from OMIM, networks from KEGG, and PDB protein structures. The analysis was performed using methods like BLAST, P21 network inference from CDKN1A, gene expression clustering, and modeling of the pathways with BioSystems. The project presents new insight on the details of the functionality of the role of P21 protein in the apoptosis regulation and role of EGFR and ERBB2 receptors in communication and signal transduction between breast cancer cells.

Role of P21 in Apoptosis Regulation of Breast Cancer Tumors

Sayeda Sakina Amir, Dr. Valentin Ilyin
Biological Sciences Program, Carnegie Mellon University Qatar

Abstract

P21 is a cyclin-dependent kinase inhibitor (cdkn) gene which promotes cell cycle arrest in response to certain stimuli. This gene inhibits the cyclin-cyclin dependent kinase 2 & 4 and acts as a regulator for the G1 check point in the cell cycle. Previous research shows that the upregulation of CDKN1A in breast cancer tumors has caused an inhibitory effect on cell apoptosis. However, not much has been found to *comprehensively* explain this gene's function in tumors. In this project, we studied the function of P21 in terms of its different splice forms, gene & protein structure, and homologs. We then looked into two of CDKN1A's protein partners (EGFR and ERBB2) which help in disrupting the cell cycle arrest function in breast cancer cells, along with CDKN1A. The data was obtained from public databases: NCBI/GenBank, GEO expression dataset, mutation analysis from OMIM, networks from KEGG, and PDB protein structures. The analysis was performed using methods like dynamic programming, BLAST, P21 network inference from CDKN1A, gene expression clustering, and modeling of the pathways with BioSystems.

Introduction

P21 (also known as Cdkn1a) is a cyclin-dependent kinase inhibitor gene found in humans. p21 is regulated by p53, a tumor suppressor protein that binds to DNA which then stimulates the production of p21 which inhibits the cyclin-cyclin dependent kinase 2 & 4 and acts as a regulator for the G1 check point in the cell cycle. The homolog of this gene can be found in house mice and is symbolized as **Cdkn1a**. The genes are conserved and thus, have the same function in humans as it does in mice. It is plausible that the gene variation can influence organism health because if the p21 gene is faulty, then the cell cycle arrest function would be compromised and could lead to the formation of tumor cells.

Previous research shows that breast cancer tumors contain higher expression levels of Cdkn1a, which is responsible for causing an inhibitory effect on cell apoptosis. Thus, we will be studying the role of P21 in apoptosis regulation in terms of the gene and protein structure. The project presents new insight on the details of the functionality of the role of P21 protein in the apoptosis regulation and role of EGFR and ERBB2 receptors in communication and signal transduction between breast cancer cells.

Methods

Information was obtained from these Public Databases:

- **NCBI/GenBank**
- **GEO Expression**
- **OMIM**
- **KEGG**
- **PDB**

Gene

There exist 5 splice variants in total and the gene itself is 10850 bases long. The gene coordinates on chromosome 6 are 2809...13658
The longest transcript is variant 3 and it's size is 2325bp.
The mRNA of variant 3 joins at coordinates
(78..111,1237..1474,7638..8087,9292..10880)

Figure 1: Gene information of p21 from NCBI.

Homologs

Species Name	Bit Score	% Identity
Mandrillus leucophaeus (drill)	376	94%
Pan troglodytes (chimpanzee)	333	100%
Pongo abelii (Sumatran orangutan)	323	98%
Macaca mulatta (Rhesus monkey)	318	97%
Rhinopithecus bieti (black snub-nosed monkey)	290	91%
Sus scrofa (Pig)	275	82%

The best match is to Mandrillus leucophaeus (drill)

Protein Analysis

This protein has only one domain, shown below.

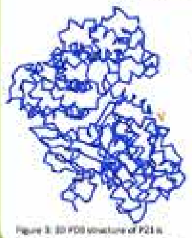

Figure 3: 3D PDB structure of P21 is shown here in blue interacting with its ligand SO4 in yellow and red.

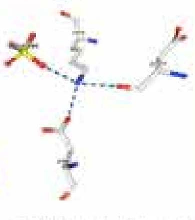

Figure 4: This image zoomed in shows the active site residues K270, D177, and D183 interacting with the ligand.

p21 Pathway in Breast Cancer

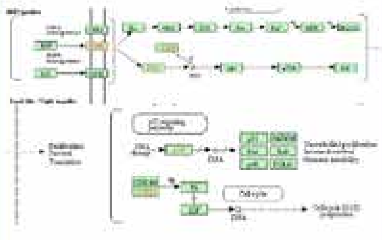

Figure 5: KEGG Pathway of p21 involving EGFR in Breast Cancer. The image on the bottom is a continuation of the one on top.

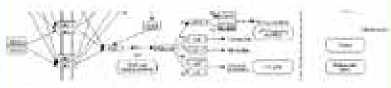

Figure 6: KEGG Pathway of p21 involving ErbB. Note, this pathway is not in Breast Cancer tumors, but it still shows the involvement of ErbB in the p21 pathway.

Linking P21 to Tumors

EGFR (also known as ErbB1) is a receptor whose ligands are cell signaling molecules that play a role in cell survival, differentiation, proliferation, and motility. Mutations in the EGFR will cause the production of abnormal EGFR proteins which will disrupt their role in signaling. Cells that sometimes have overexpressed EGFR will always upregulate the division of cells, causing tumors.

ErbB2 is a gene that encodes for a protein which binds to ligand-bound epidermal growth factor (EGF) family members, stabilizing them and enhances kinase-mediated activation of mitogen-activated protein kinases (MAPK) and phosphatidylinositol-3 kinase. This gene has two isoforms and the most common allele of this gene has Ile654/Ile655. Overexpression of this gene causes many cancers like lung, breast & ovarian tumors.

According to the data obtained from the different databases, p21 is a relatively small gene that plays a great role in cell cycle arrest. The p21 pathway from KEGG in Figure 5 shows that EGFR is linked to p21. EGFR, a surface receptor, regulates a cascade of events that lead to cell proliferation. This proliferation is then at stake for a higher probability for DNA damage, affecting p53 which then regulates p21, leading to reduced apoptosis.

As for ErbB, the KEGG pathway in Figure 6 shows that there is a relation between its signal and p21. Even though that pathway is not relevant to breast cancer tumors, recent evidence shows that p21 is a mediator of ErbB in breast cancer[3]. Both EGFR and ErbB communicate with p21 which then regulates apoptosis in breast cancer.

Acknowledgements

I would like to thank Professor Valentin Ilyin for teaching me how to use the different computational databases required to for this research.

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Carnegie Mellon University Qatar

PTEN gene encodes a ncRNA that acts as a potent tumor suppressor in breast cancer

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Category
Biological Sciences

Abstract
Breast cancer is the most common cancer in women worldwide with a high rate of mortality. In cancer, a group of genes called tumor suppressor genes are inactivated to ensure uncontrolled cell cycle progression. PTEN (Phosphatase and tensin homolog), a tumor suppressor gene, negatively regulates a pro-survival pathway, and hence is typically downregulated or deactivated in tumors via one of many known mechanisms, including mutations/deletion of PTEN gene or PTEN promoter methylation that inhibits its transcription. Another less explored mechanism of gene expression regulation is post-transcriptional, such as regulation of intron splicing. However, this mechanism is not explored for PTEN in breast cancer cells. We hypothesized that PTEN can be potentially regulated post-transcriptionally because the PTEN gene contains a minor intron. Minor introns are known to be highly regulated and tend to regulate the expression of the genes that harbor them. In this study, we explored the splicing efficiency of PTEN in MDAMB231 breast cancer cell line and its correlation with PTEN's loss of expression/function. We have shown that PTEN's minor intron is inefficiently spliced in breast cancer cells, leading to downregulation of PTEN protein expression. Interestingly, we showed that the unspliced PTEN minor intron expresses a separate gene product that has a significant effect on cancer cell growth. This study sheds the light on a novel mechanism for downregulating PTEN as well as provide a novel therapeutic target for breast cancer.

PTEN gene encodes a ncRNA that acts as a potent tumor suppressor in breast cancer

Aisha Fakhroo, Boshra Al-Sulaiti, Reem Elasad and Ihab Younis

Biological Sciences Program, Carnegie Mellon University in Qatar

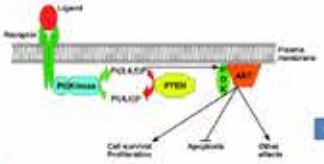
Abstract

Breast cancer is the most common cancer in women worldwide with a high rate of mortality. In cancer, a group of genes called tumor suppressor genes are inactivated to ensure uncontrolled cell cycle progression. PTEN (Phosphatase and tensin homolog), a tumor suppressor gene, negatively regulates a pro-survival pathway, and hence is typically downregulated or deactivated in tumors via one of many known mechanisms, including mutations/deletion of PTEN gene or PTEN promoter methylation that inhibits its transcription. Another less explored mechanism of gene expression regulation is post-transcriptional, such as regulation of intron splicing. However, this mechanism is not explored for PTEN in breast cancer cells. We hypothesized that PTEN can be potentially regulated post-transcriptionally because the PTEN gene contains a minor intron. Minor introns are known to be highly regulated and tend to regulate the expression of the genes that harbor them. In this study, we explored the splicing efficiency of PTEN in MDAMB231 breast cancer cell line and its correlation with PTEN's loss of expression/function. We have shown that PTEN's minor intron is inefficiently spliced in breast cancer cells, leading to to downregulation of PTEN protein expression. Interestingly, we showed that the unspliced PTEN minor intron expresses a separate gene product that has a significant effect on cancer cell growth. This study sheds the light on a novel mechanism for downregulating PTEN as well as provide a novel therapeutic target for breast cancer.

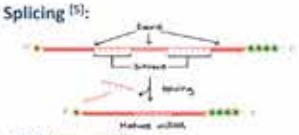
Background

- PTEN, a tumor suppressor, antagonizes AKT/PKB pathway [1].
- Splicing is a post-transcriptional regulation, where introns are removed to produce mature mRNA.
- Two types of introns, Minor and Major introns.
- ~700 minor introns, highly regulated [3].
- First intron of PTEN is a minor intron.

AKT/PKB Pathway [3]:



Splicing [5]:



PTEN pre-mRNA:

Aim

To study PTEN regulation at the splicing level, specifically its minor intron

Hypothesis

Significance

By understanding how a potent tumor suppressor gene, such as PTEN is regulated, we can provide a novel therapeutic target for breast cancer

Experimental Flow

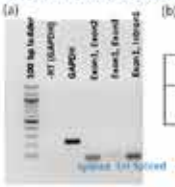
Primers Design

Downregulating PTEN's Expression

Downregulating PTEN's Activity

Results

Figure 1: PTEN is inefficiently spliced in breast cancer cells. (a) 1.5% midi agarose gel analysis of PTEN PCR products of MDAMB231 cDNA. (b) Splicing Ratio/Index of PTEN.



Splicing Ratio	0.76
Splicing Index	0.43

Figure 2: Transfection with PTEN AMO decreased PTEN gene expression. (a) Western Blot analysis of PTEN protein expression in MDAMB231 cells transfected with Control (50uM) and PTEN AMO (10,25 and 50uM). (b) 1.5% midi agarose gel analysis of PTEN PCR products of MDAMB231 cDNA.

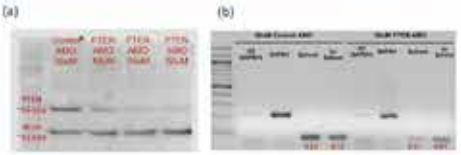


Figure 3: High doses of PTEN AMO reduced breast cancer cells viability. Growth of cells was measured using Cell Titer Glo Assay over 72 hours.

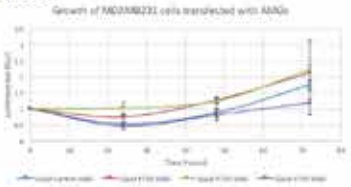


Figure 4.a: Un-spliced PTEN is a part of poly-adenylated segment. Expected fragments from nested PCR.

Figure 4.b: Un-spliced PTEN is a part of poly-adenylated segment. 1.5% midi agarose gel analysis of Nested PCR products.

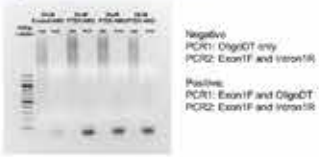
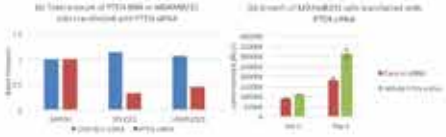


Figure 5: Downregulating PTEN increased MDAMB231 cells viability. (a) Total amount of PTEN RNA in MDAMB231 cells transfected with 300nM PTEN siRNA. (b) Growth of Cells following 48h.



Conclusion

This study provides a novel mechanism for downregulating PTEN. Also, it sheds the light on a a novel mechanism through which PTEN can act as a tumor suppressor. The un-spliced PTEN was found to be a part of a ncRNA, that decreased cancer cells viability at higher concentrations. Thus, providing a novel therapeutic target for breast cancer.

Future Work

- Overexpress PTEN ncRNA exogenously without affecting PTEN protein.
- Bioinformatics to study cleavage and polyadenylation sites (CPA) in intron1 and possible RNA binding protein (RBP) partners.

Acknowledgments/References

Nourhan Elbhaib, Bernadette Bernales, Maya Kenaldeen, Dr. Mohamed Bouazoune. This research was funded by CMUQ seed grant to Prof Ihab Younis.

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Expression and purification of dihydrofolate reductase

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Category
Biological Sciences

Abstract

Dihydrofolate reductase (DHFR) is an enzyme that is known to reduce dihydrofolic acid into tetrahydrofolic acid using NADPH as a source of hydride. The catalytic residues of DHFR have been well studied and characterized. The goal of this project is to express wild type dihydrofolate reductase (DHFR) and modified variants in BL21 cells to compare their enzymatic activities after purification. Specifically, a residue in the catalytic site of DHFR was modified (from arginine to glutamic acid) using site-directed mutagenesis and was found to reduce the enzymatic activity of DHFR. Techniques used in this project include: plasmid isolation, site-directed mutagenesis, transformation, induction of expression, protein purification, and measurement of enzymatic activity.

Expression and Purification of Dihydrofolate Reductase

Dona Ferdinando, Muhammad Nahin Khan and Annette Vincent
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Introduction

Dihydrofolate reductase converts dihydrofolate into tetrahydrofolate, a methyl group shuttle required for the *de novo* synthesis of purines, thymidylic acid, and certain amino acids. As DHFR plays an important role in folate metabolism and cancer treatment, changes in the level of DHFR expression can affect susceptibility to a variety of diseases.

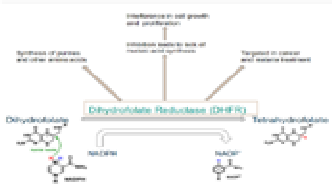


Figure 1: DHFR reaction mechanism and its function.

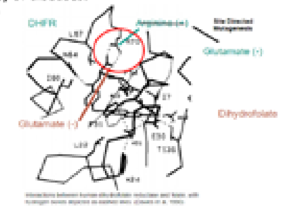


Figure 2: Mutation of DHFR resulted in Arginine → Glutamic acid using site-directed mutagenesis

Singh, A et al 2018, describe peptide based *hDHFR* inhibitor that wasdesigned on the basis of structural analysis of dihydrofolate reductase (DHFR) and tested against cancer cells. Such peptides that can occupy similar binding pocket in the *hDHFR* active site, which otherwise is occupied by folic acid/MTX and allows for the incorporation of suitable aromatic amino acids to counter the hydrophobic nature of the *hDHFR* active site.
Selectively targeting the *DHFR* active site may help to selectively target the cancer cells due to the higher concentration of the enzyme (i.e., higher expression of DHFR) than in the normal tissues.

Aim

To selectively replace amino acids (Arginine to Glutamic acid) within the catalytic site of mDHFR (DHFR in Mus musculus) using the site-directed mutagenesis and testing its effect on catalytic activity of mDHFR.

Methods

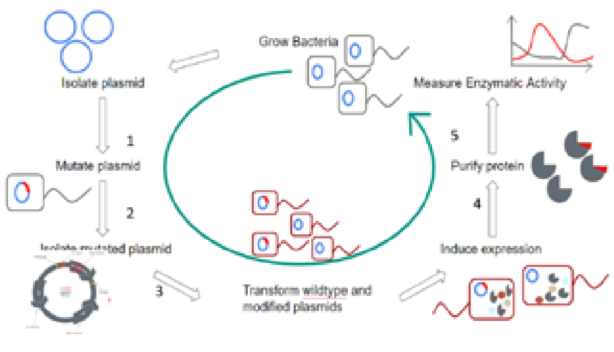
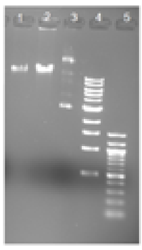


Figure 3: Flowchart of experimental strategy.

Results



Lane	Sample
1	1 kb ladder
2	100 bp ladder
3	Primer DHFR1
4	Primer DHFR2
5	1 kb ladder
6	100 bp ladder
7	No template (with DHFR1)
8	No template (with DHFR2

Figure 4: Isolation and site-directed mutagenesis of pDHFR

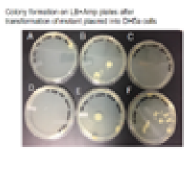


Plate	Contents
A	DHFR1 mutated plasmid (trial 2)
B	DHFR1 mutated plasmid (trial 2)
C	Wildtype plasmid (trial 1)
D	DHFR1 mutated plasmid (trial 2)
E	Wildtype plasmid (trial 2)
F	Wildtype plasmid (trial 2)

Lane	Sample
1	1 kb ladder
2	100 bp ladder
3	Midi prep
4	Mini prep

Figure 5: Transformation and isolation of mutated pDHFR into BL-21 DE3 cells

Figure 6: Sequence and Computational analysis of substrate-protein interaction

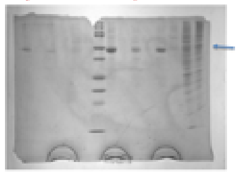


Figure 7: SDS-PAGE analysis of expression of DHFR protein (mutated and wild type) in BL-21 DE3 cells.

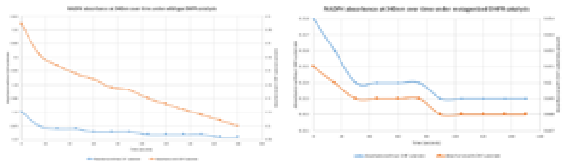


Figure 8: Enzymatic activity of wild type and mutant DHFR

Discussion

Computational analysis of mutagenesis of DHFR substrate binding site
Using foldX simulation software, we found changes in Gibbs free energy
Mutagenesis of pDHFR plasmid
Presence of a band after mutagenesis confirmed successful mutagenesis
Sequencing the wildtype and mutagenized plasmids and subsequent alignment reveals successful mutagenesis at nucleotide position 208 and 209 (from AG to GA)
Computational translation of the sequences further confirms the substitution of the 71st amino acid from arginine (R) to glutamic acid (E)
Induction and purification of wildtype and mutagenized DHFR proteins
BL21 DE3 pLysS cells transformed with wildtype pDHFR was found to be able to express DHFR within 4 hours. BL21 cells transformed with mutagenized pDHFR only expressed mutagenized DHFR after 22 hours.
Enzymatic activity assay of wildtype and mutagenized DHFR proteins
Wildtype DHFR activity was found to be 0.0140 μmolmin^{-1} mg^{-1}
Mutagenized DHFR activity was found to be reduced.

Future Directions

Mutate other sequences in regions outside the catalytic site

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MEETING OF THE Minds
Undergraduate Research Symposium

Carnegie Mellon University Qatar

Effect of EDTA on enzymatic activity of calf intestinal alkaline phosphatase

Authors
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Category
Biological Sciences

Abstract

Alkaline phosphatase (AP) is an essential enzyme found across many organisms including prokaryotes and eukaryotes, where it functions to break phosphate bonds from organic molecules with the aid of metal ion cofactors such as zinc ions. The purpose of this research was to characterize the kinetic properties of calf intestinal alkaline phosphatase (ALP) and to test the inhibitory effects of EDTA on its enzymatic activity. EDTA is a chelating agent that would bind zinc ions show the importance of zinc ion to AP activity.

Optimal enzyme concentration for ALP was found to be 0.01 U/ul based on tests across a range from 0.002 U/ul to 0.03 U/ul. The optimal substrate concentration was found to be 0.4 mM, testing across 0.05 mM to 1.0 mM. The Km and Vmax were determined to be 8.37 mM and 2.83 Abs/min respectively. Using the optimum experimentally determined concentrations, an enzyme assay was conducted using concentrations of EDTA ranging from 10-5 M to 0 M. EDTA was found to decrease the activity of ALP through allosteric cooperative inhibition.

Effect of EDTA on enzymatic activity of calf intestinal alkaline phosphatase

M. Nahin Khan Dona Ferdinando Dr Annette Vincent

Abstract

The hypothesis is that with increased concentration of EDTA, there will be a decrease in enzyme activity of calf intestinal ALP. In order to carry out this experiment an enzyme saturation curve was generated and a Michaelis-Menten plot was drawn in order to plot a Lineweaver-Burke. Km and Vmax was determined using this kinetic data.

Introduction

Alkaline phosphatase (AP) is an essential enzyme found across many organisms including prokaryotes and eukaryotes, where it functions to break phosphate bonds from organic molecules with the aid of metal ion cofactors such as zinc ions. In humans, it is expressed by several tissues of the body, with the largest amounts produced by the cells found in the liver and the bones. Overall, ALP can be found in liver, bones, intestines, pancreas and kidneys.

The purpose of this research was to characterize the kinetic properties of calf intestinal alkaline phosphatase (ALP) and to test the inhibitory effects of EDTA on its enzymatic activity. EDTA is a chelating agent that would bind zinc ions and hence show the importance of zinc ion to AP activity. We hypothesized that since ALP is a metalloenzyme and EDTA is a metal ion chelating agent, the presence of EDTA would reduce enzymatic activity of ALP.

Previous research on human intestinal alkaline phosphatase and human placental ALP showed different effects of EDTA on their enzymatic activity. Hence, we wanted to determine if calf intestinal ALP behaved similarly to human ALP or if it behaved similarly to placental ALP.

The hypothesis is that calf intestinal ALP will exhibit a decrease in enzymatic activity when subjected to EDTA, similar to human intestinal ALP.

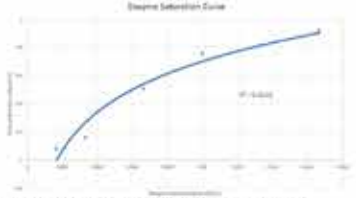
Methods and Results

1. **Enzyme Saturation Curve for Calf Intestinal Alkaline Phosphatase**

   What?
   - A set of standard enzymatic activity assays
   - Substrate concentration kept constant at 11.2mM
   - Enzyme concentration was varied from 0.002U/ul to 0.03U/ul

   Why?
   - Allows for determination of the optimal amount of enzyme to be used for further experiments
   - Too little = the rate of reaction is limited by the enzyme concentration instead of the substrate concentration
   - Too much = waste of the enzyme reagent and difficult to capture the fast rate of reaction on the spectrophotometer.

Enzyme Saturation Curve Result:

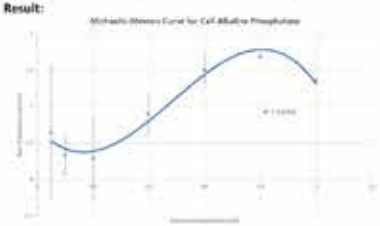


2. **Michaelis-Menten Curve for Calf Intestinal Alkaline Phosphatase**

   What?
   - A set of standard enzymatic activity assays
   - Enzyme concentration kept constant at 0.01U/ul
   - Substrate concentration was varied from 0.05mM to 1.0mM

   Why?
   - Allows for determination of kinetic properties of the enzyme, namely the Km (substrate binding affinity) and the Vmax (enzyme activity)
   - Allows for determination of the optimal substrate concentration to be used for determining the effect of inhibitor on enzyme activity

   Result:



3. **Testing the effect of EDTA concentration on enzymatic activity of Calf Intestinal Alkaline Phosphatase**

   What?
   - A set of standard enzymatic activity assays
   - Enzyme concentration kept constant at 0.01U.ul
   - Substrate concentration kept constant at 0.4mM
   - EDTA concentration varied from 10-5M to 0M

   Why?
   - Allows for determination of the effect of EDTA on the kinetic properties of the enzyme, namely the Km (substrate binding affinity) and the Vmax (enzyme activity)
   - Allows for determination of the type of inhibition that EDTA has on calf intestinal alkaline phosphatase (e.g. competitive, allosteric, etc.)

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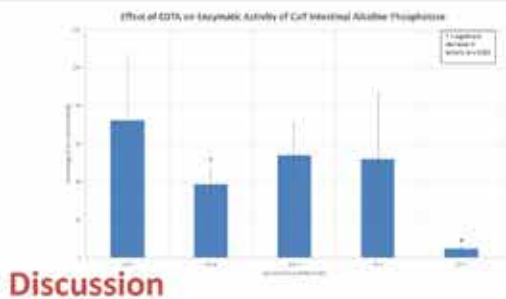

Discussion

From the results obtained from this project, three main conclusions were drawn:

1) The optimum enzyme concentration is 0.01U/ul

The enzyme saturation curve showed a hyperbolic relationship between enzyme concentration and rate of reaction. The purpose of generating this curve was to determine the optimum enzyme concentration to use in the enzyme assay with varying EDTA concentrations. From Figure 1 it is clear the the curve begins to reach a plateau at 0.01U/ul enzyme concentration and it was therefore chosen as the optimum concentration to use in future assays.

2) The optimum substrate concentration is 0.4mM

In order to determine the optimum concentration of substrate, varying substrate concentrations were tested with the enzyme concentration determined above. An S-shaped graph was obtained, from which the optimum substrate concentration was deemed to be 0.4mM. This means that alkaline phosphatase does not follow traditional Michaelis-Menton type of relation between its activity and substrate concentration; instead, the sigmoidal curve indicates a positive-cooperative allosteric relationship between the multiple substrate binding sites that the enzyme must have. Indeed, alkaline phosphatase is known to have multiple binding sites for its metal cofactors (Sharma, 2013)

3) The addition of EDTA causes a decrease in enzyme activity

After the optimum substrate concentration and enzyme concentration was determined, it was used in an assay with varying concentrations of EDTA. From Figure 3 we can coeclude that the addition of EDTA causes a decrease in enzyme activity.

Future Directions

There are multiple avenues for development in this project of which the immediate are:

1) Testing human placental ALP with varying concentrations of EDTA

This will allow for a comparison between placental and intestinal ALP which are from two different stages of development.

2) Testing pure E. Coli ALP with varying concentrations of EDTA

Literature values largely compare human placental with human intestinal however, none make the comparison between E. Coli and human ALP. We believe this will be an interesting area of research; to determine if E. Coli ALP behaves similarly to human placental or intestinal when subject to EDTA.

MEETING of the Minds Undergraduate Research Symposium

Carnegie Mellon University Qatar

The role of p38α kinase in regulating AUF1 binding to ATF3 transcripts in breast cancer

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Category
Biological Sciences

Abstract

ATF3 is a transcription factor that is overexpressed in many cancers and is involved in tumor progression. [1] Since cancer is the second leading cause of death worldwide [9], studying the factors which affect ATF3 expression levels is important. AUF1 binds to 3'UTR of ATF3 mRNA causing its destabilization. For example, upon amino acid deprivation, AUF1 is released from 3'UTR of ATF3 mRNA leading to its stabilization. [5] On the other hand, the stress activated protein kinase p38α is known to regulate ATF3 levels under persistent DNA damage. [7] Also, AUF1 binding to its target mRNAs has been shown to be regulated by p38α via an unknown mechanism. [8] We proposed that upon induction of stress, p38α kinase phosphorylates AUF1 reducing its binding to ATF3 transcripts and stabilizing ATF3 mRNA. This would lead to downstream effects such as increased reactive oxygen species levels which is a characteristic of cancer cells.[1] Our results show that ATF3 expression decreased upon induction of DNA damage with UV light and they suggest that this decrease is due to p38α activation. Our results indicate; however, that ATF3 expression increased upon induction of DNA damage with the topoisomerase II inhibitor, Doxorubicin. Also, ATF3 expression increased upon serum starvation. Thus, different methods of stress have different effects on ATF3 expression levels in cancer cells. Future work will investigate this difference on the molecular level to come up with a novel therapeutic approach that will specifically target ATF3 levels with minimal side effects.

The Role of p38α Kinase in Regulating AUF1 Binding to ATF3 Transcripts in Breast Cancer

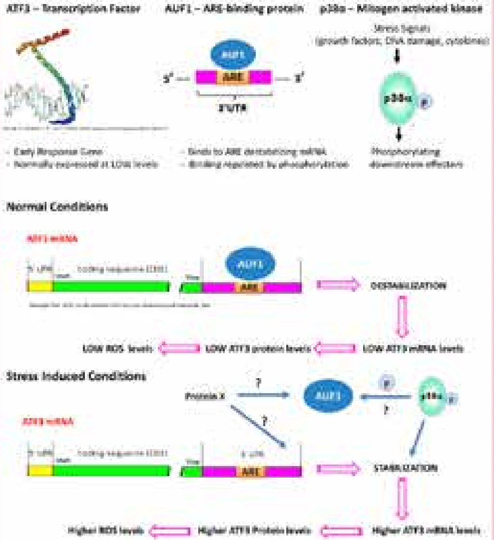
Aya Nour, Prof. Ihab Younis

Carnegie Mellon University, Biological Sciences Program, Qatar

ABSTRACT

ATF3 is a transcription factor that is overexpressed in many cancers and is involved in tumor progression.[1] Since cancer is the second leading cause of death worldwide [2], studying the factors which affect ATF3 expression levels is important. AUF1 binds to 3'UTR of ATF3 mRNA causing its destabilization. For example, upon amino acid deprivation, AUF1 is released from 3'UTR of ATF3 mRNA leading to its stabilization.[3] On the other hand, the stress activated protein kinase p38α is known to regulate ATF3 levels under persistent DNA damage.[7] Also, AUF1 binding to its target mRNAs has been shown to be regulated by p38α via an unknown mechanism.[8] We proposed that upon induction of stress, p38α kinase phosphorylates AUF1 reducing its binding to ATF3 transcripts and stabilizing ATF3 mRNA. This would lead to downstream effects such as increased reactive oxygen species levels which is a characteristic of cancer cells.[1] Our results show that ATF3 expression decreased upon induction of DNA damage with UV light and they suggest that this decrease is due to p38α activation. Our results indicate; however, that ATF3 expression increased upon induction of DNA damage with the topoisomerase II inhibitor, Doxorubicin. Also, ATF3 expression increased upon serum starvation. Thus, different methods of stress have different effects on ATF3 expression levels in cancer cells. Future work will investigate this difference on the molecular level to come up with a novel therapeutic approach that will specifically target ATF3 levels with minimal side effects.

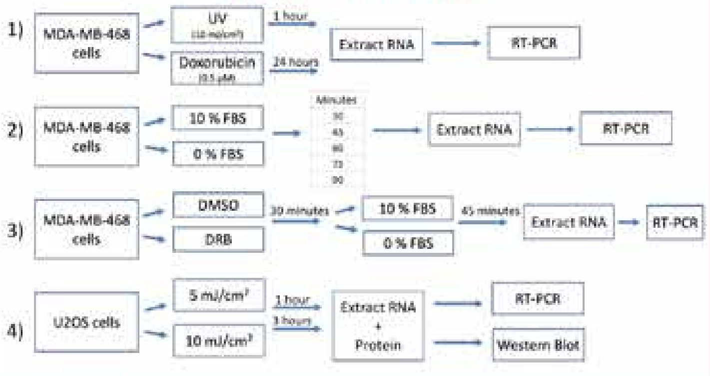
BACKGROUND



HYPOTHESIS : p38α kinase phosphorylates AUF1 (at Ser 83) thereby reducing its binding to ATF3 transcripts and stabilizing ATF3 mRNA.

SIGNIFICANCE: ATF3 levels matter because ATF3 is involved in tumor progression:
1) Involved in many pathways (p53, Wnt/β-catenin)
2) Reactive oxygen species production

EXPERIMENTAL DESIGN



RESULTS

Figure 1. ATF3 Expression in UV irradiated and Doxorubicin treated MDA-MB-468 cells. a, MDA-MB-468 cells were either irradiated with 10 mJ/cm² of UV light followed by a 1-hour incubation at 37°C or treated overnight with 0.5 μM DMSO or Doxorubicin before RNA was extracted. ATF3 3'UTR expression was measured with actin used as loading control. b, c, Band intensities were calculated using ImageJ software and normalized against actin band intensity.

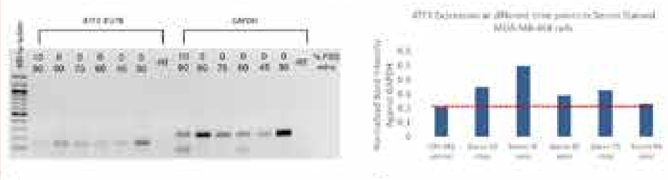


Figure 2. ATF3 Expression in untreated and serum starved MDA-MB-468 breast cancer cells. a, Serum Starved MDA-MB-468 cells were incubated in DMEM media with 0% FBS serum for 30, 45, 60, 75, and 90 minutes and untreated cells were incubated in DMEM media with 10% FBS for 90 minutes before RNA was extracted. ATF3 3'UTR expression was measured with GAPDH as loading control. b, Band intensities were calculated using ImageJ software and normalized against GAPDH band intensity.

Figure 3. ATF3 Stability in Serum Starved MDA-MB-468 breast cancer cells. a, MDA-MB-468 cells were treated with DRB or DMSO (negative control) for 30 minutes, then cells were serum starved in 0% FBS or incubated with 10% FBS for 45 minutes before RNA was extracted. ATF3 3'UTR expression was measured with actin as loading control. b, Band intensities were calculated using ImageJ software and normalized against actin band intensity.

Figure 4. ATF3 expression and p38α status in U2OS cells treated with varying doses of UV for different time points. U2OS cells were irradiated with 5mJ/cm² or 10 mJ/cm² and RNA and protein were extracted at 1 hour and 3 hours following UV irradiation. a, ATF3 3'UTR expression was measured with hu 18S as loading control. b,c, Band intensities were calculated using ImageJ software and normalized against hu 18S band intensity. d, Western blot of protein samples showing p38α, p-p38α, H2AX, and GAPDH expression.

CONCLUSIONS

- ❖ Different methods of stress have different effects on ATF3 expression levels in cancer cells.
- ❖ Knowing more about how ATF3 is regulated and the pathways involved can help us come up with a novel therapeutic approach that will specifically target ATF3 levels with minimal side effects.

FUTURE WORK

- ❖ Investigate the pathways activated when cells are stressed by DNA damage through Doxorubicin/UV
- ❖ Investigate effect of the two DNA damage methods on ATF3 mRNA stability
- ❖ Determine role of p38α in AUF1 phosphorylation and subsequent binding to ATF3 transcripts

ACKNOWLEDGEMENTS AND REFERENCES

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Meeting of the Minds Undergraduate Research Symposium

Carnegie Mellon University Qatar

Effect of aspartame on kinetics of calf intestinal alkaline phosphatase

Authors
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Category
Biological Sciences

Abstract

Alkaline phosphatase is an enzyme that catalyzes the hydrolysis of phosphate esters that are present in the extracellular space, making it essential for cell's growth. Aspartame is an artificial non-saccharide utilized as sugar in some food products. It was determined that with increasing concentration of aspartame, the activity of calf intestinal alkaline phosphatase decreases. Since aspartame is a non-competitive inhibitor, increasing aspartame concentration should only decrease the Vmax of calf intestinal alkaline phosphatase. According to a paper published by Chaudhri (2012), the expected Km and Vmax values of calf intestinal alkaline phosphatase are found to be 7.6 x 10-4M and 3.12 μmoles/min respectively using 50 mM Tris-HCl pH 11, while it is found to be 4.0 x 10-4M and 1.6 μmoles/min using 100mM glycine-NaOH at pH 9.5. Initially the optimum enzyme and substrate concentration was determined to prevent them from being limiting factors of the results obtained. Enzyme activity was analyzed by utilizing para-nitrophenylphosphate (p-NPP) as a substrate and buffer solution (0.1M Diethanolamine, 0.05mM $MgCl_2$ pH 9.8). The Vmax and Km values obtained have decreased to -0.78 and -0.085 respectively. The enzyme activity was also measured with the addition of different concentration of aspartame. The experimental result shows that the activity of CIAP is inversely proportional to with the concentration of aspartame.

The effect of aspartame on kinetics of calf intestinal alkaline phosphatase

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Abstract

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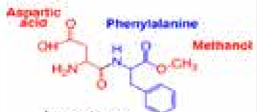
Introduction

Intestinal alkaline phosphatase (IAP) is an enzyme that plays an important role in maintaining gut homeostasis. A loss of IAP is associated with increased intestinal inflammation and dysbiosis — an imbalance of microorganisms[c]. Aspartame is an artificial sweetener that is used as a substitute for sugar in a sweet beverage. According to the research by Sarah S. Gul et al., the in vitro activity assay of IAP decreased in the presence of aspartame[e]. Sarah S. Gul et al. also showed that the activity of IAP in vivo decreased in the presence of aspartame by injecting aspartame into intestinal lumen of a mice[e]. Aspartame is likely to inhibit activity of IAP because Aspartame is composed of three molecules: L-aspartic acid, L-phenylalanine and methanol. L-phenylalanine is a known noncompetitive inhibitor of IAP that binds to both the enzyme and enzyme-substrate complex to slow down enzymatic activity as shown in figure 1[f]. Given that aspartame breaks down under basic environment, in pH 7.4 basic luminal space, aspartame is likely to breakdown into L-phenylalanine and inhibit the activity of IAP via noncompetitive inhibition.



Aspartame

In this experiment, the activity of calf intestinal alkaline phosphatase (CIAP) was measured using para-nitrophenyl phosphate(p-NPP) as a substrate to determine experimental Km and Vmax of CIAP. The experimental Km and Vmax value was compared with the literature Km and Vmax value of CIAP — 7.6 10^{-4}M and 3.12 units respectively[b]. The activity of CIAP was measured with different concentration of aspartame to find out the change in activity of CIAP in the presence of aspartame.

Methods

Determination of the Rate of Nitrophenol as function of Enzyme Concentration(ug/ul).

CIAP's optimum activity was measured by varying concentrations of the enzyme or substrate to determine the its Km and Vmax value from the initial velocity. Five varying concentration of enzyme – 3.33U, 6.67U, 8.33U, 13.3U- were tested to generate a Michaelis Menten plot to determine the optimum concentration of enzyme to utilize. Using UV Visible Spectrophotometer (Evolution™ 60S UV-Visible Spectrophotometer) at a wavelength of 410nm, 5 second interval for 3 minutes at temperature 37° Celsius with reaction buffer (0.1M Diethanolamine, 0.05mM MgCl2 pH 9.8) in a total volume of 300ul with the addition of 0.67M p-NPP to the reaction mix, which provides direct measure of dephosphorylation of p-NPP by the enzyme into nitrophenol which is absorbed at 410nm.

Similarly, five varying concentrations of substrate – 0.005mM, 0.011mM, 0.044mM, 0.055mM, 0.11mM – were tested with CIAP optimum enzyme concentration to generate a Michaelis Menten plot. Using UV Visible Spectrophotometer (Evolution™ 60S UV-Visible Spectrophotometer) at a wavelength of 410nm, 10 second interval for 3 minutes at temperature 37° Celsius with reaction buffer (0.1M Diethanolamine, 0.05mM MgCl2 pH 9.8) in a total volume of 300ul with the addition of 8.33U of enzyme to the reaction mix. One replicate is measured for each substrate.

Effect of amount of aspartame on enzyme activity.

The enzyme activity was measured with varying concentrations of aspartame – 0.000833mM, 0.00133mM, 0.002mM, 0.00266mM, and 0.003mM with CIAP's optimum enzyme concentration, 8.33U, and substrate concentration,0.055mM. Using UV Visible Spectrophotometer (Evolution™ 60S UV-Visible Spectrophotometer) at a wavelength of 410nm, 10 second interval for 3 minutes at temperature 37° Celsius with reaction buffer (0.1M Diethanolamine, 0.05mM MgCl2 pH 9.8) in a total volume of 300ul with the addition of the enzyme, substrate, and varied inhibitor. Two replicates were measured for each inhibitor concentration.

Data

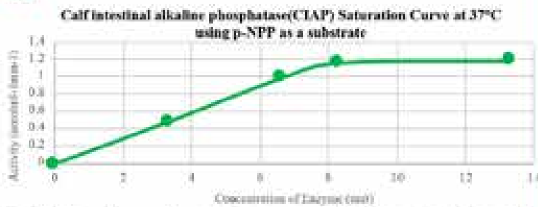


Fig.1. CIAP saturation curve. The activity of varying units of CIAP – 3.3units, 6.6units, 8.3units, 13.3 units – was measured with 5 ul of 0.67M p-NPP with reaction buffer(0.1M Diethanolamine, 0.05mM MgCl2 pH 9.8) at 37°C using UV Visible spectrophotometer at wavelength 410nm (n=1).

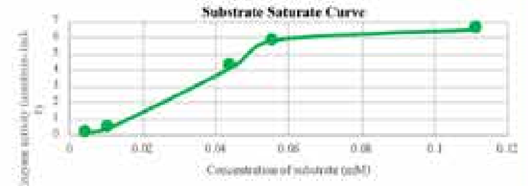


Fig. 2. Michaelis Menten plot. The activity of CIAP was measured with varying concentration of p-NPP — 0.05mM, 0.011mM, 0.044mM, 0.055mM, 0.11mM with reaction buffer (0.1M Diethanolamine, 0.05mM MgCl2 pH 9.8) at 37°C using UV Visible spectrophotometer at wavelength 410nm (n=1).

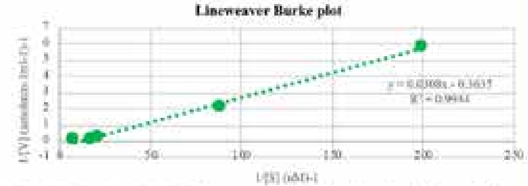


Fig. 3 Line Weaver Burk Plot. The Lineweaver Burk Plot is plotted by taking the inverse of substrate, p-NPP concentration and inverse of rate of activity of CIAP. The equation of the Lineweaver Burk plot y = 0.0308x - 0.3635 with R^2 value of 0.9944.

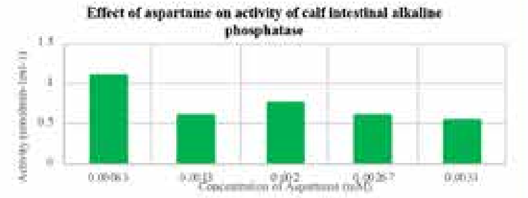


Fig. 4 CIAP activity assay with aspartame. The activity of CIAP was measured in with varying concentration of aspartame. The activity of CIAP was measured with p-NPP as a substrate and reaction buffer(0.1M Diethanolamine, 0.05mM MgCl2 pH 9.8) at 37°C using UV Visible spectrophotometer at wavelength 410nm (n=1).

Analysis and Discussion

Michaeli's Menten kinetics. In fig. 1 the enzyme saturation experiment determined that at 8.33 unit of CIAP reached maximum rate of enzyme activity. Using the unit derived from enzyme saturation experiment, the activity of CIAP was measured with varying concentration of p-NPP and reaction buffer (0.1M Diethanolamine, 0.05mM MgCl2 pH 9.8) kept at 37°C hot water tub. The measurement of enzyme activity was a sensitive task. Temperature of enzyme, degree of speed and precision at which an experimenter executed lab procedure were factors that affected the measurement of CIAP activity. In an effort to execute lab procedure consistently, experimenters were assigned with a role and repeated steps in high consistency. The activity of CIAP was measured once for each substrate concentration. The replicate of the enzyme activity should be measured to strengthen the validity of enzyme activity value and check the extent of deviation of measured activity from the mean.

Lineweaver Burk Plot. The Km and Vmax was calculated with the equation derived from Lineweaver Burk Plot as shown in figure 3. The equation is the following: y=0.0308x - 0.3635 with R^2 value of 0.9944. The experimental Km and Vmax are -0.084mM and -2.75 units respectively. The experimental Km and Vmax were not in accordance with the literature Km and Vmax values of CIAP — 7.6 10^{-4}M and 3.12 units respectively (Chaudhuri G. et al, 2013). The difference in experimental and literature value might have arose from number of factors: temperature of reaction buffer, temperature of CIAP and precision of lab technique. The CIAP is an endogenous enzyme that originates from the intestine of a calf. Under natural condition, CIAP catalyzes enzyme activity at 37 degree Celsius. According to the study by Chaudhuri et. al, the optimum temperature for the activity of CIAP is 45 degree Celsius. As such, the temperature is an important factor that affect the activity of CIAP. In an effort to mimic the natural condition of CIAP, the reaction buffer was kept at 37 degree Celsius and enzyme was kept at room temperature prior to the measurement of enzyme activity. A consistent lab technique was necessary so that every activity of CIAP have consistency. For instance, a time taken to measure enzyme activity after the addition of enzyme and how many pipet-stirs were made prior to the measurement of enzyme activity.

CIAP Activity Assay with Aspartame. With constant concentration of CIAP(8.33unit) and substrate ([p-NPP] = 0.06mM), the activity of CIAP was measured with five varying concentration of aspartame with reaction buffer(0.1M Diethanolamine, 0.05mM MgCl2 pH 9.8) as shown in figure 4. In figure 4, the highest CIAP activity (1.08 unitml^{-1} Standard Error +0.399) was measured when least amount of aspartame (0.00083mM) was present. The lowest CIAP activity (0.529 unitml^{-1}) was measured when most amount of aspartame (0.0033mM) was present. The overall trend was that the activity of CIAP decreases as concentration of aspartame increases. The inhibition of the activity of CIAP by aspartame was confirmed by figure 4. With the experimental result in figure 4, we cannot determine how aspartame inhibit the activity of CIAP — competitive or non-competitive. In the future experiment, the activity of CIAP can be measured with aspartame and varying concentration of substrate to determine the Km and Vmax.

Conclusion and Future Direction

The results obtained show proportional relationship between the activity of CIAP and the concentration of aspartame: a decrease in enzyme activity with the addition of enzyme inhibitor, aspartame. In order to determine further practical results of the effect of aspartame on alkaline phosphatase, the average uptake of aspartame concentration and the amount of alkaline phosphatase in the intestine must be taken into consideration.

The FDA acceptable daily intake (ADI) of aspartame is 50 milligrams per body weight[h]. The average body weight of adults in north America is 80.7 kg; thus, acceptable daily intake for an average adult in North America is 4.035g grams[a]. The range of alkaline phosphatase in normal serum is 20U/L-140U/L[i]. Assuming that the volume of small intestine is approximately 2 liters, the amount of alkaline phosphatase in small intestine is approximately 40U-280U. With the amount of intestinal alkaline phosphatase (IAP) present in human small intestine and ADI of aspartame, the activity of IAP can be assayed to analyze the effect of ADI of aspartame on the activity of IAP. Moreover, the activity of IAP cab be assayed with the amount of aspartame present in a commercial product such as diet coke to determine the effect of diet soda on the activity of IAP.

The result derived from the aforementioned experiments can possible reveal the extent of inhibition of aspartame on the activity of IAP. However, the results may not directly apply in a practical sense since the amount of IAP differ from people to people and the experimental results are *in vitro* assay.

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Assessing the catalytic activities of purified placental alkaline phosphatase and alkaline phosphatase from MDA.MB.231 cancer cell-line

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Category
Biological Sciences

Abstract
The goal of this project is to analyze and compare human placental alkaline phosphatase (AP) to AP derived from MDA.MB.231 cancer cell line. This was done by comparing the Vmax and Km of each isoform. In order to obtain the Km and Vmax, an enzyme saturation curve was generated to find the optimum amount of enzyme that would no longer become a limiting factor (saturation). This amount was then used to generate a substrate saturation curve, which varied the substrate concentration. A double-reciprocal plot, also known as a lineweaver-Burke Plot, was generated from the substrate saturation curve. Through this the Vmax and Km, which indicate efficiency/speed affinity of enzyme to substrate respectively, were obtained. We hypothesized that cancer AP would have higher speed and efficiency than that of placental AP, but would have the same affinity to substrate (higher Vmax value while the Km value remains constant. The Km and Vmax for placental AP were calculated to be 0.0314 mM and 1.71U/ml respectively. After lysing the cancer cells, the curves mentioned could not be generated. The Biorad Assay was then performed to quantitate the protein present in the sample, as well as a controlled enzyme activity assay of cancer AP. Through this, the amount of protein in the sample and activity of AP were obtained. In conclusion, assessing AP was unsuccessful as the amount of AP and the corresponding activity isolated from the cancer cells were minimal and not detectable.

Assessing the catalytic activities of purified placental alkaline phosphatase and MDA.MB.231 cancer cell line alkaline phosphatase

Reema Subeh, Zahra Al-Raisi and Annette Vincent
Biological Sciences Program, Carnegie Mellon University Qatar

Abstract

The goal of this project is to analyze and compare human placental alkaline phosphatase (AP) to AP derived from MDA.MB.231 cancer cell line. This was done by comparing the Vmax and Km of each isoform[a]. In order to obtain the Km and Vmax, an enzyme saturation curve was generated to find the optimum amount of enzyme that would no longer become a limiting factor (saturation)[b] . This amount was then used to generate a substrate saturation curve, which varied the substrate concentration[c]. A double-reciprocal plot, also known as a lineweaver-Burke Plot, was generated from the substrate saturation curve. Through this the Vmax and Km, which indicate efficiency/speed affinity of enzyme to substrate respectively, were obtained. We hypothesized that cancer AP would have higher speed and efficiency than that of placental AP, but would have the same affinity to substrate (higher Vmax value while the Km value remains constant)[a]. The Km and Vmax for placental AP were calculated to be 0.0314mM and 1.71U/ml respectively. After lysing the cancer cells, the curves mentioned could not be generated. The Biorad Assay was then performed to quantitate the protein present in the sample, as well as a controlled enzyme activity assay of cancer AP[d]. Through this, the amount of protein in the sample and activity of AP were obtained. In conclusion, assessing AP was unsuccessful as the amount of AP and the corresponding activity isolated from the cancer cells were minimal and not detectable[e].

Introduction

Alkaline phosphatase has an important role in disease. Recent research has proved that it can be used as a biomarker for certain diseases concerning the bone and liver. Recently, a test called the alkaline phosphatase level test has been used to test for such diseases, including certain cancers[a]. Moreover, there has been identified an important key role of alkaline phosphatase in cancers that involves its catalytic activity[a]. It has also been identified as a key factor that aids in tumor growth and metastasis of these cancers Recent research has also expanded on the possibility of using alkaline phosphatase as a predictor for breast cancer recurrence.

For this project, the catalytic activities of purified human placental alkaline phosphatase and alkaline phosphatase were compared. This is because research has shown that alkaline phosphatase can acquire new properties in disease that would enhance or inhibit its catalytic activities[a]. To compare the activities, the Km and Vmax values, which are indicative of affinity and speed/efficiency respectively, were obtained using the double reciprocal or lineweaver-Burke Plot that is generated from the substrate saturation curve. This provides a good medium for comparison as it only takes into account the catalytic activities[a]. The breast cancer cell line used was MDA.MB.231 cancer cell line[h].

Methods

First, an enzyme saturation curve was created by plotting the reaction rate of each sample in abs/min versus the enzyme concentration[a]. The enzyme used is NPP, which turns into yellow NP and phosphate upon interacting with AP. The enzyme concentration here was varied in order to find the minimum enzyme concentration that would lead to saturation in order to prevent the enzyme from being a limiting factor. This would control any limiting factors that might affect the Km and Vmax values[a]. The ideal concentration of enzyme was then used to generate a substrate saturation curve, where the substrate was now varied. From this graph, we obtained the reciprocal plot, and therefore the Km and Vmax values. The absorbance was measured at 410nm. This was only successful for placental AP[a].

As for cancer AP, first we lysed the cells using lysis buffer (mention what it contains) and glass homogenizer. The lysate was then used to create the substrate and enzyme curves but was unsuccessful. To ensure that the cells were lysed correctly, the Biorad assay was performed[d].
Using a stock of 2mg/ml of BSA, dilutions of concentrations [0, 50, 100, 150, 200, 300, 600 and 900] µg/ml were prepared. 0.1 ml of standards along with diluted Bradford reagent were added to the tubes. The tubes were vortexed and left to incubate at room temperature for 10 min. The samples were placed into cuvettes, and the absorbance was measured at 595nm using the ThermoScientific Evolution 60S UV-Visible Spectrophotometer[d]. Also, to test for AP activity, a controlled enzyme assay was performed. This was done by activity the enzyme substrate interaction at 37C for 30 min, then inhibiting it with 0.02 NaOH. The enzyme activity was then measured at 410nm[a].

Data and Results

Figure 1: Enzyme Saturation curve of varying placental alkaline phosphatase (PALP) concentration.

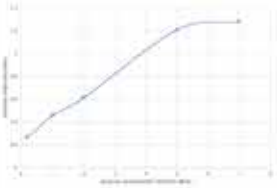

Figure 2: Substrate Saturation curve of varying p-nitrophenylphosphate (NPP) substrate concentration .

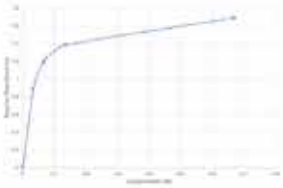

Figure 3: lineweaver-Burk Plot of the reciprocal of the substrate saturation curve (Figure 2)

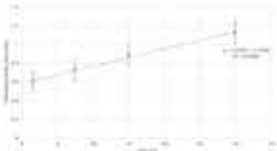

Table 1: The concentration of regular and cancer cell line AP calculated from the absorbance value at 595 nm

Sample	Absorbance	Concentration (µg/ml)
Placental Alkaline Phosphatase	0.875	673.33
Breast Cancer Lysate	0.790	604.17
Breast Cancer Pellet	0.190	104.17

Discussion

The optimum concentration activity (5U/µl) of purified placental alkaline phosphatase (PALP) enzyme was determined by varying its concentration and allowing its reaction with a constant concentration (0.67M) & volume of substrate (5µl) where it was the point at the start of plateau (figure 1). This is ideal because it means that the concentration of enzyme chosen cannot be the limiting factor in subsequent experiments[a]. The lineweaver-Burke plot was analyzed by varying the substrate concentration (0M, 0.0335M, 0.067M, 0.135M, 0.67M), in which the rate of enzyme activity was analyzed as the substrate concentration increased until the optimum concentration. The km value (affinity) of PALP was determined by the negative reciprocal of the x-intercept which was found to be 0.0314mM and Vmax (efficiency/speed) was determined by calculating the reciprocal of the y intercept the was calculated to be 1.71U/ml (figure 3). The R-squared value of the Lineweaver-Burk plot obtained is 0.9948, which indicates that the values obtained are accurate (figure 3).

Performing the same procedure of obtaining the Km and Vmax values for cancer AP were not successful, and this could possibly have been due to low cell count, unsuccessful lysis of the cancer cells or even presence of AP in very small amounts in the cancer cells. Other reasons could be due to presence of inhibitory molecules in the lysis buffer, such as EDTA, which is a well-known chelating agent which binds divalent cations and would greatly affect later experiments like the enzyme assay. To find out the reason, a series of standards [50, 100, 200, 300, 600, 900] µg/ml of BSA (Bovine Serum Albumin) were prepared and their absorbance plotted to get the equation of line y = 0.0012x + 0.065 and R-squared value 0.9795 (figure 4). The R-squared value was higher than 0.95, further strengthening the plot's validity. The concentration of the samples were then obtained from the plot to be 604.17µg/ml , which means the the cells were lysed, and the first experiments were unsuccessful due to low AP concentration or presence of EDTA[d].

After the controlled enzyme assay was performed with NPP, the abs/min values obtained were 0.06, 0.07 and 0.06. This means that there was AP present in the cancer cell lysate, but in very low amounts. This could also mean that the activity of AP was inhibited by the EDTA present in the lysis buffer, through EDTA binding divalent cations that are AP's cofactors[a] such as Mg^{2+} and Zn^{2+}.

Future Work

We would like to further purify and concentrate AP obtained from cancer cell line MDA.MB.231, in order to be able to plot the enzyme saturation, substrate saturation and lineweaver-Burk plots.

We would also like to test the effect of different inhibitors on the activity of alkaline phosphatase obtained from the cancer cells, as literature suggests that alkaline phosphatase in disease might have different inhibitors. This also applies to co-factors, and so we would like to test different cofactors.

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Near-optimal dynamic pricing strategies for selling limited inventory to rational customers

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Category
Business Administration

Abstract
This project studies the near-optimal dynamic pricing strategies for selling the limited inventory of a product over a finite horizon. Potential customers arrive stochastically over time and make rational purchasing decisions. Customers' willingness to pay is private information known only by the customer himself. The seller knows the distribution of valuations but not the actual realizations. An arriving customer only purchases if the current posted price is lower than the valuation derived from purchasing.

The goal is to devise dynamic pricing strategies that generate (near)-optimal expected revenues and have desirable computational properties.

The optimization problem of the seller is formulated as a continuous-time stochastic dynamic program, which does not admit a closed-form solution except in very special-form demand functions (cf. McAfee and Velde 2006, Gallego and van Ryzin 1994). Computational modeling and simulations are used to evaluate properties of the benchmark and proposed pricing policies.

Near-Optimal Dynamic Pricing Strategies for Selling Limited Inventory to Rational Customers

Shireen Ahmed I Fahad Bahzad I Abraham Farooqui
Professor Mustafa Akan, PhD

Abstract

This project studies the near-optimal dynamic pricing strategies for selling the limited inventory of a product over a finite horizon. Potential customers arrive stochastically over time and make rational purchasing decisions. Customers' willingness to pay is private information known only by the customer himself. The seller knows the distribution of valuations but not the actual realizations. An arriving customer only purchases if the current posted price is lower than the valuation derived from purchasing.

The goal is to devise dynamic pricing strategies that generate strong revenue performance and have desirable computational properties.

The optimization problem of the seller is formulated as a continuous-time stochastic dynamic program, which does not admit a closed-form solution except in very special-form demand functions (cf. McAfee and Velde 2006, Gallego and van Ryzin 1994). Instead we use a alternative solution paradigm and develop a self adjusting price policy based on decision rule approximation. Computational modeling and simulations are used to evaluate properties of the benchmark and proposed pricing policies.

Background

The two types of Revenue Management control are quantity based and price based. For this project, we focus on the dimension of price and how it should be managed to maximize revenues. Price has two main effects on the value function of the stochastic dynamic program:

1) Supply effect- since resources are limited a firm will look to increase (decrease) prices as supply falls (increases).
2) Demand effect- If not enough (or too many) people demand a product, the price of that product will fall (or rise) to maximize revenue.

With our model, we hope to replicate this effect without the heavy computational burden of re-optimization.

Business setting

- Single product in isolation with rational short-lived customers
- Demand is based solely on price and time, without competition and substitution impact
- Interarrival times are exponentially distributed with a mean of 2 hours (Poisson arrival process)

Method

We restrict the set of feasible price adjustments to a simple functional form, i.e., linear. The underlying probability space is kept intact.
We defined five variables which are: Capacity, Price, Revenue, % Hike, and % Depreciation as seen in Table 1. The reason we decided to use variables for these is so that we can alter them dynamically as the simulation is running and so that we can adjust them in Process Analyzer. For example, the price changes dynamically every time period and each time a customer decides to purchase and capacity is reduced by one.

Variable - Basic Process

	Name	Comment	Rows	Columns	Data Type	Clear Option	File Name	Initial Values	Report Statistics
1	Capacity				Real	System		1 rows	☐
2	Price				Real	System		1 rows	☐
3	Revenue				Real	System		0 rows	☐
4	PercentageHike				Real	System		1 rows	☐
5 ▸	PercentageDepreciation				Real	System		1 rows	☐

Table 1

Our model, as shown in Figure 1 starts with a node that creates customers based on a Poisson arrival rate with an inter-arrival time of 2 hours. Then a valuation is assigned to each customer based on an EXPO(25) distribution. Following, the model checks if we have enough capacity or not. If capacity is >= 1 it will allow the customer to pass and if not it will dispose of him/her. After passing the first decision node, there is another decision node that checks whether the assigned customer's valuation is >= the price and if it is the customer will pass on and if not the customer will be disposed. If both decision nodes are true, then we have an assign node that reduces capacity by 1, increases revenue by the price, and increases the price by a specified percentage hike. After that, the customer is disposed.

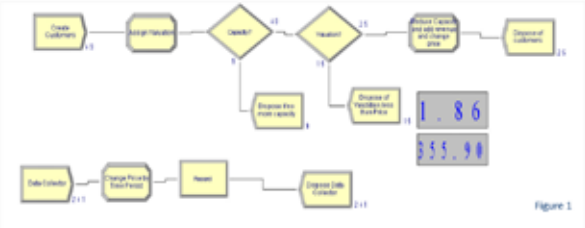

Figure 1

Experimental Design

- Run for 1 120-hour replication
- Total time period is 240 periods.
- Capacity is taken as 25
- Controls that will vary are x= Initial Price, y= Price depreciation percentage and z= price hike percentage x= 30, 50; y= 1%,3%; z= 5%,20%, as shown in Table 2

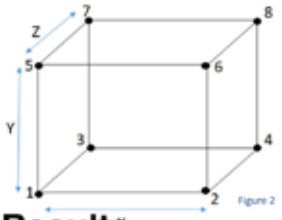

Figure 2

No.	X (Price)	Y(Depreciation)	Z (Hike)
1	30	1	5
2	50	1	5
3	30	1	20
4	50	1	20
5	30	3	5
6	50	3	5
7	30	3	20
8	50	3	20

Table 2

Results

After comparing the possible scenarios, we found that the one that generates the highest revenue is having a low depreciation percentage (1%), a high hike percentage (20%), and a high price ($50). The average revenue in this scenario with 5 replications totaled up to $585.18 and the average capacity left was 12.8, as seen in Table 3.
From the eight possible scenarios, we find that at a price of $50, Hike Percentage of 20% and Depreciation percentage of 1%, we get maximum revenue.

Depreciation Percentage	Hike Percentage	Price	Average Revenue	Average Capacity Left
1%	5%	30	537.94	0.2
1%	20%	50	585.184	12.8
3%	5%	50	248.844	0
3%	5%	30	218.218	0
1%	5%	50	580.073	2.8
1%	20%	30	624.306	10.4
3%	20%	50	534.333	0
3%	20%	30	523.463	0

Table 3

Figure 3.1

Figure 3.2

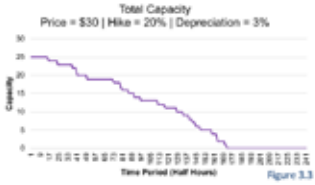

Figure 3.3

Compared to the base scenario of static, i.e., fixed price policy (where %hike and %depreciation are set to 0), dynamic pricing increases revenue by 11.8%.

Conclusion

Initial price has the smallest effect on expected cumulative revenue over the selling horizon for 1% decrease in initial price the revenue increased by $0.4 on average. Dynamic pricing is the stronger instrument for maximizing revenue. Specifically, increasing %hike by 1% leads to $11.7 more average revenue. And 1% point increase in depreciation reduces the revenue by $97.9 on average.

The effect on leftover inventory at the end of the selling season is also similar. Depreciation has a negative effect on capacity left and seems to be the most significant lever. Initial price and %hike increases capacity leftover.

This suggests that dynamic pricing is a powerful method to maximize revenue for sellers of limited capacity facing rational customers. In order to enjoy the benefits of dynamic pricing without overdoing it, firms should be less aggressive in discounts which may lead to stockouts and reduced revenue. Future work can extend the results to time-varying setting.

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MEETING OF THE Minds Undergraduate Research Symposium
Carnegie Mellon University Qatar

Supporting students development of self-authorship and reflective judgement

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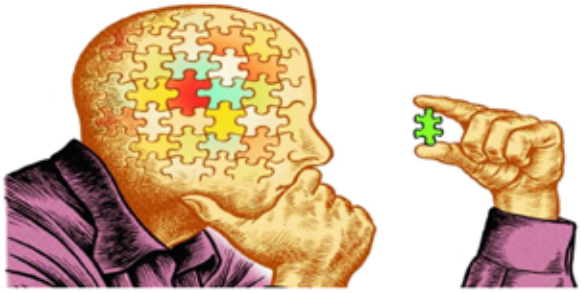
Category
Business Administration

Abstract

Our research aims to answer: Can learning support aid student performance on a classroom assessment technique aimed at developing self-authorship and reflective judgement? Self-authorship (Baxter-Magolda et al., 2010) and reflective judgement (King & Kitchener, 1994) enables students to take what they learn, make a personal critical judgment and apply this information in real life situations. Self-authorship and reflective judgment, together, was operationally defined as student's ability to make a claim, support it with a reason, and demonstrate self-reflection in their response to a classroom assessment technique called an Exit Ticket. Exit Tickets are slips of paper students submit at the end of each class with responses that meet three criteria: (a) provide a claim (critical, creative, or curious insight about a class concept) (b) support the claim with a reason (c) self-reflect on the claim/reason through personal application. A substantial response met all of the three criteria. We conducted an in classroom intervention study in the Organizational Behavior (70-311) class offered in CMU-Q during the Spring 2019 semester. Our intervention involved asking students to complete an Exit Ticket with scaffolding (i.e., a posted question) and students were provided with regular feedback. In order to assess whether or not there was any impact on student performance on the Exit Tickets we compared the scores of the Exit Tickets of the Fall 2018 (n = 28) semester, where no scaffolding was provided, with Spring 2019 (n = 21), where scaffolding was provided. We used information from the first half of each of the semesters to do this.

We compared the scores of the Exit Tickets across both semesters by using between-subject and within-subject designs. As we implemented our intervention in Spring 2019, we predicted that the Exit Ticket scores would be higher when compared to that of Fall 2018. Additionally we predicted that student performance would improve over time. The results supported our predictions that scaffolding aids student performance in an assessment targeted at developing self-authorship and reflective judgement.

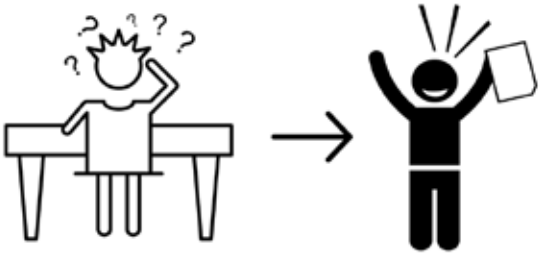


CAN STUDENTS LEARN TO THINK FOR THEMSELVES?

Zeina Darwiche, Cecile le Roux,
Marisella Rodriguez and Chad Hershock

WHY DO WE CARE?

Students can think independently by using self-authorship (Baxter-Magolda et al., 2010) and reflective judgment (King & Kitchener, 1994). This type of thinking supports students to take what they learn, make a personal critical judgement and apply this information in real life situations. Thereby, students can maximize their potential in various situational contexts.

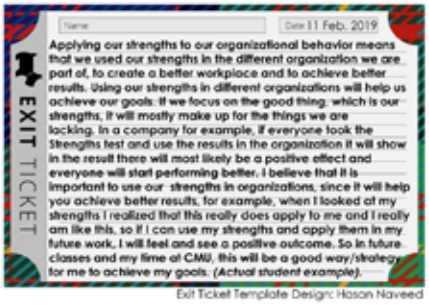


WHAT DID WE DO?

Each class, students in 70 311 completed an Exit Ticket: a classroom assessment of student learning submitted at the end of class. It was noticed that students in F18 performed better when completing an Exit Ticket with scaffolding (i.e., a posted question) as opposed to without scaffolding. In S19, this study explored the impact of a scaffolded Exit Ticket on student grades. It was predicted that students' Exit Ticket performance would improve with scaffolding as compared to no scaffolding. It was also predicted that student performance would improve over time.

	First Half of the Semester	Second Half of the Semester
Fall 2018	No Exit Ticket Question	Exit Ticket Question
Spring 2019	Exit Ticket Question	No Exit Ticket Question

THE INTERVENTION GUIDES STUDENTS TO DEVELOP THEIR OWN THOUGHTS



Exit Ticket Template Design: Hasan Naveed

WHAT DID WE FIND?

How well do students perform on exit tickets with and without scaffolding in the first half of the semester?

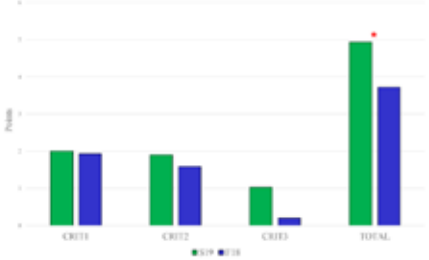


Figure 1. F18 vs. S19 All Exit Tickets: Rubric Components and Total Score

* Independent samples t-test

N	μ
F18 = 28	3.40
S19 = 21	4.74

$p < .05$ (2-tailed), $SD = 2.0$

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Data Analysis Support: Michael Melville

Does student performance change over time in the first half of the semester?

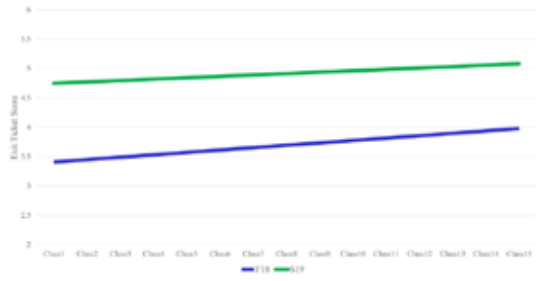


Figure 2. Change Trajectories for F18 vs. S19 Students (total score)

MEETING OF THE Minds
Undergraduate Research Symposium
Carnegie Mellon University Qatar

Two-sided matching with random utility and outside options

Authors
Anthony Lo, Fariza Shiyap, Xinyu Ma

Advisor
Mustafa Akan

Category
Business Administration

Abstract

Matching models are used to model a variety of economic also economic allocations, like workers to jobs, students to schools, tenants to dorm rooms, etc. The canonical model of one-to-one matching with strict preferences (Gale and Shapley 1962) includes two finite sets of agents on the two sides of the market. The complete list of preferences for all agents are known a priori. The famous Gale-Shapley Deferred Acceptance Algorithm is proven to result in a stable matching and terminates in a finite number of rounds. The goal of this project is to extend this algorithm to uncertain preferences.

Two Sided Matching with Random Utility

Anthony Lo Fariza Shiyap Xinyu Ma Advisor: Prof. Mustafa Akan

Motivation and Background

Matching models are used to model a variety of economic also economic allocations, like workers to jobs, students to schools, tenants to dorm rooms, etc.)

Preferences Gale and Shapley (1962) canonical model of one-to-one matching with strict preferences include two finite sets of agents on the two sides of the market.

The complete list of preferences for all agents is known a priori. The famous Gale-Shapley Deferred Acceptance Algorithm is proven to result in a stable matching and terminates in a finite number of rounds.

The goal of this project is to extend this algorithm to uncertain preferences.

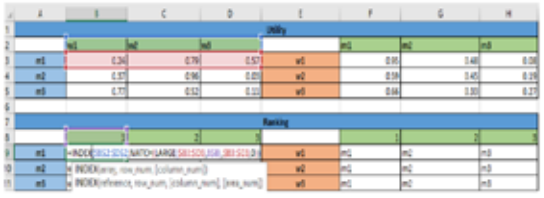
Spreadsheet Implementation of the Deferred Acceptance Algorithm

Matching is stable if it is

- Individually rational: for each x, x (weakly) prefers her match to being alone,
- Pairwise stable: no pair of men and women can block the matching:

Gale-Shapley algorithm:

- Each man has a list of of women from top to bottom.
- Each woman has a notebook with all the others she got.
- The algorithm through rounds,
- At the beginning of round 0
- Each man start with the complete list of preferences,
- Each woman starts with her own name in the notebook.



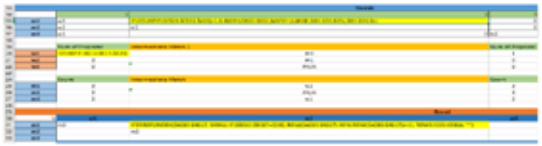
- Utility=Rand() or riskUniform(0.1,0.9)+risklognorm(0.1,0.5)
- As shown above we used the index, match, and large function to help us rank the ranking for both man and woman.

Deferred Acceptance Algorithm with Random Utility

Implement the Gale-Shapley Deferred Acceptance Algorithm on this model:

- 3 men and women
- U_{ij} = average utility of man i for matching with women j
- V_{ij} = average utility of women j for matching with men i
- δ_i = idiosyncratic utility shock of man I for each women type
- γ_i = idiosyncratic utility shock of man I for each women type
- δ_i, γ_i Are log-normally distributed

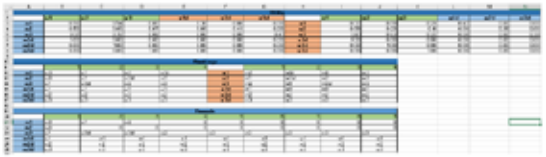
The utility of an individual i of type x for alternative y is xy +epsilony, where epsilony ~ PX



- In the first round, man proposes to woman with the highest utility.
- Use the countif function in the second table above to keep track of number of proposal.
- The last table keeps track of the number of proposals to a woman, and ranks them by woman's utility for the man.

Extension to Uncertain Outside Options

Implement the Gale-Shapley Deferred Acceptance Algorithm considering the outside options. (i.e, utility of remaining unmatched)



To incorporate outside options into the model, we artificially created agents on the other side of the marketplace who strictly prefer to match with a particular agent, giving him/her their outside option utilities. Running the deferred acceptance algorithm results in some agents being matched with their outside options, which are drawn from a Normal Distribution.

Consider the case that the outside options are correlated with someone's desirability in the marketplace

Draw outside options randomly from a Normal distribution with mean 0.5 and std. Dev. 0.2 where this variable is positively correlated with the average utility of all women j have for man I compared to the other men in the market. We set the correlation of the outside option with average utility of women for men to 0.5.

Model with Option Correlations

- We observe the likelihood of remaining single increases

Stochastic State Transition Model

The matching markets in the real world are dynamic. That is, new agents arrive, existing matched pairs leave, and as a result, the state of the system evolves. We introduce a stochastic transition model with a probability transition matrix from one state to the other. Assume that the utilities can take only finite number of possible values between 0 and 1 with increments of 0.1.

- The utility for the option of being single is 0.646 based on the probability of 0.66 of being single.
- The table above is used to obtain the average utility for the two groups (single or not single).
- At the end of the matching process, if man or woman is match with his or her corresponding dummy, we denote '1' at the single column. The utility column next to the single column is used to obtain the corresponding utility of the match.
- The calculate the option price for being single we used the binomial option method, where the probability of being single is obtained by taking numerical expectations in @Risk.

Results

- We demonstrate how to Implement the Deferred Acceptance Algorithm in a spreadsheet environment using standard functions alone.
- We extend the deterministic spreadsheet model to allow for uncertain matching utilities by adding idiosyncratic utility shocks for each agent on one side of the market for each agent type on the other side of the market.
- We further expand the model to take into account the possibility of agents' outside options (i.e., the utility of remaining unmatched) as random variables.
- We calculate the expected utilities and probabilities of remaining unmatched for the aforementioned models by running Monte-Carlo simulations.

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Carnegie Mellon University Qatar

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Design of service points in queuing networks

Authors
Madhvi Menon, Menatalla Mahmoud

Advisor
Mustafa Akan

Category
Business Administration

Abstract

Many retail services are designed as multi-phase services, where the auxiliary servers perform some basic tasks in earlier phases to allow more time on the value-added tasks later on. Motivated by the produce weighing practice of a large grocery store chain in Qatar, we investigate the performance of a two-phase service system under two scenarios:

1) Current design (Dedicated scale area): The fresh produce is weighed by dedicated servers before the customer proceeds to the check-out with all other items.

2) Alternative Design: The check-out counters are equipped with scales that can weigh produce and, hence, can process the entire shopping cart of the customer.

The two designs are compared in terms of expected waiting times at each service phase, expected queue lengths and the space requirements, and the effect of waiting time on probability of customer purchase.

In the rough-cut analysis, approximate queueing formulae are used to estimate the mean waiting times. The effect of parameter uncertainty is evaluated via Monte-Carlo simulation. In the exact dynamic queueing analysis, a discrete-vent system simulation is constructed to calculate the performance metrics as well as their distributions. We also investigate whether waiting time is an effective price discrimination tool in the current design, which incentivizes shoppers with high waiting cost to buy pre-packaged goods at higher prices to skip the produce weighing queue.

The insights carryover to more general settings. Our findings can be used to improve the process flow in other multi-phase services such as expert services (e.g., physicians and lawyers).

Design of Service Points in Queueing Networks

By: Madhvi Menon and Menatalla Mahmoud

Project Advisor: Professor Mustafa Akan

Objective

Many produce services in hypermarkets are designed as multi-phase services (i.e. weighting tables near the produce section and the cashier counters), where the auxiliary servers (weighting tables) perform some basic tasks in earlier phases to allow more time on the value-added tasks later on (cashier counter). We will be taking into account the space requirements and price discrimination.

The objective of this project is to optimize the service provided to those buying produce items at popular supermarkets, such as Carrefour.

Methodology

Used a Monte Carlo simulation to formulate our rough-cut model.

Used a discrete event system simulation on the dynamic model.

Data Source

1. Observations/Time study conducted on sites
2. Publicly available resources for customer traffic periods

Performance Metrics

The two designs, current and alternative, are compared in terms of expected waiting times at each service phase, expected queue lengths and the space requirements , and the effect of waiting time on probability of customer purchase.

Current Design

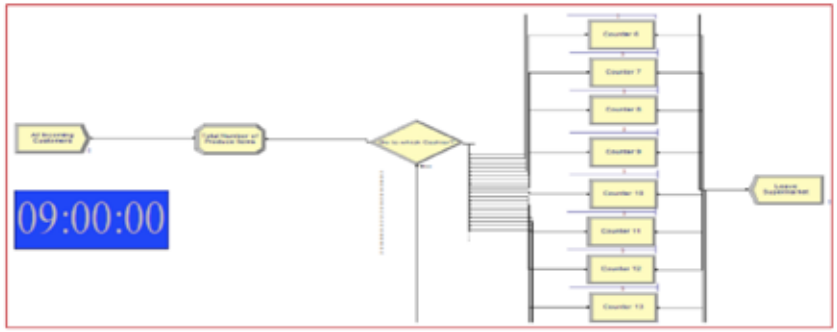

Figure 1 – Arena Snapshot of Current Design Model

The fresh produce is weighed by dedicated servers before the customer proceeds to the check-out with all other items. The produce sections are equipped with two weighing tables and have the same efficiency when serving customers.

Through our analysis, this design leads to price discrimination as customers might be tempted to buy packaged goods due to the long waiting lines at the weighing tables.

Total Service Time/Customer: 0.35 mins
Waiting Time at Weighing Table: 0.19 mins
Waiting Time at Cashier: 0.88 mins

Alternative Design

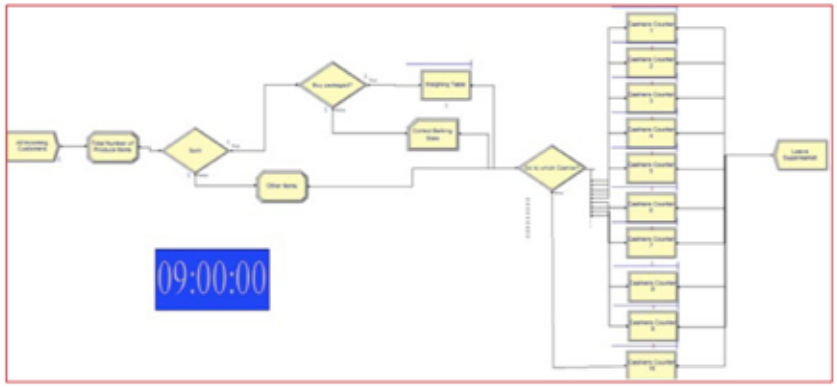

Figure 2 – Arena Snapshot of Alternative Design Model

The cashier counters are equipped with scales that can weigh produce and, hence, can process the entire shopping cart of the customer.

Total Service Time/Customer: 1.21 mins
Waiting Time at Cashier: 0.0582 mins

Results

As we can see, the **Alternative Design** is **optimal** as it gives the least amount of waiting time for customers in the system (0.0582 minutes). Given our recommendation we calculated the space required for each customer at the waiting queues to be 0.462 m^2.

Conclusion and Future Iterations

The insights carryover to more general settings. Our findings can be used to improve the process flow in other multi-phase services such as expert services (e.g., physicians and lawyers).

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Carnegie Mellon University Qatar

Re-expression of BRCA1 using targeted DNA demethylation in breast cancer cells

Author
Youssef Kanbour

Advisor
Ihab Younis

Category
Computational Biology

Abstract

In eukaryotes, upon the transcription of a gene into a pre-mRNA, the pre-mRNA first matures, leaves the nucleus, then gets translated into a functional protein in the cytoplasm. This entire process of converting the information stored as DNA into a functional protein contains many regulation steps, but the very first step is dictated by the activity of the promoter. Because tumor suppressor genes inhibit the growth or survival of tumor cells, their expression levels are highly regulated (inhibited) in cancer cells. For example, the BRCA1 gene is a tumor suppressor that encodes a protein which functions in DNA damage repair, a process that is mis-regulated in cancer. One approach cancer cells use to silence the expression levels of tumor suppressor genes such as BRCA1, is through the hypermethylation of CpG islands found in the promoter. Hypermethylation of these CG rich regions ultimately prohibits transcription factor binding leading to a decrease in their expression levels. For this project, we aimed to synthesize a protein-DNA complex (herein we call it iyk) which can target CpG islands of specific genes. The protein component of this complex is a DNA demethylating enzyme (TET1), whereas the DNA component functions to guide TET1 to specific promoters (BRCA1 promoter in this case) by base pair complementarity. We hypothesize that by expressing iyk in cancer cells, BRCA1 will be re- expressed providing the cells with potent tumor suppressor activity and ultimately leading to their demise. This approach should provide a novel potential therapy with reduced side effects.

Re-expression of BRCA1 using Targeted DNA Demethylation in Breast Cancer Cells

Youssef Kanbour[1], Ihab Younis[2]

1. *Carnegie Mellon University in Qatar, Computational Biology Program*
2. *Carnegie Mellon University in Qatar, Biological Sciences Program*

Abstract

In eukaryotes, upon the transcription of a gene into a pre-mRNA, the pre-mRNA first matures, leaves the nucleus, then gets translated into a functional protein in the cytoplasm. This entire process of converting the information stored as DNA into a functional protein contains many regulation steps, but the very first step is dictated by the activity of the promoter. Because tumor suppressor genes inhibit the growth or survival of tumor cells, their expression levels is highly regulated (inhibited) in cancer cells. For example, the BRCA1 gene is a tumor suppressor that encodes a protein which functions in DNA damage repair, a process that is mis-regulated in cancer. One approach cancer cells use to silence the expression levels of tumor suppressor genes such as BRCA1, is through the hypermethylation of CpG islands found in the promoter. Hypermethylation of these CG rich regions ultimately prohibits transcription factor binding leading to a decrease in their expression levels. For this project, we aimed to synthesize a protein-DNA complex (herein we call it iyk) which can target CpG islands of specific genes. The protein component of this complex is a DNA demethylating enzyme (TET1), whereas the DNA component functions to guide TET1 to specific promoters (BRCA1 promoter in this case) by base pair complementarity. We hypothesize that by expressing iyk in cancer cells, BRCA1 will be re-expressed providing the cells with potent tumor suppressor activity and ultimately leading to their demise. This approach should provide a novel potential therapy with reduced side effects.

Background

- In cancers, Tumor Suppressor Genes (TSGs) are silenced by the methylation of CpG islands in their promoters.
- TET1 is a DNA demethylating enzyme that can remove methyl groups from CpG islands of TSGs.
- BRCA1 is a TSG whose promoter is methylated in Breast Cancer.
- In this project, we engineered a TET1 fusion protein that can be targeted to specific CpG islands such as the one in BRCA1's promoter.
- Once the cDNA sequence of the fusion protein is cloned in a mammalian expression vector, the expressed fusion protein will bind to a biotinylated guide DNA, where the guide DNA will help deliver the whole complex (iyk) BRCA1's CpG island by base pair complementarity.
- Re-expressing such TSGs in this specific manner is a potential new therapeutic approach with minimized side effects.

Goal

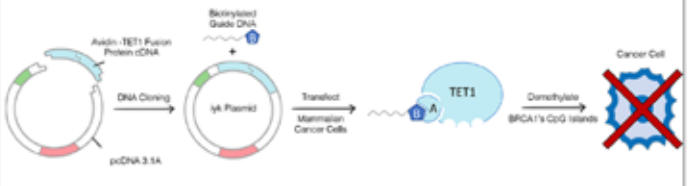

Figure 1a – The Goal of Our Project – We aimed to create a recombinant plasmid (Avidin-TET1), that when transfected into mammalian cancer cells alongside a biotinylated guide DNA, the recombinant plasmid would able to express a fusion protein that binds to the biotinylated guide DNA via an Avidin-Biotin interaction allowing TET1 to demethylate specific CpG islands in the genome leading to the re-expression of some important tumor suppressor genes such as DNA damage repair genes like BRCA1. Information on iyk's molecular functionality can be found in Figure 1b.

How iyk works

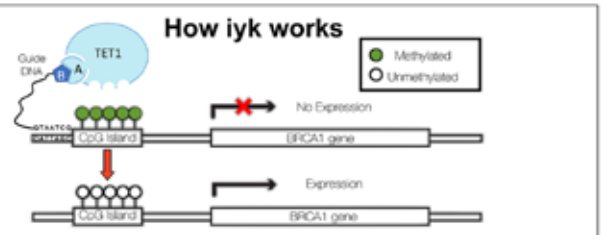

Figure 1b – Functionality and Specificity of iyk – The targeting of the DNA demethylating enzyme (TET1) BRCA1's CpG island in its promoter region is made possible by linking it to a guide DNA that binds to the region of interest by base pair complementarity. TET1 and the guide DNA are linked by an intermediary Avidin-Biotin Link. The treatment of cancer cells with this drug, in our case MDA-MB-231 breast cancer cells which have silenced BRCA1 by promoter hypermethylation, will lead to the re-expression of BRCA1 as demonstrated in the figure and thus lead to the demise of the cancer cells. Note: This complex/drug can be altered and be used on other silenced genes, given a well-designed Guide DNA.

Experimental Design

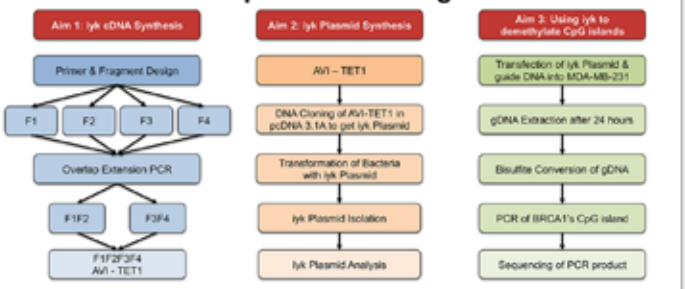

Figure 2 – The Three Aims of Our Project – Aim 1 represents the steps required in order to synthesize the cDNA sequence of Avidin-TET1 fusion protein. Aim 2 represents the steps required to synthesize iyk plasmid, which is the plasmid that expresses the protein component of iyk. In Aim 3, our goal is be to combine the protein & DNA components of iyk and transfect them into MDA-MB-231 cancer cells where we would be measuring the epigenetic changes in BRCA1's promoter.

iyk cDNA synthesis

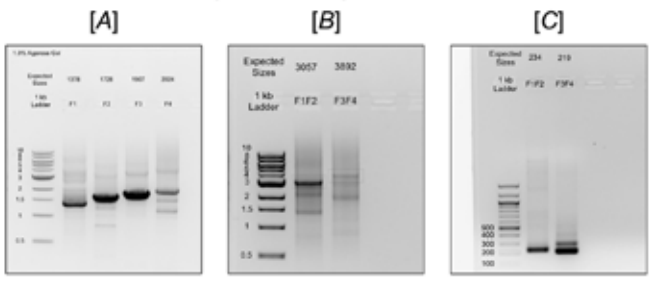

Figure 3a – PCR of fusion protein cDNA sequence – [A] Extension PCR of cDNA Fragments F1, F2, F3 and F4 (ordered from IDT) in order to create a 20bps overlap between each adjacent fragment required for Overlap PCR. [B] Overlap PCR of F1 with F2 and F3 with F4 to get F1F2 and F3F4. [C] PCR confirming the identity of the Fragments F1F2 and F3F4. *Conclusion: we successfully optimized OE-PCR to generate fragments F1F2 and F3F4.*

gDNA Bisulfite Conversion

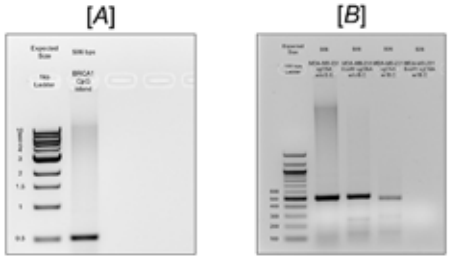

Figure 3b – Optimization of Bisulfite Conversion methodology – [A] PCR amplification of BRCA1's CpG Island confirming high quality gDNA extraction and proper primer design. [B] PCR testing the different methods of shearing DNA in preparation for bisulfite conversion. sgDNA: physical shearing of genomic DNA using a 18-gauge syringe. EcoRI sgDNA: shearing genomic DNA with EcoRI restriction enzyme. B.C: Bisulfite conversion. *Conclusion: physically shearing the gDNA gives us high quality DNA for B.C of gDNA.*

Discussion and Conclusions

- Our goal was to synthesize a DNA-protein complex guided to BRCA1's hypermethylated CpG islands in its promoter towards their demethylation, leading to BRCA1 gene re-expression.
- After optimizing OE-PCR for iyk cDNA synthesis by using different High Fidelity DNA polymerases and different thermocycling conditions, we successfully generated fragments F1F2 and F3F4 as seen in figure 3a. We are still aiming towards the synthesis of the continuous fragment F1F2F3F4.
- The size of iyk cDNA could be massively reduced (from 6.9 kb down to 2kb) by only incorporating the Catalytic Domain (CD) cDNA of TET1 required for DNA demethylation and the 45 bps cDNA motif of Avidin sufficient for Biotin binding.
- The bisulfite converted sgDNA will be sent for sequencing to measure the methylation state of BRCA1's CpG island prior iyk treatment. From our initial assumption that BRCA1 expression levels in MDA-MB-231 are low do to hypermethylated CpG islands, we expect most of the C's to be methylated in the 506 bps sequence that we have amplified in figure 3b.
- Ultimately, we believe that the synergy created by using CRISPR (genome editing) alongside iyk (epigenetic editing) would allow us to alter any genome by first, inserting or removing any desired gene in the genome using CRISPR and second, by regulating the transcription of the gene using iyk. This will allow us to dictate the fate of any cell since the fate of any cell is directly effected by the RNA and proteins it expresses.

Acknowledgments and References

- CMUQ Seed Grant to Professor Ihab Younis
- Biological Sciences Junior Ayşe Haruka Açikbaş (2018)

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Carnegie Mellon University Qatar

Code translation for implementing a functional assertion engine in SML

Authors
Sameer Ahmad, Julian Sam

Advisor
Giselle Reis

Category
Computer Science

Abstract

In today's technology-driven environment, software is becoming a driving force for growth and innovation for businesses and, as a consequence, consumers. However, we are often plagued with buggy software that hinders our ability to use it correctly. A popular method of combating this issue that has been adopted by existing software is the idea of design by contract (DbC), also known as contract programming (Meyer, 1992). This is a strategy of designing software that provides us with a method to ensure correctness and reliability of code at every module. It involves placing assertions (statements that are expected to be true) at different points in code that help prove reliability of various operations. However, many languages lack this feature which leaves a gap in the reliability of the language when it comes to reasoning about code. One such language is Standard Meta Language (SML). SML is a functional programming language widely used among researchers, as well as in the development of automated theorem proving and formal verification software (Tofte, 2009). We will be implementing an assertion engine to facilitate a DbC strategy in the New Jersey implementation of SML, in order to help verify correctness of code.

An Assertion Engine for a Functional Programming Language

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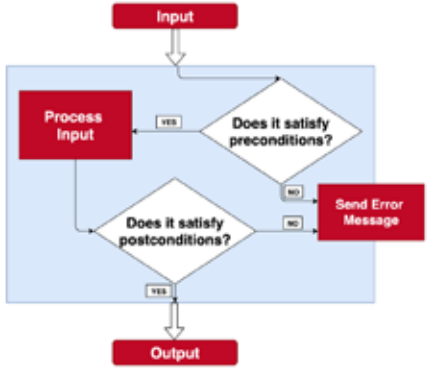
Undergraduate Research Symposium

Background

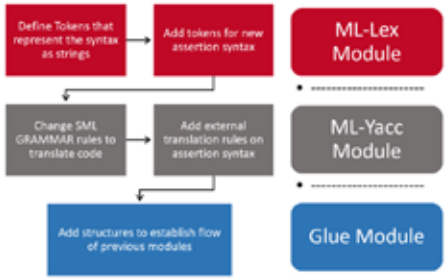
In today's technology-driven environment, software is becoming a driving force for growth and innovation for businesses and, as a consequence, consumers. But, for software engineering-based companies to release global scale software, it is quintessential that programmers know exactly how the code is executed. Therefore, to motivate the practice of developing **safe** code, a popular method of resolving this issue that has been adopted by existing software is the idea of design by contract (DbC), also known as contract programming. This is essentially a strategy of designing software that provides programmers with a method to ensure correctness and reliability of code at every module.



Design By Contract (DbC)

Our project focuses on providing a third-party assertion engine to facilitate the DbC strategy to help verify correctness of code. SML is a widely used programming language for the development of proof assistants and theorem provers and, in this field, program execution seems to be quite computationally exhaustive and processing rigorous. Since SML lacks an extension that allows it to support DbC programs, we strive to create an assertion engine that will serve this very purpose.

Methods and Procedures



Translation Flow Scheme

To create our assertion engine, we modified the SML compiler to identify and work with assertion statements. The first step was to create a lexer with the help of **ML-Lex**, a lexer for the ML programming language. Our lexer differs from the standard SML lexer in that it adds our reserved keywords for assertions: "REQUIRES" and "ENSURES". The first stage of the lexer uses regular expressions to categorize each set of characters into specific tokens. The second stage of the lexer notifies the parser what is currently identified.

The parser is created with the help of **ML-Yacc**, a parser generator written for the ML programming language. The parser generator takes as input a grammar, which is a set of rules defining valid sequences of tokens. Unlike the standard SML parser, which would interpret the expressions using added levels of complexity, our parser simply translates these expressions to represent equivalent SML code. To translate the reserved tokens for our assertion syntax, we incorporated code needed to execute these assertions that would adhere to the rules of the original SML GRAMMAR. The **glue module** combines the standalone operations of the lexer and the parser. This newly generated code is interpreted in the SML interpreter.

Assertion Engine Syntax

To use the engine, an assertion block can be represented using the following syntax. Each block is a series of one or more boolean expressions connected by SML boolean connectives.

"(*!" - This token opens an assertion block.
"REQUIRES" - This token initiates a precondition
"ENSURES" - This token initiates a postcondition.
"!*)" - This token closes the assertion block.

To refer to the output of the function in your ENSURES boolean expressions, use the variable name 'result'.

```
(*!
    REQUIRES: n >= 5 andalso b > 0
    ENUSRES: result >= 5
!*)
fun pow (n: int, b as 0: int) = 1
  | pow (n, b) = n * pow (n, b-1)

(*!
    REQUIRES: n >= 0
    ENUSRES: result = (n % 2 == 0)
!*)
fun even (n as 0: int): bool = true
  | even n = odd (n-1)
and
(*!
    REQUIRES: n >= 0
    ENUSRES: result = (n % 2 == 1)
!*)
fun odd (n as 0: int): bool = false
  | odd n = even (n-1)
```

Simple Assertion Usage Example

Results

```
(*!
    REQUIRES: n > 0
!*)
fun fibonacci (n: int): int =
  if n < 3 then 1
  else fibonacci (n-1) + fibonacci (n-2)

val a = fibonacci(0)
val a = fibonacci(10)
val a = fibonacci(~3)
```

```
File Parsed Successfully!
Standard ML of New Jersey v110.79 [built: Tue Aug  8 23:25:20 2017]
- -
uncaught exception Fail [Fail:  Function fibonacci Error: Requires Failure in line 2 ]
  raised at: stdIn:3.135-3.196
-
```

```
(*!
    REQUIRES: n > 0
    ENSURES: result > 0
!*)
fun fibonacci (n: int): int =
  if n = 3 then ~1
  else if n < 3 then 1
  else fibonacci (n-1) + fibonacci (n-2)

val a = fibonacci(3)
```

```
File Parsed Successfully!
Standard ML of New Jersey v110.79 [built: Tue Aug  8 23:25:20 2017]
- -
uncaught exception Fail [Fail:  Function fibonacci Error: Ensures Failure in line 3 ]
  raised at: stdIn:3.261-3.321
-
```


Advisor: Giselle Reis

Sameer Ahmad & Julian Sam

Carnegie Mellon University Qatar

IRg: A distributed graph-based framework for information retrieval

Author
Omar Khattab

Advisor
Mohammad Hammoud

Category
Computer Science

Abstract:

Efficient and scalable query evaluation is crucial for web-scale research and development in Information Retrieval (IR). While high-level parallel abstractions have enjoyed notable success in other domains of data science, existing IR platforms do not provide a unifying and optimizing framework for developing retrieval models. To facilitate developing and exploring custom models efficiently at large scale, we propose IRg, a distributed graph-based framework for IR. IRg presents an intuitive abstraction, which encodes terms and documents as well as their statistics and relationships in a graph structure, and allows expressing retrieval models in terms of message passing between vertices in this graph. Alongside, IRg presents an efficient distributed engine, which automatically optimizes and parallelizes retrieval models implemented using the corresponding abstraction.

Carnegie Mellon University Qatar

IRg: A Distributed Graph-based Framework for Information Retrieval

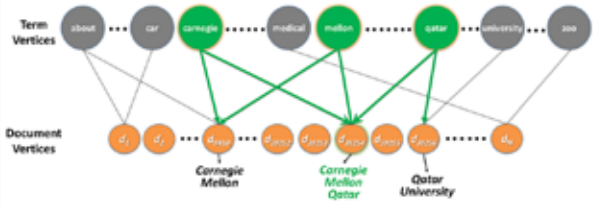
Omar Khattab
Adviser: Mohammad Hammoud

MOTIVATION

- Large-scale **Information Retrieval** (IR) is central to many applications like Web search and e-commerce search
- IR researchers and practitioners need to prototype and evaluate their **retrieval models** *at large scale*—but building or extending a *scalable* search engine to test custom models requires tremendous engineering efforts
- We propose ***IRg***, a novel framework that enables IR experts to *easily develop* and *efficiently deploy* their own retrieval models at scale

THE *IRg* ABSTRACTION

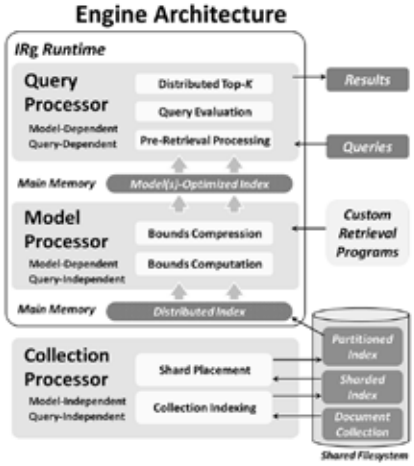
- *IRg* allows developers to express retrieval via *message passing* in a **Term–Document Graph**



- Developers' code is provided to *IRg*'s **engine** for *automatic* optimization and parallelization

THE *IRg* ENGINE

Engine Architecture



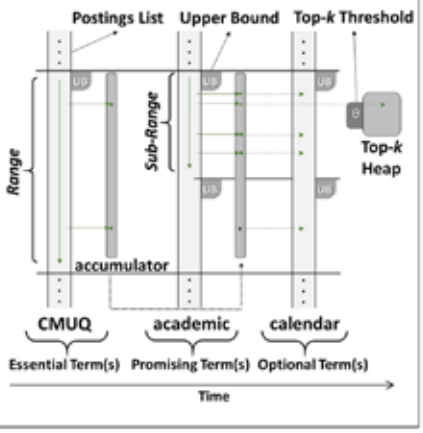
Query Processor: Employs a novel *RAAT 3-way MaxScore* (*R3M*) algorithm that *generically* applies robust *pruning*, significantly reducing retrieval latency for custom models

Model Processor: Aggressively optimizes the search index for user-implemented models

Collection Processor: Efficiently distributes and compresses the index for scalable retrieval

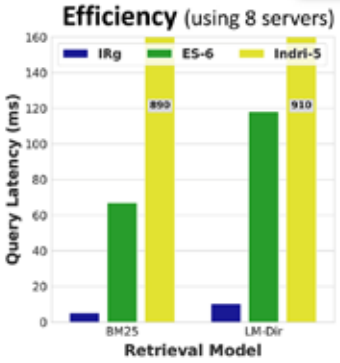
An Illustration of R3M

(for query "CMUQ academic calendar")



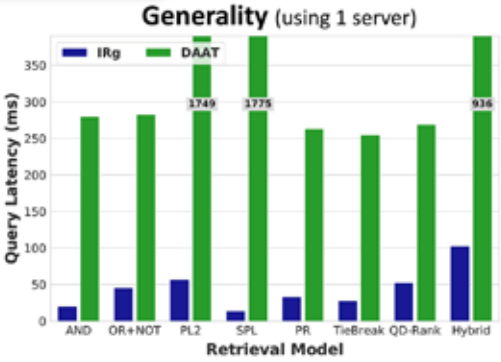
EXPERIMENTAL EVALUATION

Efficiency (using 8 servers)



Scalability (using 16 servers)

Generality (using 1 server)



IRg delivers substantial speed up to *custom* retrieval models at large scale, without impacting search accuracy!

Acknowledgements. We would like to thank our collaborators Dr. Tamer Elsayed, Reem Suwaileh, Dr. Yousuf Ahmad, and Dr. Mucahid Kutlu for their insights and feedback and their contributions to the experimental evaluation of and data pre-processing for *IRg*. This work was made possible by NPRP grant # 7-1330-2-483.

Educating girls in Qatar: Toward enhancing technology use in public schools

Author
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Advisor
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Category
Information Systems

Abstract:

This research analyzes the usage of technology in the public single-gender education system in Qatar. Specifically, this research examines how several factors can influence the e-learning in Qatar such as teacher's attitude, student's motivation and technologies support availability. In this research, I conducted user studies and problem analysis to further my understanding of the issue. Through conducting user studies, including participant interviews (initial, cued-recall and retrospective), analysis of curriculum and questionnaires distribution to students in the secondary school of Qatar. The results of the research show potential reasons why a disconnect occurs between fully equipped technological classrooms and their lack of use of it. The user studies methods uncover Qatar specific insights focused on user goals, tasks, and importantly the barriers that might prevent teachers or students from exploring these technologies. The data reveals uniquely designed interventions that move toward solving the problem.

Despite ongoing best faith efforts on all sides,

69%

of high school female students in Qatar are unhappy with the use of techonolgy in public schools (Schools in Qatar, 2017).

Why: Background on the Problem:

This research focuses on where the disconnect occurs between the availability of technology resources and the dissatisfied users of these technologies. Specifically, the three stakeholders of technology use are: students, teachers and Qatar's government. As Qatar is clearly invested in bringing new technologies into the classroom, most students are open to learning through technology (Palmer, 2016), and our work suggests teachers have a positive attitude where is the disconnect?

Studies done in Qatar looked into either student, teacher, or government perspectives. Despite the governments best efforts, an observational study from 2016 suggests that technology is rarely or never used in Qatar's classrooms (Palmer, 2016). In trying to address this gap, Ramzi (2017) reports driving forces that can reform the educational system such as school autonomy, accountability, variety, and choice. Outside of Qatar, Domingo & Garganté (2016) propose four dimensions for any technology adoption: ease of use, cost, personalization and usefulness.

These solutions might contribute to Qatar's solution. But from our design perspective, with its focus on synthesizing all stakeholders, our work suggests effective communication and personalization are integral aspects of technology adoption.

Educating Girls in Qatar: Toward Enhancing Technology Use in Qatar's Public Schools

Al-Dana Al-Mohannadi Advisor: Professor Susan Hagan

Research Method:

Literature Review:

- Successful Models of Technology (Fu & Hwang 2018, Evrim 2014)
- Teachers & Students Attitude toward IT (Koehler & Mishra 2009, Ronau 2012, Domingo & Garganté 2016)
- Qatar's Current Model of Technology (Ramzi 2017, Palmer 2016, Brewer 2007)

Created by Vigorn from Noun Project

Questionnaire Distribution to high school girls:

- Total distribution: 15
- Semantic differential, multiple choice and open-ended questions
- The questionnaire takes 10 minutes to fill out

Created by Gan Khoon Lay from Noun Project

Interview with teachers & ministry representative

- 4 Teacher participants
- 1 Ministry representative interview discussed K-12 goals including thoughts on opportunities and challenges

Created by Delwar Hossain from Noun Project

Data Analysis:

Quantitative Analysis:

Statistical analysis of the questionnaire shows 29% of students do not find an advantage in the use of technology in the classroom. 8 out of 15 reported that they rarely use laptops or computers in classrooms. Potential reasons could be accessibility issues as one student who states, "we have one ipad for the whole group and two or three girls are just solving" Notably, the most popular task among high school students is conducting research with laptops and tablets.

Qualitative Analysis:

Thematic content analysis identified users' **goals** and the **tasks**. Further, the data reveals **barriers** that prevent users from performing needed tasks.

- **Goals:** Teachers have a positive attitude towards technology, want to personalize its use while maintaining some traditional methods.
- **Tasks:** Action verbs: preparing lessons, using smart boards, grading students work.
- **Tools:** Included smart boards, tablets, mobile phones and LMS.
- **Barriers:** Teachers believe students need to be "pushed" to use technology. Further, they found personalization interests suffered because of lack of time.
- **Gate Opening:** Statements that suggest solutions include requests for additional online training, building IT infrastructure, and personalizing technology.
- **Information Gaps:** Future research would explore 'black box' statements.

Implications:

The western findings on personalization are reflected in this research, both directly in the qualitative data and indirectly in quantitative data. Specifically, from the qualitative data, **"I would like to try, more of online courses, or online teaching websites instead of actual technologies."**

From the quantitative data this research speculates that the number of students who do not find value in technologies might experience that because teachers have not yet made technology their own. For instance, a student stated **"During visual lessons when some people outside school come to observe our class, [technology] is really fun and at times challenging,"** which might suggest personalization was prioritized for that special event.

Future Work:

The research suggests a move toward establishing an internal communication platform targeted between the ministry and the teachers. The common value between all stakeholders, is the students education. Hence, future work could be targeted towards understanding how to best design and implement an open line of communication between the teachers and the ministry. As well as facilitate the implementation by monitoring the application.

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Carnegie Mellon University Qatar

What does the eye say?

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Category
Information Systems

Abstract

Cross-cultural communication is a fundamental practice in the modern, globalized world. Studies surrounding this by H.A. Elfenbein and N. Ambady (2002) examined the prominence of In-group advantage in emotion recognition. In a cultural sense, the concept of In-group advantage describes how emotional communication tends to be more accurate when both the expresser and perceiver originates from the same cultural group. This study aims to extend on this concept of in-group advantage and investigate if cultural group homogeneity is more important than the factor of cultural assimilation. To achieve this, the participants were split into two groups. One group consists of local Qatari students while the other group consists of students originating from a Western background. In this study, participants were asked to judge a set of western-centric expressions in the form of photos (Baron-Cohen, 2001). Results show that Qatari locals judged emotions of Western eyes equally as well as the in-group, suggesting that students in Education City are good at perceiving emotions in Western faces. Further research can explore whether this effect is limited to Qataris in Education City, and can explore if Westerners living in Qatar are equally as good at judging emotions in Qatari faces.

WHAT DOES THE EYE SAY?

Faiq Defiandry | Jennifer Bruder
Information Systems | Arts & Sciences

INTRODUCTION

This study explores in-group and out-group effects. Belonging to a cultural group provides in-group advantages for judging emotional expressions in faces of that group (Elfenbein & Ambady, 2002).

Studies show that miscommunications arise due to errors in interpreting emotions between different cultural groups (Elfenbein & Ambady, 2002). Because EC is a multicultural environment, it is important to explore cultural differences that can affect communication. We explored how well Qatari Education City (EC) students can judge emotional expressions in Western faces compared to Western EC students.

METHOD OF OBSERVATION

Population: 20 Qatari and 15 Western EC students
Measurements:
1) Accuracy in judging emotions in 36 validated photos of Western eyes (Figure 1, Baron-Cohen, 2001)
2) Completion of the Empathy Quotient Test (EQT, Baron-Cohen, 2004). The EQT is a theory of mind measure. In this study it is used as a control measure to ensure that the accuracy of responses to emotions in Western eyes reflects the ability of emotion recognition and is not related to generalised theory of mind deficits

Figure 1: Example of Emotional Eye Stimuli

Hypothesis: Due to extensive contact with Western populations, Qatari EC students and Western EC students will be equally proficient in judging emotions depicted in Western eyes.

RESULTS

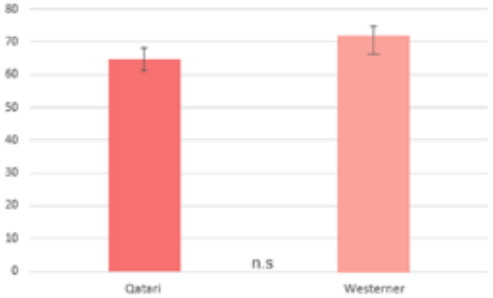

Figure 2: Percentage of Correct Responses to Eye Stimuli

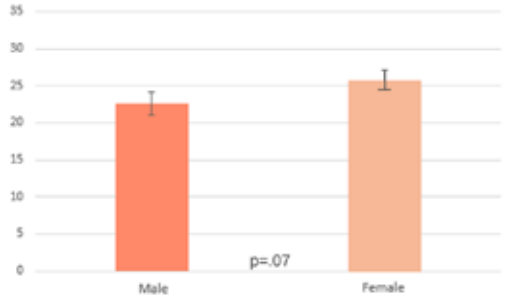

Figure 3: Total Correct Responses by Gender

1) Figure 2: As predicted, Qatari EC students showed an in-group advantage. They judged Western eyes as accurately as Western participants (p=0.15).
2) Figure 3: Data were collapsed and analysed post-hoc for gender differences. Females performed marginally better (47.2%) than males (42.3%), (t(33)=1.89, p=0.07).
3) EQT: Three participants (1 Qatari, 2 Westerners) with below average empathy scores on the EQT were removed from analysis. Qatari and Western participants had equal EQT scores (Qatari: 45.5, (SD: 10.3); Western: 45, SD:9.7). EQT correlated moderately with accuracy of judging emotions in eyes (r=0.36).

CONCLUSION

Qatari EC students judged emotions of Western eyes equally as well as the in-group, suggesting that EC students are good at perceiving emotions in Western faces.
A trend for gender differences was observed where females were better at emotional judgments than males, which replicates previous findings (Baron-Cohen et al., 2015).
Further research can explore whether this effect is limited to Qataris in EC, and can explore if Westerners living in Qatar are equally as good at judging emotions in Qatari faces.

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MEETING OF THE Minds Undergraduate Research Symposium
Carnegie Mellon University Qatar

Effect of language direction on spatial cognition

Authors
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Advisor
Jennifer Bruder

Category
Information Systems

Abstract

Previous research demonstrates how the languages one speaks influence how they perceive the world, affecting both visual perception and visual-spatial cognition. For example, bilinguals generally out-perform monolinguals on cognitive inhibition tasks, like the Simon Task or the Stroop Task, by displaying better cognitive control mechanisms (Blumenfeld & Marian, 2014). Furthermore, performance and perceptual differences related to language reveals differential brain activity between monolinguals and bilinguals on spatial stroop tasks (Bialystok, Craik, et al., 2005). Research has also explored the effects direction of language have on spatial perception (Blumenfeld & Marian, 2103). Specifically, effects of native language direction differences between left-to-right (e.g. French) and right-to-left (Arabic e.g.) languages, show that right-to-left language speakers are biased towards the right visual hemifield compared to the left-to-right language speakers (Fagard & Dahmen, 2003). In the Qatari context, our study aims to explore whether right-to-left language (Arabic) speakers will show similar effects when compared to left-to-right language speakers. Study participants are CMU-Q bilingual speaking students who either read and write in only left-to-right languages (e.g. English and French) or read and write in English and Arabic. Therefore, with respect to the Simon Task, we hypothesize that the attention to space given by right to left language speakers will in fact be different than that of the left to right language speakers. We predict that based on the language of communication in this area, right to left language speakers will answer the incongruent trials of the Simon Task more accurately than left to right language speakers, due to their longer exposure to both the right (primary language) and the left (English secondary language) hemifields.

Effect of Language Direction on Spatial Cognition

Exploring the effects of reading and writing language direction on performance on visual tasks in the Middle Eastern context

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Information Systems

Danish Memon
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Jennifer Bruder
Department of Arts and Sciences

Introduction

- Language affects how people perceive the world[1]
- Language direction (left-to-right (L-to-R) or right-to-left (R-to-L) affect visual-spatial perception[2,3]
- The Simon Effect[4]: Cognition is optimal (i.e. reaction times are faster; accuracy is higher) when the words "left" or "right" are spatially congruent with the motor response (see Figure 1). However, this effect has only been reported in L-to-R languages

- **Hypothesis:** Direction of written language direction will influence visual-spatial perception

Methods

- Participants were students at CMU-Q
- The R-to-L group could read and write in Arabic
- The L-to-R group could read and write in L-to-R languages only, like English (Table 1)
- Both groups were controlled for age (range: 18-25) and handedness (right-handed only)
- Both groups performed a variation of a visual Simon Task (Figure 1). The Arabic group performed the task in Arabic
- Accuracy and reaction time were measured

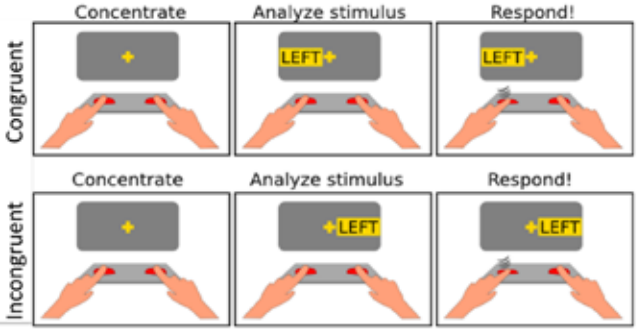

Figure 1: *A visual Simon Task*

The Simon effect: Response accuracy is usually higher and response times faster when stimulus appears in the same relative direction as the response.

Type of language	Congruent	Incongruent
Arabic Group (n=18)	30 randomized trials	30 randomized trials
English Group (n=15)	30 randomized trials	30 randomized trials

Table 1: *Independent & Dependent variables*

Results

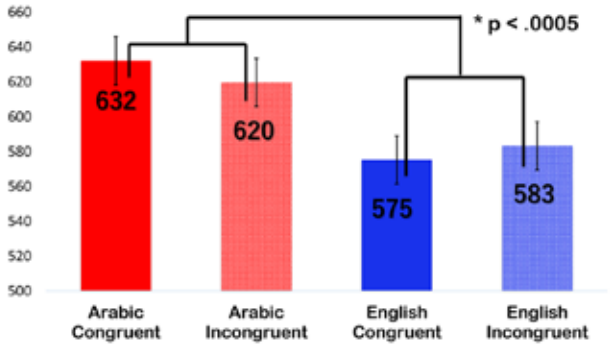

Figure 2: *Average Response times (ms)*

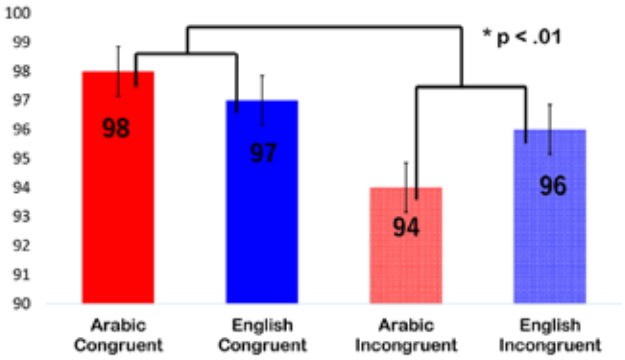

Figure 3: *Accuracy (Percentage Correct)*

- Overall, reaction times for the English Group were significantly faster (Figure 2, $F(2,1982)=14.67$, $p < .0005$; Wilk's $\Lambda=0.96$, partial $\eta 2 = .01$)
- Across groups, accuracy for congruent vs. incongruent conditions was higher, demonstrating a common Simon Effect (Figure 3, $F(1,1983)=8.18$, $p < .01$; partial $\eta 2 = .004$)

Discussion

- The Simon Effect was reproduced across groups: spatially congruent trials were more accurate than incongruent trials
- Our results suggest that bi-directionality of written languages increases cognitive load and leads to delays in speed of responses to space and language judgements

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Postgraduate Posters

An oracle hierarchy for small one-way finite automata

Authors

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Category

Postgraduate

Abstract

We introduce a polynomial-size oracle hierarchy for one-way finite automata. This generalizes the polynomial-size alternating hierarchy for one-way finite automata with a bounded number of alternations; and relies on an original definition of what it means for a nondeterminsitic automaton to access an oracle, which we carefully justify. We prove that our hierarchy is strict and that the first level already contains problems outside the polynomial-size alternating hierarchy. We then identify five restrictions to our oracle-automaton, under which the oracle hierarchy is proved to coincide with the alternating one, thus providing an oracle-based characterization for it. We also show that, given all others, each of these five restrictions are necessary for this characterization.

An Oracle Hierarchy for Small One-Way Automata

Malek Anabtawi, Sabit Hassan, Christos Kapoutsis, Mohammad Zakzok
Carnegie Mellon University in Qatar

Motivation.

The Polynomial-Time Hierarchy

Problem. *Given a partially filled Sudoku board, fill the rest of it so that each row, each column, and each square contains each one of the digits 1-9 exactly once.*

5	3			7				
6			1	9	5			
	9	8					6	
8				6				3
4			8		3			1
7				2				6
	6					2	8	
			4	1	9			5
				8			7	9

Question. How hard is this problem?

- **Answer 1**. Nobody knows of an efficient algorithm to solve this problem
- **Answer 2**. It is in Level 1 of the ***Polynomial-Time Hierarchy***.

Answer 2 is much more informative.

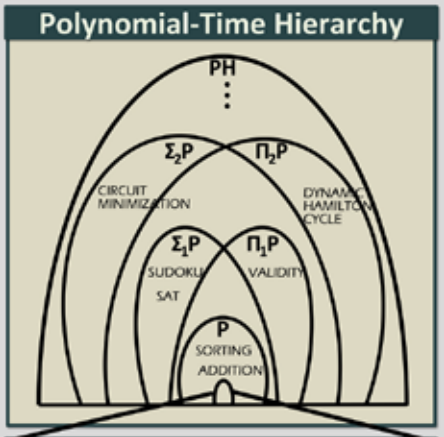
The ***Polynomial-Time Hierarchy (PH)*** is important for describing how hard a computational problem is.

- Infinitely many levels.
- The harder a problem is, the higher the level of the hierarchy where it belongs.
- Multiple equivalent definitions using: (1) *polynomial-time oracle Turing machines, (2) polynomial-time alternating Turing machines, (3) polynomial-time predicates, and (4) second-order logic.*
- A simpler 1FA analog of the *PH* exists. It can us help gain intuition about the *PH*.

Question. What are the 1FA analogs of the PH under each of these definitions? Are they also equivalent?

Contribution. Analogs under (2), (3), (4) already treated. In this work:

- Analog of the PH under (1);
- Prove that it is strictly stronger than the analog of the PH under (2).
- Restrict it to match other analogs.



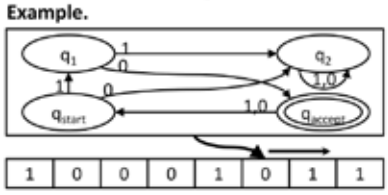
Prior work.

The Poly-Size Alternating Hierarchy

Definition: An alternating finite automaton (AFA), consists of:

- A set of states, Q.
- A partition of Q into U and E, the universal and existential states, respectively.
- A set of input symbols, Σ.
- A transition function, $\delta: Q \times \Sigma \rightarrow Q$.
- A start state, q_{start}.
- An accept state, q_{accept}.

Example.



Definition.

The polynomial-size alternating hierarchy is a collection of classes. Let $k \geq 0$ be the number of alternations:

- $1\Sigma_k$ is the class of problems that can be solved using an AFA which has alternating level $\leq k$ and starts in an existential state.
- $1\Pi_k$ is the respective class that starts in a universal state.

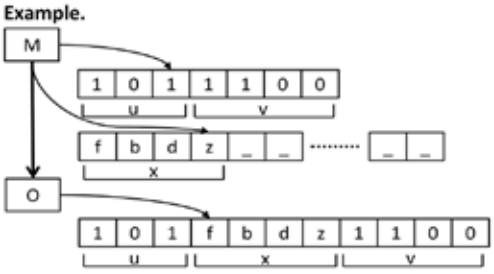
The Oracle Finite Automaton Hierarchy

First, we give a natural definition of *oracle finite automata* (OFA) and define the powerful OFA hierarchy on top of it.

Definition.

A OFA, *M*, has a one-way write-only oracle tape, access to an oracle O, and is able to: **(1)** ask multiple questions to *O*, **(2)** write on the oracle tape at any point in its computation, **(3)** write an indefinitely long string on the oracle tape, and **(4)** ask "*uxv*" to *O*.

Example.



Definition.

The powerful OFA hierarchy is the collection of classes: $1\hat{\Delta}_0 = 1\hat{\Sigma}_0 = 1\hat{\Pi}_0$, and for all $k \geq 0$:

$$1\hat{\Delta}_{k+1} = 1\mathrm{D}^{1\hat{\Sigma}_k},$$
$$1\hat{\Sigma}_{k+1} = 1\mathrm{N}^{1\hat{\Sigma}_k},$$
$$1\hat{\Pi}_{k+1} = \mathrm{co}1\hat{\Sigma}_{k+1}.$$

Theorem.

The strong OFA hierarchy is strict. That is, for all $k \geq 0$:

$$1\hat{\Sigma}_k \neq 1\hat{\Sigma}_{k+1}$$

Theorem.

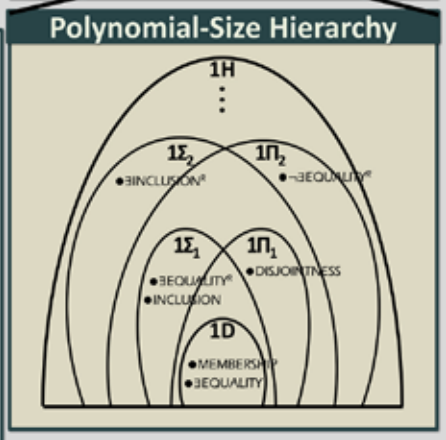
The strong OFA hierarchy is strictly stronger than the polynomial-size alternating size hierarchy.

$$1\mathrm{H} \subset 1\hat{\mathrm{H}}$$

Open Problem.

For every level, are the opposing classes equivalent?

$$1\hat{\Sigma}_k = 1\hat{\Pi}_k$$



Restricting Oracle Finite Automata

To create an analogous oracle-based hierarchy to the polynomial-size alternating hierarchy, we restricted the OFA model in five ways. An oracle OFA *M* is said to be:

1. **Many-one:** If it makes one query and returns the answer of the query.
2. **Synchronous:** If it only prints on the oracle tape in the same step as it asks a query to the oracle.
3. **Laconic:** If the query tape can only contain one symbol.
4. **Omitting:** If, at every query with partition *u*, *v*, and query tape string *x*, the query string omits the prefix *u*, and is thus only asking "*xv*" to the oracle *O*.
5. **Query-deterministic:** If, at every query, for every possible prior state-symbol-move triple, there is ≤ 1 string that can be printed on the query tape.

The Weak OFA Hierarchy

We define the weak OFA hierarchy using an OFA with all five restrictions shown in the left panel.

Definition.

The weak OFA hierarchy is the collection of classes:

$1\tilde{\Sigma}_0 = 1\tilde{\Pi}_0$, and for all $k \geq 0$:

$$1\tilde{\Sigma}_{k+1} = 1\mathrm{N}^{1\tilde{\Pi}_k},$$
$$1\tilde{\Pi}_{k+1} = \mathrm{co}1\tilde{\Sigma}_{k+1}.$$

Theorem.

The hierarchy is contained within 1DFAs of elementary size.

$$1\tilde{\mathrm{H}} \subseteq \mathrm{e}^{1\mathrm{D}}$$

Theorem.

The weak OFA hierarchy represents the polynomial-size hierarchy. That is, For all $k \geq 0$:

$$1\tilde{\Sigma}_k = 1\Sigma_k$$
$$1\tilde{\Pi}_k = 1\Pi_k$$

Proof Sketch.

By induction on *k*, we prove that every level in the weak oracle hierarchy $1\tilde{\Sigma}_k$ ($1\tilde{\Pi}_k$) can simulate the corresponding level in the polynomial-size hierarchy $1\Sigma_k$ ($1\Pi_k$) and vice versa.

Corollary.

The weak OFA hierarchy is strict. For all $k \geq 0$:

$$1\tilde{\Sigma}_k \neq 1\tilde{\Sigma}_{k+1}$$

This follows directly from the previous theorem and [3], where Geffert prove that the polynomial-size hierarchy is strict.

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MADAR Twitter user dialect identification

Authors

Houda Bouamor, Nizar Habash, Sabit Hassan, Kemal Oflazer

Category

Postgraduate

Abstract

We present a dataset of Twitter profiles and corpus of tweets created for the Multi Arabic Dialect Applications and Resources (MADAR) project. The corpus has two components. First is a collection of Twitter user profiles who have tweeted with hashtags of different countries in the Arab Region. The twitter profiles are annotated for their countries of origin. We provided the annotators with guidelines for the annotation process. A high Inter-annotator Agreement shows that the annotation can be trusted. Second is a collection of tweets (publicly shared as tweet-ids) from each of the Twitter user profiles. The tweets are automatically annotated with dialectal probabilities using the MADAR dialect identification system which classifies Arabic dialects into 25 cities plus Standard Arabic. This corpus of tweets alongside their dialectal probabilities can be used for Arabic Dialect Identification. We also present results of our initial experiments on automatically predicting countries of Twitter users by utilizing Tweets posted by the users.

MADAR Twitter User Dialect Identification

Houda Bouamor, Nizar Habash, Sabit Hassan, Kemal Oflazer

1. Motivation: Arabic Dialect Identification

- Arabic is rich in dialectal diversity across different regions of the Arab World.

English: The price is high, isn't it?
Modern Standard Arabic: ؟ أليس كذلك ، هذا سعر مرتفع
Cairo: ؟ ده غالي ، مش كده
Doha: ؟ هذي غالي ، صح

- Dialect Identification can be helpful for tasks such as sentiment analysis or author profiling.
- Users on social media such as Twitter share their thoughts with a diverse range of dialects.

2. Our Contributions:

- Construct an unbiased and diverse dataset by collecting Tweets from different countries of Arab World.
- Build models that can predict countries of Twitter users from their Tweets.

Example of a Tweet. Can we predict country of the user from Tweets?

Twitter Profile

3. Data Collection and Annotation:

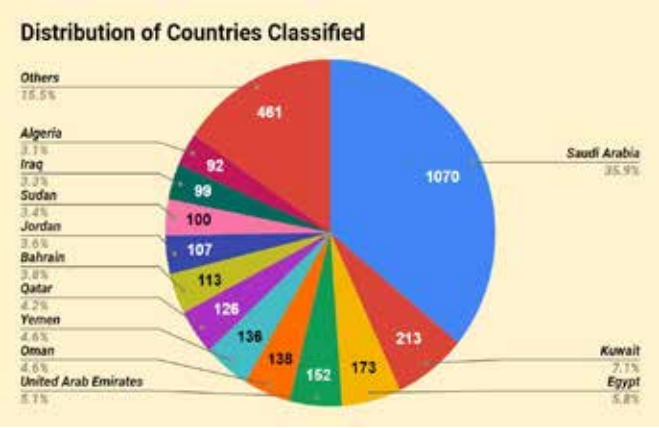
- We collect 2980 Twitter users who have posted Tweets using hashtags of 22 countries from the Arab Region.
- We collect up to 100 Tweets from each of the users.
- The Twitter profiles are annotated by 3 annotators for their countries.
- We collect up to 100 Tweets from each of the users.

Inter-Annotator Agreement:

Annotator 1 - Annotator 2	Annotator 1 - Annotator 3	Annotator 2 - Annotator 3
83.66	75.7	81.11

Average Cohen Kappa: 80.16
Fleiss Kappa: 80.15

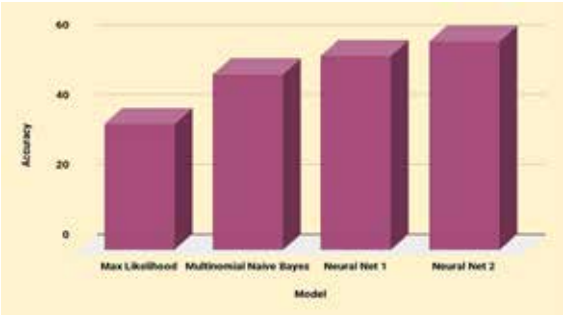
4. Distribution of annotated data:



- The ratio of Twitter users from Saudi Arabia (35.9%) is significantly high.
- Some countries have low ratio of Twitter users. 10 countries (Others) together contribute to 15.5% of the countries data

5. Initial Experiments:

- Trained Neural Networks and Machine Learning models to predict countries of Twitter users from Tweets



6. Future Work:

- Improve the performance of the current models
- Shared task hosted at WANLP 2019: https://sites.google.com/view/madar-shared-task/home

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Qatar National Research Fund
NPRP 7-290-1-047

ARAP – Author profiling and its application for market segmentation

Authors

Anis Charfi, Syed Mehdi, Esraa Mohamad

Category

Postgraduate

Abstract

This poster will present the ARAP – Author profiling (AP) tool for Arabic, which is based on machine learning and language resources that were developed in the context of the ARAP project. The poster shows how author profiling can be used to analyze the market segmentation by analyzing an organization's social media Arabic speaking followers. The AP tool can analyze the public tweets by the followers of an organization's Twitter account and categorize the organizations' followers based on age group, gender, and Arabic dialect. Such information would be useful for organizations to understand their customers/ followers' base and would also enable targeted marketing. For the purposes of this demo, we took Ooredoo as example organization that could leverage the AP tool.

ARAP – Author Profiling Tool for Customer Segmentation

Anis Charfi, Syed Mehdi, Esraa Mohamad
Information Systems, Carnegie Mellon University in Qatar

Tool Overview

The ARAP Arabic author profiling tool is a machine learning based tool that allows to profile the known or unknown author of Arabic text with respect to gender, age, language variety (dialect), by analyzing the Arabic text written by the user or the user's public tweets (in case the user's Twitter id is known). The tool is available through a Web user interface and also as a REST API.

Figure 1: Author Profiling Tool Result Web UI

{"Profile":{"LVIConfidence":0.57,"ProfileImg":"https://pbs.twimg.com/profile_images/1113166576649666816 5/h12OSYCC_normal.jpg","NNNConfidence":0.9876676786130071,"UserName":"i_alghanim2","GenderConfidence":1,"Bio":"الرابطة القطرية للأميين إدارة مجلس فرع #TheQPA @ جامعاً أعاظم لم طالب أبي بن علي) قليلك إلا عالماً أعاظم ولم قليلي إلا)","Gender":"male","NNN":"NAS","Tweets":{"Tweet":[{"Text":"RT @Banna_Aldarwish: بن عُمر مع \nفقال المؤمنين ؟ أمير يبكيك يا ما رفاقه احد له فقال قلبية أ فبكى بكاء الرعد صوت الخطاب عبد وحصول الشرطة نقودات الدولية المطولة في اليوم مساء البشركي نظيره القطري على الشرطة صنخب فوز u2026","Date":"Sat\ب Apr 13 09:04:56 UTC 2019"},{"Text":"RT @qaqa1976: مبروك الاول المركز منتخبنا على \u2026","Date":"Sat Apr 06 15:48:11 UTC 2019"},{"Text":"RT @b0_diary: الكورية التعريبات يمفضن الجنيات وقع https://t.co/Vp8ZFnjrOY","Date":"Sat Apr 06 12:19:27 UTC 2019"},{"Text":"RT @Just_ali84: الغياني فيه مع الفكر خلق \n@Moezali_ https://t.co/g3JtdeJMvB","Date":"Sun Mar 31 19:45:02 UTC 2019"},{"Text":"RT @binnahar85: تركيا!\n\إذا كان بهيان عه بكلمة يتكلم لم لكنه وإسرائيل، أدان إيران قبل قطيه، القمة بهيان صور فعلى تركيا يذكر لم العربية الدول u2026","Date":"Sun\ Mar 31 18:23:21 UTC 2019"},{"Text":"RT @silentrealistic: \n'\n\n\h أمامويهيه ربيعة ميادة\h لكم و لنا # https://t.co/jeO8LYxH4X","Date":"Sun Mar 31 18:08:15 UTC 2019"},{"Text":"RT @binnahar85: ولبس رئيس دولة كلمة كانت القبط كلمة أبو الغيظ، أبو كلمة ألقاه، الأمير سمو بخروج جدا صعدت u2026","Date":"Sun\نعمي Mar 31 18:07:43 UTC 2019"},{"Text":"RT @AbdulazizSay: والأخوة #تونس العربية\n\الوفادي\n\إحتراماً ل إحتراماً ل #تونس قارئ\n العربية https://t.co/o1oF8W568W","Date":"Sun Mar 31 14:14:05 UTC 2019"},{"Text":"RT @NasserFAlKhater: في يتواجه لم الذي العمل فريق يبالغي الي حقكم.....و توفيكم فكر لا كلمة u2026","Date":"Sat\.نعطي Mar 30 11:04:33 UTC 2019"},{"Text":"RT @3body_bb: الخطالني خالد واخوي الكيميس على الاشادة مع ....و أيضاً ....نشجع نجعل..نعطي الصورة.....نجعا https://t.co/P4GOhDu9Nc","Date":"Wed Mar 27 14:04:14 UTC 2019"}]},"ProcessingTime":2164,"FullName":"Ibrahim Alghanim","Age":"35+","AgeConfidence":0.9999866067136729,"LVI":"QATAR","Location":"Doha - Qatar"}}

Figure 2: Author Profiling Tool Result API

Author Profiling Tool for Customer Segmentation

ARAP – Author Profiling (AP) tool can be used to analyze the customer segmentation by profiling the organization's Arabic speaking followers on social media. The AP tool can analyze the public tweets by the followers of an Organization's Twitter account and categorize the users based on Age Group, Gender, and Arabic Dialect. Such information is useful for organizations to understand their prospects and customers and enables targeted marketing. Below, we present the process and the results of using the AP tool to analyze Ooredoo's followers on Twitter.

Using Twitter API retrieved the last 5000 followers

Filter users with at least 60% Arabic tweets and at least 100 tweets.

AP API on the final set of 3460 users

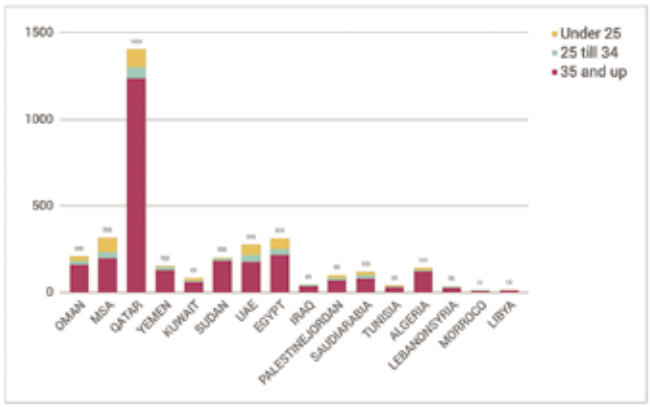

Figure 3: Author Profiling Tool Age and Dialect Distribution

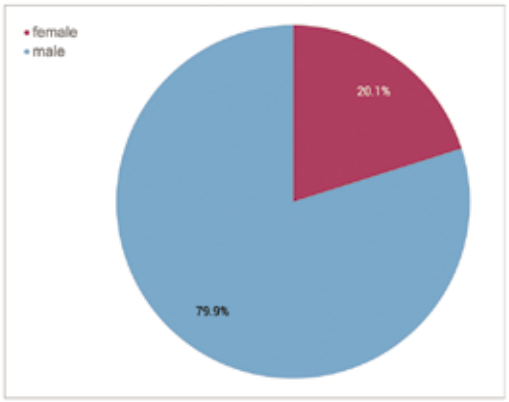

Figure 4: Author Profiling Tool Gender Distribution

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Carnegie Mellon University Qatar

Deception detection in Arabic text

Authors
Anis Charfi, Esraa Mohamad, Syed Mehdi

Category
Postgraduate

Abstract

With the widespread of the internet online sources, they gain a lot of deception content from many users. Deception is a very common phenomenon, and people have been interested in how to accurately detect deception for much of human history. This has motivated the need for methods to automatically detect and identify deceptive content. In the framework of the ARAP project, we translate the open deception Seven Truth and Seven Lies corpus (Perez-Rosas and Mihalcea (2015)) to Arabic and used it to automatically detect deception. Then, we used machine learning classification techniques in order to identify of deceit in short texts. The corpus contains about 7000 sentences annotated as truth or lie.

Deception Detection in Arabic Text

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INTRODUCTION & MOTIVATION

- The Seven Truth and Seven Lies corpus (Perez-Rosas and Mihalcea (2015))
- Developed using Amazon Mechanical Turk: Each worker was asked to contribute seven lies and seven truths, on topics of their choice.
- We translated that corpus to Arabic and built a supervised-learning classification models to classify the lies and truths in Arabic.

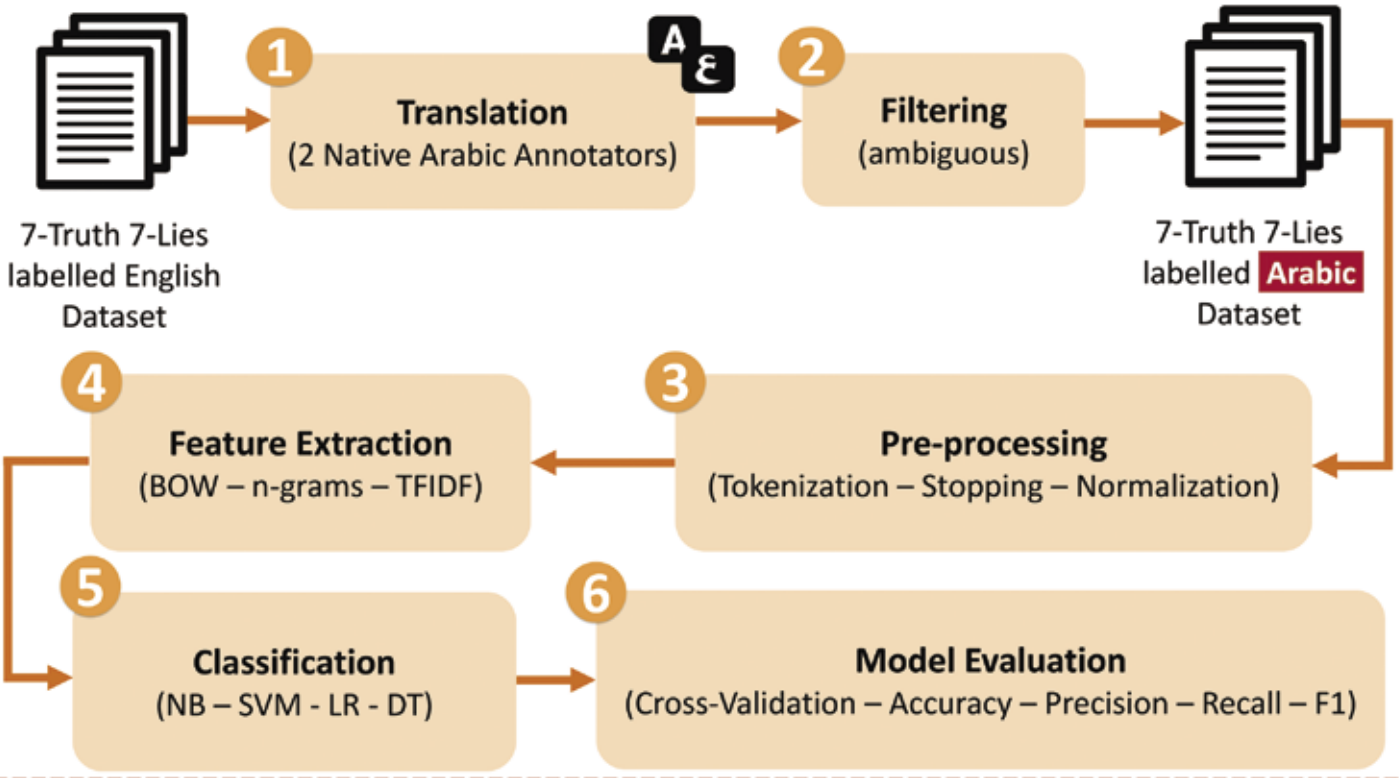
Applications

Rumors Control

Fake News Detection

Trustful Community

APPROACH



RESULTS

- Binary classification [Truth, Lie]
- 10-fold cross-validation
- BOW with HP = 1500 tokens
- TF-IDF with min_df = 4 & max_df = 0.25

CONCLUSION

- We have an Arabic dataset of truths and lies with over 7000 sentences
- Best accuracy of 62% with the Logistic Regression classifier and using tf-idf with bi-gram features
- As future work, we will try BOW with Okapi BM25 weighting schema features and W2V features

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Supporting students writing case analysis in information systems and organizational behavior

Authors

Silvia Pessoa, Maria Pia Gomez Laich, Thomas Mitchell, Michael Maune

Category

Postgraduate

Abstract

This poster reports on a collaboration between applied linguists and professors in Information Systems and Organizational Behavior to support the writing of case analyses. We describe our application of several linguistic and knowledge models of writing from Systemic Functional Linguistics (SFL) and Legitimation Code Theory (LCT) to the teaching of analytical argument. These models describe language patterns for analysis and argument, the structure of the case analysis genre, and the amount and kind of information from the disciplinary theory needed to meet the expectations of the assignment. Using these models, the linguists developed and conducted writing workshops for the students to prepare them for their writing assignments.

We argue for the effectiveness of these models in helping students succeed in their writing by drawing on data collected from a high and low-graded case analyses from students in these courses. We show how the students who were successful on the assignments were more on topic, used the specific topic of the paper to do their analysis, and engaged in sophisticated reasoning and use of specific language patterns to effectively accomplish the goal of the assignment. We conclude by showing some of the teaching principles that may help future students in their case analysis writing.

Supporting Students Writing Case Analyses in Information Systems & Organizational Behavior

Introduction

The purpose of our study is to improve the quality of technical and professional communication skills in the fields of Information Systems (IS) and Organizational Behavior (OB).

Through collaboration between applied linguists and IS and OB faculty, we aimed to develop undergraduate students' ability to write a case analysis, a genre that is a common requirement for students in IS and business programs. This genre involves examining a case, identifying an organization's problem(s), and proposing a solutions using concepts from IS or OB.

Figure 1.
Rubric used to assess case proposals in OB class.

Methods

Collaborating with IS & OB professors to:

- Prepare case analysis workshops for IS students using models from linguistics and sociology of education
- Develop a clear and comprehensive rubric to assess student writing
- Collect students' drafts and provide them with individual feedback using the rubric

Results

High-graded students reflected a **solid command** of the main writing characteristics for the **case analysis genre**, such as:

- Following the stages of a case analysis
- Accurately and succinctly stating the purpose of the case analysis and an overarching claim
- Using expanding and contracting resources to support their thesis
- Analyze the case through specific concepts from their discipline as outlined by the professor

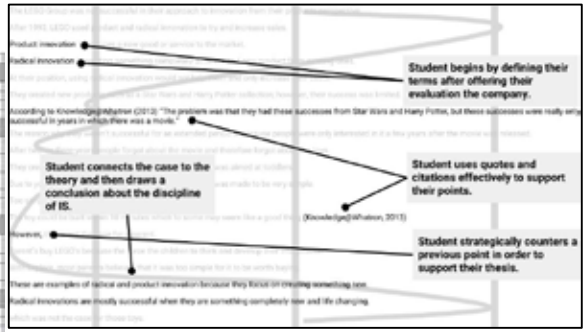

Figure 2.
Example of a low-graded case analysis from IS with annotations.

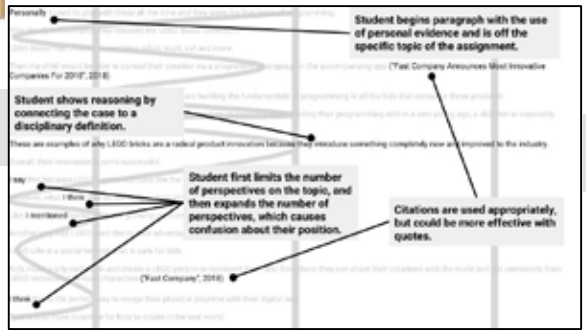

Figure 3.
Example of a high-graded case analysis from IS with annotations.

Lessons Learned

Instruction and feedback should focus on effective use of:

- Appropriate disciplinary concepts as a lens for analyzing the specifics of the case
- Reasoning concepts, like logical relations, so that students connect the case to the disciplinary framework and draw conclusions about both
- Expanding and contracting resources through making sure that expanding perspectives is followed by narrowing the perspective to the student's thesis

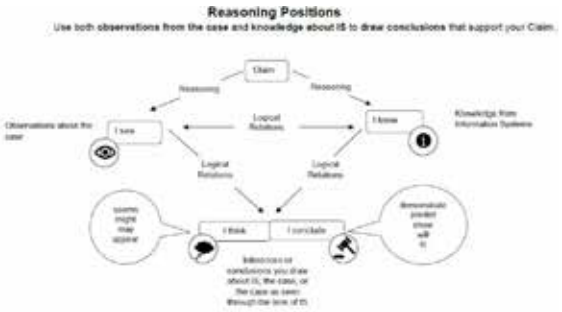

Figure 4.
Example of workshop materials used to teach reasoning in IS class.

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