Gene Expression Transcription Pogil Answer Key

Gene Expression Transcription Pogil Answer Key gene expression transcription pogil answer key is an essential resource for students and educators seeking to understand the fundamental processes involved in gene expression, particularly transcription. Mastering this topic is crucial for comprehending how genetic information is converted into functional products like proteins. This article provides a comprehensive overview of gene expression transcription, the importance of Pogil activities in learning, and tips for utilizing answer keys effectively to enhance understanding. Understanding Gene Expression and Transcription What Is Gene Expression? Gene expression is the process by which the information encoded in a gene is used to synthesize a functional gene product, typically proteins or RNA molecules. This process is tightly regulated and fundamental to cellular function, development, and response to environmental stimuli. Gene expression involves multiple steps, including transcription and translation, each controlled by various factors. The Role of Transcription in Gene Expression Transcription is the first step in gene expression, during which a segment of DNA is copied into messenger RNA (mRNA). This process involves several key components: DNA template strand: The strand of DNA that is read by RNA polymerase to synthesize mRNA. RNA polymerase: The enzyme responsible for synthesizing mRNA from the DNA template. Promoter regions: Specific DNA sequences that signal where transcription should begin. Transcription factors: Proteins that assist or regulate the binding of RNA polymerase to DNA. The process of transcription results in an mRNA strand that is complementary to the DNA template, which then moves on to translation to produce proteins. Gene Expression Transcription Pogil Activities What Is Pogil? Pogil (Predict-Observe-Explain-Live) activities are student-centered, inquiry-based learning 2 exercises designed to promote active engagement with scientific concepts. In the context of gene expression transcription, Pogil activities guide students through exploring the mechanisms of transcription, understanding key components, and applying their knowledge to different scenarios. Purpose of Pogil Activities in Learning Transcription These activities help: Develop critical thinking skills by encouraging students to predict outcomes based on their prior knowledge. Observe experimental data or diagrams to build understanding. Explain concepts in their own words, enhancing retention. Apply knowledge to new situations, promoting transfer learning. Using the Gene Expression Transcription Pogil Answer Key Effectively Understanding the Importance of the Answer Key The Pogil answer key serves as a guide for educators and students to check their responses, understand misconceptions, and reinforce correct concepts. It provides detailed explanations that clarify complex processes involved in transcription. Strategies for Utilizing the Answer Key To maximize learning: Attempt first: Students should try to answer questions independently before1. consulting the answer key. Compare responses: Review your responses against the answer key to identify2. areas of misunderstanding. Focus on explanations: Pay close attention to the explanations provided, as they3. often contain additional insights. Use as a learning tool: Rather than just copying answers, use the key to4. understand the reasoning behind each response. Common Topics Covered in the Pogil Answer Key The answer key typically addresses: The initiation of transcription, including promoter recognition. The role of RNA polymerase and transcription factors. 3 The steps of elongation and termination. Differences between prokaryotic and eukaryotic transcription. The significance of regulatory sequences and factors. Key Concepts in Transcription to Remember Promoters and Transcription Factors Promoter regions are DNA sequences that indicate where RNA polymerase should bind to start transcription. Transcription factors are proteins that facilitate or inhibit this binding, controlling gene expression levels. RNA Synthesis During elongation, RNA polymerase moves along the DNA, synthesizing a complementary RNA strand by adding ribonucleotides in the 5' to 3' direction. Termination of Transcription Transcription ends when RNA polymerase encounters a termination signal, leading to the release of the newly formed mRNA. Differences in Transcriptional Regulation In prokaryotes, transcription is often regulated by operons and repressor proteins, whereas eukaryotic transcription involves complex enhancer and silencer sequences, along with a multitude of transcription factors. Additional Resources for Learning Transcription Visual Aids and Diagrams Utilize diagrams that depict the transcription process step-by-step, highlighting the roles of different molecules and sequences. Interactive Simulations Online tools and simulations can help students visualize how RNA polymerase interacts with DNA, initiates transcription, and synthesizes RNA. Practice Questions and Quizzes Engaging with practice questions, especially those aligned with Pogil activities, consolidates understanding and prepares students for assessments. 4 Conclusion Mastering gene expression transcription is fundamental to understanding molecular biology and genetics. The Pogil answer key is a valuable tool that supports active learning, helps clarify complex concepts, and strengthens students' grasp of the transcription process. By engaging deeply with Pogil activities and using the answer key thoughtfully, students can develop a solid foundation in gene regulation and expression, preparing them for more advanced studies in biology. Final Tips for Students and Educators Always attempt to answer questions independently before consulting the answer key. Use the explanations in the answer key to deepen your understanding rather than merely copying answers. Combine Pogil activities with other resources like videos, textbooks, and laboratory experiments for a comprehensive learning experience. Encourage collaborative discussions among students to enhance comprehension and critical thinking. With these strategies and resources, mastering gene expression transcription becomes an achievable and engaging goal for students, fostering a lasting understanding of one of biology's most vital processes. QuestionAnswer What is the main purpose of the Pogil activity on gene expression transcription? The purpose is to help students understand the process of transcription, how genes are expressed, and the key components involved in transcription within cells. How does transcription differ from translation in gene expression? Transcription is the process of copying a gene's DNA sequence into mRNA, while translation is the process of decoding the mRNA to assemble a protein. What role do RNA polymerase enzymes play in transcription? RNA polymerase enzymes are responsible for synthesizing the mRNA strand by reading the DNA template strand during transcription. Why is understanding gene expression important in biology? Understanding gene expression helps explain how cells differentiate, respond to environmental signals, and how genetic information leads to specific traits and functions. What are the key steps involved in the process of transcription as outlined in the Pogil activity? The key steps include initiation (binding of RNA polymerase to DNA), elongation (synthesis of mRNA), and termination (release of mRNA from the DNA template). 5 How does the Pogil answer key assist students in mastering gene transcription concepts? The answer key provides detailed explanations and correct responses to guide students in understanding each step and concept involved in transcription. What are some common misconceptions about gene transcription that the Pogil activity aims to address? Common misconceptions include confusing transcription with translation, thinking DNA is directly translated into proteins without mRNA, and misunderstanding the roles of different enzymes involved. How can understanding the Pogil answer key improve students' grasp of gene regulation? It clarifies the mechanisms controlling gene expression, including how transcription factors and environmental signals influence transcription levels. Are there visual aids or diagrams included in the Pogil activity to help explain transcription? Yes, the activity typically includes diagrams and visual aids that illustrate the steps of transcription and the components involved, enhancing comprehension. How does practicing with the Pogil answer key prepare students for assessments on gene expression? It helps students reinforce key concepts, understand correct procedures, and develop confidence in explaining the transcription process, leading to better performance on tests and quizzes. Gene Expression Transcription Pogil Answer Key: A Comprehensive Guide to Mastering the Fundamentals Understanding gene expression transcription pogil answer key is essential for students and educators aiming to grasp the intricacies of molecular biology. This resource serves as a valuable tool in reinforcing concepts related to how genes are transcribed into RNA, a fundamental step in gene expression. Whether you're navigating your way through a Pogil activity or seeking clarity on transcription mechanisms, this guide will provide a thorough breakdown of key concepts, common questions, and strategies for mastering the material. --- What Is Gene Expression and Why Is Transcription Important? Gene expression is the process by which information from a gene is used to synthesize functional gene products, typically proteins. It involves multiple steps, with transcription being the first and crucial phase. During transcription, a segment of DNA is transcribed into messenger RNA (mRNA), which then serves as a template for protein synthesis during translation. Key points: - Transcription converts genetic information from DNA to RNA. - It occurs in the nucleus of eukaryotic cells and the cytoplasm of prokaryotic cells. - It is tightly regulated to ensure proper cellular function and response to environmental signals. --- The Structure of a Transcription Pogil Activity A typical Pogil activity on transcription guides students through: - The identification of the roles of different DNA regions (promoters, coding regions, terminators). - The understanding of how RNA polymerase initiates and elongates the mRNA strand. - The differentiation between DNA and RNA nucleotide pairing. - The recognition of factors influencing transcription regulation. An answer key for such activities provides detailed Gene Expression Transcription Pogil Answer Key 6 explanations for each question, clarifying misconceptions and reinforcing correct understanding. --- Key Concepts in Transcription Covered by the Pogil Answer Key 1. The Role of Promoters in Transcription Initiation Promoters are specific DNA sequences located upstream of the gene that signal RNA polymerase where to begin transcription. The most common promoter in eukaryotes is the TATA box. Important points: - Promoters contain specific sequences recognized by transcription factors. - Binding of RNA polymerase and transcription factors to the promoter forms the transcription initiation complex. - The promoter position determines where the RNA synthesis begins. 2. The Process of RNA Polymerase Binding and Initiation RNA polymerase binds to the promoter region, unwinding the DNA strands to access the template strand. The process involves: - Formation of the transcription initiation complex. - Initiation of RNA synthesis at the +1 site. - Directionality of transcription (from 3′ to 5′ on DNA, producing 5′ to 3′ mRNA). 3. Elongation of the mRNA Strand

Once initiated, RNA polymerase moves along the DNA, synthesizing the mRNA complementary to the DNA template strand: - Nucleotides are added in the 5′ to 3′ direction. - Base pairing rules: A with U (in RNA), T with A, C with G, G with C. - The DNA unwinds ahead of the polymerase and rewinds behind it. 4. Termination of Transcription In prokaryotes, specific sequences signal the end of transcription, causing the RNA polymerase to detach. In eukaryotes, transcription termination involves complex mechanisms, including cleavage and polyadenylation signals. --- Using the Pogil Answer Key Effectively Step-by-Step Approach: 1. Review the Activity: Read through the initial questions to assess your understanding. 2. Attempt the Questions Independently: Try answering before consulting the answer key. 3. Compare and Learn: Use the answer key to check your responses, paying attention to explanations and reasoning provided. 4. Clarify Misconceptions: Focus on questions where your answers differed from the key. 5. Summarize Key Takeaways: Write down important points to reinforce learning. Common Questions and Their Clarified Answers | Question | Typical Student Answer | Correct Explanation (from Answer Key) | |------------|--------------------------|--------------------------------------- -| | What is the function of the promoter? | It helps RNA polymerase bind to DNA. | Correct! The promoter contains specific sequences that attract RNA polymerase and initiate transcription. | | Why does RNA polymerase only synthesize in the 5′ to 3′ direction? | Because of the way nucleotide bases pair. | Yes, the enzyme adds nucleotides to the 3′ end of the growing RNA, so synthesis proceeds 5′ to 3′. | | What signals the end of transcription? | A stop codon. | Not quite. Stop codons signal the end of translation, not transcription. In prokaryotes, specific terminator sequences signal termination; in eukaryotes, polyadenylation signals are involved. | --- Strategies for Mastering Gene Transcription - Visualize the Process: Use diagrams and models to understand how RNA polymerase interacts with DNA. - Memorize Key Terms: Promoter, terminator, transcription factors, RNA polymerase, template strand. - Relate Concepts: Connect transcription to translation, gene regulation, and overall gene expression. - Practice with Flashcards: Gene Expression Transcription Pogil Answer Key 7 Reinforce vocabulary and process steps. - Work Through Practice Questions: Use the Pogil answer key to check understanding and develop confidence. --- Common Mistakes and How to Avoid Them - Confusing DNA and RNA roles: Remember DNA is the template, and RNA is the product. - Misunderstanding directionality: Always think in terms of 5′ to 3′ synthesis. - Overlooking regulation points: Recognize the importance of promoters, enhancers, and repressors. - Assuming eukaryotic and prokaryotic transcription are identical: Understand differences in mechanisms and regulation. --- Conclusion: Mastering Transcription with the Pogil Answer Key The gene expression transcription pogil answer key is an invaluable resource for students seeking a deeper understanding of how genetic information is transcribed into RNA. By actively engaging with the questions, reviewing detailed explanations, and applying strategies to reinforce learning, students can confidently grasp the fundamental mechanisms of transcription. Remember, mastering these concepts not only enhances exam performance but also builds a strong foundation for exploring more advanced topics in molecular biology and genetics. --- Final Tips for Success - Regularly revisit key concepts and diagrams. - Collaborate with peers to discuss challenging questions. - Seek clarification from teachers or tutors when needed. - Connect transcription to broader biological systems and processes. Embark on your learning journey with confidence, knowing that resources like the gene expression transcription pogil answer key are designed to guide you toward mastery. gene expression, transcription, pogil, answer key, biological processes, DNA, RNA, protein synthesis, genetics, learning resources

Gene TranscriptionTranscription and translationTranscriptionTranscription FactorsIn Vitro Transcription and Translation ProtocolsElucidation of Regulatory Mechanisms of Transcription and Coupled MRNA ExportGene TranscriptionGene TranscriptionTranscriptionTranscription and TranslationTranscription FactorsInferring Properties of Transcription from Stochastic Gene ExpressionNon-canonical Transcription Initiation: the Expanding Universe of Transcription Initiating SubstratesSingle Molecule and Synthetic Biology Studies of TranscriptionIs Symmetrical Transcription a General Mechanism of Gene Expression?Measuring Gene Expression NoiseA Laboratory Guide to in Vitro TranscriptionA Factor Analysis Approach to Transcription Regulatory Network Reconstruction Using Gene Expression DataA Factor Analysis Approach to Transcription Regulatory Network Reconstruction Using Gene Expression DataA Factor Analysis Approach to Transcription Regulatory Network Reconstruction Using Gene Expression Data R. J. White U Satyanarayana William M. Brown Guido Grandi Geetha Durairaj Steve J. Higgins B. D. Hames Ronald C. Conaway B. D. Hames Paul J. Higgins Marc Samuel Sherman Bradley Michael Zamft Yaffa Beck Albert Lin Felipe Sierra Wei Chen (Ph. D.) □ □ Wei Chen

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Substrates Single Molecule and Synthetic Biology Studies of Transcription Is Symmetrical Transcription a General Mechanism of Gene Expression? Measuring Gene Expression Noise A Laboratory Guide to in Vitro Transcription A Factor Analysis Approach to Transcription Regulatory Network Reconstruction Using Gene Expression Data A Factor Analysis Approach to Transcription Regulatory Network Reconstruction Using Gene Expression Data A Factor Analysis Approach to Transcription Regulatory Network Reconstruction Using Gene Expression Data *R. J. White U Satyanarayana William M. Brown Guido Grandi Geetha Durairaj Steve J. Higgins B. D. Hames Ronald C. Conaway B. D. Hames Paul J. Higgins Marc Samuel Sherman Bradley Michael Zamft Yaffa Beck Albert Lin Felipe Sierra Wei Chen (Ph. D.) Wei Chen*

TRANSCRIPTION IS THE FOCUS OF MUCH CUTTING EDGE RESEARCH AS BEFITS ITS ESSENTIAL PLACE IN BIOLOGY THE ESTABLISHED LINK BETWEEN DEFECTS IN GENE TRANSCRIPTION AND MANY HUMAN DISORDERS HAS FUELLED CONSIDERABLE ACTIVITY IN THE BIOMEDICAL ARENA PARTICULARLY CANCER RESEARCH THIS CONCENTRATION OF ATTENTION HAS UNCOVERED A MYRIAD OF FACTORS INVOLVED IN TRANSCRIPTION AND THE LITERATURE IS NOW RIFE WITH JARGON AND COMPLEXITY GENE TRANSCRIPTION MECHANISMS AND CONTROL AIMS TO DEMYSTIFY THE SUBJECT FOR A NON EXPERT AUDIENCE PROVIDING A GUIDED TOUR AROUND THE COMPLEX MACHINERY OF THE TRANSCRIPTIONAL APPARATUS AND DISCUSSING HOW THE VARIOUS FACTORS ACHIEVE THEIR FUNCTIONS BY FOCUSING ON GENERAL PRINCIPLES AND ILLUSTRATING THESE WITH A SELECT GROUP OF EXAMPLES MANY OF WHICH ARE LINKED TO HUMAN DISEASES THE AUTHOR CONVEYS THE INTRICACIES OF TRANSCRIPTIONAL CONTROL IN AN ACCESSIBLE MANNER WITH THE FIRST CHAPTER PRESENTING AN OVERVIEW OF GENE EXPRESSION THIS IS A STAND ALONE TEXT IDEAL FOR ADVANCED LEVEL UNDERGRADUATES AND POSTGRADUATES IN BIOLOGY BIOCHEMISTRY AND MEDICAL SCIENCES IT WILL ALSO APPEAL TO RESEARCH SCIENTISTS WHO REQUIRE A BROAD CURRENT PERSPECTIVE ON THIS RAPIDLY MOVING AND COMPLEX FIELD PROVIDES A BROAD AND ACCESSIBLE INTRODUCTION TO GENE TRANSCRIPTION UP TO DATE COVERAGE OF THE MAJOR TOPICS IN A RAPIDLY EVOLVING FIELD ILLUSTRATES THE LINKS BETWEEN ABERRANT TRANSCRIPTION AND HUMAN DISEASE EXPLAINS THE JARGON ASSOCIATED WITH TRANSCRIPTION FACTORS

TRANSCRIPTION AND TRANSLATION TRANSCRIPTION AND TRANSLATION

KNOWLEDGE OF TRANSCRIPTION HAS MOVED FORWARD AT A FURIOUS PACE OVER RECENT YEARS AND AN UNDERSTANDING OF THE PROCESSES INVOLVED IN GENE REGULATION AND EXPRESSION HAS BECOME AN ESSENTIAL ELEMENT IN BIOCHEMISTRY GENOME BIOLOGY MOLECULAR BIOLOGY AND MOLECULAR GENETICS IN THIS TIMELY BOOK THE AUTHORS PRESENT AN ACCESSIBLE YET COMPREHENSIVE COVERAGE SUITABLE FOR STUDENTS AT A SENIOR UNDERGRADUATE LEVEL AND FOR POSTGRADUATES NEEDING AN OVERVIEW OF THE CURRENT STATE OF PLAY IT COVERS A NUMBER OF PERTINENT EXAMPLES OF TRANSCRIPTION SYSTEMS FOR EUKARYOTES AND PROKARYOTES INDICATES METHODS FOR STUDYING TRANSCRIPTION AND SURVEYS THE WHOLE TOPIC OF TRANSCRIPTION FROM MANY PERSPECTIVES

SINCE THE PUBLICATION OF THE FIRST EDITION FIVE YEARS AGO A WIDE RANGE OF NEW METHODOLOGIES HAVE BEEN DEVELOPED TO FACILITATE STUDIES ON BOTH ISOLATED PARTS OF THE GENOME AND THE GENOME AS A WHOLE THIS NEW EDITION HAS BEEN UPDATED AND EXPANDED SO THAT IT PROVIDES A COMPREHENSIVE GUIDE TO THE METHODS CURRENTLY AVAILABLE TO CHARACTERIZE THE FUNCTION AND ACTIVITY OF AN INDIVIDUAL TRANSCRIPTION FACTOR ALL THE ORIGINAL CHAPTERS HAVE BEEN FULLY UPDATED OR REWRITTEN AND ADDITIONAL CHAPTERS COVER THE USE OF IN VITRO TRANSCRIPTION ASSAYS ANALYSIS OF CHROMATIN STRUCTURE USE OF THE GENOMIC BINDING SITE ASSAY AND ANALYSIS OF TRANSCRIPTION FACTOR MODIFICATIONS AS WITH THE PREVIOUS EDITION THE BOOK STARTS WITH A SERIES OF CHAPTERS CONCERNED WITH CHARACTERIZING THE PROTEINS BINDING TO A SPECIFIC DNA SEQUENCE AND THEN A CHAPTER ON MORE DETAILED CHARACTERIZATION OF THE PROTEIN ITSELF THE NEXT TWO CHAPTERS DESCRIBE THE ISOLATION OF CDNA CLONES ENCODING A TRANSCRIPTION FACTOR USING OLIGONUCLEOTIDES PREDICTED FROM PROTEIN SEQUENCE AND SCREENING OF A CDNA EXPRESSION LIBRARY CHAPTER 6 DEALS WITH IDENTIFICATION OF TRANSCRIPTION FACTORS BASED ON SEQUENCE HOMOLOGY ANALYSIS BY BOTH EXPERIMENTAL SCREENING AND DATABASE SEARCHES CHAPTER 7 IS A NEW CHAPTER THAT DESCRIBES METHODS OF IDENTIFYING THE TARGET GENES OF A PREVIOUSLY UNCHARACTERIZED FACTOR THE NEXT CHAPTERS DEAL WITH ANALYSIS OF TRANSCRIPTION FACTOR FUNCTION CHAPTER 8 DEALS WITH GENERAL TECHNIQUES AND THEN THE FOLLOWING CHAPTERS COVER THE SPECIALIZED TECHNIQUES OF IN VITRO TRANSCRIPTION ASSAYS USING TRANSCRIPTIONALLY ACTIVE NUCLEAR EXTRACTS DERIVED FROM RAT BRAIN AND ANALYSIS OF THE EFFECT OF TRANSCRIPTION FACTORS ON CHROMATIN STRUCTURE THE FINAL CHAPTER DESCRIBES METHODS FOR DETECTING THE PHOSPHORYLATION AND GLYCOSYLATION STATE OF TRANSCRIPTION FACTORS

THIS BOOK IS A HIGHLY ANTICIPATED UPDATE OF THE PREVIOUS EDITION IT PROVIDES MOLECULAR BIOLOGY LABORATORIES WITH THE MOST POWERFUL TECHNIQUES FOR EXPLOITING IN VITRO TRANSCRIPTION AND TRANSLATION SYSTEMS IT HAS BEEN COMPLETELY UPDATED WITH NEW CHAPTERS AND TOPICS

GENE EXPRESSION IS DRIVEN BY THE COMBINED ACTIONS OF NUMEROUS PROTEINS THAT ACT IN A REGULATED FASHION ONE OF THE CENTRAL PROCESSES IN GENE EXPRESSION IS THE EXPORT OF THE SYNTHESIZED MRNA FROM NUCLEUS TO the cytoplasm where it gets translated into proteins this journey of mrna is mediated by numerous factors and receptors that get loaded onto the transcribing mrna in a co transcriptional fashion mutations of many export factors are directly linked to various diseases thereby stressing the importance of tight regulation in mrna export thus one of my thesis aims had been focused on understanding the regulatory mechanisms of mrna export by an f box protein mdm30 two recent studies have implicated the role of ubiquitylation in mrna export via hect domain containing e3 ligases like tom1 and rsp5 my work has revealed the role of an f box protein mdm30 component of ring domain containing scf skp cullin f box family of e3 ligases in mrna export thereby implicating the role of ubiquitylation in export via non hect e3 ligase for the first time my results show that mdm30 ubiquitylates sub2 an integral component of the trex transcription export complex which is involved in mediating proper nuclear export of mrna in addition to its role in mrna export trex complex is also involved in transcription hence during the course of my research i have also worked on understanding the regulation of transcription especially we focused on the role of sus1 a component of trex 2 complex in regulation of transcription further since sense transcription and coupled mrna export is likely to be regulated by antisense transcripts or antisense transcription it is important to understand how antisense transcription is regulated to ultimately control mrna export therefore i have also worked on understanding the regulation of antisense transcription our results show that activator and gtfs general transcription factors are involved in antisense transcription initiation however antisense transcription is initiated independently of the sense transcription collectively the outcomes of my thesis research provide important insights on the regulatory mechanisms of transcription and coupled mrna export

this volume provides detailed practical guidance on some of the most important techniques relating to gene transcription the mechanisms of this research should be of interest to researchers in cell and molecular biology

presents a coherent account of many productive lines of investigation organized as a series of mini reviews that focus on major research areas including studies on the structure and mechanisms of action of bacterial viral and eukaryotic rna polymerases and the transcription factors that control their activities each review provides a brief but up to date account of the progress of research in a particular area a discussion of the major issues and questions driving that research and a brief description of the evolving approaches and technologies used to address those questions annotation copyright by book news inc portland or

in the last few years significant breakthroughs in transcription research expanded our appreciation for the complexity of molecular controls on gene expression in mammalian cells in transcription factors methods and protocols experts in the field describe state of the art approaches that investigators can use to probe critical mechanisms underlying transcription factor nuclear cytoplasmic trafficking as well as to assess the functional impact of post translational modifications on transcription factor function the chapters are written by prominent scientists many of whom developed these methods and highlight protocols that focus on specific transcription factor family members with particular relevance to human disease composed in the highly successful methods in molecular biologytm series format each chapter contains a brief introduction step by step methods a list of necessary materials and a notes section which shares tips on troubleshooting and avoiding known pitfalls comprehensive and current transcription factors methods and protocols compiles the latest techniques for elucidating controls on transcription factor intracellular localization and activity and consequently is unlike any other methods based text on transcriptional regulation today

single cell experiments reveal considerable fluctuations in the expression of both mrna and protein among genetically identical cells these fluctuations manifest as remarkably broad distributions of expression what does the nature of stochastic expression intimate about the mechanisms underlying transcriptional regulation to understand how specific cis regulatory mechanisms manifest as noise in expression we tackled two key methodological deficiencies 1 a lack of assumption free approaches for fitting mechanistic models to protein distributions and 2 methods for isolating biochemical noise intrinsic from noise due to cellular environment extrinsic to fit the standard stochastic protein model without assumptions we constrained a search algorithm with solutions to the model s higher order moments the resulting algorithm enables efficient discovery of solution ensembles representing every kinetic scheme consistent with an observed protein distribution i find that measurement of protein and mrna degradation rates should permit estimation of the macroscopic rate constants governing gene on off transitions transcription and translation from distribution shape alone i also found that higher order moments of intrinsic noise separate naturally from their extrinsic counterparts in principle enabling intrinsic stochasticity to be estimated by comparing expression of strains containing a variable number of identical genes to test both frameworks we assembled s cerevisiae strains expressing one or multiple copies of gfp reporter genes driven by the heat shock responsive promoter ssa1 in contrast to previous studies we find that stochastic expression from ssa1 resists decomposition into intrinsic and extrinsic components degradation rates appear constant across the population while transcription rates vary extensively with cellular volume leading us to predict that a large fraction of noise arises from extrinsic mrna fluctuations consistent with this hypothesis perturbations to transcription rate dramatically impact the balance of protein noise together these data argue for models of stochastic expression that explicitly incorporate fluctuating inputs into transcription

rna polymerase can utilize coenzymes such as nad as initiating substrates in vitro and also likely in vivo and this has implications for regulation of gene expression

the horizons of biology are ever expanding from the discernment of the detailed mechanisms of enzyme function to the manipulation of the physiological processes of whole organisms and ecosystems single molecule studies allow for the characterization of the individual processes that comprise an enzyme s mechanochemical cycle through standardization and generalization of biological techniques components and knowledge synthetic biology seeks to expand the scale of biological experiments and to usher in an age of biology as a true engineering science in which those studying different hierarchical levels of sophistication need not start from the fundamental biochemical principles underlying all biological experiments here we report our findings on the processes governing transcription and its role in gene expression through the use of both single molecule and synthetic biology methods we have established a promoter free factor free method of initiation of transcription by the mitochondrial rna polymerase in saccharomyces cerevisiae rpo41 through the use of synthetic oligonucleotides to imitate the hybridization geometry of rpo41 during active transcription using this system we have established that a sub micromolar ntp concentration is appropriate for non saturating transcriptional runoff assays we have optimized the transcription buffer and found that 10 mm mgcl2 40 mm kcl and 10 mm dtt are sufficient for robust transcription stability studies show that rpo41 loses approximately 30 of its activity during each freeze thaw cycle and that the pre formed elongation complex loses transcriptional activity with a half life of 7 4 1 5 hr through the use of optical trapping techniques we have established a method to monitor the transcription of individual rpo41 molecules in real time this has allowed us to measure the kinetic rates of nucleotide incorporation by the enzyme km 22 13 mm 1 and vmax 25 2 5 bp s both of these rates are more similar to those of the main nuclear rna polymerase in the same organism rna polymerase ii pol ii than to that of the t7 rna polymerase despite the fact that rpo41 is a single subunit rna polymerase with homology to those of the t odd bacteriophage and no discernable homology to pol ii furthermore like pol ii and the e coli rna polymerase transcription by rpo41 consists of periods of processive transcription interspersed with periods of pausing we have also observed retrograde motion of rpo41 during pauses termed backtracking a process that has not been reported in phage like rna polymerases we have performed single molecule assays of transcription by both pol ii and rpo41 on templates of differing base pair composition and found that in general the characteristics of pausing are attenuated in templates of higher gc content specifically the frequency of pausing is decreased in gc rich templates as is the average pause duration the distribution of pause durations is correspondingly shifted to shorter pauses on gc rich templates we discuss two mechanisms by which template composition may affect pausing 1 movement of the backtracked transcription bubble is affected by differences in the base stacking energies from the disrupted created dna dna and rna dna base pairs at the ends of the bubble and 2 secondary structure of the nascent rna upstream of the backtracked transcription bubble imposes an energetic barrier to its backward movement we give in silico evidence that it is the latter mechanism incorporation of this secondary structure energy barrier an energy penalty into a model of transcriptional pausing by backtracking allows for statistical fits of the mean pause densities mean pause durations and the distribution of pause durations for each enzyme on each template furthermore incorporation of the energy penalty allows for fitting of the pause characteristics for a given enzyme using a single enzyme specific hopping rate k0 that is independent of template and a single template dependent energy penalty term delta grna which is enzyme independent for rpo41 we find that k0 the hopping rate of the backtracked enzyme along dna without rna secondary structure is 5 4 1 8 s 1 while it is 2 9 0 3 s 1 for pol ii furthermore the average energy penalty due to the nascent rna delta grna on the at rich template used in this study is 0 7 0 1 kt while it is 0 8 0 1 kt for random dna and 1 0 0 1 kt for gc rich dna in order to confirm that it is the secondary structure of the rna that is the cause of the energy penalty we performed the same single molecule transcription assays in the presence of rnase a an enzyme that digests unprotected rna in both single stranded and double stranded form the pausing characteristics of all traces on all templates in the presence of rnase a are statistically indistinguishable from those on at rich dna without rnase indicating that the rnase digested enough of the nascent rna to disrupt any secondary structure protection of the 5 region of

THE NASCENT RNA BY STERIC INTERACTIONS BETWEEN THE POLYMERASE AND THE RNASE PREVENTED FULL DEGRADATION OF THE RNA AND THUS ALLOWED FOR SOME BACKTRACKING THIS STRONGLY SUPPORTS THE NEW MODEL PRESENTED HERE OF MODULATION OF TRANSCRIPTIONAL PAUSING BY SECONDARY STRUCTURE OF THE NASCENT RNA IN CONTRAST TO THE DETAILED AND ISOLATED NATURE OF SINGLE MOLECULE TRANSCRIPTION WE ALSO PERFORMED A SYNTHETIC BIOLOGY PROJECT INVOLVING RPO41 THE INTENT OF THIS PROJECT WAS TO INVESTIGATE THE PLAUSIBILITY OF THE CREATION OF A TRANSCRIPTIONALLY INDEPENDENT MITOCHONDRION AND BY EXTENSION A MINIMAL CELL BY MOVEMENT OF THE MITOCHONDRIAL TRANSCRIPTIONAL MACHINERY FROM THE NUCLEAR TO THE MITOCHONDRIAL GENOME THUS WE PERFORMED IN VIVO MITOCHONDRIAL TRANSFORMATION OF YEAST CELLS WITH A SYNTHETIC CONSTRUCT CONTAINING THE GENE ENCODING FOR RPO41 WE REPORT THAT WE HAVE SUCCESSFULLY INTEGRATED SAID SYNTHETIC GENE INTO THE MITOCHONDRIAL GENOME AND HAVE SEEN ITS EXPRESSION TO THE TRANSCRIPTIONAL LEVEL FURTHERMORE WE ARE FAIRLY CONFIDENT THAT THE FULL INTACT MRNA OF THE SYNTHETIC GENE IS BEING CREATED WITHIN THE MITOCHONDRIAL MATRIX WE HAVE NOT BEEN ABLE TO DETECT EXPRESSION OF THE PROTEIN PRODUCT OF THE INTEGRATED SYNTHETIC CONSTRUCT NOR HAVE WE BEEN ABLE TO ISOLATE A STRAIN THAT EXHIBITS ITS EXPRESSION IN THE ABSENCE OF THE WILD TYPE NUCLEAR COPY BECAUSE THE LENGTH OF RPO41 IS LONGER THAN ANY OTHER PROTEIN SYNTHESIZED WITHIN THE MITOCHONDRIAL ORGANELLE WE HAVE BEGUN EXPERIMENTS TO DETERMINE THE MAXIMAL POLYPEPTIDE LENGTH ABLE TO BE TRANSLATED BY THE MITOCHONDRIAL RIBOSOME AND ASSOCIATED COFACTORS

THIS DISSERTATION A FACTOR ANALYSIS APPROACH TO TRANSCRIPTION REGULATORY NETWORK RECONSTRUCTION USING GENE EXPRESSION DATA BY WEI CHEN ⍰ ⍰ WAS OBTAINED FROM THE UNIVERSITY OF HONG KONG POKFULAM HONG KONG AND IS BEING SOLD PURSUANT TO CREATIVE COMMONS ATTRIBUTION 3 0 HONG KONG LICENSE THE CONTENT OF THIS DISSERTATION HAS NOT BEEN ALTERED IN ANY WAY WE HAVE ALTERED THE FORMATTING IN ORDER TO FACILITATE THE EASE OF PRINTING AND READING OF THE DISSERTATION ALL RIGHTS NOT GRANTED BY THE ABOVE LICENSE ARE RETAINED BY THE AUTHOR ABSTRACT RECONSTRUCTION OF TRANSCRIPTION REGULATORY NETWORK TRN AND TRANSCRIPTION FACTOR ACTIVITY TFA FROM GENE EXPRESSION DATA IS AN IMPORTANT PROBLEM IN SYSTEMS BIOLOGY CURRENTLY THERE EXIST VARIOUS FACTOR ANALYSIS METHODS FOR TRN RECONSTRUCTION BUT MOST APPROACHES HAVE SPECIFIC ASSUMPTIONS NOT SATISFIED BY REAL BIOLOGICAL DATA NETWORK COMPONENT ANALYSIS NCA CAN HANDLE SUCH LIMITATIONS AND IS CONSIDERED TO BE ONE OF THE MOST EFFECTIVE METHODS THE PREREQUISITE FOR NCA IS KNOWLEDGE OF THE STRUCTURE OF TRN SUCH STRUCTURE CAN BE OBTAINED FROM CHIP CHIP OR CHIP SEQ EXPERIMENTS WHICH HOWEVER HAVE QUITE LIMITED APPLICATIONS IN ORDER TO COPE WITH THE DIFFICULTY WE RESORT TO HEURISTIC OPTIMIZATION ALGORITHM SUCH AS PARTICLE SWARM OPTIMIZATION PSO IN ORDER TO EXPLORE THE POSSIBLE STRUCTURES OF TRN AND CHOOSE THE MOST PLAUSIBLE ONE REGARDING THE STRUCTURE ESTIMATION PROBLEM WE EXTEND CLASSICAL PSO AND PROPOSE A NOVEL PROBABILISTIC BINARY PSO FURTHERMORE AN IMPROVED NCA CALLED FASTNCA IS ADOPTED TO COMPUTE THE OBJECTIVE FUNCTION ACCURATELY AND FAST WHICH ENABLES PSO TO RUN EFFICIENTLY SINCE HEURISTIC OPTIMIZATION CANNOT GUARANTEE GLOBAL CONVERGENCE WE RUN PSO MULTIPLE TIMES AND INTEGRATE THE RESULTS THEN GCV LASSO GENERALIZED CROSS VALIDATION LEAST ABSOLUTE SHRINKAGE AND SELECTION OPERATOR IS PERFORMED TO ESTIMATE TRN WE APPLY OUR APPROACH AND OTHER FACTOR ANALYSIS METHODS ON THE SYNTHETIC DATA THE RESULTS INDICATE THAT THE PROPOSED PSOFASTNCA GCV LASSO ALGORITHM GIVES BETTER ESTIMATION IN ORDER TO INCORPORATE MORE PRIOR INFORMATION ON TRN STRUCTURE AND GENE EXPRESSION DYNAMICS IN THE LINEAR FACTOR ANALYSIS MODEL FOR IMPROVED ESTIMATION OF TRN AND TFAS A LINEAR BAYESIAN FRAMEWORK IS ADOPTED UNDER THE UNIFIED BAYESIAN FRAMEWORK BAYESIAN LINEAR SPARSE FACTOR ANALYSIS MODEL BLSFM AND BAYESIAN LINEAR STATE SPACE MODEL BLSSM ARE DEVELOPED FOR INSTANTANEOUS AND DYNAMIC TRN RESPECTIVELY VARIOUS APPROACHES TO INCORPORATE PARTIAL AND AMBIGUOUS PRIOR NETWORK STRUCTURE INFORMATION IN THE BAYESIAN FRAMEWORK ARE PROPOSED TO IMPROVE PERFORMANCE IN PRACTICAL APPLICATIONS FURTHERMORE WE PROPOSE A NOVEL MECHANISM FOR ESTIMATING THE HYPER PARAMETERS OF THE DISTRIBUTION PRIORS IN OUR BLSFM AND BLSSM MODELS WHICH CAN SIGNIFICANTLY IMPROVE THE ESTIMATION COMPARED TO TRADITIONAL WAYS OF HYPER PARAMETER SETTING WITH THIS DEVELOPMENT REASONABLY GOOD ESTIMATION OF TFAS AND TRN CAN BE OBTAINED EVEN WITHOUT USE OF ANY STRUCTURE PRIOR OF TRN EXTENSIVE NUMERICAL EXPERIMENTS ARE PERFORMED TO INVESTIGATE OUR DEVELOPED METHODS UNDER VARIOUS SETTINGS WITH COMPARISON TO SOME EXISTING ALTERNATIVE APPROACHES IT IS DEMONSTRATED THAT OUR HYPER PARAMETER ESTIMATION METHOD IMPROVES THE ESTIMATION OF TFA AND TRN IN MOST SETTINGS AND HAS SUPERIOR PERFORMANCE AND THAT STRUCTURE PRIORS IN GENERAL LEADS TO IMPROVED ESTIMATION PERFORMANCE REGARDING APPLICATION TO REAL BIOLOGICAL DATA WE EXECUTE THE PSO FASTNCAGCV LASSO ALGORITHM DEVELOPED IN THE THESIS USING E COLI MICROARRAY DATA AND OBTAIN SENSIBLE ESTIMATION OF TFAS AND TRN WE APPLY BLSFM WITHOUT STRUCTURE PRIORS OF TRN BLSSM WITHOUT STRUCTURE PRIORS AS WELL AS WITH PARTIAL STRUCTURE PRIORS TO YEAST S CEREVISIAE MICROARRAY DATA AND OBTAIN A REASONABLE ESTIMATION OF TFAS AND TRN DOI 10 5353 TH B4961778 SUBJECTS FACTOR ANALYSIS GENETIC TRANSCRIPTION REGULATION STATISTICAL METHO

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