

Cell-type-specific germline immune variants as predictors of immunotherapy response

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Inherited genetic variation in the immune system influences cancer progression and patient response to immunotherapy, but the specific immune cell populations mediating these effects remain unclear. This project aims to investigate how germline immune regulatory variants contribute to immunotherapy response by integrating cell-type-specific expression quantitative trait loci (eQTLs) and chromatin accessibility data to identify the immune cell populations most relevant to treatment response. eQTLs from the Database of Immune Cell Expression (DICE) and OneK1K datasets were filtered to retain variants associated with immune regulatory genes, and linkage disequilibrium expansion was performed to identify linked regulatory variants. The expanded variants were integrated with immune cell-type-specific chromatin accessibility data generated by the assay for transposase-accessible chromatin using sequencing (ATAC-seq). Statistical testing of accessibility across stimulated immune cell types identified 647 variants located in regions with cell-type-specific patterns of chromatin accessibility. After matching these variants to an immunotherapy-treated patient cohort and filtering, 614 variants were retained for machine learning analysis. XGBoost models were trained using all variants and cell-type-specific variant sets to predict immunotherapy response. Cross-validation showed modest predictive signal from germline variation alone, with a tuned all-variant area under the receiver operating characteristic curve (AUC) of 0.611 and cell-type-specific models with the highest cross-validation AUCs reaching approximately 0.60. Model performance was positively associated with the number of variants included in each cell-type feature set, suggesting that differences in feature-set size may contribute to observed differences in performance between cell types. These results suggest that cell-type-specific germline regulatory variation may be relevant to immunotherapy response, but further validation is needed to determine whether particular immune cell populations provide unique predictive information.

Toward a mechanistic understanding of Daple regulation by cytoplasmic dynein-1

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The spatiotemporal regulation of signaling pathways via G proteins is essential for healthy cellular function. In addition to G protein-coupled receptors, non-receptor activators have emerged as important regulators of G protein signaling. One such activator, Daple, facilitates guanine exchange in G proteins, thereby regulating noncanonical Wnt signaling, a pathway critical for cell polarity, migration, and cancer progression. However, the mechanisms governing Daple's regulation, subcellular localization and signaling activity remain poorly understood. Recent work from the Kendrick lab identified Daple as a cytoplasmic dynein-1 (dynein) activating adaptor, potentially linking dynein transport to cell signaling. This project investigates the structural basis of Daple-G protein binding as well as the functional consequences of Daple-dynein interactions on G protein activation. Using bioluminescence resonance energy transfer (BRET) assays and cross-linking approaches we examined how mutations that disrupt Daple-dynein interactions affect the separation of G protein subunits. We found that disruption of the Daple-dynein interaction reduced Daple-mediated G protein activation, while optimization of BRET conditions enabled consistent detection of this activity. Additionally, we established conditions for cross-linking Daple-Gai3 complexes, providing a foundation for future structural analysis of this interaction. These advances provide an experimental pipeline to define how dynein-mediated transport spatially regulates Daple-mediated G protein activation.

Opposing regulation of androgen receptor stability by RNF113A and PSMD14

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Uncontrolled androgen receptor (AR) signaling is a major driver of prostate cancer. Although androgen deprivation therapy initially suppresses advanced disease, nearly all patients with metastatic prostate cancer eventually develop castration-resistant disease. Defining mechanisms that regulate AR protein stability may reveal new therapeutic opportunities. Unbiased genetic screens identified the E3 ligase RNF113A as a driver of AR proteasomal degradation, and PSMD14, a deubiquitinating proteasomal subunit, as a stabilizer of agonist-bound AR. In purified-protein pulldown assays, androgen-response element DNA reduced AR-RNF113A binding, suggesting that RNF113A recognizes the AR DNA-binding domain (DBD). AlphaFold 3 modeling predicted specific RNF113A-contacting residues within the AR DBD. Mutating these residues prevented RNF113A binding, whereas transplanting them onto the estrogen receptor increased RNF113A binding, further localizing the recognition site to the AR DBD. To evaluate PSMD14-mediated regulation, we measured AR ubiquitination following chemical inhibition of PSMD14, which restored agonist-bound AR ubiquitination and degradation. We then combined endogenous PSMD14 knockdown with overexpression of PSMD14 point mutants. Flow cytometric analysis of an AR degradation reporter identified candidate residues required for receptor stabilization. Together, these findings support an androgen-dependent RNF113A-PSMD14 axis that regulates AR ubiquitination and stability.

SNAP: An SSL-based neural network for automated pseudo-time generation

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Infertility affects one in six people worldwide, yet fertility clinics cannot reliably predict which embryos will develop successfully for in-vitro fertilization (IVF). Embryo viability is assessed using preimplantation genetic testing, which requires an invasive biopsy and can be confounded by mosaicism, or by morphological grading, which is subjective and shows low inter-observer agreement for some features. Computer vision offers a non-invasive and objective alternative by extracting quantitative developmental information directly from embryo images. Our long-term aim is to read developmental potential from a single image. Here, we address a critical first step: determining whether developmental time itself can be recovered from static embryo morphology. Cleavage timing correlates with embryo viability and, unlike viability, can be directly measured from time-lapse recordings, providing an objective ground truth for model training. We therefore ask whether a single label-free image can reveal where a zygote lies within its first cell cycle and how accurately this information can be recovered relative to existing approaches applied to identical data. We find that a single image can classify an embryo's cleavage timing to within 1.3 hours across the roughly 18-hour span of the first cell cycle, and does so more accurately than all seventeen alternatives we scored on the same images, including two published embryo staging schemes and three frontier general-purpose large language models.

Characterization of the organ systems and vasculature of sleeping and exercised glassfrogs

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Glassfrogs are a type of frog that turn transparent while sleeping as a form of camouflage. Previous work has found that glassfrogs enter this state by storing ~89% of their red blood cells (RBCs) in their liver. However, the anatomical changes in glassfrogs that occur during camouflage are not yet documented. Furthermore, it is unknown how the flow of oxygenated blood could change to account for a lower concentration of RBCs in circulation during camouflage. Recent work has found that after blue-light induced release of stored RBCs, the anterior abdominal vein (ABV) appears oxygenated, which indicates that a branch of this vein connects directly to the heart. To determine the ABV's branches and the organ anatomy, we analyzed MicroCT images of both the sleeping and exercised glassfrogs, and created organ and blood vessel models with 3D slicer. We found significant changes in structure, size and positioning in the frog's kidneys, lungs, stomach and liver. Additionally, we tracked the ABV through the liver and found possible connections between the heart and the vein, which

suggest an alternative path for circulation during camouflage. These models and observed changes of the glassfrogs' organs and vasculature provide the basis for further exploration of glassfrogs' mechanism of camouflage.

Determining the differential progression of specific aneuploidies at the onset of gastrulation

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Early human development demonstrates a remarkable tolerance for aneuploidy with over 80% of blastocyst embryos displaying mosaicism, and recent single-cell RNA-sequencing studies suggest that mosaic aneuploidy persists to at least week 26. Previous studies investigating mosaic aneuploidy have discovered a lineage specific depletion of aneuploidy, especially in embryonic lineages, but have primarily focused on reversine-induced aneuploidy, a system that generates chaotic, non-specific aneuploidies which are non-representative of in vivo development. However, due to ethics of culturing human embryos past day 14, the progression of aneuploidy in human development is largely unknown in gastrulation, a key developmental milestone in the black box of development. By utilizing human embryonic stem cells as a model for the epiblast lineage, we study the progression of aneuploidy through early gastrulation. Here, we compared trisomy 12 and trisomy 21 cell lines, reversine-treated cells, and mixed euploid:aneuploid chimeras in the lumenoid model. By staining for the EMT marker Brachyury to assess progression through the primitive streak, we found trisomy 12 lumenoids showed reduced brachyury expression when compared to trisomy 21, reversine-treated, and euploid lumenoids. Furthermore, chimeric conditions tended to show a decreased proportion of brachyury-positive cells within each lumenoid.

Understanding the mechanisms behind sex-biased trophectoderm advantage in preimplantation development using stem cell-based embryo models

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In vitro fertilization (IVF) is commonly associated with a higher male-to-female birth ratio compared to natural conception, but the developmental mechanisms underlying this sex bias remain poorly understood. During early development, both mouse and human embryos form the blastocyst, a structure comprising three lineages: epiblast (EPI), primitive endoderm (PrE), and trophectoderm (TE). The TE, which gives rise to the placenta, has emerged as a key focus of how sex-specific differences arise. Meglicki et al. (in revision) demonstrated that male mouse embryos exhibit accelerated TE development relative to females, specifically in high glucose culture, from the morula stage onwards. To further investigate the relationship between sex and TE function, this study utilized blastoids, stem cell-based embryo models that recapitulate key features of the natural blastocyst. Male and female TSC lines were used to model the TE and combined with pre-established male and female ESC lines, representing the EPI, and inducible Gata4-expressing cells, representing the PrE. Blastoids with different combinations of male and female lineages were generated to determine how the sex of each lineage influences early developmental fitness. Blastoid formation and cavitation efficiency as well as the composition and organization of the resulting cell lineages were compared across sex combinations.

Microfluidic approaches to modeling the placenta-brain axis utilizing human organoids

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The placenta plays a central role in fetal development by regulating the exchange of nutrients and oxygen while secreting hormones, growth factors, cytokines, and other signalling molecules that influence the developing brain. Increasing evidence suggests that communication along the placenta-brain axis plays an important role in both normal neurodevelopment and the pathogenesis of neurological disorders; however, the underlying mechanisms remain poorly understood due to the limited access to human development and physiological differences of animal models. This project aimed to develop and establish a 3D printed microfluidic co-culture platform capable of modelling interactions between human trophoblast and cerebral organoids under controlled culture conditions.

A dual-chamber microfluidic device was designed and fabricated using stereolithographic 3D printing, undergoing iterative optimization of both the device design and fabrication process to produce reproducible devices suitable for biological applications. The chip was designed with unidirectional flow driven by gravity, with resistance channels to control flow rate between the compartments. Two compartments are included in this design to accommodate human placental trophoblast organoids and cortical organoids. Methods for chip fabrication, post-processing, and assembly were established, including the development of a sealing strategy to maintain sterile culture conditions while permitting controlled communication between the two organoid compartments. We have pioneered a microfluidic device platform in the Karvas Laboratory with reproducible workflows for the design, fabrication, and use of 3D printed microfluidic co-culture devices, introducing a new experimental capability that can be adapted for future organ-on-a-chip applications.

By providing a more physiologically relevant model of the placenta-brain axis, this platform offers a foundation for future studies investigating how placental signalling influences fetal neurodevelopment. The fabrication workflow established during this project introduces a new experimental capability within the Karvas Laboratory and provides a reproducible approach that can be adapted and expanded in future axes-on-a-chip research. As the platform continues to be developed and validated, it has the potential to support studies of both normal placental function and pregnancy-related disorders, contributing to a better understanding of the role of the placenta in fetal brain development.

Cross-modality reasoning enabled by multimodal AI agents for diabetic progression inference

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Artificial intelligence (AI) is increasingly being explored as a tool for analyzing and interpreting complex biomedical data. Diabetes studies produce heterogeneous patient data, from clinical diagnostics to islet hormone secretion dynamic profiles and to imaging of molecular features. However, integrating these distinct data modalities into a cohesive interpretation remains challenging. Large language model (LLM)-based AI agents may facilitate the integration of these diverse data sources and support the identification of cross-modal relationships relevant to diabetes research. Here, we developed a memory-augmented, multimodal multi-agent framework to systematically reason across heterogeneous patient data, identify relationships between clinical and functional phenotypes, and support researchers in interpreting complex patient cases. Extensive benchmarking was performed to characterize the performance and behavior of various agent architecture-LLM model pairings across computational efficiency, diagnostic accuracy, reasoning quality, consistency, and diagnostic uncertainty. We further evaluated whether the multi-agent system could leverage previously characterized cases stored in a memory database to infer characteristics of a deliberately withheld modality from the remaining patient information with modality-masking experiments.

Investigating self-replicating droplets

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Programmable DNA nanostar condensates provide a model system for studying how molecular interactions control phase separation. Motivated by the broader goal of developing self-replicating condensates, we investigated how different types of linker-mediated interactions can produce temperature-dependent changes in phase behavior. Linker strands, previously used to mediate interactions between otherwise non-interacting DNA-functionalized structures, can be designed so that temperature changes alter their binding configurations and effective interaction strengths. We developed a mean-field free-energy framework to characterize how nanostar concentration, linker availability, and binding energetics influence the stability of mixed and demixed phases. Guided by this analysis, we explored temperature-responsive linker nanostructures and computationally screened DNA sequences whose binding energetics vary over the desired temperature range. Together, these results provide a theoretical and design framework for controlling phase behavior in DNA nanostar condensates through programmable linker interactions.