Quick Guide

Friday, November 22nd 8:00 – 17:00

Saturday, November 23rd 9:00 – 11:30

Kyushu Institute of Technology (Kyushu Tech.)
Tobata Campus
GYMLABO
1-1 Sensui-cho, Tobata-ku, Kitakyushu-shi, Fukuoka, Japan

Organized by

BIOMOD INSTITUTE

BIOMOD Institute

Sponsored by

Molecular Cybernetics

株式会社 新興精機 Shinkouseiki Co.,Ltd.

北九州市 CITY OF KITAKYUSHU

KCV 公益財団法人 北九州観光コンベンション協会 Kitakyushu Convention & Visitors Association

Molecular Cybernetics, JSPS Grant-in-Aid for Transformative Research Areas (A), City of Kitakyushu,

Shinkouseiki Co., Ltd., City of Kitakyushu / Kitakyushu Convention & Visitors Association

Schedule (all based on JST, Japan Standard Time)

Thursday, November 21st, 2024 from 16:00 to 19:45

- Pre-Team Check-In at Kyushu Tech
- Practice session, 15 minutes per team (by prior arrangement, see page 4)

Friday, November 22nd, 2024

08:00	**Doors Open @ GYMLABO,– Please do not arrive earlier than 08:00**
08:00–09:00	**Team Check-In (Breakfast available to all registered participants)** **Connection check by online presenters (Team #1 & #2)**
09:00–09:10	Welcome — Opening Remarks
09:10–09:30	Team #1 — Imperial BIOMOD Team **[Online]** 0:10–0:30 (GMT)
09:30–09:50	Team #2 — UW Biomod **[Online]** Nov.21 19:30–19:50 (UTC-5)
09:50–10:10	Team #3 — Team NoKo
10:10–10:40	**Group Photo and Break**
10:40–11:00	Team #4 — Carbonova
11:00–11:20	Team #5 — Nano_JLU
11:20–11:40	Team #6 — OhioMOD
11:40–12:00	Team #7 — KRNM-SPH
12:00–13:10	**Lunch Break**
13:10–13:30	Team #8 — USYD UFOLD
13:30–13:50	Team #9 — Team Sendai
13:50–14:10	Team #10 — DNADetect - BIOMOD UCalgary
14:10–14:30	**Break**
14:30–14:50	Team #11 — Team Kansai
14:50–15:10	Team #12 — BioRegen
15:10–15:30	Team #13 — YOKABIO
15:30–15:50	**Break**
15:50–16:10	Team #14 — UBC BIOMOD
16:10–16:30	Team #15 — Team Tokyo Tech
16:30–16:50	Team #16 — NTU_Taiwan
16:50–17:00	Closing Remarks
17:30–	Banquet @ Welfare Facility {Build. #55 in campus map (p.5)}

Saturday, November 23rd, 2024

09:00	**Doors Open @ GYMLABO,– Please do not arrive earlier than 09:00**
09:00–10:00	**Breakfast available to all registered participants**
10:00–11:30	**Awards & Closing Ceremony**
(11:30–12:30)	(BIOMOD Mentors & Committee Members Meeting)

Presentation Details

All team presentations will take place in GYMLABO at Kyushu Institute of Technology (Tobata Campus).

The **TOTAL** time allotted to each team is **20 minutes**. This includes laptop setup (**1 min**), present slides (**10 min**), audience questions (**~7 min**), and exiting the stage + buffer time (**2 min**). The time limits will be strictly enforced, so rehearsing in advance is strongly recommended to avoid being cut off. The room is equipped with a projector and screen. *Please remember to bring your PC, laser pointers, converters, adapters, cables, etc., as these will **not** be provided by BIOMOD.* (Please note that each team needs to use your PC and connect to WiFi for your presentation.)

Teams are required to **share screen via Zoom** for the presentation slides. (See page 6 for Network and Wi-Fi information.) A shared screen will be projected in the hall. A camera crew will also live broadcast the presentation on-site.

Zoom Address (Thursday-Saturday)
https://kyutech-ac-jp.zoom.us/j/81671540291?pwd=uRD1hpDEbAH1aISk2uGSr0kBcvYvAN.1

Practice Session (Thursday)

You may sign up for a practice session at the hall from the form below (first-come, first-serve basis; **for on-site presentation teams**):
https://docs.google.com/spreadsheets/d/1h70l4vbN3jmiGLIwloccjtXFyJ3xwK8o8n3UBfli82A/edit?usp=sharing
Please enter your team name in a desired time slot (please do not edit/move other team's information without permission). Practice sessions are strictly limited to **15 minutes per team**. Thanks in advance for arriving on time, and finishing in the time allotted.

For **online presentation teams**, we will test your connection and screen sharing during short break and group photo session on **Friday (8:00 JST)**, so please join the Zoom address above.

Registration/Check-in

All teams are required to check-in for BIOMOD on Thursday, Nov. 21 or Friday. Nov. 22. The presentation practice session (see above) also secures your check-in time on Thursday. Checking in on Thursday is preferred; please come and check in by all members of the team at the time specified in the practice slot spreadsheet.

Getting to BIOMOD

Location: Kyushu Institute of Technology (Kyushu Tech)
Tobata Campus
GYMLABO

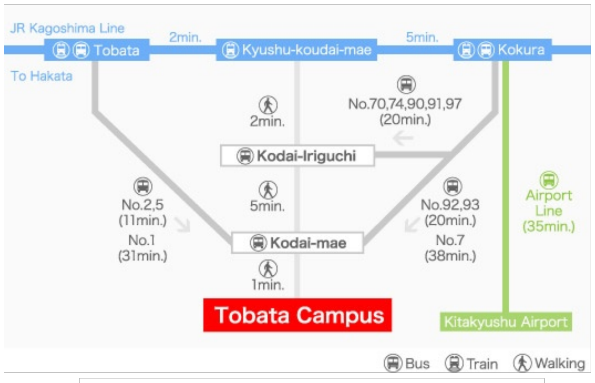
Address: 1-1 Sensui-cho, Tobata-ku, Kitakyushu-shi, Fukuoka, Japan

Google Maps: https://maps.app.goo.gl/vJ56CyCNvcnBiaA87

Transportation to Kyushu Tech

Kyushu Tech (Tobata campus) is located in Kitakyushu-city. You will get off at Kyushukodai-mae station. From the station, it will take 20 min to GYMLABO by walk (~6 min to the main gate).
https://www.kyutech.ac.jp/english/about/map/tobata.html

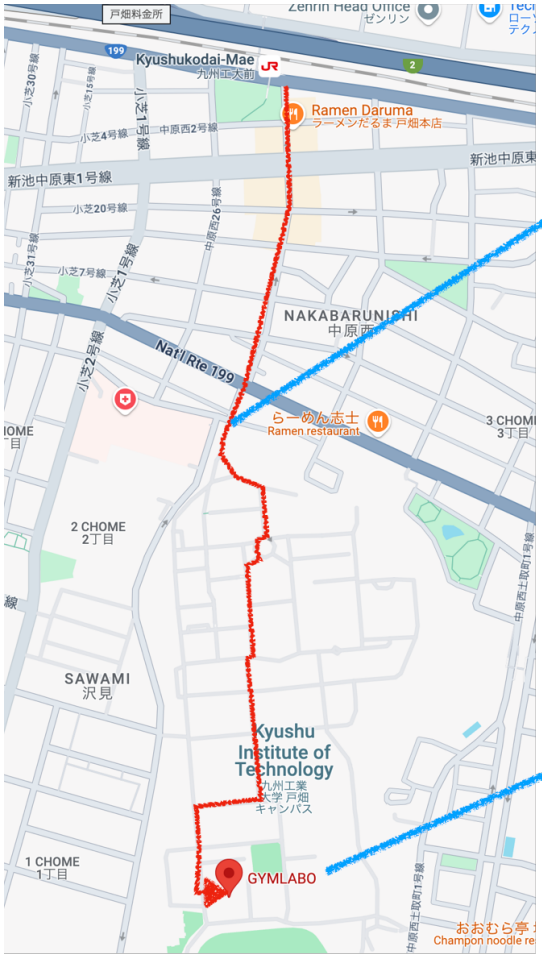
Google Maps' Transit function is quite reliable. You may find the fastest route from your hotel to the campus.



from Tokyo
From Tokyo Station to Kokura Station by Shinkansen (4 h 40 min.)
From Kokura Station to Kyushukodaimae Station by train (6 min.)
From Kyushukodaimae Station to Kyutech on foot (6 min.)

Once at Kyushu Tech Campus – Getting to GYMLABO

Once you arrive at Kyushukoudai-mae station, you can walk to the main gate. It takes about 6 minutes to get there. Enter the main gate and walk south. The gym-like building is GYMLABO (a renovated gym, #60 on the campus map. See page 5).



Accommodation

We recommend a hotel around Kokura station. It is a 5-minute train ride to Kyushu-kodai-mae station. If the hotels around Kokura Station are all booked up, you could also consider staying near stations along the JR Kagoshima Line. This is an easy option because you will not have to change trains. There are also several budget hotels around Kurosaki station.
If all of the hotels are unavailable mentioned above, we recommend the Hakata station area in Fukuoka city. You can get to Kokura station in less than 20 minutes by Shinkansen.

Building Security

You must wear your BIOMOD ID badge at all times while on campus. *If you forget your BIOMOD ID badge you will have to return to your hotel to get pick it up as additional ID badges cannot be printed on-site.*
Please do not arrive early. Building doors will not be unlocked before 08:00 on Friday and 09:00 on Saturday.

Network and Wi-Fi

At BIOMOD2024 we ask you to use the eduroam network. Before traveling to BIOMOD, we recommend that you prepare your eduroam account in your institution. If you are unable to set up an eduroam account in your institution, we will issue you with an individual guest account, which you can request at the reception desk.:

Meals

We have arranged the following meal services for all participants. Please review the schedule and location details below:

1. ***November 22 (Fri) – Breakfast***
    - ***Time:*** *08:00 - 09:00*
    - ***Location:*** *GYMLABO*
    - ***Details:*** *Light breakfast for all participants.*
2. ***November 22 (Fri) – Lunch***
    - ***Time:*** *12:00 - 13:30*
    - ***Location:*** *GYMLABO*
    - ***Details:*** *Bento lunch provided for all participants. Dietary restrictions will be accommodated (see details below).*
3. ***November 22 (Fri) – Evening Social Event***
    - ***Time:*** *17:30 -*
    - ***Location:*** *University Cafeteria*
    - ***Details:*** *Buffet-style dinner with sushi. Mainly non-alcoholic drinks will be served, along with alcoholic beverages.*
4. ***November 23 (Sat) – Breakfast***
    - ***Time:*** *09:00 - 10:00*
    - ***Location:*** *University Cafeteria*
    - ***Details:*** *Buffet-style breakfast for all participants.*

Dietary Accommodations

For the Bento lunch on November 22, we have considered various dietary needs and allergies. Please note the following:

Halal, ***Mushroom Allergy***, ***Dairy-Free***, ***Vegetarian***, ***Seafood Allergy***, ***Mango Allergy***, ***Nut Allergy***

For the buffet, please be mindful of other participants and take only one serving at a time. You are welcome to return for additional servings once everyone has been served.

VR Cell World @ VRChat by Daisuke Inoue

At the networking event, we are planning an activity where you can experience cellular VR. Put on a VR headset and enjoy immersing yourself in the world inside a cell.

Judging

Our aim is to maintain an open and fair process for evaluating each team's performance. Teams will be scored by judges using a point system. The judging process will have two stages: online content (worth up to **75 points**) and live presentation (worth up to **25 points**).

Online content scoring

- Maximum combined score for online content is **75 points** (= 50 points for wiki + 25 points for internet video)
- Scores will be determined by a range-voting system.
- Five (5) judges will be randomly assigned to each team.
- Judges will evaluate each project according to a rubric (see below) and assign a point value in each category.
- Highest and lowest judges' total scores are excluded to remove outliers.
- Three remaining judges' scores will each weighted by 1/3 and combined to determine the total scores for each category.

Website (up to 50 points)

Project Idea (20 points)

- Relevance: Has the team made a strong case that their project idea is scientifically and/or technologically interesting? **(5 points)**
- Specification: Are the project goals well-defined? (i.e. Does the team explicitly state what criteria need to be met in order to consider the project a success?) **(5 points)**
- Feasibility: Was the proposed solution feasible? (i.e. Was it reasonable to expect that the solution could be implemented by a BIOMOD team in one summer?) **(5 points)**
- Merit: Is the proposed solution a good one? Is it particularly elegant or innovative? **(5 points)**

Project Documentation (20 points)

- Clarity: Is the project description well-written and easy to understand? Does it include the background and motivation of the project, methods, results, and discussion? Are the figures easy to understand? **(10 points)**
- Transparency: Are all of the raw experimental data and source files easily accessible? Would it be straightforward to attempt to reproduce the team's results? **(5 points)**
- Layout: Is the team's project page arranged in a clear and logical fashion? **(5 points)**

Project Execution (10 points)

- Execution: Did the team accomplish what they set out to do? **(10 points)**

YouTube video (up to 25 points) (clarified 3 min. limit in accordance with the detailed description on biomod.net)

- Overall impact: Was the video interesting? Did you want to watch more than once? **(10 points)**
- Clarity: Was the project described in a simple and clear manner that could be easily understood by a wide audience? **(10 points)**
- Production: Was the video duration 3 minutes or less? Was the sound and video high quality? Were the images focused and scaled properly? **(5 points)**

Presentation scoring (up to 25 points)

- Content: Were the slides clear and easy to understand? Did the project narrative have a logical flow, with clearly stated goals and results? **(10 points)**
- Delivery: Did the speaker(s) give a well-rehearsed, well-paced presentation? Did the speaker(s) engage with the audience and maintain good eye contact? **(10 points)**
- Impact: Was the presentation interesting? fun? clever? memorable? **(5 points)**

Judges

- Judges were selected from a pool of BIOMOD faculty mentors and outside experts.
- Mentors will not evaluate their own team.

Award Categories

The following prizes will be awarded:

Top Prizes

- Grand prize = 1st highest total combined points from wiki + video + presentation
- 1st runner up = 2nd highest total combined points from wiki + video + presentation
- 2nd runner up = 3rd highest total combined points from wiki + video + presentation

Category awards

- Best Website = 1st place, 2nd place, 3rd place
- Best YouTube Video = 1st place, 2nd place, 3rd place
- Best Presentation = 1st place, 2nd place, 3rd place
- Audience Favorite = 1st place, 2nd place, 3rd place

Project awards

- Bronze: Team satisfied all minimum requirements for judging (i.e. Submitted a complete Project wiki and uploaded a YouTube video)
- Silver: Satisfied criteria for Bronze, plus at least one device (part of the system) in the team's design is worked as expected.
- Gold: Satisfied criteria for Silver, and overall point score from Wiki + YouTube + Jamboree presentation are in top 50% of all teams.

Special Awards

- Molecular Robotics Award
- Best ELSI Practice Award
- Best Team T-shirt Award

Organizer Contact Info.

In case of emergency, please email/call the organizer:

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BIOMOD (Steering Committee)
Email: info@biomod.jp

Team Abstracts

Imperial BIOMOD Team 2024 - DNA Origami PETase-MHETase Cascade

Of the 450 million tonnes of plastic produced globally every year, less than 10% is estimated to be recycled. Insufficient and expensive recycling technologies limit the proportion of plastics which can be repurposed into new materials, while lack of economic incentive has encouraged the uptake of less sustainable waste management strategies.

The IsPETase-MHETase cascade is a new low energy recycling technology which is able to degrade PET to useful products through the manipulation of cellular machinery. Enzymes IsPETase and IsMHETase are immobilised on DNA origami modular nanotubes to enhance degradation of PET via metabolite channelling.
These products can be separated and used for the generation of new materials (terephthalic acid) or repurposed for a variety of commercial applications including antifreeze and break-fluid products.
We hope that our technology may be used in the development of novel, low energy plastic recycling techniques.

Team #2: UW BIOMOD

ANAlyze: Enhancing Systemic Lupus Erythematosus Detection with DNA Origami-Based Biosensors

Systemic Lupus Erythematosus (SLE) is a chronic autoimmune disease characterized by the production of antinuclear antibodies (ANAs) that mistakenly target the body's own cells. Accurate and early diagnosis of SLE is crucial for effective management and treatment. Current diagnostic methods are time consuming and subjective, leading to delayed intervention. In this project, we develop a novel biosensor utilizing DNA origami technology to enhance and streamline ANA detection, laying the groundwork to develop a more reliable diagnostic tool for SLE. DNA origami allows for the precise assembly of nanoscale structures, enabling the creation of highly specific and sensitive biosensors. Our biosensor is designed to bind selectively to ANAs, inducing conformational change to produce a measurable fluorescence signal. This signal directly correlates with the presence and concentration of ANAs in the bloodstream, to provide timely feedback.This innovative approach offers several advantages over traditional diagnostic methods, such as increased sensitivity, higher specificity, and faster detection times. By improving the reliability of the detection process, this biosensor holds the potential to significantly improve patient outcomes. Further, the modular nature of the DNA origami platform allows for future adaptability in detecting other autoimmune biomarkers, expanding its clinical relevance.

BGB: Bilayer-Gel-Bilayer system ~Development of a triple-layered capsule structure that can form nuclear-like pore using DNA artificial cytoskeleton~

Team NoKo

Liposomes are expected to be applied as reactors in drug delivery systems and molecular robotics by encapsulating drug molecules or autonomous systems constructed by designed biomolecules. Conventional liposomes are sensitive to external stresses such as pH changes and osmotic shock and collapse easily in such environments. This prevents the application of liposomes in vivo and the natural environment, thus the increase of the strength of the liposome membrane is essential for further applications. Previous research has increased the stability of liposomes by using DNA hydrogel cytoskeleton or PEG-modified lipid. However, DNA hydrogel inside the membrane and PEG modification on the membrane might inhibit molecular transportation through liposome membranes.

Here, we developed the “BGB (Bilayer-Gel-Bilayer)” structure to solve this problem. BGB is a capsule-like structure consisting of three layers: inner lipid bilayer layer, DNA hydrogel layer, and outer lipid bilayer layer. The DNA hydrogel is packed by inner and outer bilayers. For the formation of BGB, first, liposomes are prepared with positive-charged lipids (DOTAP). Next, DNA strands that form Y-motif hydrogel are mixed with the liposome solution. DNA strands can be attached to the liposome membrane by electrostatic interactions, and the formed hydrogel will pack the liposome membrane. We call this precursor “BG (Bilayer-Gel)”. This BG structure is used as the inner solution for the preparation of the new liposome. This process leads to the formation of the BGB structure. Furthermore, we propose a pore formation mechanism on the BGB structure. DNA origami nanostructures inserted in the middle layer of DNA hydrogel induce membrane fusion between the inner and outer bilayers. Polymerized DNA origami raises the inner bilayer to form specific membrane curvature, and the bilayer is expected to form tubes. Then, membrane fusion will be caused by shrinking the membrane distance between the inner and outer bilayers. We have achieved the preparation of DOTAP-containing liposomes including DOTAP, BG structure, and BDB structure. We will evaluate the stability of the BGB structure on the external stresses and confirm pore formation using DNA origami in future experiments. Our BGB structure increases the

stability of liposomes without the interference between the DNA hydrogel and the inner solution. Moreover, DNA origami for the pore-formation is protected by DNA hydrogel, thus the formed pore is expected to be stable in various environments compared with conventional pore-formation systems such as using DNA nanostructures. Our BGB structure will expand the application field of liposomes under more extreme conditions.

BIOMOD Project Abstract: Carbonova

The increasing concentration of atmospheric carbon dioxide is a major driver of climate change, creating an urgent need for effective carbon capture, utilization, and storage (CCUS) technologies. CCUS involves capturing carbon dioxide from industrial emissions and either converting or storing it to mitigate its environmental impact. Carbonic anhydrase (CA), an enzyme that catalyzes the rapid conversion of carbon dioxide into bicarbonate ions, plays a key role in enhancing the efficiency of this process. Using genetic engineering, the CA gene from *Saccharomyces cerevisiae* was expressed in *Pichia pastoris*, a widely used host for recombinant protein production due to its ability to ensure proper protein folding, its capacity to grow to high cell densities, and its secretion of recombinant proteins into the external environment. This simplifies downstream processing and purification. The enzyme was subsequently immobilized onto carboxyl-functionalized ferromagnetic nanoparticles, enhancing its stability, reusability, and separation efficiency. This immobilized CA system offers a sustainable approach to carbon dioxide capture and utilization, with a major application in algal biofuel production in photobioreactors. When added to the photobioreactor, the immobilized CA is expected to improve algal growth rate due to more efficient carbon utilization. In conclusion, this research contributes to carbon recycling and environmental sustainability.

Title: Photothermal-responsive DNA origami-Gold nanorobot for precise drug delivery：**A remote conformational switch of DNA origami**

DNA nanorobotics with various conformational switches, including azobenzene, toe-hold mediated strand displacement reaction and i-motif, hold a great promise as nanoscale machines with controllable structures and functions. These switches empower DNA nanorobots to execute sophisticated and complex tasks. However, achieving remote control of DNA origami robots in vivo using current switches still poses challenges, such as the low stability of molecular switches within biological environments, slow responsiveness, and the limited signal penetration within biological entities. Herein, we design an in vivo remotely and real-time controllable nanorobot with near-infrared photothermal-responsive switches by utilizing DNA origami coupled with gold nanorods. In this study, we utilized photoexcitated large-size gold nanorods as our switch to induce conformational changes in DNA origami. By modifying the design of DNA origami to accommodate large-size gold nanorods and irradiating it with near-infrared light to excite surface plasmon resonance and generate heat that denatures heat-sensitive DNA on the origami structure, we initiate conformational change enabling remote photoresponsive regulation of its internal cargo. By adjusting the size of gold nanorods and utilizing near-infrared light which has good penetration effects on the human body as an activation source. Therefore, we address the challenges of remote control and real-time responsiveness in vivo for DNA nanorobots. Additionally, equipping these nanorobots with tumor-targeting monoclonal antibodies allows for near-infrared light activation specifically at tumor sites, facilitating precise, targeted cancer therapy.

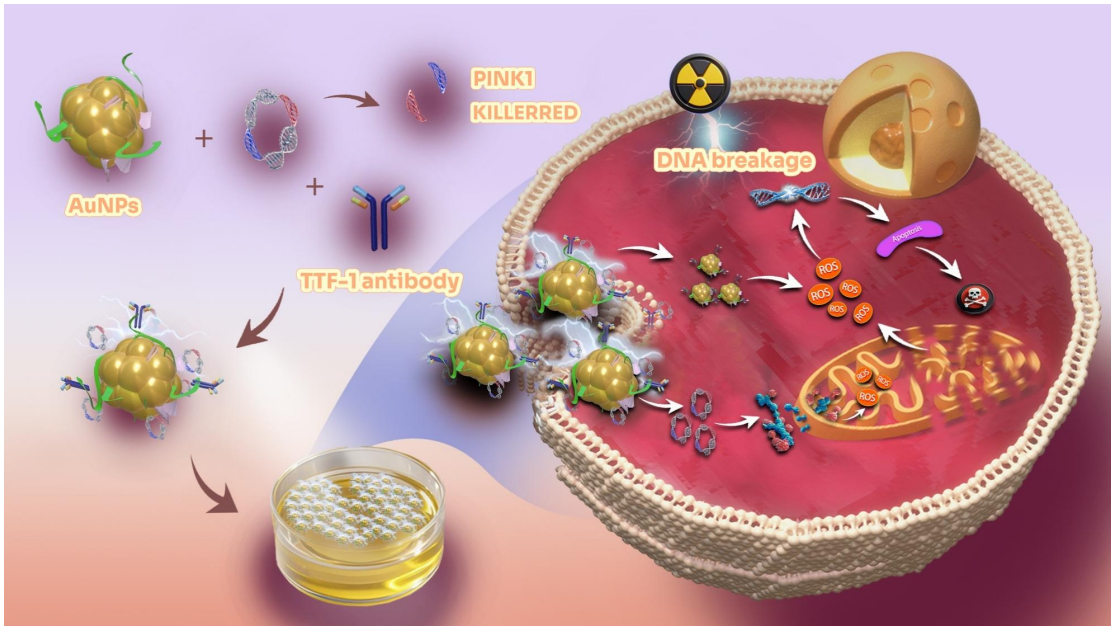
Project Cyclone

Deoxyribonucleic acid (DNA) nanotechnology has been developing rapidly in recent years through the introduction of various structural designs of DNA origami/tile assemblies. However, studies of DNA origami cell interactions are limited due to the complexity of cell structures. Synthetic cells can be used for studies of individual cell functions with external control mechanisms. This project aims to design a reconfigurable DNA origami 3-arm nanostructure as the basic building block of a hierarchical nanomachine assembly. The team has designed a 3-arm structure with a uniform length of 50 nm for each arm. The structure is reconfigurable by switching the connection profile between the three arms to a single-stranded, double-stranded, or cross-over connection. The reconfigurability of the 3-arm structure allows it to change the angle between arms, allowing the higher-order assembly to generate motion or ultimately change the structure's overall configuration. Future work for this project includes polymerization of the 3-arm structure and control of the system through reconfiguration. Potential applications of this nano-system include synthetic cell studies such as forming a contractile ring to assist in cell mitosis and membrane deformation in endo-/exocytosis. This structure can also be used for future research in non-cellular systems to create adaptive membranes for ionic channel.

Crimson Crusader: A novel radiosensitize system targeting lung cancer cells

Lung cancer holds the top position in terms of both incidence and mortality rates globally. The non-small cell lung cancer (NSCLC) is a particularly aggressive subtype that constitutes approximately 85% of all lung cancers. Radiotherapy plays a crucial role in the neoadjuvant treatment of NSCLC; however, the outcomes remain less than optimal. Recently, mitochondria and the reactive oxygen species (ROS) have emerged as the important cancer therapeutic targets. KillerRed (KR), a genetically encoded dimeric red fluorescent protein photosensitizer, generates a substantial amount of ROS under visible light, to induce cancer cell destruction. Regarding of the unique advantages of gold nanomaterials (AuNPs) in drug delivery and radiotherapy sensitization, targeted therapy could be achieved through its specific abilities to deliver drug to the site of action.

In our study, we initially constructed a recombinant expression plasmid containing the targeting sequence of Pink1 (PTEN-induced putative kinase 1) to localize KR to ROS-producing mitochondria using recombinant gene technology. This construct was then bound to the stable Au nanoclusters through charge interactions. Additionally, to endow this system with tumor-targeting capabilities, we conjugated the nanoparticles with the cancer cell-targeting antibody TTF-1 via amidation reactions, resulting in the multifunctional nanosystem AuNPs-TTF1-Pink1-mtKR, named as Crimson Crusader. Finally, the lung cancer cells were treated with Crimson Crusader and ionizing radiation, followed by the examination of the intracellular ROS generation and the killing of the cells to confirm the Crimson Crusader has more effective functions of cancer cell targeting and radiosensitizion, expecting to provide a promising new strategy for the clinical treatment of lung cancer.



Astrobubble: Modulating Immune Responses in Space with DNA Origami Nanostructures

As humanity pushes the boundaries of space exploration, one of the most critical health challenges for astronauts is the deterioration of the immune system in space.

We aim to solve this by engineering a DNA origami nanostructure designed to modulate immune responses dynamically. DNA origami is a technique where DNA strands are folded into specific nanoscale shapes. Inspired by pre-existing designs using the Hoberman circle, our therapeutic device is engineered to transition between open and closed states triggered by stimuli such as radiation or chemical signals. CpG oligonucleotides — short DNA sequences known to influence immune activity – will be attached to the device, and their spacing will change with device transitions. The objective is to create a nanodevice that navigates the bloodstream and precisely modulates immune function by manipulating CpG spacing. This DNA origami-based system will present significant advancements in biomedical engineering and increase our knowledge of immune system regulation.

Team #9: Team Sendai

In general, molecules have a single function. If it becomes possible to selectively bring together multiple types of dispersed molecules in one place, it will be possible to create molecular complexes that serve as highly functional reaction fields. Therefore, long-distance interactions between molecules are extremely important. However, achieving long-range interactions in the molecular world is not easy. Electrostatic interactions and van der Waals forces weaken rapidly with distance. We developed a method called “Bidirectional Nano Grabbers” which connects and attracts distant substances.

In this project, the goal is to attract the target substance, which is in Brownian motion, to the prepared base substance by forming short paths between them. The path consists of three types of molecules: molecules that bind to the base substance, molecules that bind to the target substance, and molecules that connect the two. First, we design the base substance and the target substance to connect by forming paths from both directions. By forming paths from both directions, it becomes easier to connect them than forming paths from only one direction. Furthermore, in order to form the shortest paths, we add a function of restricting the bond angle between molecules and introduce molecules that break bonds. The effectiveness of these mechanisms is demonstrated by simulation, followed by molecular design and validation, showing that it is possible to connect and attract distant substances.

De Novo Design of a Self-Assembling Modular Biosensor

UCalgary BIOMOD
DNADetect

On a molecular level, the pathophysiology of a disease precedes diagnosis, which is detrimental because it delays intervention. The use of Magnetic Resonance Imaging to detect biomarkers is time-consuming, costly, and poses a high radiation risk to patients. Non-invasive biosensors are a rising solution; however, their current use of protein receptors limits the range of targets that can be detected. To address these issues, we propose the design of a self-assembling molecular sensor using DNA origami nanopores, catalytic RNA strands or aptazymes, and toehold-mediated strand displacement (TMSD) for target detection.

Our DNA origami nanopore is designed for on-demand structural modifications to accommodate aptazymes of various biomarkers or targets. The nanopore is functionalized with a TMSD mechanism for reversible aptazyme attachment. This conserves the structure of the sensor while allowing for the detection of multiple targets by enabling exchange of a target-specific aptazyme for another. The nanopore-aptazyme construct, inserted into large unilamellar vesicles loaded with Sulforhodamine B dye, uses the aptazyme as a gate to control dye leakage. Target binding induces self-cleavage of the aptazyme, opening the gate and increasing the fluorescence to indicate target detection.

This self-assembling sensor offers a potential solution for early biomarker detection and could be used for site-specific cargo release, artificial cell components, and portable analysis technology in the future.

Team Kansai

BIOMOD Abstract

A ninja is a person or group of persons who use ninjutsu. Ninja infiltrate enemy strongholds to gather information or destroy them. It is said that the real role of the ninja is to reduce fighting and prevent war. The ninja's ability to sneak in and assassinate anywhere is like a liposome formulation for a drug delivery system. However, the problem with liposomes is their low stability.

The DNA Origami method utilizes the self-assembly of DNA and is expected to be applied to various fields as a technology to construct nm-sized structures. By folding a single long ring of scaffolded DNA with many short stapled DNAs, a variety of shapes can be created, ranging from flat to three-dimensional structures.

It has been reported that DNA origami can be adsorbed onto planar lipid bilayers by electrostatic interactions mediated by divalent cations. For example, this can be applied to form DNA origami lattice patterns on the membrane. Not only lipid bilayers, but also DNA droplets generated by self-assembly of DNA branching structures can be coated with DNA origami to improve the surface stability of the droplets. With these references, we thought that we could create a lattice pattern of DNA origami on the lipid bilayer of liposomes to enhance the stability of liposomes.

The underwear woven into a chain-like pattern that ninja always wear is called chain mail. It is highly protective against attacks by blades and can prevent the blades from touching the skin. In this project, we will attempt to fabricate a chainmail-like mesh bag using DNA origami. We will replace the chainmail to protect the ninja with DNAorigami-net, which we believe will be stronger by covering the liposomes with DNAorigami-net.

One of the DNAorigami structures, 6-Helix Bundle, will be used. When this is mixed with liposome solution, 6HB is expected to adsorb randomly in the membranous form of liposomes. However, a reticular network will not be formed if this is not done. Therefore, the streptavidin-biotin interaction is used. Biotin has the property of binding to tetrameric proteins, and streptavidin has the ability to bind up to four biotin molecules. By modifying biotin with 6HB and adsorbing it to the liposome membrane, and then adding streptodiamine, it is believed that 6HBs can bind to each other in a net via the streptavidin-biotin interaction. Next, we confirm that the DNA origami - net is formed.

Team #12: BioRegen

INJECTABLE HYDROGEL DESIGNED FROM MESENCHYMAL STEM CELLS AS A TREATMENT FOR INTERVERTEBRAL DISC DEGENERATION

Intervertebral disc degeneration (IVDD) is characterized by the loss of disk height and volume due to decreased proteoglycan synthesis and lower protein degradation. It can occur from adolescence to old age, with a higher incidence in 80% of men and 65% in women aged 40 years, being asymptomatic in most cases. IVD functions as stabilizers, composed of an extracellular matrix called pulposus nucleus (PN) on the inside and a fibrous ring on the outside, both of which are collagen compounds that provide tensile strength. The hyaline of the intervertebral cyst formed by chondrocytes and extracellular matrix (ECM) allows solutes to be transported from vertebra to disc.

IVDD has been healed in dependence of its gravity, where the early stage focuses on pain reduction by physiotherapy or oral drugs NSAID type. More advanced phases evolve into severe nerve compression, which is treated by ablative measures. However, these treatments do not reverse the tissue degenerative process, *de novo* treatments develop biomaterials capable of replacing the function of PN.

BioRegen focuses on developing a therapy for the regeneration of ECM from PN using an injectable hydrogel compound capable of gelifying under corporeal conditions. This hydrogel is formed by ECM of porcine origin cellularized with exosomes and chondrocytes differentiated from mesenchymal cells of adipose tissue. Exosomes can inactivate the NLRP3 pathway and regulate pyroptosis of PN cells; and chondrocytes overexpressing COLx genes and integrins to induce increased collagen production in PN. This system could be replicated potentially in other inflammation related pathologies.

Note: *The name of our team "Bio Explorers" was recently changed for* ***BioRegen***

The development of technologies for molecular transport mechanisms that function in micro-compartments such as liposomes, as well as methods for controlling their flow rate, is crucial for the advancement of molecular robotics. Generally, the flow rate of molecules through a pore structure depends on inner radius and length of pore. Thus far, the regulation mechanisms of molecular transport through tubular DNA nanostructures have primarily focused on the inner radius of pore. However, none of the studies have focused on altering both the inner radius and length simultaneously. In this study, we propose a dual-controlling mechanism for regulating the inner radius and length in tubular DNA nanostructures. We designed Kresling-Inspired Tubular DNA nanostructure(KIT-D), based on an origami technique known as Kresling folding. This technique is a pattern composed of multiple parallelograms, each connected along its edges to form a tubular shape. By folding each parallelogram along its diagonal, the structure can be compressed, reducing the inner radius and the length of the tube as it twists and closes. KIT-D is a DNA origami nanostructure that utilizes Kresling folding. Parallelogram-shaped units are connected to each other by single-strand DNA hinges. The transformation of KIT-D is regulated by DNA hybridization, in which the degree of opening is fixed with specific single-strand DNA signals. The dual-control mechanism allows for regulation of the material flow inside the tube through synchronous changes in both inner radius and length. By allowing for such detailed control over the flow rate, this mechanism opens up new possibilities for the design of molecular-level robotics and nanomachines. Ultimately, it is expected to enhance the efficiency and functionality of these systems by providing more versatile and accurate control over their operations.

AND-IE Box: A DNA Origami-Based Immunotherapy For Prostate Cancer

As the second most common cancer in males, prostate cancer presents significant therapeutic challenges. Leading conventional immunotherapies, such as CAR-T-cell therapies, face obstacles in tumour site infiltration due to insufficient tumoural antigen heterogeneity and concentration. To address these issues, we developed the *AND-IE box*, a DNA nanostructure. AND-IE incorporates two aptamers that specifically bind to overexpressed receptors, prostate-specific membrane antigens (PSMAs), thus serving as targeted immunotherapy. Upon binding to both PSMAs, AND-IE unhinges to reveal an anti-CD3 antibody, which activates T-cells through the T-cell receptor (TCR)-CD3 complex. The activated T-cells then trigger an adaptive immune response.

The AND-IE box was designed in Cadnano© and demonstrated high conformational stability in CanDo©. Additionally, Haddock© simulations indicated favourable docking of the anti-CD3 antibody to the AND-IE. Computationally, AND-IE demonstrated high binding affinity for prostate cancer when delivered intratumorally.

Gel electrophoresis will be used to validate the formation of the box. Additionally, an ELISA assay will verify the binding of the AND-IE box to PSMA. To finalize the design, anti-CD3 antibody docking is mediated by NTA–Ni^{2+}–histidine interactions in the box cavity. We aim to advance this design toward a future immunotherapeutic, with the potential to enhance current treatment strategies.

Nano Pill Bug

Team Tokyo Tech

In recent years, early detection technologies based on the analysis of miRNA expression patterns have emerged for various diseases, including cancer. The miRNAs targeted by these methods are limited to those protected from RNases by proteins or lipids, leaving unprotected miRNAs underexplored. Here we propose a device the “Nano Pill Bug”, that focuses on protecting miRNAs from degradation by RNases. Inspired by pill bugs, this device captures miRNA and rolls up to protect it.

The structure consists of two DNA origami sheets and several stopper DNA strands connecting them. Each sheet has a two-layer structure, and its bend is formed by differences in the number of bases, creating a tube-like shape. These two bent sheets are held in place by pre-arranged stopper DNA, which maintains the flat structure by balancing the tension between the sheets. When the stopper DNA is replaced by miRNA through a strand displacement reaction, the tension in the sheets is released, and the miRNA is encapsulated within the cylindrical structure, protecting it from degradation by RNase.

We are developing Nano Pill Bug, a nanorobot that protects miRNAs that are present in minute quantities as the planar structure curls up fast. By extending the functionality of the Nano Pill Bug to target small molecules such as DNA and proteins, the Nano Pill Bug has the potential for a wide range of applications.

Team: NTU_Taiwan

Project Title: DNA Origami: A Novel Approach for Targeted Brain Tumor Therapy

Abstract:

In this study, we employed a novel approach for targeted brain tumor treatment by designing a triangular DNA origami structure that serves as a nanocarrier for antitumor drugs. To enhance drug delivery, the structure is equipped with an aptamer designed to facilitate interaction with and penetration through the blood-brain barrier (BBB). Our current experiments utilize a microfluidic model simulating the BBB, incorporating mouse bend.3 endothelial cells and ALT astrocytes to closely mimic barrier conditions. We expect that these DNA nanocarriers will advance drug delivery systems capable of traversing the human BBB, with potential applications in neuro-oncology.

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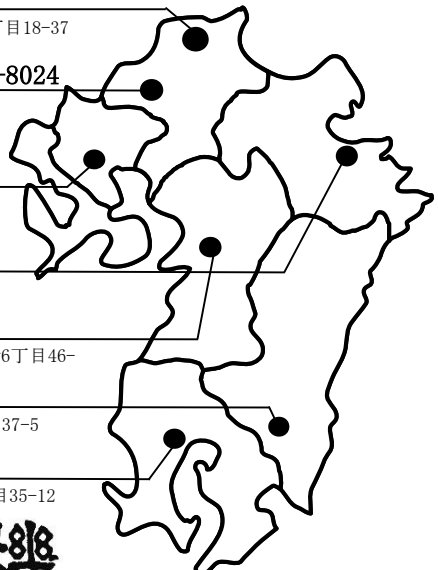

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