卒研生向け
論文紹介の仕方

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論文とは？

- 研究者間のコミュニケーションツールとして最もよく使われるもの
- この200年～300年の間メインのツールとして利用されてきており、「型（構成）」が確立している
- 学術雑誌に投稿して、他の研究者が審査する過程(査読 Peer Review)を経て、採択されたら出版される

なぜ論文紹介をするか？

- 論文の「型（構成）」を知る。科学のお作法を知る
- 自分が卒論を書く時にどのような構成で書けばよいのか学ぶ
- 科学技術英語2単位のため
- 理系の学生として、理系のメインコミュニケーションツールである論文とは何かをそもそも知る
- 院進の人は、口述試験、面接試験のため
- パワーポイントの使い方の学習
- プレゼンの練習。専門的知識を他の人へどう伝えるか？
- 専門的知識を得ること自体は、今回は優先度は低い

論文の「型（構成）」

- 概要(Abstract)
  - 概要はその論文の内容を簡潔に説明したもの。この概要の文章だけがネットに出回る。世界中の研究者は論文本体ではなくこの概要だけを読むことが多い。したがって概要は最も重要。短い時間で内容全体がわかるように自己完結していなければならない
- イントロダクション (Introduction)
  - 背景知識を説明して、先行研究を紹介しながら、自分の研究が重要であることを読者に納得させる
- 方法 (Methods)
  - 数式や図を使用して、手法などをコンパクトに説明する。過不足なく、再現性がとれる情報を書く
- 計算結果と考察 (Results and Discussion)
  - 結果を図を一枚一枚説明しながら、それぞれの結果の自分なりの解釈(どうしてそうなったか)で書いている
- おわりに (Concluding Remarks)
  - 今回の研究内容と結果をコンパクトに、また今後の波及効果（展開）について

概要

End-to-end differentiable blind tip reconstruction for noisy atomic force microscopy images

Yasuhiro Matsunaga[1]✉, Sotaro Fuchigami[2], Tomonori Ogane[1] & Shoji Takada[2]

重要性

Observing the structural dynamics of biomolecules is vital to deepening our understanding of biomolecular functions. High-speed (HS) atomic force microscopy (AFM) is a powerful method to measure biomolecular behavior at near physiological conditions. In the AFM, measured image profiles on a molecular surface are distorted by the tip shape through the interactions between the tip and molecule. Once the tip shape is known, AFM images can be approximately deconvolved to reconstruct the surface geometry of the sample molecule.

重要性

Thus, knowing the correct tip shape is an important issue in the AFM image analysis. The blind tip reconstruction (BTR) method developed by Villarrubia (J Res Natl Inst Stand Technol 102:425, 1997) is an algorithm that estimates tip shape only from AFM images using mathematical morphology operators. While the BTR works perfectly for noise-free AFM images, the algorithm is susceptible to noise. To overcome this issue, we here propose an alternative BTR method, called *end-to-end differentiable* BTR, based on a modern machine learning approach. In the method, we introduce a loss function including a regularization term to prevent overfitting to noise, and the tip shape is optimized with automatic differentiation and backpropagations developed in deep learning frameworks. Using noisy pseudo-AFM images of myosin V motor domain as test cases, we show that our end-to-end differentiable BTR is robust against noise in AFM images. The method can also detect a double-tip shape and deconvolve doubled molecular images. Finally, application to real HS-AFM data of myosin V walking on an actin filament shows that the method can reconstruct the accurate surface geometry of actomyosin consistent with the structural model. Our method serves as a general post-processing for reconstructing hidden molecular surfaces from any AFM images. Codes are available at https://github.com/matsunagalab/differentiable_BTR.

現状の問題点

何をしたか

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自己完結している

イントロダクション

Atomic force microscopy (AFM) is a unique technique for imaging structures of sample molecules bound to a surface at ambient conditions[1]. Recently, high-speed AFM (HS-AFM) with dramatically faster imaging rates (up to tens of frames per second) has been developed, enabling us to directly observe biomolecules in action[2,3]. HS-AFM can investigate detailed structure–function relationships in biomolecules that cannot be observed with other methods, and it has been established as one of indispensable techniques in modern biophysics. Example applications of HS-AFM include myosin V walking along an actin filament[4], rotary catalysis of F_1-ATPase[5], structural dynamics of intrinsically disordered protein[6], and the functional dynamics of CRISPR-Cas9 in action[7]. Currently, the spatial resolutions of HS-AFM instruments are ~ 2 nm in the lateral direction and ~ 0.15 nm in the vertical direction to the AFM stage[8].

Importantly, separately from the resolution of the image profile, the resolutions of the obtained sample surface information are further limited by the tip geometry and the tip-sample interactions. The relationship among the tip geometry, the image profile, and the sample surface is shown in Fig. 1a and b. When the tip is sufficiently thin, the obtained image profile is nearly the same as the sample surface (Fig. 1a). On the other hand, when the tip is blunt compared to the scale of samples, the image profile is blurred from the sample surface (Fig. 1b). Once the tip shape is known, algorithms, called *erosion*, have been proposed to "deconvolve" the image profile for reconstructing approximate sample surface geometry[9–11]. Thus, to reconstruct the surface geometry of the sample molecule, it is crucial to know the tip shape accurately. Tip shape estimation is also important for inferring 3D molecular structures from AFM images. In the recent analysis of AFM images, pseudo-AFM images are emulated from 3D molecular structures (obtained with different experimental or computational techniques, e.g., X-ray crystallography or molecular dynamics simulations) and an assumed tip shape and then compared with

自分の研究の重要性を納得させる。そのために、
背景知識、関連のある先行研究を紹介

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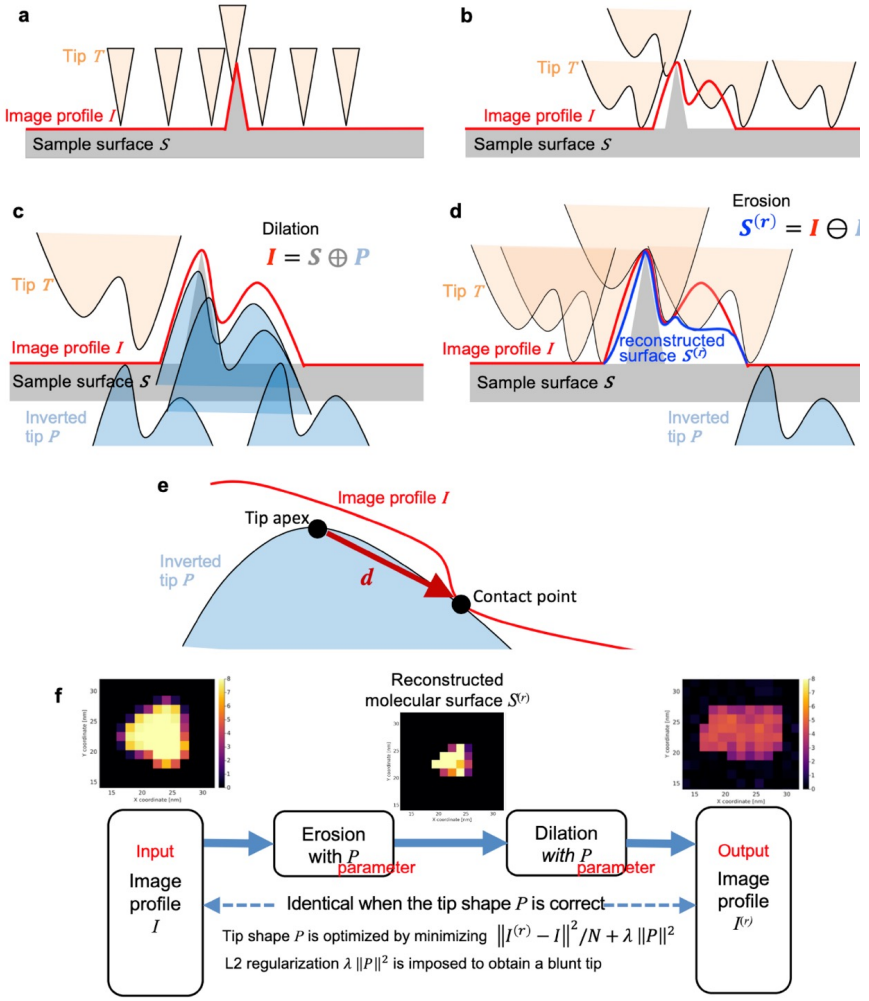
方法 Methods



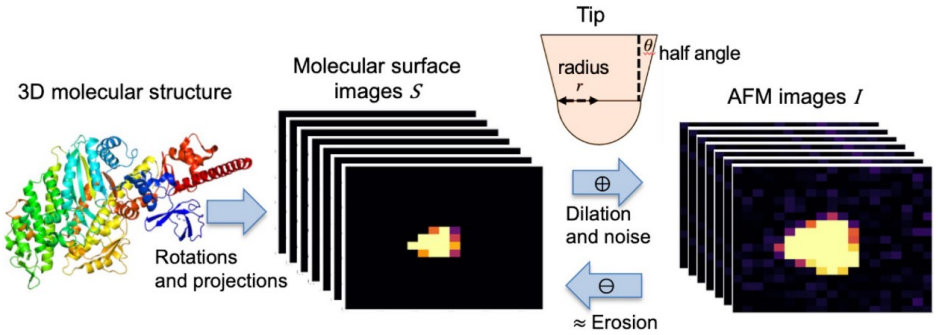


Figure 2. Schematic of twin experiments. From the structural model, the images of molecular surfaces are generated. Then, the molecular surfaces are converted to pseudo atomic force microscopy (AFM) images by dilation with a given tip shape. These molecular surfaces and tip shape are used as ground truths in the experiments. The 3D molecular structure is drawn with PyMOL[39].

In the following, the end-to-end differentiable BTR is introduced (Fig. 1f). The fact that the tip reconstruction is affected by noise in the original BTR can be regarded as an overfitting problem in machine learning. In machine learning, a typical approach to prevent overfitting is to use an appropriate loss function including a regularization. The point of our idea is to statistically determine whether or not to carve the tip from the entire AFM data according to a loss function that takes noise into account, rather than individually determining whether or not to carve the tip from each dent in the image profile, as in the original BTR. Following the original BTR, our differentiable BTR is based on the relation $I \circ P = I$, but the condition is relaxed because the equality does not hold in the presence of noise. Specifically, assuming that the noise is spatially independent Gaussian noise, the loss function to be minimized would be the mean square error,

$$\mathrm{MSE}(p) = \frac{1}{N}\|i^{(r)} - i\|^2 = \frac{1}{N}\sum_{x,y}\left(i^{(r)}(x,y) - i(x,y)\right)^2$$

where N is the number of pixels, and $\|\|^2$ is the L2 norm. $i^{(r)}$ is an image created by the opening $I^{(r)} = I \circ P = (I \ominus P) \oplus P$ with some tip shape P. Expanding $i^{(r)}(x,y)$ by morphology operators, it can be written as

$$i^{(r)}(x,y) = \max_{u,v}\left[\min_{u',v'}\left[i\left(x-u+u', y-v+v'\right) - p\left(u',v'\right)\right] + p(u,v)\right]$$

The differentiable BTR finds the optimal tip shape $p(u,v)$ by minimizing the mean square error over tip shape through its gradient. Because max and min functions appeared in dilation and erosion seem to be non-differentiable at first glance, we treat this problem following the implementations of Max Pooling layer in convolutional neural networks (CNNs). For example, consider a one-dimensional tip shape p_u with u discretized by the index of pixels. The gradient of $h = \max[p_1, p_2, p_3]$ is, if p_3 is the maximum, $\partial h/\partial p_1 = 0$, $\partial h/\partial p_2 = 0$, $\partial h/\partial p_3 = 1$, which is equivalent to argmax function. In image processing, some studies try to apply morphology operators to

再現性がとれる量の情報を図や数式を用いてコンパクトに解説。

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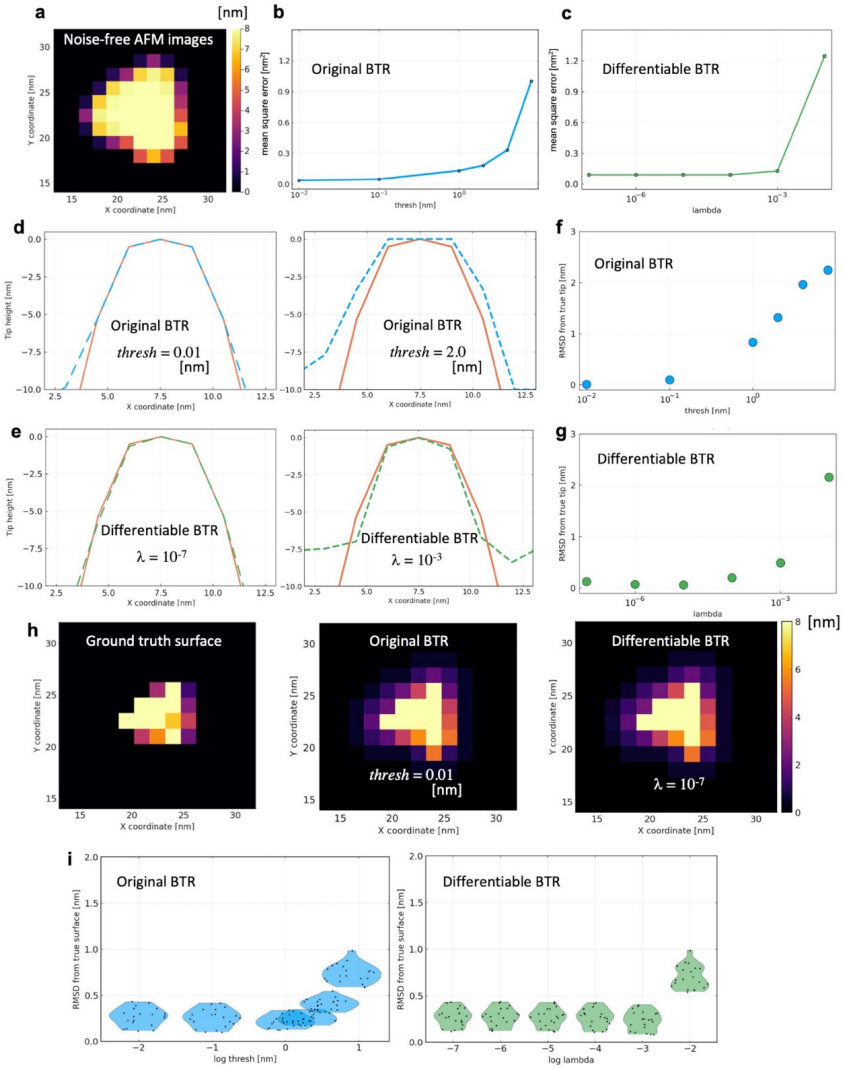
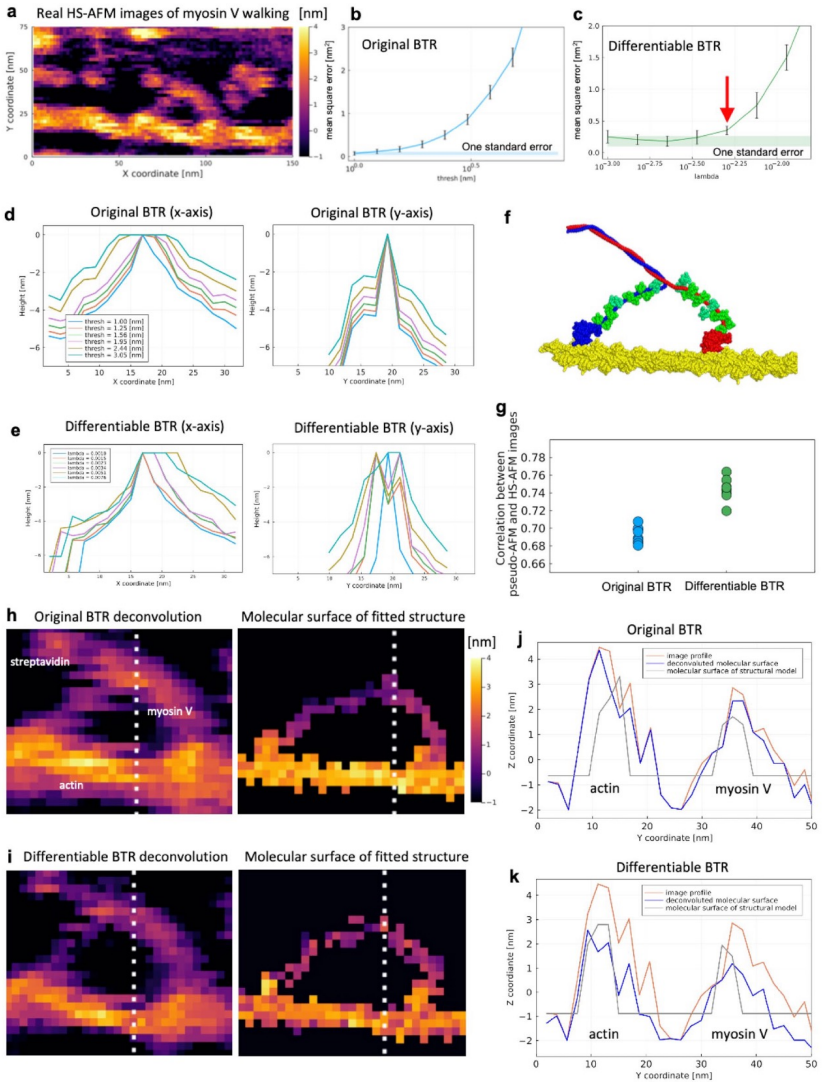
結果 Results





結果の図がメイン。読者が客観的に判断できるように見やすく載せる。

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考察 Discussion

Discussion

In this study, we have proposed the end-to-end differentiable BTR method based on a loss function including a regularization term considering noise in AFM image to avoid overfitting. The loss function is systematically optimized following the framework developed by the recent advances in deep learning studies. The results of the twin experiments showed that the differentiable BTR is more robust to noise than the original BTR. Finally, demonstration of real HS-AFM data shows that the differentiable BTR reconstructs tip shapes and the deconvoluted molecular surface is consistent with the structural model. Since the method is a quite general, we expect that the method would become a routine method for analyzing noisy AFM images as well as HS-AFM data.

まとめ

The twin experiments have shown that reconstructed tip shapes with the differentiable BTR are less dependent on the parameter λ. For relatively small noise ($\sigma = 0.3$ nm), any small λ values ($\lambda \ll 1$) are expected to reproduce tip shapes close to the ground truth one. For relatively large noise ($\sigma = 1.2$ nm), small λ may reconstruct unreasonably thin tip shapes even with the differentiable BTR. In such cases, the one standard error rule is recommended to reconstruct reasonably thick tip shapes. As with the case of neural network frameworks, there are several hyperparameters in the optimization process in the differentiable BTR. For example, if the number of epochs is too large, that increases the chance to overfit to noise, while too small epochs result in insufficient learning. Using techniques such as early stopping, or hyperparameter optimization tools[44], may help to choose appropriate hyperparameters.

As mentioned in Introduction, Tian et al. proposed an improved regularization scheme against noise in the framework of original BTR[27], and Jóźwiak et al. simplified Tian's idea to the case of standard AFM tips[28]. In this scheme, the update procedure of tip shape becomes more conservative, and it is thus expected to be robust against noise, as shown by Jóźwiak et al.[28]. Supplementary Fig. 3 shows the results of the original BTR using this regularization scheme for noisy single-tip and double-tip twin experiments. As expected, the regularization scheme improves the estimation and reconstructs blunter tip shapes compared to the original BTR. However, the RMSDs from the ground truth tip shape and molecular surface show that the differentiable BTR still outperforms the original BTR with the regularization scheme.

Since this study mainly targets the analysis of HS-AFM data, pseudo and real AFM data consisting of 20–30 frames were mainly analyzed. To check the dependence of reconstruction accuracy on the number of frames, we conducted noisy single-tip twin experiments using 1, 10, and 100 pseudo-AFM frames (Supplementary Fig. 4). Although the accuracies of tip and surface reconstructions are comparable in both BTRs for a single frame, the differentiable BTR outperforms the original BTR for 10 and 100 frames. An only drawback of the differentiable BTR here is that the computation time is much longer than the original BTR, especially for the analysis of 100 frames. For example, in our twin experiment (the single-tip case, 20 frames), the original BTR took 0.02 s to estimate the tip shape of a specific *thresh* while the differentiable BTR took 5.7 s for a specific λ with Intel Xeon Gold 6330 CPU (without multi-threads). Implementing GPU kernels for dilation and erosion, or using smooth functions instead of max and min, would accelerate the optimization.

提案手法の限界

A concern with differentiable BTR is the relationship between ground truth tip shape and erosion. To reconstruct the tip accurately with differentiable BTR, erosion must be a good approximation to the inverse function of dilation. If the ground truth tip has a cone shape, the half angle must be small for the erosion to approximate the

まとめと、提案手法の限界や、今後の展望が述べられている

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論文を読む上でのポイント

- 概要(Abstract)が最も重要。ここはゆっくり読む
- イントロダクションと手法は勉強。頑張って読む。
  - 何が重要か？
  - 何が問題なのか？
  - この研究の何が新しいのか？
  - 手法のアイデアは何か？
- 結果は図が自分で理解できればよい。著者らの解釈は話半分程度でもよい(著者らの解釈は先入観が入っていることがあるため。図・結果が全て)

読むテクニック

- パラグラフライティングを意識して読もう
  - 論文はパラグラフライティング = 1つのパラグラフに1つの話題
  - 大抵はパラグラフの最初の文に言いたいことが書いてある。そこを重視して読もう
- 翻訳サービスは遠慮なく使おう
  - DeepL、Google translate、ChatGPTやそのプラグイン
  - 英語にハードルを感じると思いますが、何本か読んでくると論文を読みこなす上では実は英語は問題でなく、科学的知識が一番重要であることに気づきます
- わからなくてもとりあえず最後まで一気に読み通そう
  - 一度止まってしまうと、ずっとそこで止まったままになってしまいます
  - わからなかった箇所は読み通した後で振り返る

余裕があればSupporting Informationを読もう

- 近年の研究手法はどんどん複雑化しており、普通の論文のフォーマットだとページ数がとんでもないことになる
- 細かな計算の条件は論文本体ではなく、論文のホームページに付録として置いてある「Supporting Information」に書いてある
- 手法の細かな内容を知るにはSupporting Informationを読む必要がある
- 全部読む必要はない。実験条件・計算条件で気になった箇所のチェックに読む程度でOK。

わからなかった箇所の調べ方

- その箇所の関連で引用されている他の論文を読んでみよう
  - ただし、きりがないので、該当論文の必要な箇所だけ読む
- Teamsで教員や先輩へ聞こう。遠慮することはありません。そのための学費です。

論文の探し方

- Google Scholarを使おう
- 商用誌は大学からアクセスしないとPDFがとれないことがある。論文によってはオープンアクセスになっており無料でPDFがとれる
- プレプリントサーバから探そう
  - 査読を通過する前の論文の置き場所。全て無料だが査読を経ていないので注意が必要。
  - arXiv
  - bioRxiv

論文管理

- アプリを使って論文を管理しよう
  - ZoteroかPaperpileがおすすめ。iPad版もあるのでiPadで読むことができる
  - Word等での論文執筆の際に、参照作成で威力を発揮
  - 成川先生の動画が参考になります

    文献管理ツールを使って研究の進展とアウトプットを加速する

    https://togotv.dbcls.jp/en/20220806.html

紹介の仕方

- 論文の型（構成）を意識して紹介しよう
- 紹介の流れ
  - 概要を説明（論文の全体像。自己完結している）
  - 背景、なにが問題か？、この研究の重要性
  - 手法説明
  - 結果説明。図は全て説明しよう（図を貼り付けるだけでも構いません）
  - 著者らの解釈の紹介は間違っている・先入観が入っていることが多いのでそこそこで良い。図・結果が全て。
  - 展望の紹介は必須ではない

- 作成したらTeamsのmeetingチャネルへアップロードして共有。
  ファイル名は
  「日付8桁_論文題名一部_発表者の名前.pptx」
  例）20230624_DeepTICA_松永.pptx

紹介のためのスライド作成

- 論文紹介ではスライド作成に時間を使う必要はない。極端な話、スライドを作らずに、論文のPDFを直に見ながらの紹介でもかまわない
- スライドへは必要な箇所だけを英語のままスクショして作っても構わない。特に結果の図は、図を貼り付けるだけで構わない
- （一方で、自分の研究のスライドは丁寧に時間をかけて何度も何度もブラッシュアップしていくように作る）
- スライドを作る場合は、先輩のスライドをテンプレートとして使おう
- パワーポイントの使い方は早水先生のYouTubeが参考になります

PowerPointスライド作成実演ライブ ①必修編
https://www.youtube.com/live/zMp3BrIakOY?feature=share