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## Quantitative Big Imaging

author: Kevin Mader date: 16 April 2015 width: 1440 height: 900 css: ../common/template.css transition: rotate  
ETHZ: 227-0966-00L

## Groups of Objects and Distributions

### Course Outline

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- 19th February - Introduction and Workflows
- 26th February - Image Enhancement (A. Kaestner)
- 5th March - Basic Segmentation, Discrete Binary Structures
- 12th March - Advanced Segmentation
- 19th March - Applying Graphical Models and Machine Learning (A. Lucchi)
- 26th March - Analyzing Single Objects
- 2nd April - Analyzing Complex Objects
- 16th April - Spatial Distribution
- 23rd April - Statistics and Reproducibility
- 30th April - Dynamic Experiments (K. Mader and A. Patera)
- 7th May - Scaling Up / Big Data

- 21th May - Guest Lecture, Applications in Material Science
- 28th May - Project Presentations

## Literature / Useful References

### Books

- Jean Claude, Morphometry with R
  - Online (<http://link.springer.com/book/10.1007%2F978-0-387-77789-4>) through ETHZ
  - Buy it (<http://www.amazon.com/Morphometrics-R-Use-Julien-Claude/dp/038777789X>)
- John C. Russ, "The Image Processing Handbook", (Boca Raton, CRC Press)
  - Available online (<http://dx.doi.org/10.1201/9780203881095>) within domain ethz.ch (or proxy.ethz.ch / public VPN)
- J. Weickert, Visualization and Processing of Tensor Fields
  - Online (<http://books.google.ch/books?id=ScLxPORMob4C&hl=PA220&ots=mYleQbaVXP&dq=&pg=PA220#v=onepage&q&f=false>)

### Papers / Sites

- Voronoi Tesselations
  - Ghosh, S. (1997). Tessellation-based computational methods for the characterization and analysis of heterogeneous microstructures. Composites Science and Technology, 57(9-10), 1187-1210
  - Wolfram Explanation (<http://mathworld.wolfram.com/VoronoiDiagram.html>)
- Self-Avoiding / Nearest Neighbor
  - Schwarz, H., & Exner, H. E. (1983). The characterization of the arrangement of feature centroids in planes and volumes. Journal of Microscopy, 129(2), 155-169.
  - Kubitscheck, U. et al. (1996). Single nuclear pores visualized by confocal microscopy and image processing. Biophysical Journal, 70(5), 2067-77.
- Alignment / Distribution Tensor
  - Mader, K. et al (2013). A quantitative framework for the 3D characterization of the osteocyte lacunar system. Bone, 57(1), 142-154
  - Aubouy, M., et al. (2003). A texture tensor to quantify deformations. Granular Matter, 5, 67-70. Retrieved from <http://arxiv.org/abs/cond-mat/0301018> (<http://arxiv.org/abs/cond-mat/0301018>)
- Two point correlation
  - Dinis, L., et. al. (2007). Analysis of 3D solids using the natural neighbour radial point interpolation method. Computer Methods in Applied Mechanics and Engineering, 196(13-16)

## Previously on QBI ...

- Image Enhancement
  - Highlighting the contrast of interest in images
  - Minimizing Noise
- Understanding image histograms
- Automatic Methods
- Component Labeling
- Single Shape Analysis
- Complicated Shapes

## Outline

- Motivation (Why and How?)
- Local Environment
  - Neighbors
  - Voronoi Tesselation
  - Distribution Tensor
- Global Environment
  - Alignment
  - Self-Avoidance
  - Two Point Correlation Function

## Metrics

We examine a number of different metrics in this lecture and additionally to classifying them as Local and Global we can define them as point and voxel-based operations.

### Point Operations

- Nearest Neighbor
- Delaunay Triangulation
  - Distribution Tensor
- Point (Center of Volume)-based Voronoi Tesselation
- Alignment

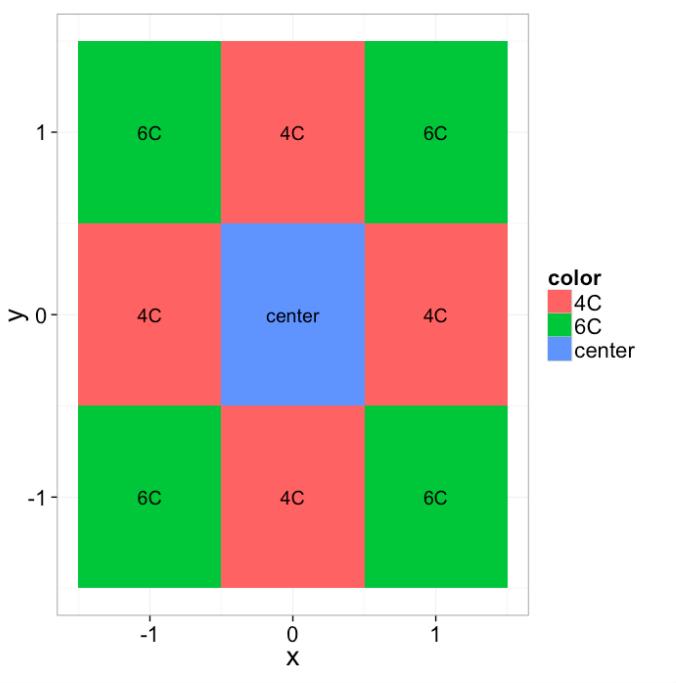
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x	y	z	
1		3	4
2		0	2
2		3	2
3		1	4

### Voxel Operation

- Voronoi Tesselation
- Neighbor Counting
- 2-point (N-point) correlation functions

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## What do we start with?

Going back to our original cell image

1. We have been able to get rid of the noise in the image and find all the cells (lecture 2-4)
2. We have analyzed the shape of the cells using the shape tensor (lecture 5)

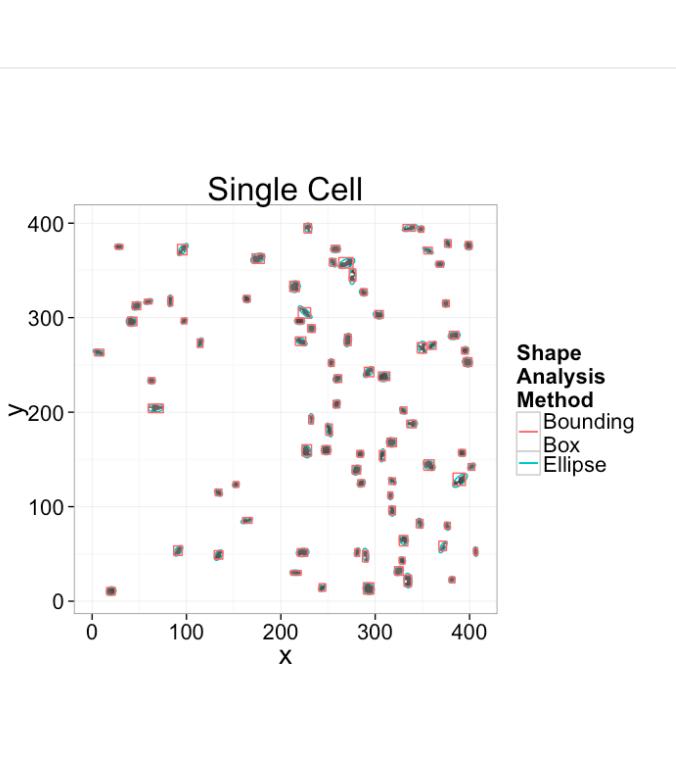
3. We even separated cells joined together using Watershed (lecture 6)

We can characterize the sample and the average and standard deviations of volume, orientation, surface area, and other metrics

## Motivation (Why and How?)

With all of these images, the first step is always to understand exactly what we are trying to learn from our images.

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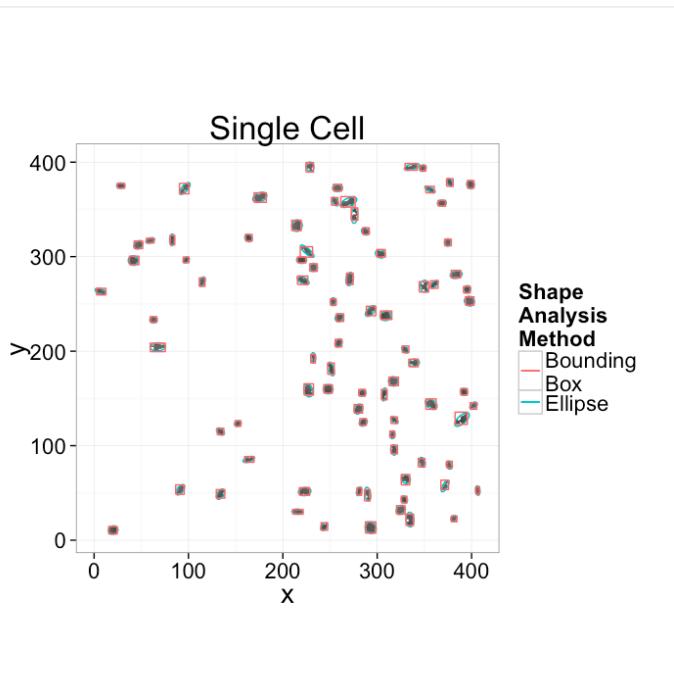


1. We want to know how many cells are alive
  - Maybe small cells are dead and larger cells are alive → examine the volume distribution
  - Maybe living cells are round and dead cells are really spiky and pointy → examine anisotropy
2. We want to know where the cells are alive or most densely packed

- We can visually inspect the sample (maybe even color by volume)
- We can examine the raw positions (x,y,z) but what does that really tell us?
- We can make boxes and count the cells inside each one
- How do we compare two regions in the same sample or even two samples?

## Motivation (continued)

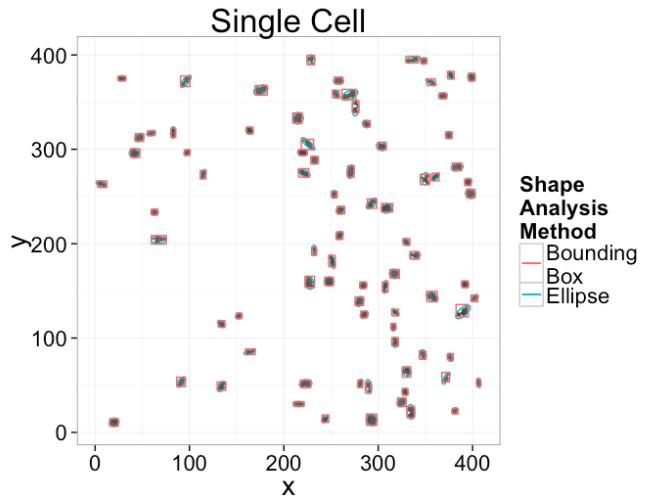
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- We want to know how the cells are communicating
  - Maybe physically connected cells (touching) are communicating → watershed
  - Maybe cells oriented the same direction are communicating → average? orientation
  - Maybe cells which are close enough are communicating → ?

• Maybe cells form hub and spoke networks → ?

## Motivation (continued)



- We want to know how the cells are nourished
  - Maybe closely packed cells are better nourished → count cells in a box?
  - Maybe cells are oriented around canals which supply them → ?

## So what do we still need

- A way for counting cells in a region and estimating density without creating arbitrary boxes
- A way for finding out how many cells are near a given cell, it's nearest neighbors
- A way for quantifying how far apart cells are and then comparing different regions within a sample
- A way for quantifying and comparing orientations

## What would be really great?

A tool which could be adapted to answering a large variety of problems

- multiple types of structures
- multiple phases

## Destructive Measurements

With most imaging techniques and sample types, the task of measurement itself impacts the sample.

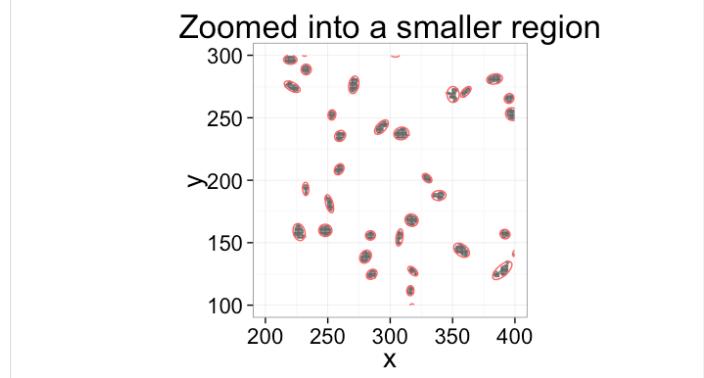
- Even techniques like X-ray tomography which claim to be non-destructive still impart significant to lethal doses of X-ray radiation for high resolution imaging
- Electron microscopy, auto-tome-based methods, histology are all markedly more destructive and make longitudinal studies impossible
- Even when such measurements are possible
  - Registration can be a difficult task and introduce artifacts

## Why is this important?

- techniques which allow us to compare different samples of the same type.
- are sensitive to common transformations
  - Sample B after the treatment looks like Sample A stretched to be 2x larger
  - The volume fraction at the center is higher than the edges but organization remains the same

## Ok, so now what?

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x	y	vx	vy
20.19	10.69	-0.95	-0.30
20.19	10.69	0.30	-0.95
293.08	13.18	-0.50	0.86
293.08	13.18	-0.86	-0.50
243.81	14.23	0.68	0.74
243.81	14.23	-0.74	0.68

...

So if we want to know the mean or standard deviations of the position or orientations we can analyze them easily.

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	Min.	1st Qu.	Median	Mean	3rd Qu.	Max.
x	6.90	215.70	280.50	258.20	339.00	406.50
y	10.69	111.60	221.00	208.60	312.50	395.20
Length	1.06	1.57	1.95	2.08	2.41	4.33
vx	-1.00	-0.94	-0.70	-0.42	0.07	0.71
vy	-1.00	-0.70	0.02	0.04	0.71	1.00
Theta	-180.00	-134.10	-0.50	-4.67	130.60	177.70

- But what if we want more or other information?

## Simple Statistics

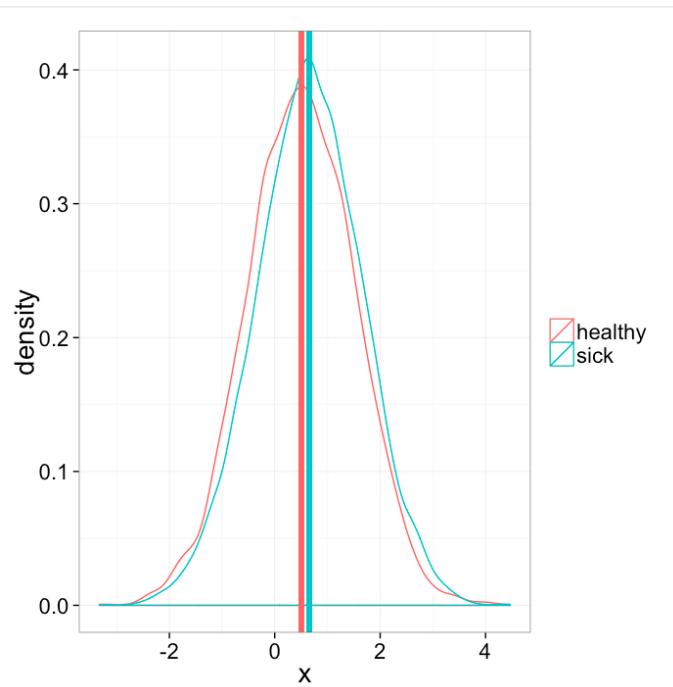
When given a group of data, it is common to take a mean value since this is easy. The mean bone thickness is 0.3mm. This is particularly relevant for groups with many samples because the mean is much smaller than all of the individual points.

### but means can lie

- the mean of  $0^\circ$  and  $180^\circ = 90^\circ$
- the distance between  $-180^\circ$  and  $179^\circ$  is  $359^\circ$
- since we have not defined a tip or head,  $0^\circ$  and  $180^\circ$  are actually the same

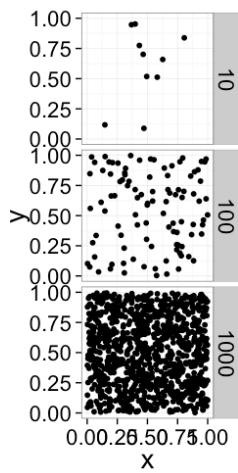
### some means are not very useful

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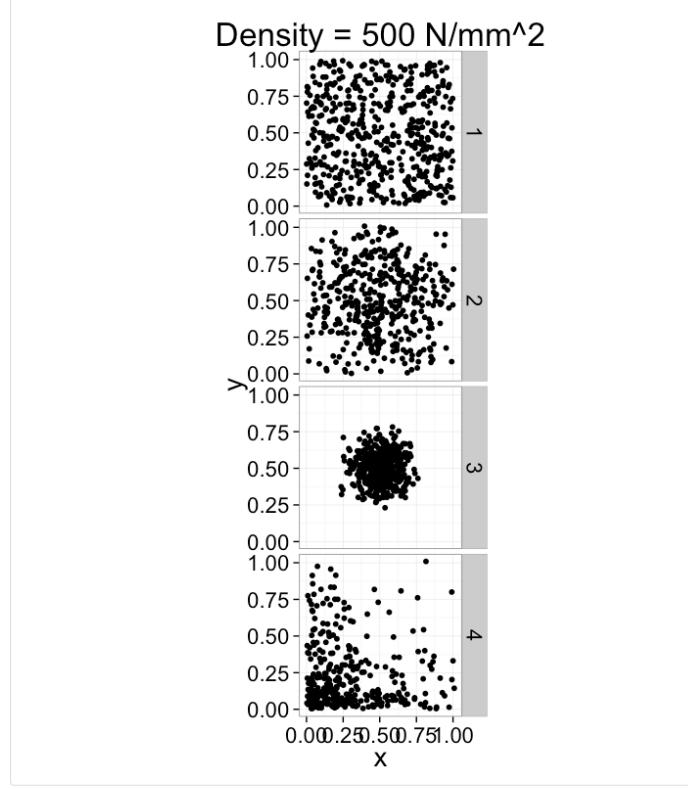
## Calculating Density

One of the first metrics to examine with distribution is density → how many objects in a given region or volume.  
It is deceptively easy to calculate involving the ratio of the number of objects divided by the volume.



It doesn't tell us much, many very different systems with the same density and what if we want the density of a single point? Does that even make sense?

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# Neighbors

## Definition

Oxford American → be situated next to or very near to (another)

- Does not sound very scientific
- How close?
- Touching, closer than anything else?

## Nearest Neighbor (distance)

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Given a set of objects with centroids at

$$\mathbf{P} = [\vec{x}_0, \vec{x}_1, \dots, \vec{x}_i]$$

We can define the nearest neighbor as the position of the object in our set which is closest

$$\vec{\text{NN}}(\vec{y}) = \operatorname{argmin}(\|\vec{y} - \vec{x}\|) \forall \vec{x} \in \mathbf{P} - \vec{y}$$

We define the distance as the Euclidean distance from the current point to that point, and the angle as the

$$\text{NND}(\vec{y}) = \min(\|\vec{y} - \vec{x}\|) \forall \vec{x} \in \mathbf{P} - \vec{y}$$

$$\text{NN}\theta(\vec{y}) = \tan^{-1} \frac{(\vec{\text{NN}} - \vec{y}) \cdot \vec{j}}{(\vec{\text{NN}} - \vec{y}) \cdot \vec{i}}$$

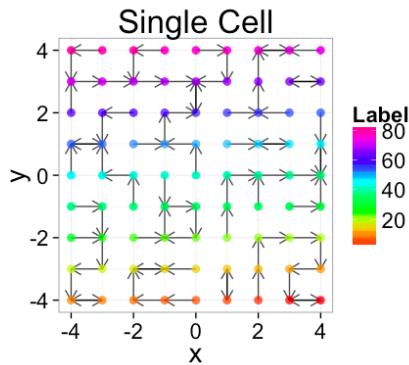
## Nearest Neighbor Definition

So examining a simple starting system like a grid, we already start running into issues.

- In a perfect grid like structure each object has 4 equidistant neighbors (6 in 3D)
- Which one is closest?

We thus add an additional clause (only relevant for simulated data) where if there are multiple equidistant neighbors, a nearest is chosen randomly

This ensures when we examine the orientation distribution ( $\text{NN}\theta$ ) of the neighbors it is evenly distributed



## In-Silico Systems

For the rest of these sections we will repeatedly use several simple in-silico systems to test our methods and try to better understand the kind of results we obtain from them.

- Compression

- The most simple system simply involves a scaling in every direction by  $\alpha$
- $\alpha < 1$  the system is compressed
- $\alpha > 1$  the system is expanded

$$\begin{bmatrix} x' \\ y' \end{bmatrix} = \alpha \begin{bmatrix} x \\ y \end{bmatrix}$$

- Shearing

- Slightly more complicated system where objects are shifted based on their location using a slope of  $\alpha$

$$\begin{bmatrix} x' \\ y' \end{bmatrix} = \begin{bmatrix} 1 & \alpha \\ 0 & 1 \end{bmatrix} \begin{bmatrix} x \\ y \end{bmatrix}$$

## In-Silico Systems (Continued)

- Stretch

- A non-evenly distributed system with a parameter  $\alpha$  controlling if objects bunch near the edges or the center. A maximum distance  $m$  is defined as the magnitude of the largest offset in x or y
- + Same total volume just arranged differently

$$\begin{bmatrix} x' \\ y' \end{bmatrix} = \begin{bmatrix} \operatorname{sign}(x) \left( \frac{|x|}{m} \right)^{\alpha} m \\ \operatorname{sign}(y) \left( \frac{|y|}{m} \right)^{\alpha} m \end{bmatrix}$$

- Swirl

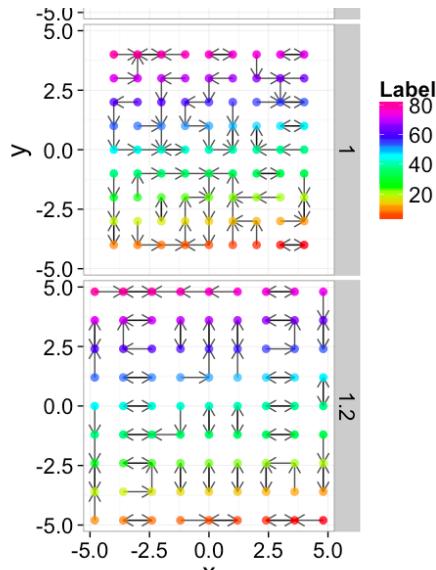
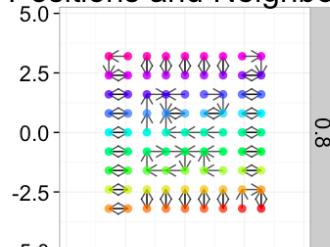
- A transformation where the points are rotated more based on how far away they are from the center and the slope of the swirl ( $\alpha$ ),

$$\theta(x, y) = \alpha \sqrt{x^2 + y^2}$$

$$\begin{bmatrix} x' \\ y' \end{bmatrix} = \begin{bmatrix} \cos \theta(x, y) & -\sin \theta(x, y) \\ \sin \theta(x, y) & \cos \theta(x, y) \end{bmatrix} \begin{bmatrix} x \\ y \end{bmatrix}$$

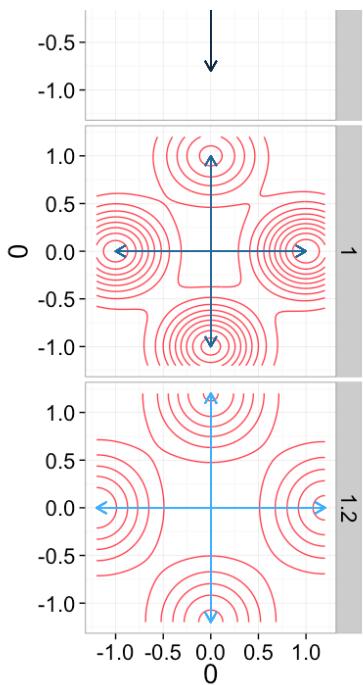
## Examining Compression

### Positions and Neighbors



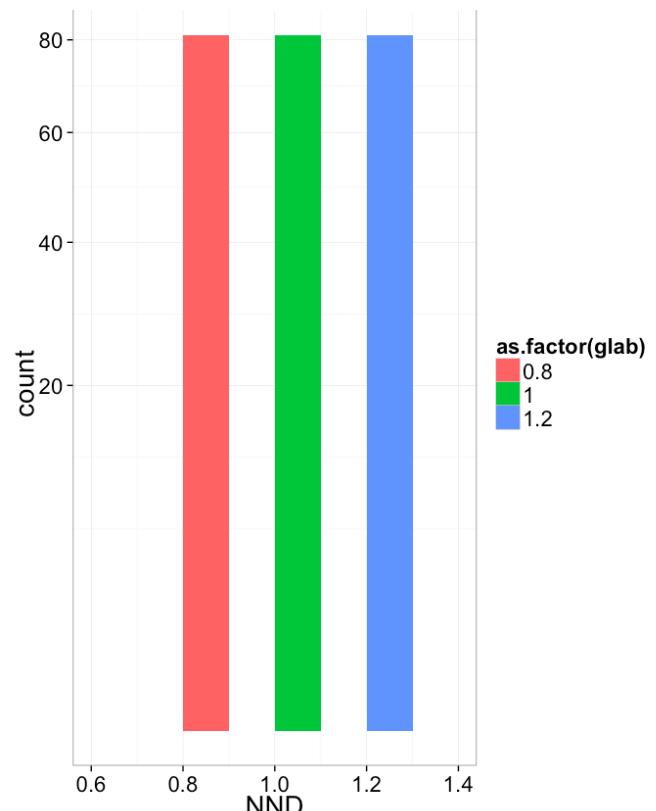
### Nearest Neighbor Orientations



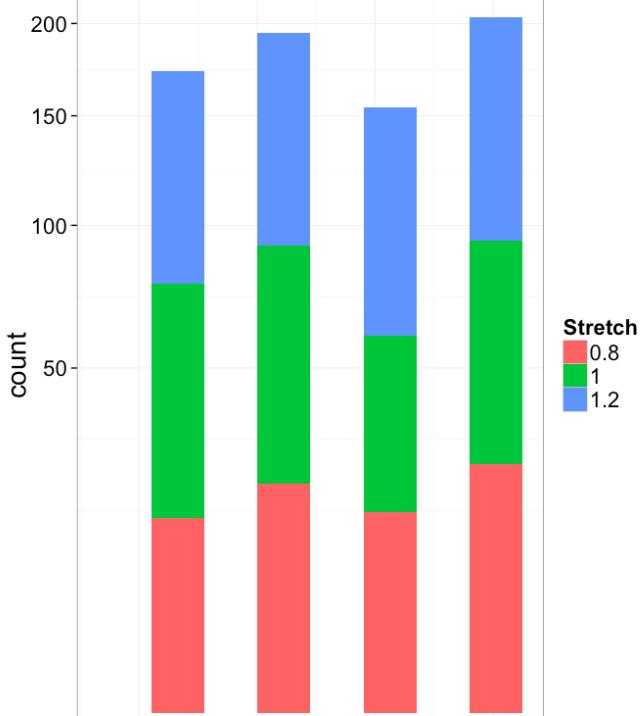


**Compression Distributions**

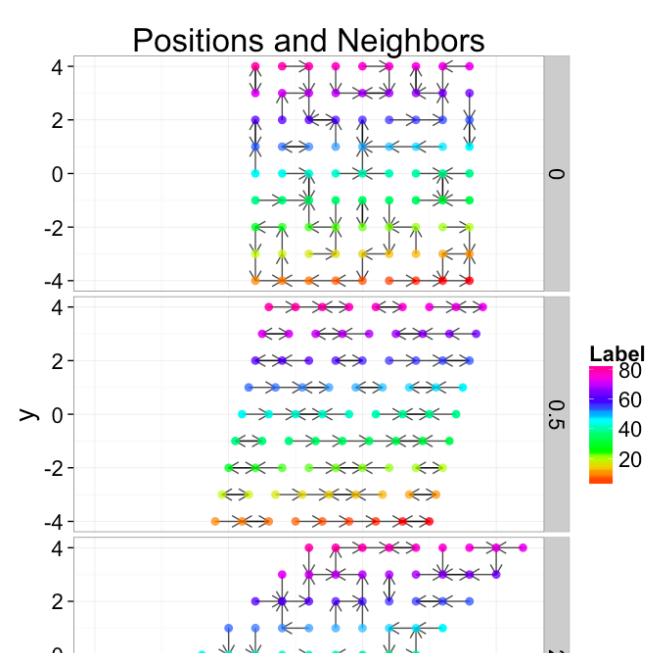
NN Distances

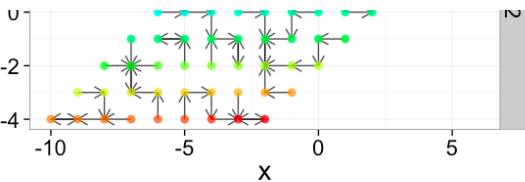


**NN Orientation**

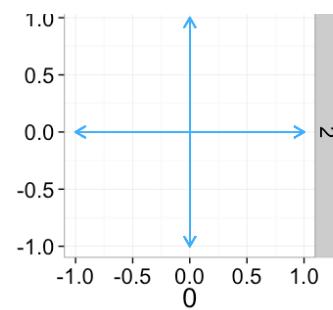
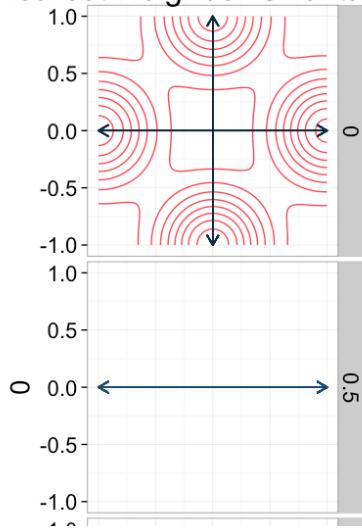


**Examining Different Shears**

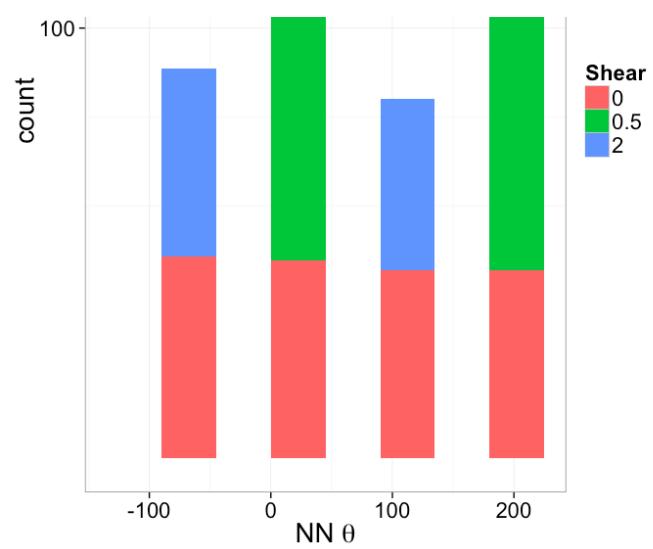
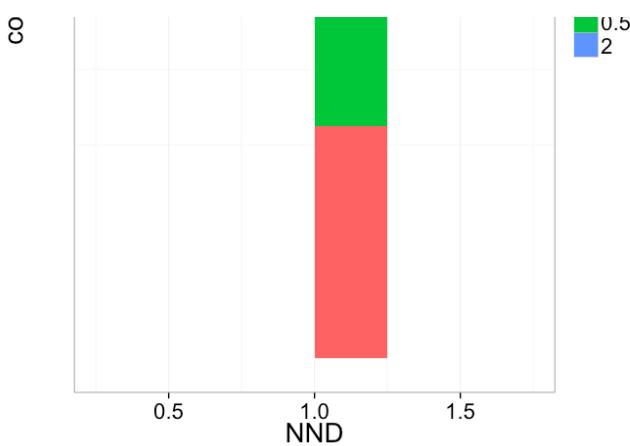
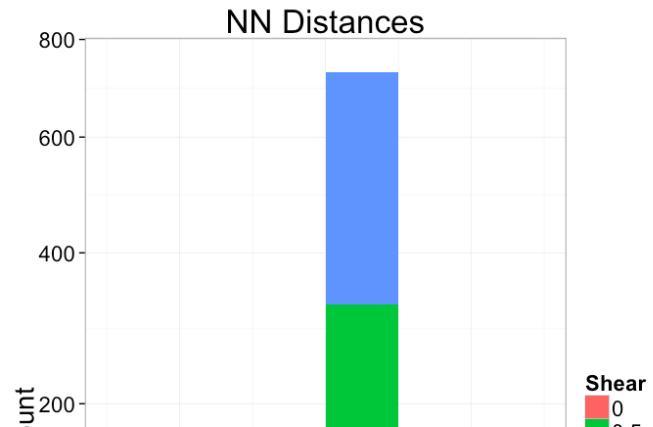




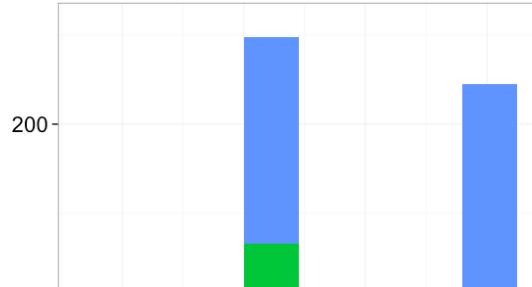
Nearest Neighbor Orientations



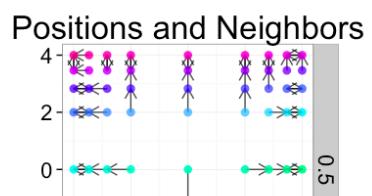
Shear Distributions

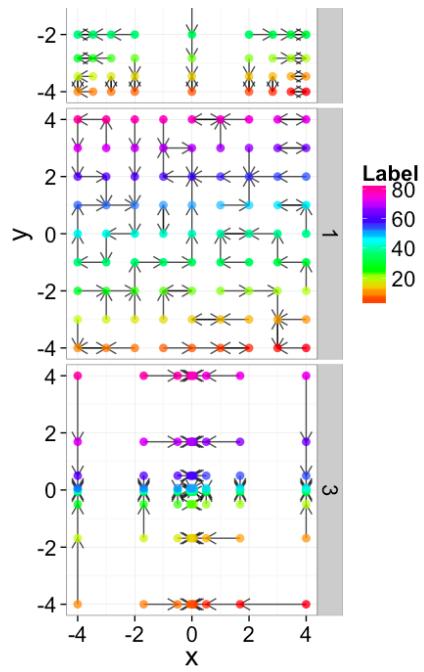


NN Orientation

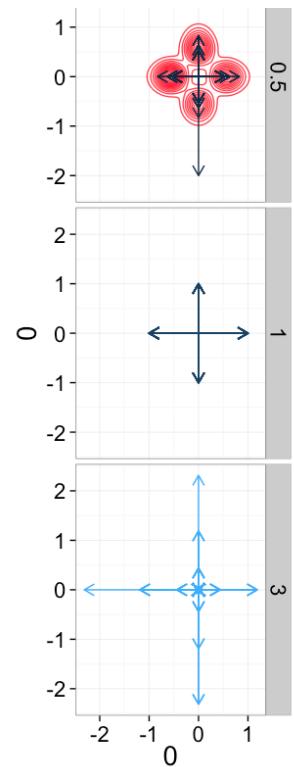


Examining Different Stretches

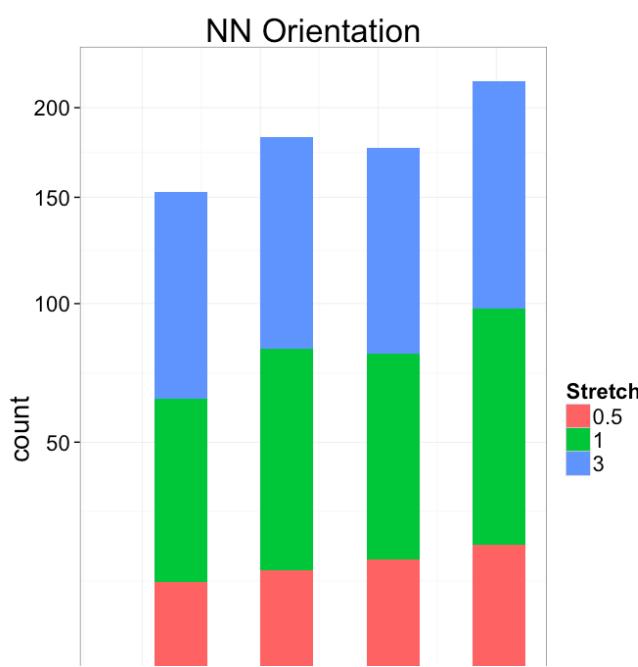
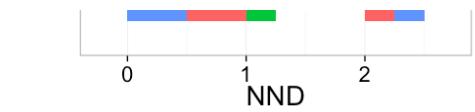
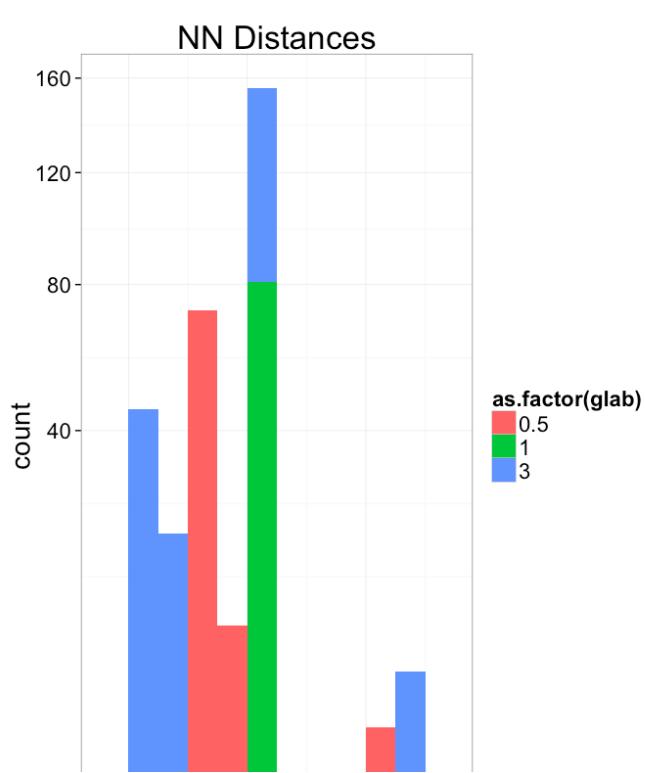


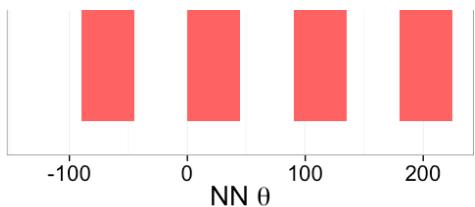


Nearest Neighbor Orientations



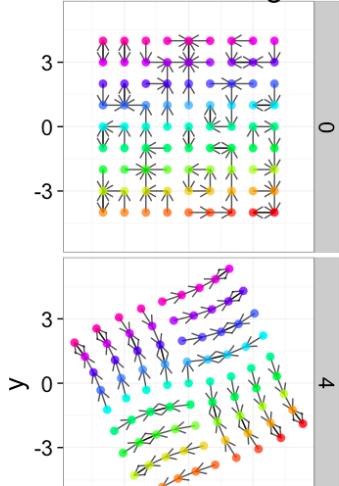
## Stretch Distributions



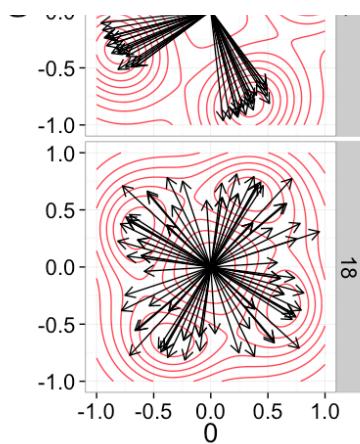
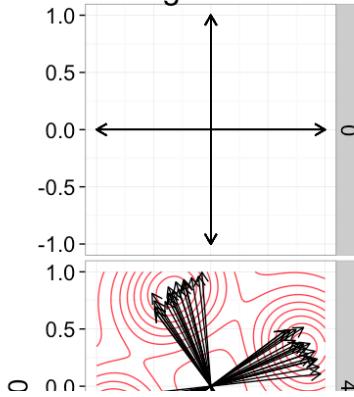


## Examining Swirl Systems

Positions and Neighbors

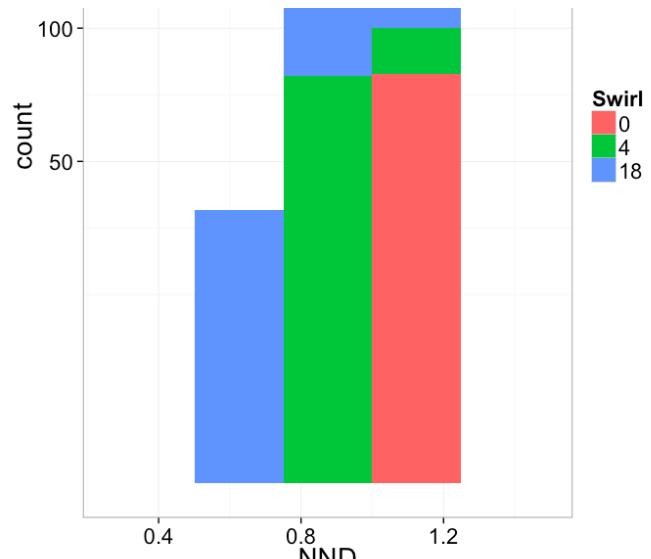


Nearest Neighbor Orientations



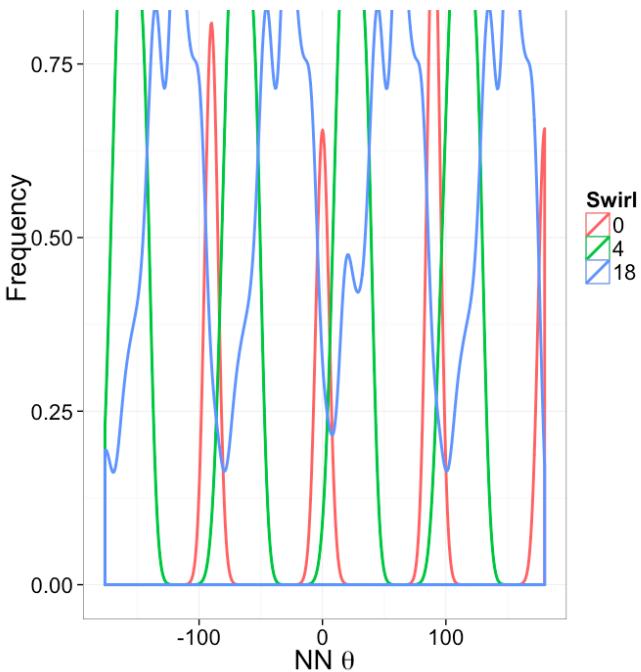
## Swirl NN Distributions

NN Distances



NN Orientation





2. Single outlier objects skew results
3. We only extract one piece of information
4. Difficult to create metrics
  - Fit a peak to the angle distribution and measure the width as the "angle variability"?

Luckily we are not the first people to address this issue

## Random Systems

Using a uniform grid of points as a starting point has a strong influence on the results. A better approach is to use a randomly distributed series of points

- resembles real data much better
- avoids these symmetry problems
  - $\epsilon$  sized edges or overlaps
  - identical distances to nearby objects

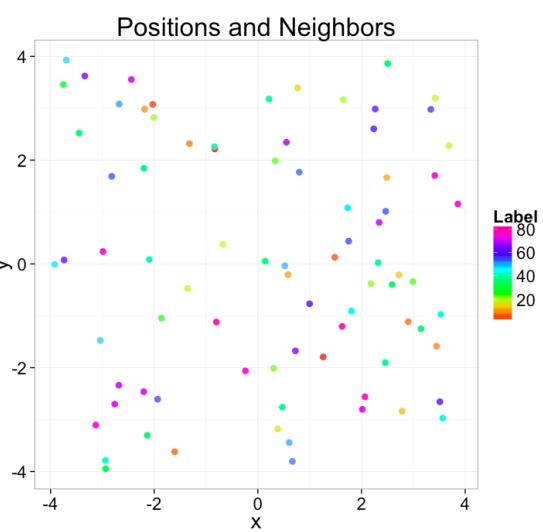
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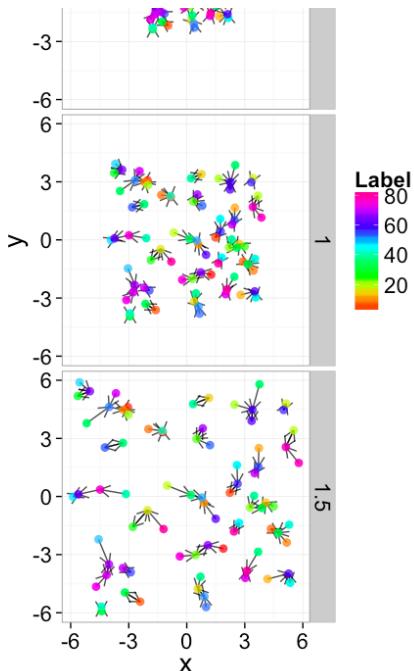
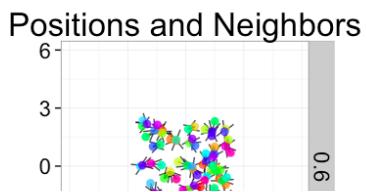
## What we notice

We notice there are several fairly significant short-comings of these metrics (particularly with in-silico systems)

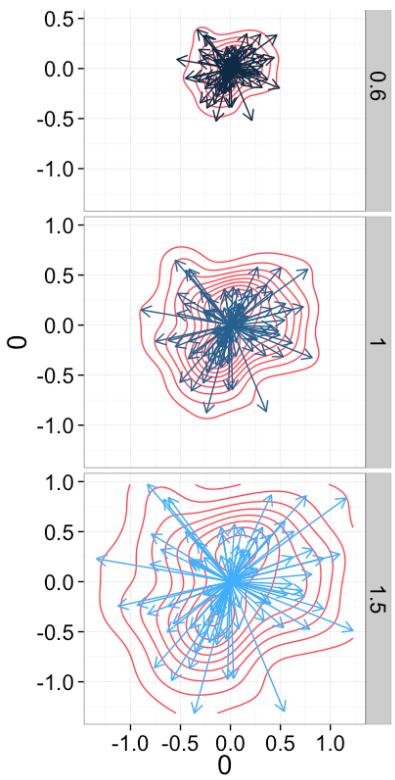
1. Orientation appears to be useful but random
  - Why should it matter if one side is 0.01% closer?



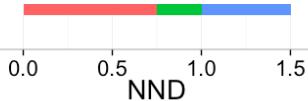
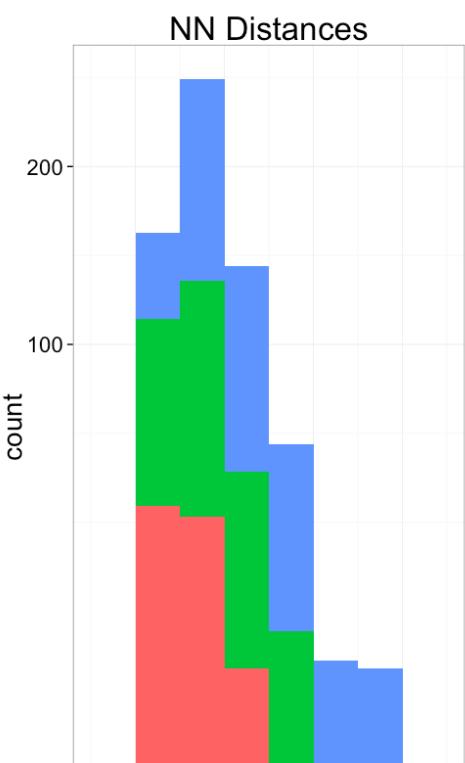
## Examining Compression



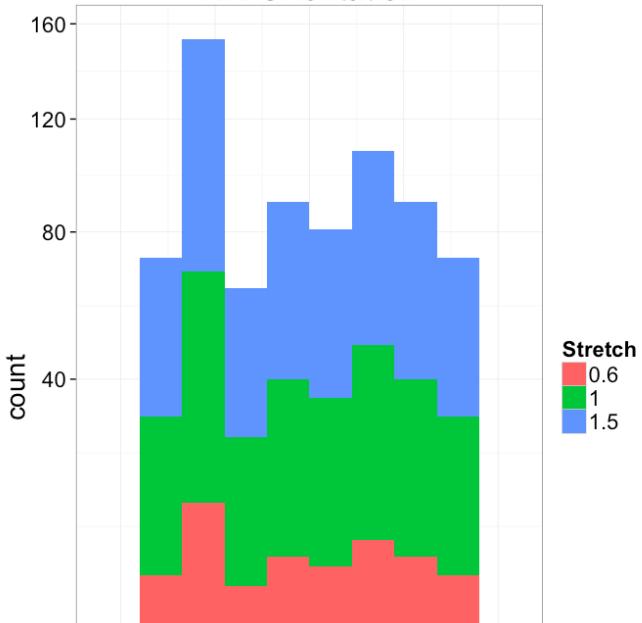
## Nearest Neighbor Orientations



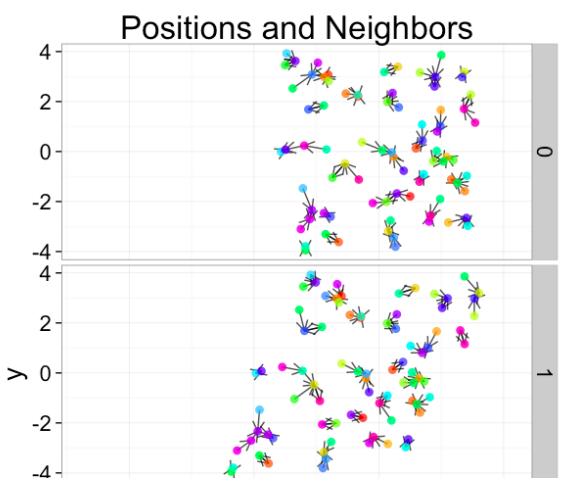
## Compression Distributions

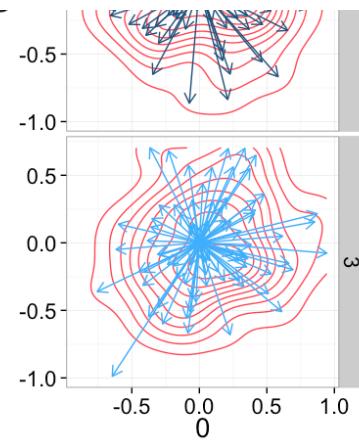
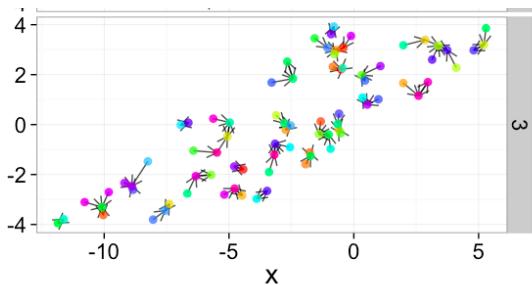


NN Orientation

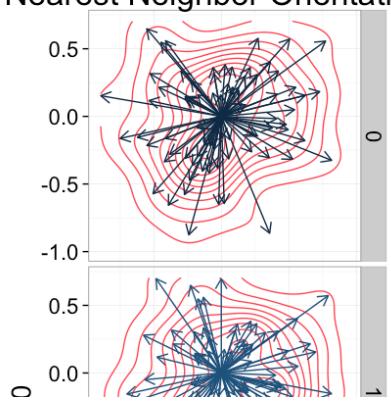


## Examining Different Shears



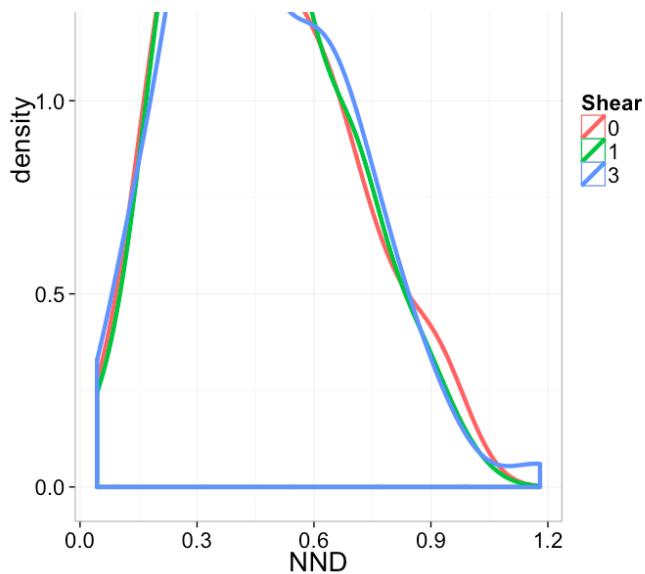


Nearest Neighbor Orientations

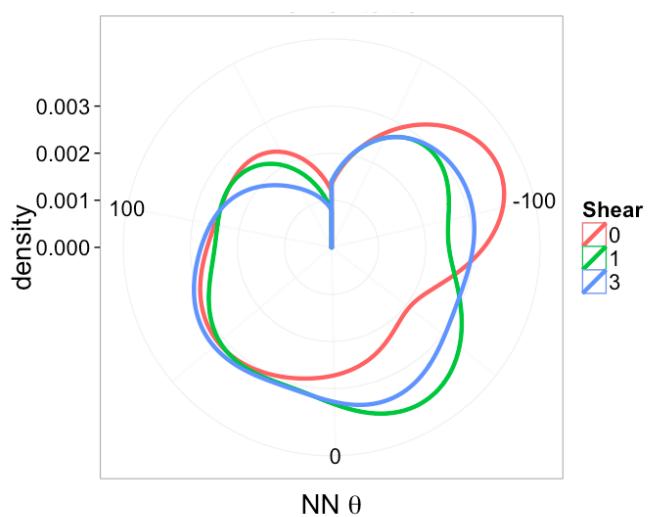


## Shear Distributions

NN Distances

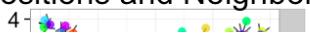


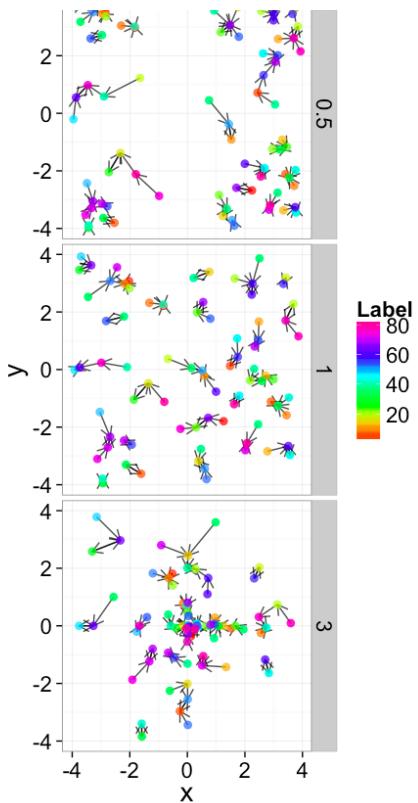
NN Orientation



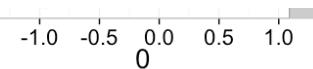
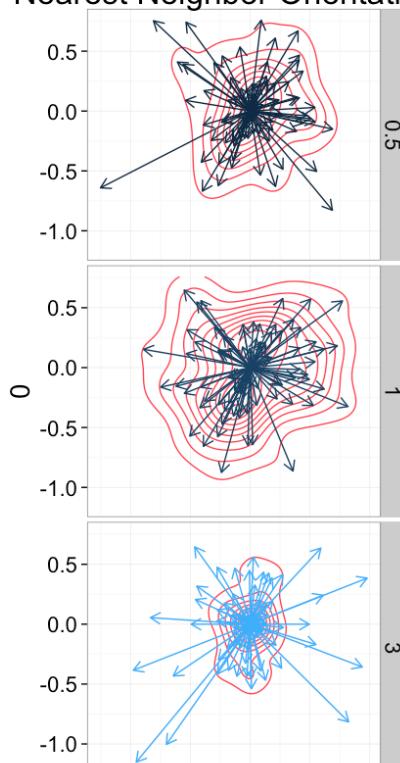
## Examining Different Stretches

Positions and Neighbors

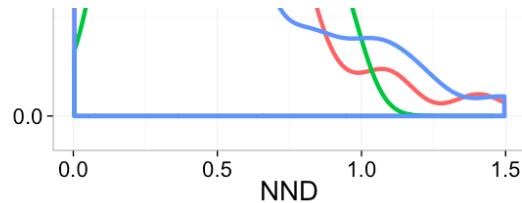
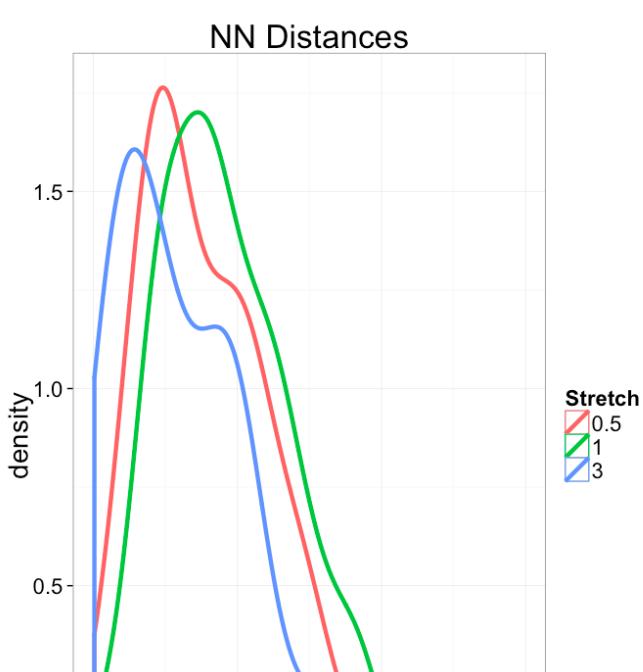




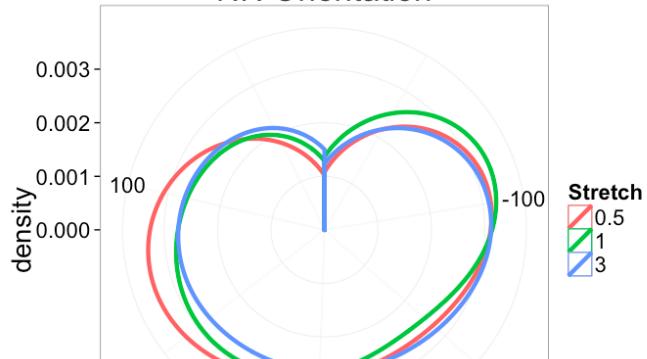
Nearest Neighbor Orientations

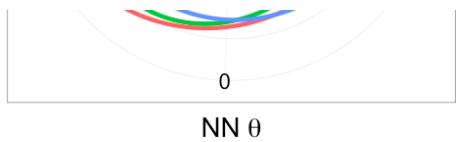


## Stretch Distributions



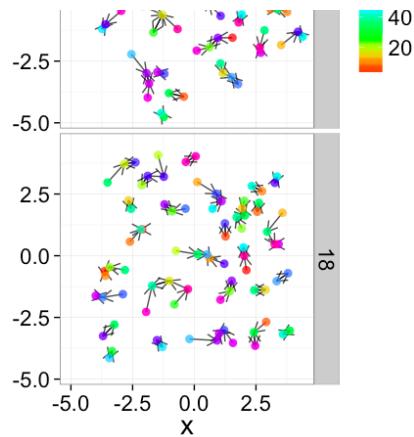
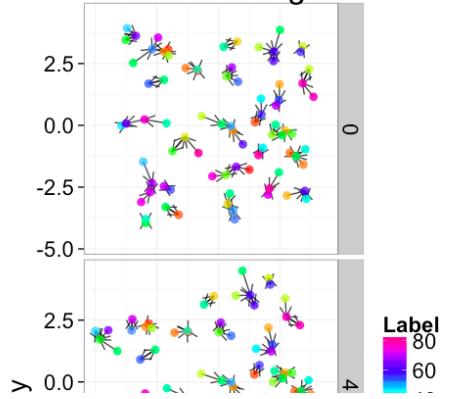
NN Orientation



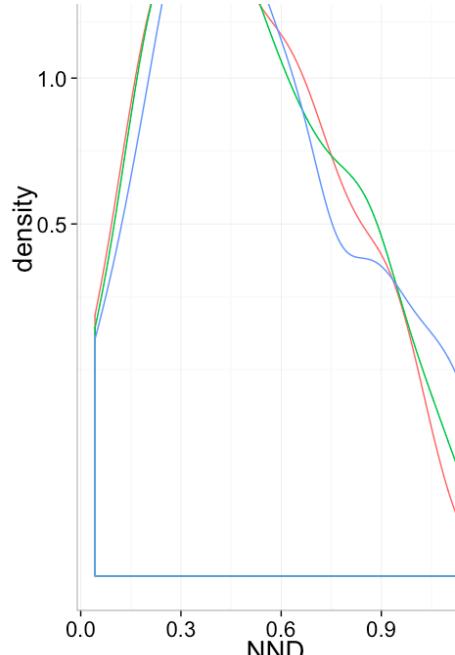
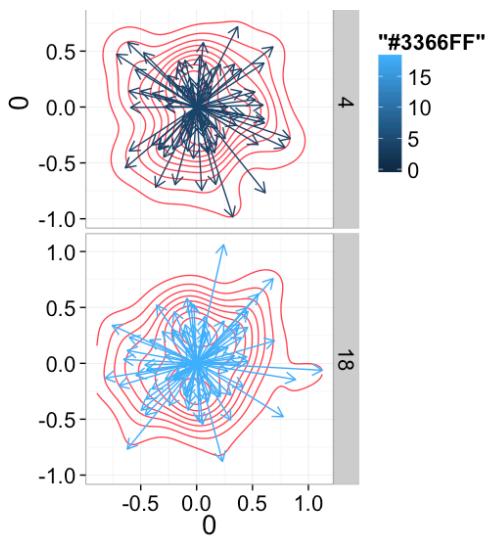
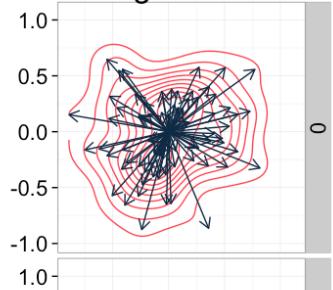


## Examining Swirl Systems

Positions and Neighbors

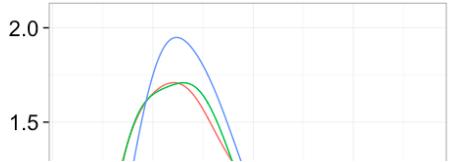


Nearest Neighbor Orientations



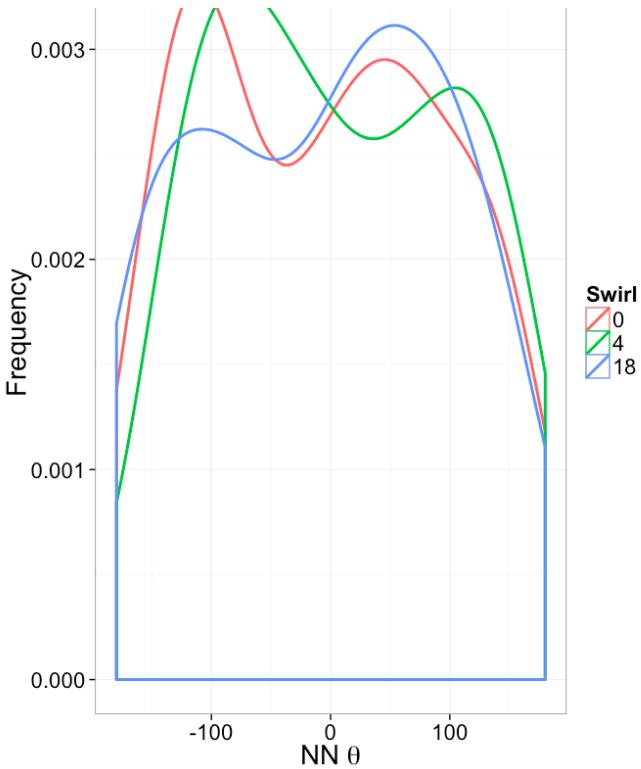
## Swirl NN Distributions

NN Distances



NN Orientation

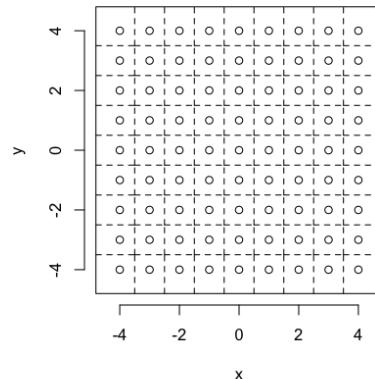




## Voronoi Tesselation

+/-. R Code

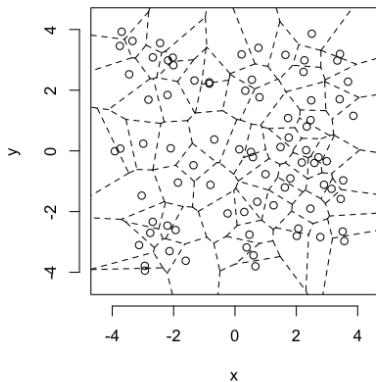
Voronoi tessellation is a method for partitioning a space based on points. The basic idea is that each point  $\vec{p}$  is assigned a region  $R$  consisting of points which are closer to  $\vec{p}$  than any of the other points. Below the diagram is shown in a dashed line for the points shown as small circles.



We call the area of a region ( $R$ ) around point  $\vec{p}$  its territory.

The grid on the random system, shows much more diversity in territory area.

+/-. R Code



## Calculating Density

Back to our original density problem of having just one number to broadly describe the system.

- Can a voronoi tessellation help us with this?
- YES

With density we calculated

$$\text{Density} = \frac{\text{Number of Objects}}{\text{Total Volume}}$$

with the regions we have a territory (volume) per object so the average territory is

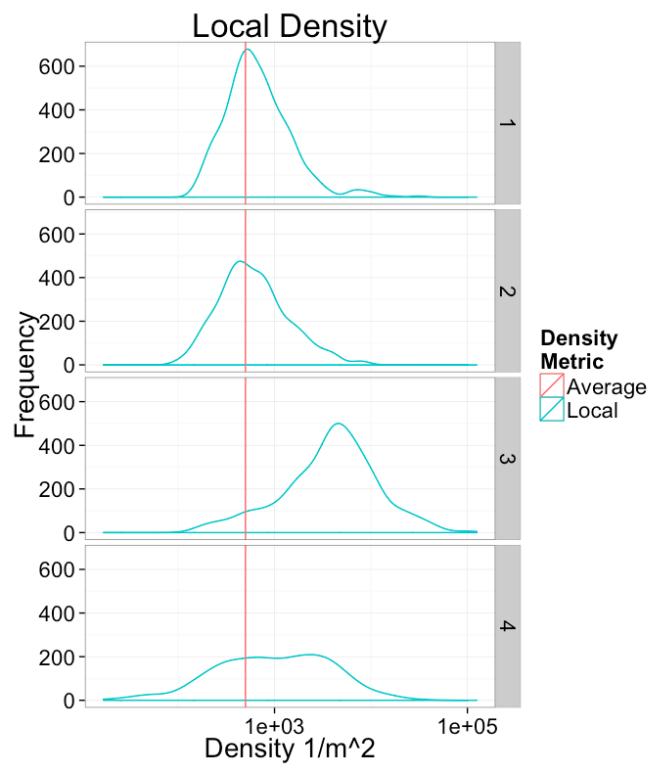
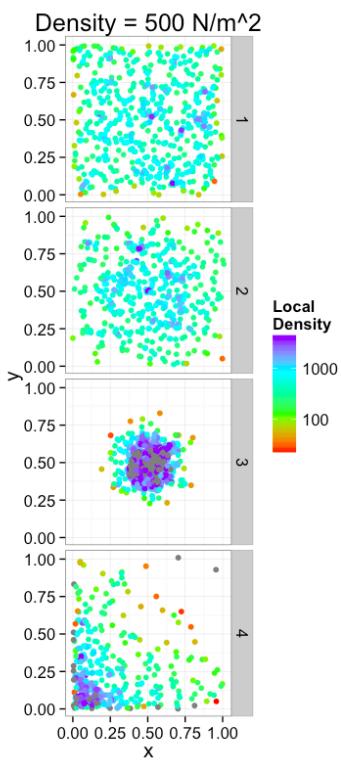
$$\bar{\text{Territory}} = \frac{\sum \text{Territory}_i}{\text{Number of Objects}} = \frac{\text{Total Volume}}{\text{Number of Objects}} = \frac{1}{\text{Density}}$$

So the same, but we now have a density definition for a single point!

$$\text{Density}_i = \frac{1}{\text{Territory}_i}$$

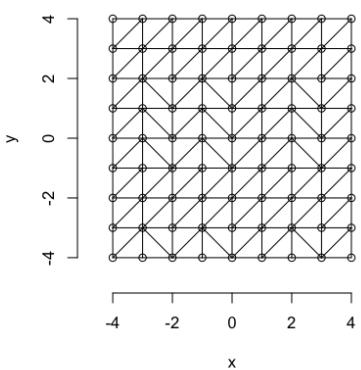
## Density Examples

+/-. R Code



## Delaunay Triangulation

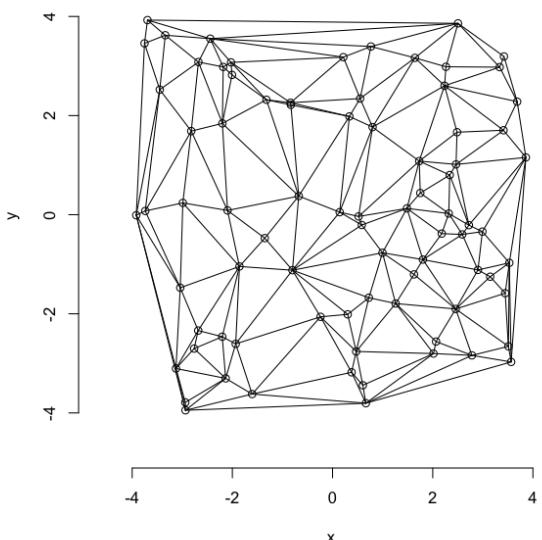
A parallel or *dual* idea (<http://mathworld.wolfram.com/DelaunayTriangulation.html>) where triangles are used and each triangle is created such that the circle which encloses it contains no other points. The triangulation makes the *neighbors* explicit since connected points in the triangulation correspond to points in our tessellation which share an edge (or face in 3D)



We define the number of connections each point  $\vec{v}_j$  has the Neighbor Count or Delaunay Neighbor Count.

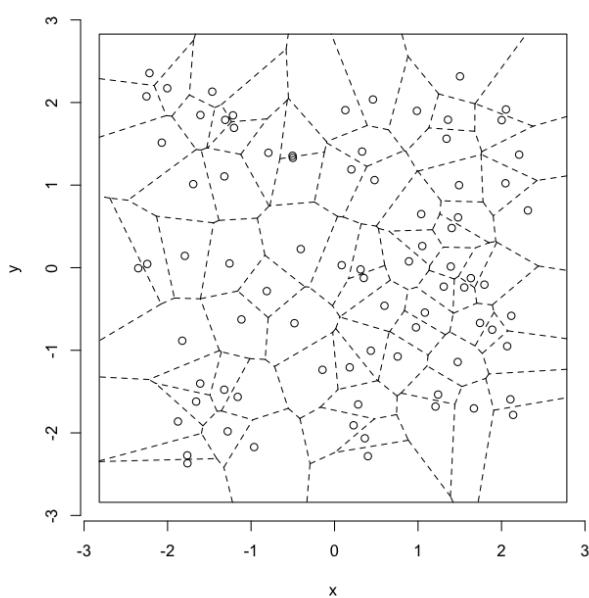
The triangulation on a random system has a much higher diversity in neighbor count

+/- R Code



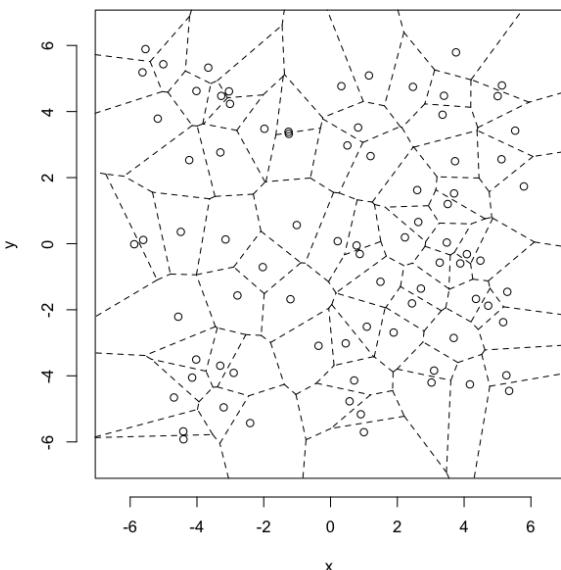
## Compression System

Compression



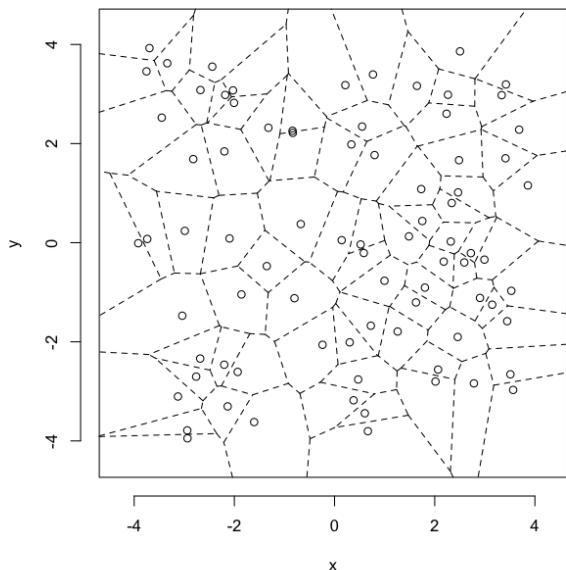
[+/- R Code](#)

Tension



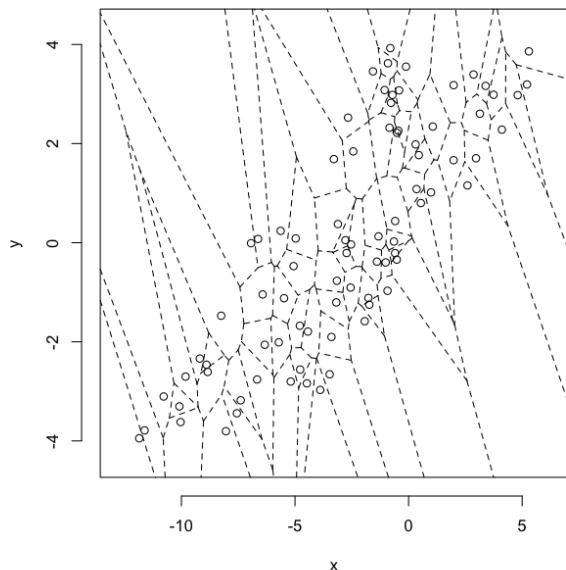
## Shear System

Low Shear



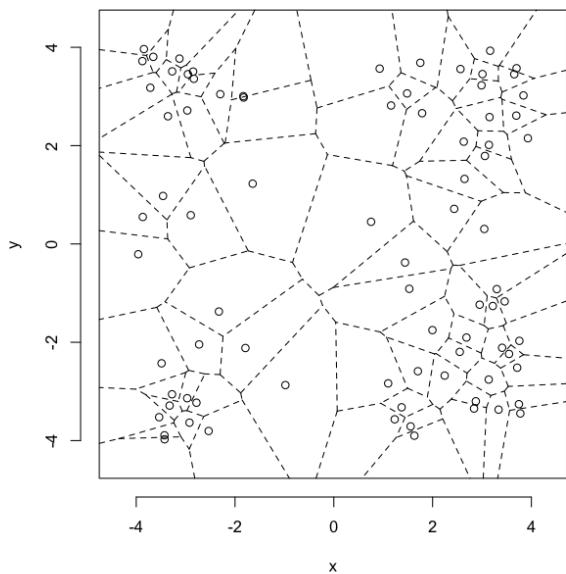
[+/- R Code](#)

High Shear



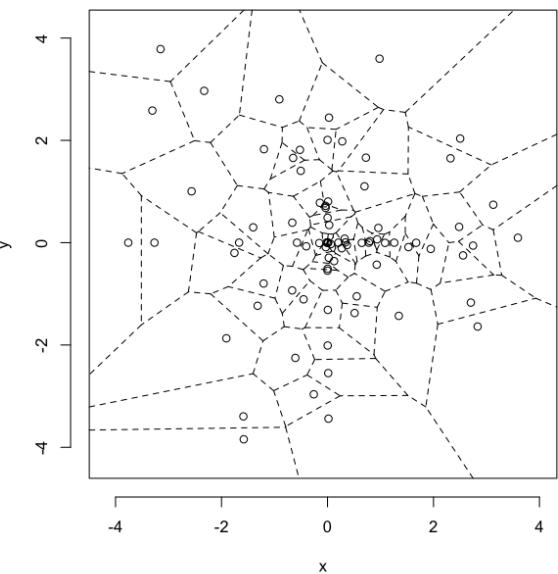
## Stretch System

Low Stretch



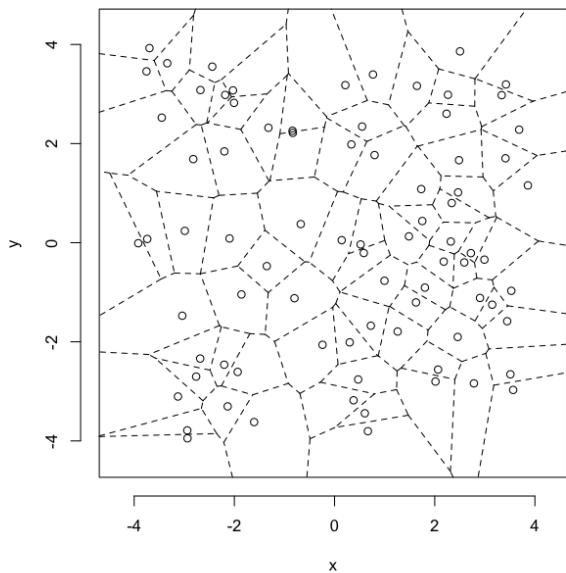
[+/- R Code](#)

**Highly Stretched System**



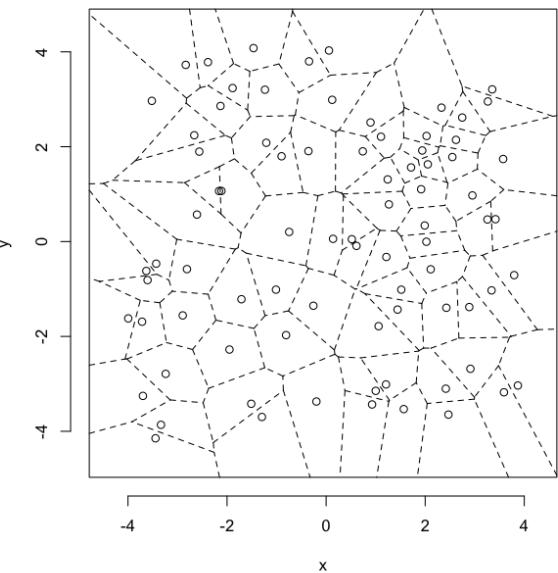
## Swirl System

**Low Swirl System**



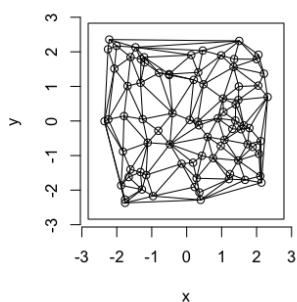
[+/- R Code](#)

**High Swirl System**



## Neighborhoods

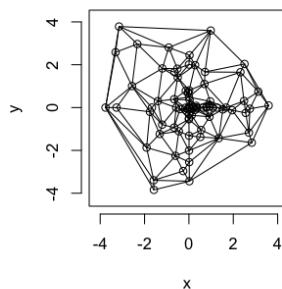
**Compression**



+/- R Code

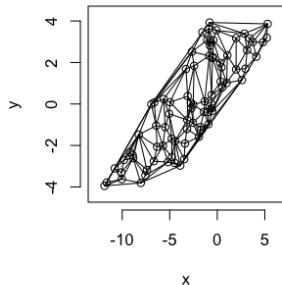
### Stretch

+/- R Code



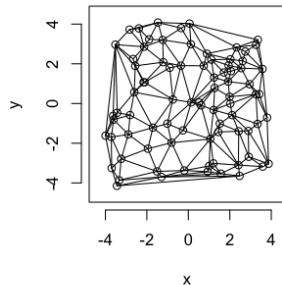
### Shear

+/- R Code



### Swirl

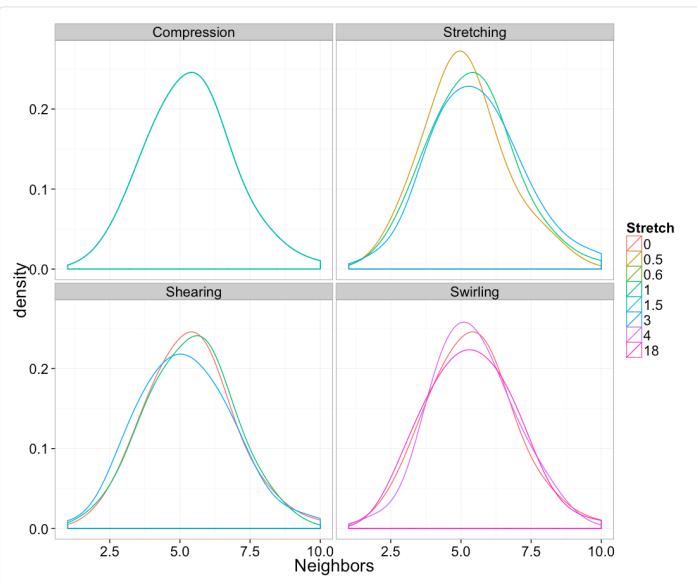
+/- R Code



### Neighbor Count

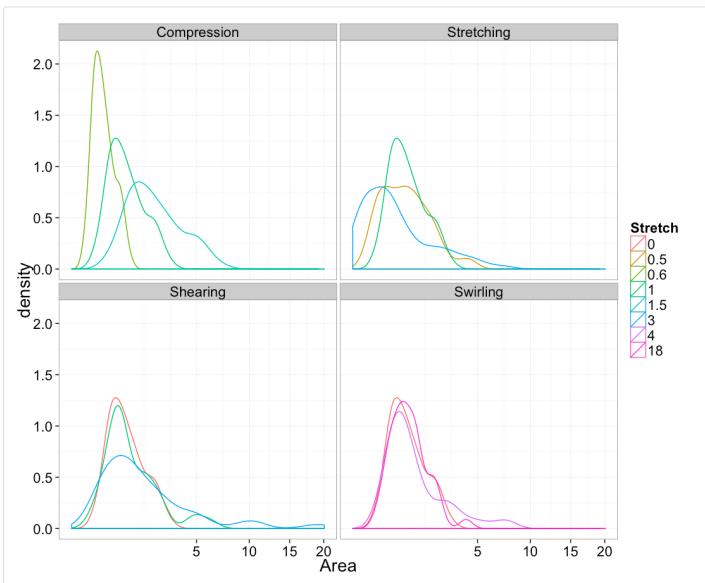
+/- R Code

+/- R Code



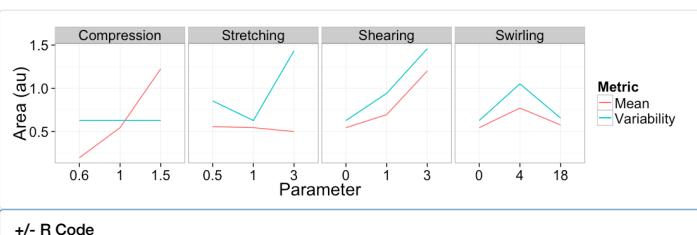
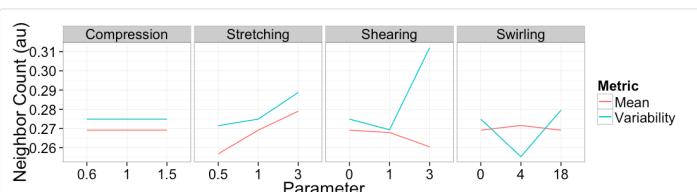
## Volume

+/- R Code



## Mean vs Variability

+/- R Code



- but in order to have a reasonable understanding of the behavior of a system we need to sample many of them.

## Understand metrics as a random system + a known transformation

- We take **mean** values
- Also **coefficient of variation** ([CV](http://en.wikipedia.org/wiki/Coefficient_of_variation) ([http://en.wikipedia.org/wiki/Coefficient\\_of\\_variation](http://en.wikipedia.org/wiki/Coefficient_of_variation))) values since they are "scale-free"

+/- R Code

## Understanding Metrics

In imaging science we always end up with lots of data, the tricky part is understanding the results that come out. With this simulation-based approach

- we generate completely random data
- apply a known transformation to it  $\mathcal{T}$
- quantify the results

We can then take this knowledge and use it to interpret observed data as transformations on an initially random system. We try and find the **rules** used to produce the sample

## Where are we at?

We have introduced a number of "operations" we can perform on our objects to change their positions

- compression
- stretching
- shearing
- swirling

We have introduced a number of metrics to characterize our images

- Nearest Neighbor distance
- Nearest Neighbor angle
- Delaunay Neighbor count
- Territory Area (Volume)

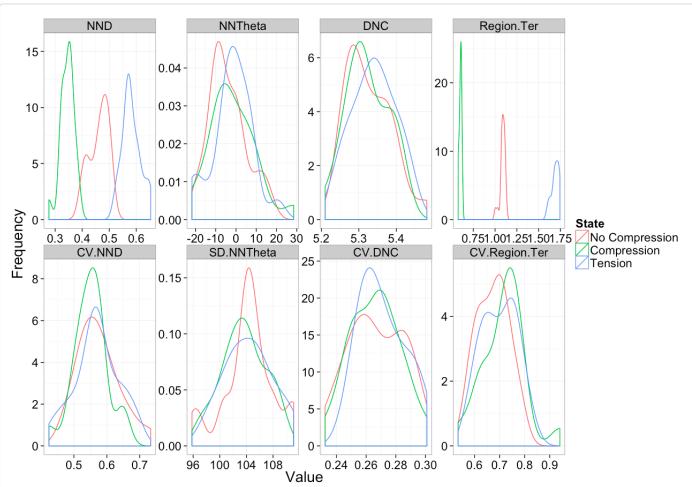
A single random systems is useful

## Examples

1. Cell distribution in bone
  - Cell position appears random
  - Metrics  $\neq$  Random Statistics
  - Cells are consistently *self-avoiding* or *stretched*
2. Egg-shell Pores
  - Pores in rock / egg shell appear random
  - Metrics  $\neq$  Random Statistics
  - Pores are also *self-avoiding*

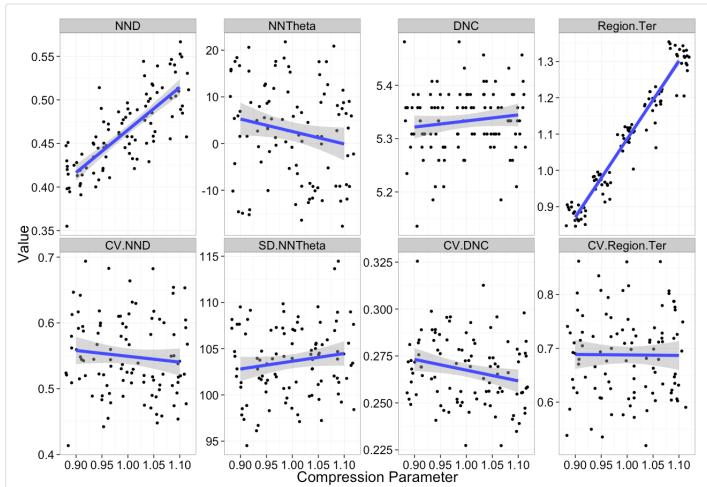
## Compression

+/- R Code



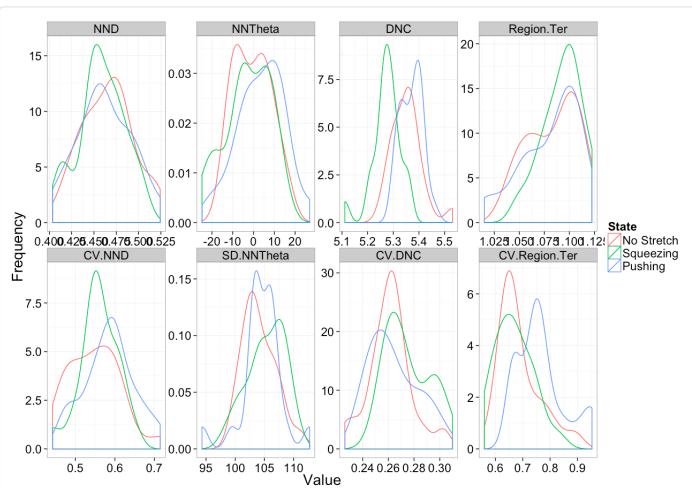
## Compression Sensitivity

+/- R Code



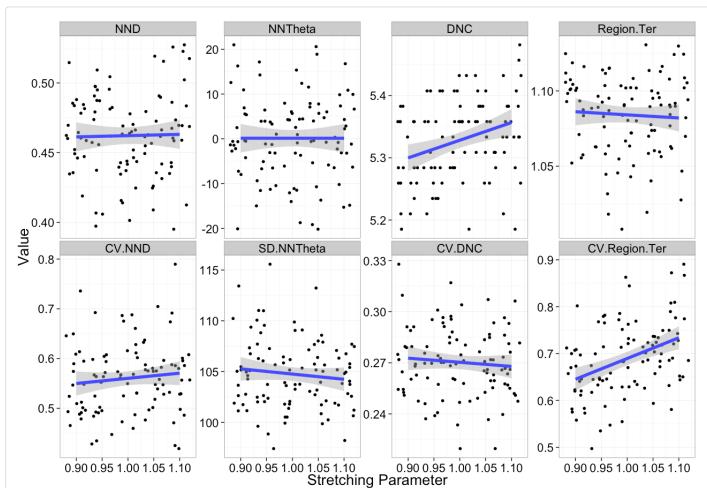
## Stretching

+/- R Code



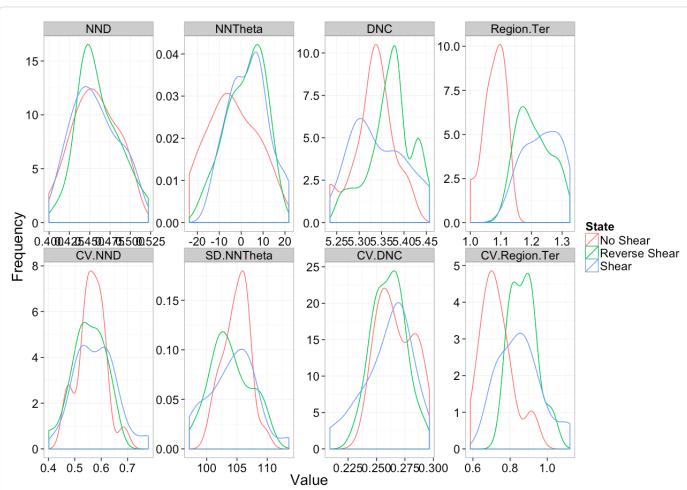
## Stretching Sensitivity

+/- R Code



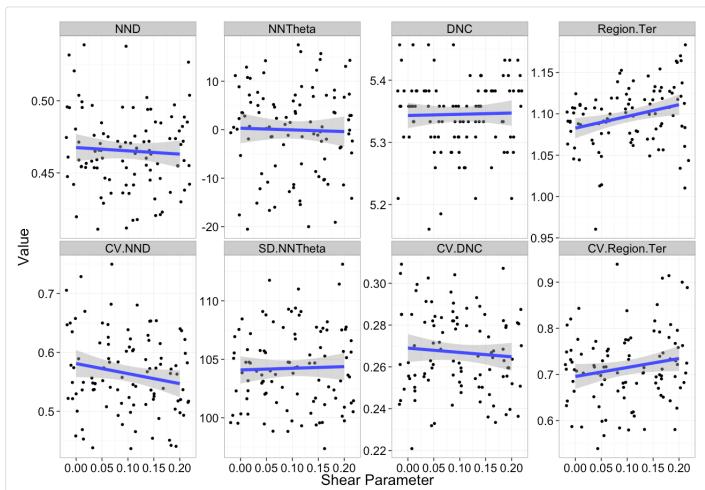
## Shearing

+/- R Code



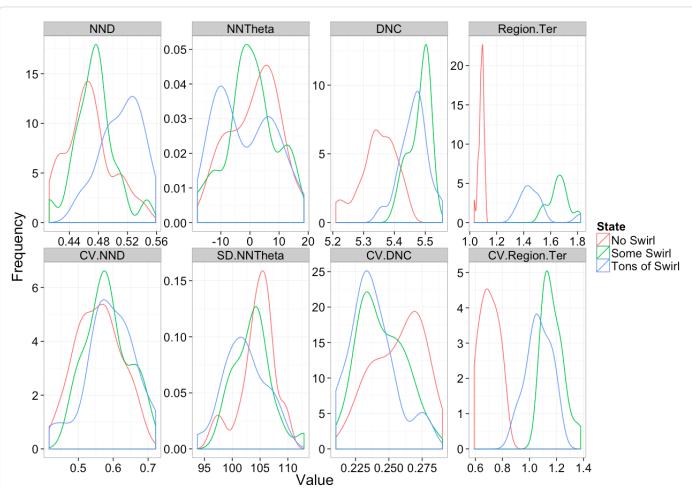
## Shearing Sensitivity

+/- R Code



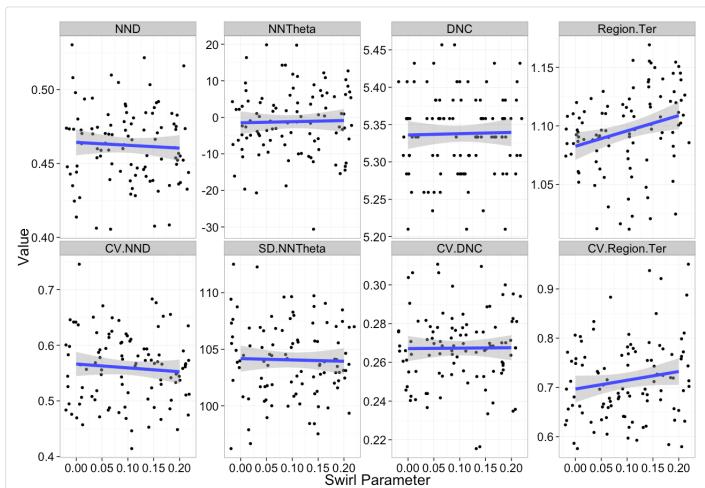
## Swirling

+/- R Code



## Swirling Sensitivity

+/- R Code



## Self-Avoiding

From the nearest neighbor distance metric, we can create a scale-free version of the metric which we call *self-avoiding coefficient* or *grouping*.

The metric is the ratio of

- observed nearest neighbor distance NND
- the expected mean nearest neighbor distance ( $r_0$ ) for a random point distribution (Poisson Point Process) with the same number of points (N.Obj) per volume (Total.Volume).

$$r_0 = \sqrt{\frac{\text{Total.Volume}}{2\pi \text{N.Obj}}}$$

Using the territory we defined earlier (Region area/volume) we can simplify the definition to

$$r_0 = \sqrt{\frac{T_{\text{er}}}{2\pi}}$$

$$\text{SAC} = \frac{\text{NND}}{\sqrt{\frac{T_{\text{er}}}{2\pi}}}$$

## Distribution Tensor

So the information we have is 3D why are we taking single metrics (distance, angle, volume) to quantify it.

- Shouldn't we use 3D metrics with 3D data?
- Just like the shape tensor we covered before, we can define a *distribution* tensor to characterize the shape of the distribution.
- The major difference instead of constituting voxels we use edges
  - an edge is defined from the Delaunay triangulation
  - it connects two neighboring bubbles together
- We can calculate distribution for a single bubble by taking all edges that touch the object or from a region by finding all edges inside that region

We start off by calculating the covariance matrix from the list of edges  $\vec{v}_{ij}$  in a given volume  $\mathcal{V}$

$$\vec{v}_{ij} = \vec{\text{COV}}(i) - \vec{\text{COV}}(j)$$

$$\text{COV}(\mathcal{V}) = \frac{1}{N} \sum_{\text{VCOM}(i) \in \mathcal{V}} \begin{bmatrix} \vec{v}_x \cdot \vec{v}_x & \vec{v}_x \cdot \vec{v}_y & \vec{v}_x \cdot \vec{v}_z \\ \vec{v}_y \cdot \vec{v}_x & \vec{v}_y \cdot \vec{v}_y & \vec{v}_y \cdot \vec{v}_z \\ \vec{v}_z \cdot \vec{v}_x & \vec{v}_z \cdot \vec{v}_y & \vec{v}_z \cdot \vec{v}_z \end{bmatrix}$$

## Distribution Tensor (continued)

We then take the eigentransform of this array to obtain the eigenvectors (principal components,  $\vec{\Lambda}_{1\dots 3}$ ) and eigenvalues (scores,  $\lambda_{1\dots 3}$ )

$$\text{COV}(I_{id}) \longrightarrow \begin{bmatrix} \vec{\Lambda}_{1x} & \vec{\Lambda}_{1y} & \vec{\Lambda}_{1z} \\ \vec{\Lambda}_{2x} & \vec{\Lambda}_{2y} & \vec{\Lambda}_{2z} \\ \vec{\Lambda}_{3x} & \vec{\Lambda}_{3y} & \vec{\Lambda}_{3z} \end{bmatrix} * \underbrace{\begin{bmatrix} \lambda_1 & 0 & 0 \\ 0 & \lambda_2 & 0 \\ 0 & 0 & \lambda_3 \end{bmatrix}}_{\text{Eigenvalues}} * \begin{bmatrix} \vec{\Lambda}_{1x} & \vec{\Lambda}_{1y} & \vec{\Lambda}_{1z} \\ \vec{\Lambda}_{2x} & \vec{\Lambda}_{2y} & \vec{\Lambda}_{2z} \\ \vec{\Lambda}_{3x} & \vec{\Lambda}_{3y} & \vec{\Lambda}_{3z} \end{bmatrix}^T$$

The principal components tell us about the orientation of the object and the scores tell us about the corresponding magnitude (or length) in that direction.

## Distribution Anisotropy

Visual example

- Tensor represents the average spacing between objects in each direction  $\approx$  thickness of background.
- Its interpretation is more difficult since it doesn't represent a *real* object



From this tensor we can define an anisotropy in the same manner as we defined for shapes. The anisotropy defined as before

$$\text{Aniso} = \frac{\text{Longest Side} - \text{Shortest Side}}{\text{Longest Side}}$$

- An isotropic distribution tensor indicates the spacing between objects is approximately the same in every direction
  - Does not mean a grid, organized, or evenly spaced!
  - Just the same in every direction
- Anisotropic distribution tensor means the spacing is smaller in one direction than the others
  - Can be regular or grid-like
  - Just closer on one direction

## Distribution Oblateness



From this tensor we can also define oblateness in the same manner as we defined for shapes. The oblateness is also defined as before as a type of anisotropy

$$\text{Ob} = 2 \frac{\lambda_2 - \lambda_1}{\lambda_3 - \lambda_1} - 1$$

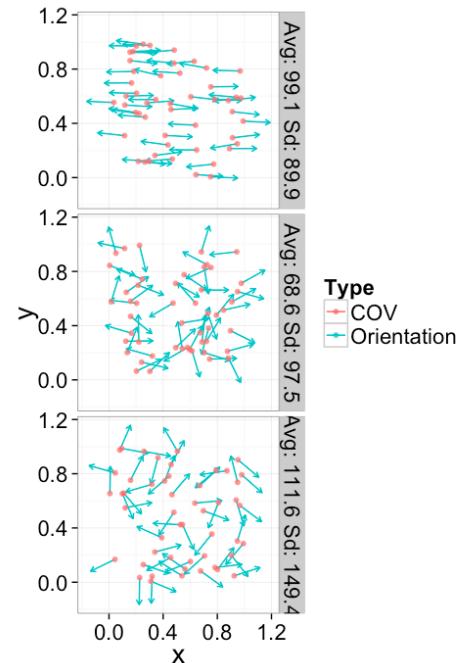
- Indicates the pancakeness of the distribution
- Oblate means it is very closely spaced in one direction and far in the other two
  - strand-like structure
- Prolate means it is close in two directions and far in the other

- sheet-like structure

## Orientation

The shape tensor provides for each object 3 possible orientations (each of the eigenvectors). For simplicity we will take the primary direction (but the others can be taken as well, and particularly in oblate or pancake shaped objects the first is probably not the best choice!)

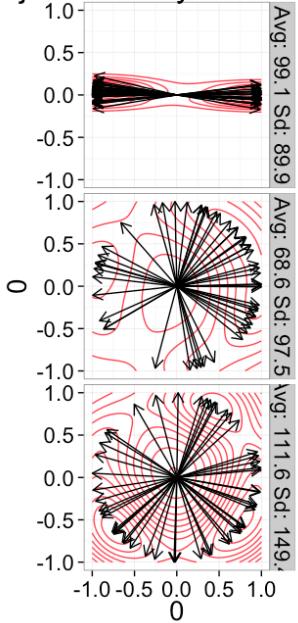
+/- R Code



## Orientation

Since orientation derived from a shape tensor / ellipsoid model has no heads or tails. The orientation is only down to a sign  $\rightarrow = \leftarrow$  and  $\uparrow = \downarrow$ .

## Object Primary Orientations



This means calculating the average and standard deviation are very poor descriptors of the actual dataset. The average for all samples below is around 90 (vertical) even though almost no samples are vertical and the first sample shows a very high (90) standard deviation even though all the samples in reality have the same orientation.

+/- R Code

Angle.Variability	Mean.Angle	Sd.Angle
5.729578	99.14070	89.91135
62.864789	68.63894	97.47078
120.000000	111.58325	149.44605

The problem can be dealt with by using the covariance matrix which takes advantage of the products which makes the final answer independent of sign.

## Alignment Tensor

We can again take advantage of the versatility of a tensor representation for our data and use an *alignment tensor*.

- The same as all of the tensors we have introduced
- Except we use the primary orientation instead of voxel positions (shape) or edges (distribution)
- Similar to distribution it is calculated for a volume ( $V$ ) or region and is meaningless for a single object

$$\vec{v}_i = \vec{\Lambda}_1(i)$$

$$COV = \frac{1}{N} \sum_{\forall COM(i) \in V} \begin{bmatrix} \vec{v}_x \vec{v}_x & \vec{v}_x \vec{v}_y & \vec{v}_x \vec{v}_z \\ \vec{v}_y \vec{v}_x & \vec{v}_y \vec{v}_y & \vec{v}_y \vec{v}_z \\ \vec{v}_z \vec{v}_x & \vec{v}_z \vec{v}_y & \vec{v}_z \vec{v}_z \end{bmatrix}$$

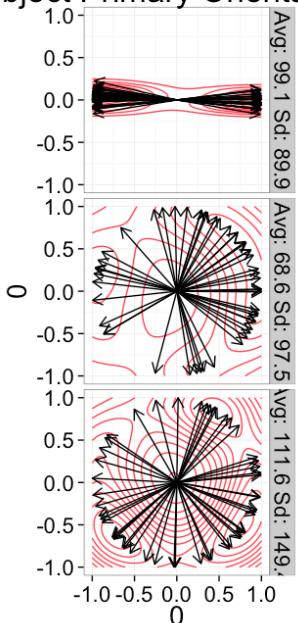


## Alignment Tensor: Example

Using the example from before

+/- R Code

## Object Primary Orientations



## Alignment Anisotropy

Anisotropy for alignment can be summarized as degree of alignment since very anisotropic distributions mean the objects are aligned well in the same direction while an isotropic distribution means the orientations are random. Oblateness can also be defined but is normally not particularly useful.

$$Aiso = \frac{\text{Longest Side} - \text{Shortest Side}}{\text{Longest Side}}$$



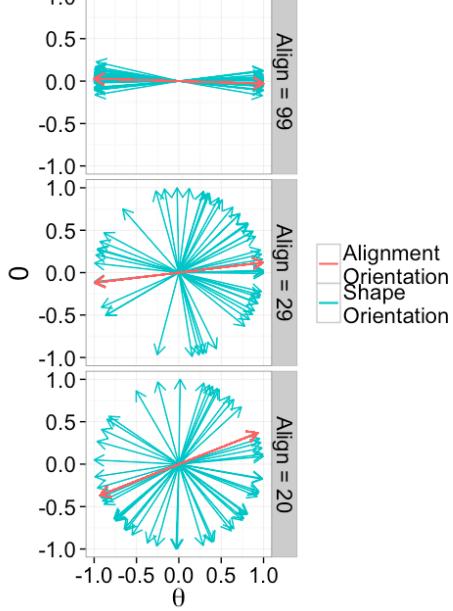
## Alignment Anisotropy Applied

+/- R Code

Show some tensor stuff here

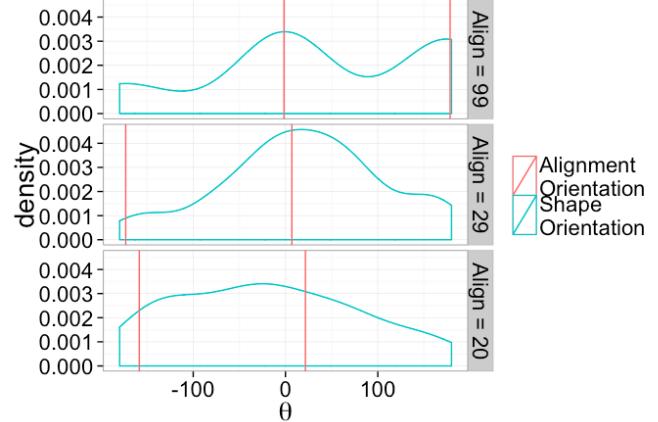
+/- R Code

## Object Primary Orientations



+/- R Code

## Histogram vs Orientation



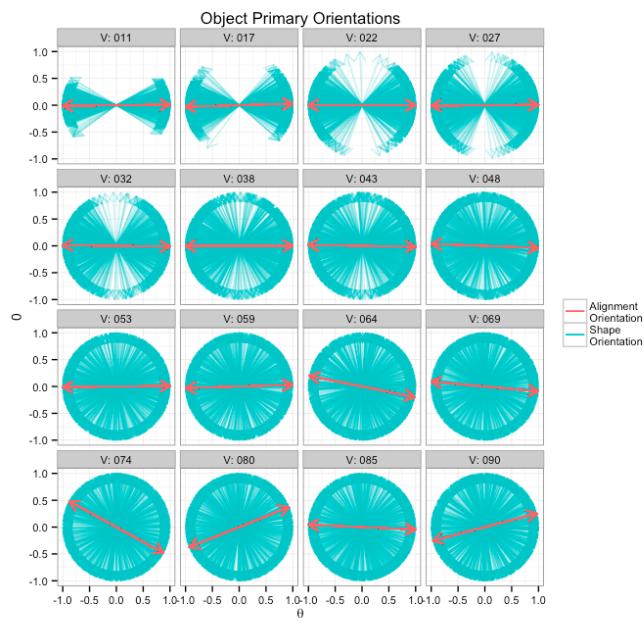
+/- R Code

Angle.Variability	Mean.Angle	Sd.Angle	Alignment
5.729578	99.14070	89.91135	99.21383
62.864789	68.63894	97.47078	28.88033
120.000000	111.58325	149.44605	19.56784

## Alignment for many samples

+/- R Code

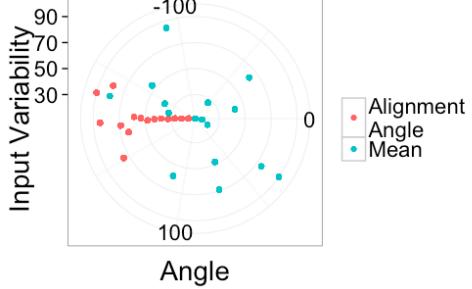
### Object Primary Orientations



Angle Accuracy

+/- R Code

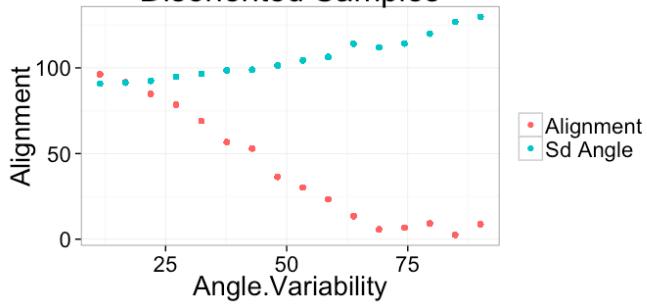
## Alignment Angle vs Mean Angle for Increasingly Disoriented Samples



Variability Accuracy

+/- R Code

## Alignment vs Sd for Increasingly Disoriented Samples



- Where are cells located in reference to canals (peaks in  $F(\vec{r})$  function,  $A$  is a point inside a canal  $B$  is the cells)

## Other Approaches

### K-Means

K-Means can also be used to classify the point-space itself after shape analysis. It is even better suited than for images because while most images are only 2 or 3D the shape vector space can be 50-60 dimensional and is inherently much more difficult to visualize.

### 2 Point Correlation Functions

For a wider class of analysis of spatial distribution, there exist a class of functions called *N-point Correlation Functions*

- given a point of type  $A$  at  $\vec{x}$
- what is the probability of  $B$  at  $\vec{x} + \vec{r}$
- How is it useful
  - A simple analysis can be used to see the spacing of objects (peaks in the  $F(\vec{r})$  function,  $A$  and  $B$  are the same phase).