## Introduction to Professional Scientific Communication Prof. S. Ganesh Department of Biological Sciences & Bioengineering Indian Institute of Technology, Kanpur

## Lecture – 15 How to Prepare Figures

So, welcome back to week four lectures. So, we have been discussing about the results section as to how you can put together your results section for a manuscript or a thesis. And we have been discussing about how do you generate the figures what are the ways by which you should make the schematics, figures and how they are helpful in explaining your results. So, in continuation of the discussion, in this lecture, we are going to look at some of the guidelines for how to make very good figures.

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It could be bar diagram, it could be figures, it could be schematics, so you are going to look into some of those issues. Again as the title of the course say these are all introduction. So, it does not really get into every point that needs to be addressed, but it gives you overall you know a guidelines as to what are the point that you should look at and you have to appreciate that you have to give a lot more thoughts when you prepare images, figures and schematics. So, it is just an introduction to this topic.

So, I have shown here in this particular you know slide which again I obtained from the publisher Springer. The figures and tables what you call as display items are often the

quickest way to communicate large amount of complex information that you would be that would be complicated otherwise to explain in the text right, so that is what we are continuing to discuss. So, we are going to look at you know how you make images how do you make data plots and so on.

So, for you know when you make images be sure to include scale bars. So, you may have for example, taken a microscopic image of a cell or tissue or you know a part of the wing of a butterfly whatever it is, but you need to give a scale bar saying that what is the magnification right, so that is very, very important. This one such data point that you have to keep in mind.

Consider labeling important items, so if because you are looking at a small of segment of a lot of data otherwise you would have understood, but you have zoomed in to such a high magnification that you do not know what it is or you want reader to look at a particular you know segment of the image. So, you want to give an image in a label that with an arrow or asterisk or something you know, so therefore, you can appreciate what you want to say. Indicate the meaning of different colors and symbols used right. This statement is made assuming that the colors and symbols you have used have got some meaning, so that is something that you come back little later.

You know. So, you have for example, a color image where in a cell is you know stained using different flourochrome which gives you red, green and blue and so on. Now, so each one what does it mean each color what does it mean. So, you have to identify that with a label. So, you want to say the red means mitochondria. So, you want to write mitochondria in red color. Therefore it convey the red signal that you find in that image is mitochondria or if it is a particular protein that you have stained with the red color dye then you want to use that name and blue meaning say nuclei or whatever it is. So, you need to identify them it is important.

And if you are use some symbol for example, you have an you know high magnified view of a cell and there are some say mitochondria, some are normal looking some or abnormal looking. So, you want to put an asterisk next to the abnormal looking mitochondria, and then right in the legend that the asterisk indicate an abnormal mitochondria with regard to the size, shape or how does it look like and so on. So,

therefore, you have to you know indicate the meaning of the colors and symbols in the figure legend we will see little later.

For data plots, when you talk about bar diagram and others we sure to label all the access you have y-axis, you have x-axis, you have to say what the value mean there. You know it could be time, it could be temperature, it could be protein level and so on, but you have to exactly explain as to what it mean. Specify units for quantities. For example, you have given some number 20, 40, 60, 80, what does that mean right, is it milligram per some volume or in molarity or fold change, what is that you have to say that. Label all curves and data sets in your bar diagram or in a line diagram you have multiple lines you know what each line indicate that needs to be explained in the figure.

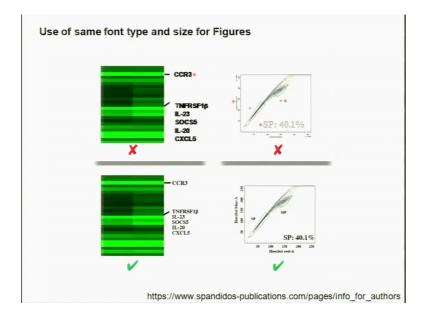
Use legible font size. You know you may have a big bar diagram, but you are you know whatever values or labels that you are adding, it is too small then when it is printed you cannot even read it right, so that becomes difficult you know to convey what information you want convey. Therefore, it should be legible. It should not be too big either. So, it has to have some relation that you need to know and what size I should give and so on.

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So, there are a large number of reading materials available for how to make figures and so on. So, what I am showing here is the figure instruction for accepted manuscript this is from a general called general of clinical investigation likewise most of the publishers have detailed instruction. So, you may want to go and consult a journal that publishes more of your kind of work, so that would tell you how to make figures for your kind of work. So, I am just giving you one such example. So, what I am going to do is to give examples from this journal, but I am also given you an another link at the bottom which from the science magazine that also gives you more information for more multidisciplinary area as to how to make figures and a schematics and so on.

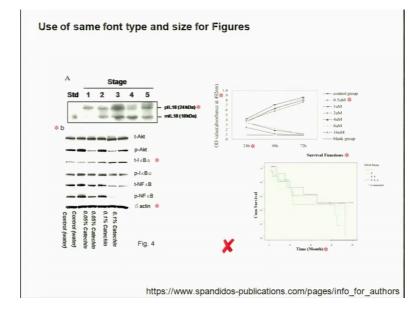
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I am going to give you some examples here some from some things right. So, you can see use of same font type and size for figures. You can see the figure on the top, it is the figure is almost identical except that the how they are labeled is what we are going to look into. The one on the top identified by the red cross mark is that this is not good. You can see that you know the label that are used in bold font IL 23 for example, you cannot even differentiate them the font is so horrible. And then again in the scatter plot on the right side, you have font that are not the professional font you can you know it is a kind of a font that you know more in a banner and things like people use. And you cannot even read them all that are identified by for example, the asterisk the red asterisk. You cannot read.

So, you on the left side you have has a large font size; on the right you have such a small font size both are belonging to the same figure group. So, it is not good. So, that is what corrected the one panel that is shown below identified by the green tick mark is that now all fonts are more or less similar size, and you are able to read them. They are legible;

therefore, they are better. So, this is again an example as to how you can make good figures and how you make labels.



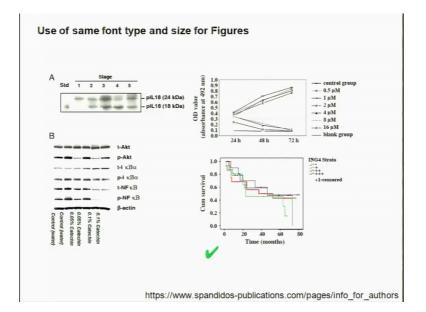
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I want to show you a few more examples. Use of the same font type and size for figures this is what it is. Now, you can see that these are group figure you know A, B. And you can see that the one that are identified by asterisk red asterisk you can see that 24 let us see that the one other top right it says 24 kda right, and there is an asterisk there. If you can guess why there is because that is not correct way because after 24 you have to give a space because you know kilo dalton 24, there it has to be a break and space should be there. So, these are minute things where people miss out.

And again you the figure has got figure a and figure b, figure A is with capital letter, figure b is identified by small letter b you can see that. Again it does not really make sense, it should be you know as for that general it should be either small letter or capital letter. And then again you can see that in each of the label for the different proteins that are identified t Akt or you know NF kappa B and so on here the Greek letter kappa alpha all these things or in a font that are very different from the rest of the font. So, it is not correct right. And for example, now you how would you corrected, this is the corrected figure. So, you can see now 24 kda you have a gap in between all these you know there labels that have been changed therefore it is better.

Now, let us look into the figure on the right side. Now, you can see that again, you see that in the value on the y-axis for the line diagram, you have .0, .1, .2, .3, .4 now that is not correct you have to have them as 0.1, 0.2 that is the best way to show them. And second, it says OD values in bracket absorbance at 492 nano meter. Now, you will find before the bracket, there is no space after 492 nano meter; there is no space right. You have to introduce this space and that has been corrected here now you can see that this is how it is given. Now, you go on the right side now control group now each line you know it is difficult to even decipher because the symbols are not very legible. And each line you are saying what it is you can see here 0.5 micro molar right. This micro you are reading it, because it is not micro; it is u, it is a common mistake people make.

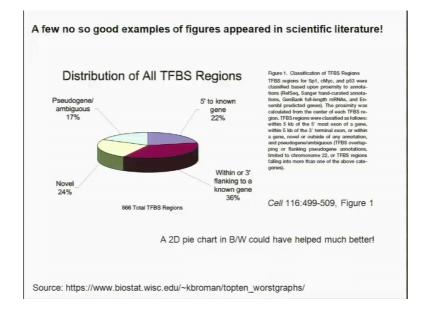
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So, it has to be the correct symbol now you can see that you can appreciate it is 0.5 and then there is a space and then micro molar right. So, you have to have you know you have to give a lot of attention when you are making figures as to that you make them in accurate way. Now, you are coming back to the figure that is given on the right side lower corner. And you can see the time which says month right. And then and then you have all the data points that are very very thin that you cannot read. Now, these are better.

Now, for example, there are lines you cannot read. Now, you have lines that are more legible and you have said that time it is months, because it is 20, 40, 60, 80 it goes in plural therefore, it should be set in months. So, it really talks about the survival of certain

organisms that is what this plot says. And it now you can understand the listen or even each one of the data points lines what they mean it looks much much better as compared to the previous one.



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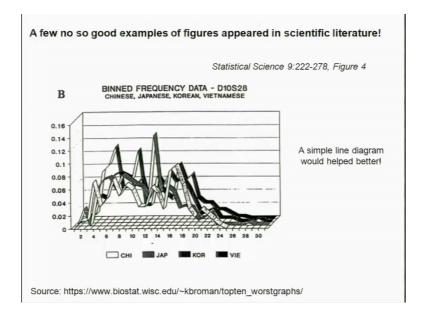
Now, you will look into some of those examples, these examples are not so good examples. In fact, if you go to the site that is given below, these are sites where in they have looked into some of the research publications, some of them even published in very reputed journal, but that they show it as a bad example right for how to make figures. But that else you to understand how you should not make figures right. So, this is figure that appeared in the paper in journal a cell consider would be one of the reputed journal which talks about distribution of you know some TFBS region right, some sites that is not worried about it. And some sequences that are present in a located in various parts of the genome that is not worry what it is.

But what is the problem with this figure? The figure is that when you have this data point there is no need for putting this pie chart in a two-dimensional or three-dimensional way right. So, the 3D really, really is not required here a 2D pie chart in black and white could have really help you to understand better. The reason is now in a 3D the for example, even for the group that is shown on the left side what is called novel 24 percent it has got two different shades of the color just to you know appreciate the 3D effect. You know, but it we mean you know there is a three different two different value there you

know for a reader who is not trying to it. And second you know the region that is present at the back. So, how do you bring 3D effect because even if there is a road, you are looking at in the 3D aspect then the road that is near to you that segment of the road is much wider as compared to the segment at the back, so that will be much narrower as compared to that. So, you give a 3D effect.

Here also the pie chart you have given a 3D effect such that the one that at the back for example, five a prime to known gene 22 percent. Now, this is you know twenty two percent is you shown in has little narrow to give you the depth right that could be misleading because for you to look at it, it may look like smaller. So, it is not necessary here. So, you could have easily you know presented the figure in two-dimensional way and simple you know pattern could have really helped it. So, that would help the reader to understand the difference, after all what you are trying to say is the relative proportion of the segments of the genome where this particular site is present. So, a 2D would have helped much or better.

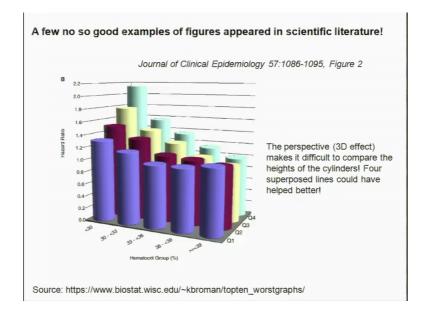
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The other example is now you can see that this is a kind of a line diagram instead of plotting a simpler line diagram. Again the authors have plotted a ribbon diagram, each ribbon now indicates the data point or trend of particular explain to a group or whatever here is a population. Now, what is difficulty here is that you cannot really because of the again the depth that they are trying to show in this three-dimensional plot, you cannot

compare one line with the other. Now, you know each of them look very, very similar. So, simple line diagram with each line having a different for example, a dotted line or continuous line or a different color would have made easier for the reader to understand the difference in the line you know between the data points, again this is one of the bad examples.

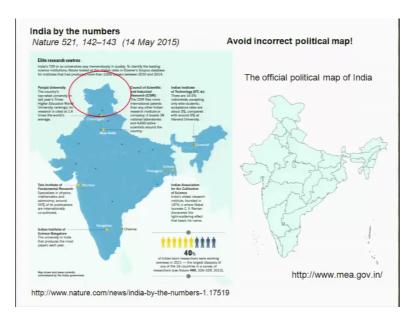
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Again this looks so beautiful, because these are all cylinders that are packed one against each other the beautiful shadow and gives you a three-dimensional view. But again you know if as a reader is extremely difficult to compare one cylinder with the other especially when you want to differentiate the height difference right. The perspective that is 3D effect makes it difficult to compare the height. For example, you consider in the 36, 39 where is second last from the right side if you see you know there are four cylinders in that particular bin. And what is difficult is that can you compare the height of Q 1 with Q 3 although the Q 3 looks taller, but because of the three-dimensional depth that you have given it looks taller, but probably it is the same height.

So, I cannot really appreciate that right. So, it could have been split into for example, three different you know figures stacked one over the other, and or a bar diagram one next to each other you know it could have made you know the comparison much, much simpler if you have used say two-dimensional you know bar diagram. So, again these are all some of the issues that you need to really look at it.

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I am going to talk about even you know at times we miss out. This is a figure that I am just going to what I am showing use from the nature issue which talks about research centers in the country which are the centers that are publishing you know very good papers and so on. And they talk about India by numbers as to the number of you know publications that are coming from various cities, in various institute that are located in the city and so on. It is a very good way of saying things as to for example, Punjab University top ranked university, and what is the publication and TF for Mumbai what is the publication, IIC and then so on.

So, now you have look up it, it looks so good, but you know it has a problem. I do not know how many of you are able to locate the problem. The problem is this. So, when you are publishing any political map, these are a political map. They are not geographical map basically it says the whatever the map that is shown by the nature of publication is a map, the blue filled regions was considered to be Indian territory right which is not accurate. Because it did not include a majority of the Jammu and Kashmir which the Indian government in as per our government as per our believe it is part of India, but is occupied by other countries. Therefore, when you publish any paper from India, if you are using a political map you know you cannot just borrow any political map that is present in the Google you know it just search it and use it that is not accurate. For example, what is shown on the right side is a political map as given by the Indian

government this is from the Ministry of External Affairs, if you go this is a political map which correctly displays the Indian Territory.

There are dispute area with the dispute for the other countries as far as Indian government is concerned, this is the Indian territory and that is what should be shown whenever he report any results that uses the map and you want to publish it in any journal. So, you have to use the correct official political map of India, so that you need to know that you should not do any such mistake with regard to any country that you do. So, you have to go for that particular country, and what is the correct you know political map, you need to make sure.

But you have a problem with that one can use always what is called a geographical map which really does not identify the territory based on the political feature, it is mostly based on the geographical. It is a neutral, so that you do not get into any trouble with regard to one party claiming one and other party claiming something different. So, you have to be careful when you use maps as to what information you wish to convey. And therefore, which map is accurate for that kind of a information that you wish to convey there are to be careful right.

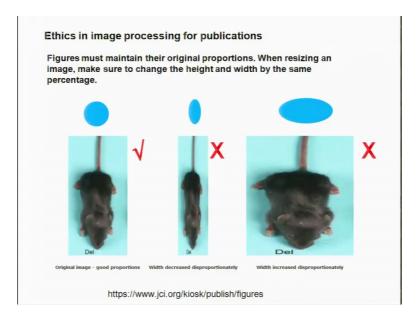
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So, when you make figures you know, there are a large number of important point that you need to consider right. So, what I am going to talk about now in a few slides is ethics in image processing for publication. So, because these are all often digital images these days right. So, the digital image is you have to process, you may have to crop the image to show only a small segment or you may have to you know adjust the contrast to highlight the differences and so on. So, when you do that such kind of image processing, there are certain guidelines which you need to follow right, so that is what shown here.

And there are a large number of sites especially the journal sites when you go they give you all the guidelines, one I am showing is for the nature publication. So, likewise you can go and do for such for every journal that you wish to publish. It is called the image integrity and standards right. So, you are going to look into some of those. So, ethics in image processing for publications, so the one of the important element is that figures must maintain their original proportion. When resizing an image make sure to change the height and width by the same percentage. For example, you have an image which may be a 3 size image left to the original dimension, but you cannot publish an a 3 sized image in a journal article or you cannot put that in your thesis. It has to be probably one-tenth of the size that you can publish or you can you want to put it in your thesis.

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So, how do you make it smaller? So, the relative proportion of the journals in the overall the image can be you know made into smaller by 90 percent, 80 percent when you do so the height and width also should be of the same proportion. What is shown here is the picture of a mouse and the mouse you can that is the size, but you can you know by

dragging it only the width if you reduce, the animal may look leaner; or if you stretch it, it may look fatter right. So, this is not the correct in our display right.

So, that is what is shown on the top there is a circle and if you make it you know you know thinner then it becomes owl shape, and if you stretch it again it becomes owl shape, but in other side. So, you have to when you make the size smaller, you have to make sure that it is you know the proportion is maintained therefore, it the relative features of the image is always you know maintain that way. It is very extremely important because that is how you know all these images you know they represent what you have seen right, but it could be much smaller than what you have seen. For the relative this you know points in that any section of that you may should not vary, it should be proportionally smaller or proportionally bigger.

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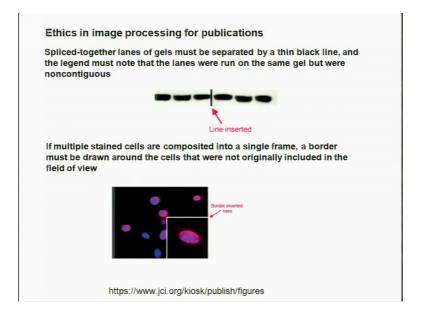


The other aspect we have to look at is that when you have image, when your process and you are allowed to do the you know enhancement of the image that is you can do overall you know you can bring more contrast have reduce the contrast that is allowed right. Because you know you have the same image, if you have taken with longer exposure it the image me look different. If you user shorter exposure, the image may look different for example, you can use your cell phone take an image in the noon and then in the night then they look very different right. Same object may be looked very different depending on how much light intensity that is there and the object that you are seen.

But you can do you know even an image that you obtained in the night, you can open it in any image processing software, you can make it you know much brighter right. But when you do that, you do it to the overall image right you are not changing any particular in a section of the image; you know you are not altering it. So, that is so overall if you apply any contrast enhancement it is ok, you know, but you cannot do that only for certain reasons.

So, what is shown here is that you know some image which is manipulated meaning you can see that on the right hand side that lower corner on the image identically image shown on the right side with a red circle that these dots are removed using some image processing software. What do you need remember is that any image that you manipulate it can be easily identified because these are all digital images? The way you make it the way you can also find what you have done. So, if you think that you are smart that you are able to you know make the change and send it, and there is a similarly smart way to look at whether any such manipulation is done or not, quickly people will find out. Therefore, you want to be careful you do not want to do such kind of a manipulation right.

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There are other ethics present images for publication. For example, if you have gel images right. So, you have two different gels and you have gotten the images. Now, you want to combine them together because they are represents a different time code or

whatever. Now, you need to you know identify that they represent two different you know gel image. So, what is shown here is that there is a line that is shown and line inserted there is an arrow and then there is an text written that is to show that the image on the left is different from the image on the right side. They are not done at the same time that is what identified by the line. So, spliced together lanes of gel must be separated by a thin black line, therefore, you know that they are not from the same you know gel and it should be said so in the legend as well, you cannot hide that information.

And second if you may have an image for example, I have taken microscopic image of you know various organisms that are there in that like one shown here in the spherical objects. Now, one of that spherical object I want to blow it, make it bigger and then show it there. So, if I have bring that, so I should demarcate that particular you know section of the image by a line. Therefore I know that this is not the same as part of the rest of the group it is different you know you have either your magnified or you brought something from elsewhere. And you were putting here again that kind of a you know line should be drawn to show that these two are not of the same magnification or the same field and so on. It is very very important when you make composite images you want to identify such image corrections right is important you need to show that.