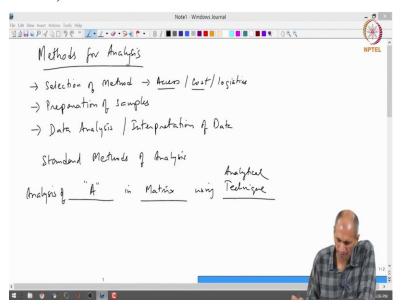
Environmental Quality: Monitoring and Analysis Prof. Ravi Krishna Department of Chemical Engineering Indian Institute of Technology-Madras

Lecture No. 19 Analysis Methods – Introduction and Water Quality Parameters

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Okay, so today we are talking about the analysis method just an overview, because each one of these methods will take a long time to completely understand. So, at the level of for environmental analysis one needs to know 2 things the objective of doing this is to a selection of method and then it also helped in your preparation of samples. In other words, you are you can tailor your sampling methods to the instrument that is being based on the selection of the method of analysis.

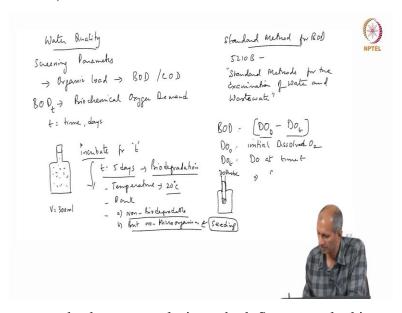
Because there are a large number of options available and sometimes the selection of method is governed by access to the instrument and also the cost and the logistics of doing it. Sometimes it may be very difficult to do some analysis, sometimes you do not have access to it was really expensive. Second, it may also help you in sampling methodology. So, preparation of if you know the method, what are the methods available? You need to you can also prepare then the third is data analysis what we call us interpretation of the data.

This requires you to know something about the instrument. So you do not need to know the how the instrument works and how it can be optimized, you need to know at least what it is giving you and whether it is reasonable it is what you are looking for, it is reasonable, it is completely pointing in some direction that mislead you. So, you have to know a little bit of the overview of the instrumentation for this purpose.

So this is the objective of doing it not to give you complete information about the working of an instrument. So it will take you an entire course to just look at 1 particular type of instrument to understand the intricate things. So our goal is not to do that. It is just to give some overview of what are available and why it is used simple so this is a mistake. So, in that context so, and we will also look at different analysis. So, last class as I mentioned.

So, we have standard methods and I will go over that again of analysis. So, the 3 things are important in the standard methods is the analysis of our measurement of the pollutant in matrix using an analytical instrument using a technique. So, this is what says you know these 3 things that constitutes 1 complete methodology that is used to doing it.

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So, we will go from we go back to our analysis method. So, we are looking at water quality, the way we are going to do this is first we will I will just do certain things that are specific to water

and restrict to air and then we will move on to something that is more generic. It applies to all matrices. And then looking at examples of some of these so what a quality so we are looking at the first thing we look in water quality is screening parameters.

Screening parameters for organic load, we are looking at BOD and COD mainly so, BOD is, we had discussed briefly earlier is biochemical oxygen demand. We had discussed some of this before let us go over this again biochemical oxygen demand usually there is a time below the series it can be anything, but typically time in whatever units typically these days, but it can be anything. So, what it the way the BOD works is there is a standard method for it the standard method is using one of these.

One of the methods standard methods for release is method number 5210 from compilation this is called as a standard methods for examination of water and wastewater. I think this is there are methods to large number of methods in this I will show you the website. So in this method according to this method, and is very generic method and there is a, what is known as a BOD bottle.

It is so designed in such a way that this is a typical volumes of this bottle is about 300 ml. This is all standard. So, you have to understand that this you can do this whichever way you want. But because it is a standard method, the objectives of many of the standard methods is for people to be able to use it without any prior knowledge of this particular thing. So, for example, I can do a BOD analysis in whichever way I want as long as I understand what I am doing and why I am doing.

But a lot of people who are just or just come into blindly they will analyze the sample. For them the standard methods gives you all instructions step by step instruction you follow blindly it should give you the correct answers, you do not have to apply your mind to it. So, there is something called as a BOD bottle, which has the volume of 300 are you completely feel it? No space. The reason it is called a BOD bottle is because the measurement of BOD is essentially the dissolved oxygen at initial time equals to 0- dissolved oxygen at some time.

This is essentially the biological oxygen demand. It measures 0 this initially DO is 0 is initial. So the BOD analysis the weight done is a 300 ml sample bottle you take a sample from somewhere taking water from a river or a lake or any anything you fill this up. So, you have a the analysis of BOD is dissolved oxygen at the 0- dissolved oxygen at some time for DO0 is the initial do or dissolved oxygen and DOt is the DO at time t what we do usually is the there is a DO probe that is placed in the into the water.

So this BOD bottle is so designed that the standard DO pro goes nicely in this without allowing any air to pass in or any oxygen to escape. And then you measure and then you take it out. So in this you can also see that there is a lot of chances of error associated with were as talking about oxygen. So, there is if the reaction is going on here, oxygen can also be consumed and oxygen can get infused. So the idea is, you close it, you open it could be probe inside measure, wait for the do DO reading to show up.

And once you are satisfied with the reading, you take it out and close the bottle again and keep it somewhere. This is the idea. So in this time, there is a last you have to check for that. What is your assume the water is not going out so that is it the other thing. So the so you have to estimate how much water is going on this as you are pulling in and out, some water will stick to the probe and come out. So, out of 300 ml if people are determining what.

So usually what they do is they will fill it up, they will almost overflow it then they will take out slowly or sometimes you can just leave it as it is, but it is not convenient. So DO probe is expensive. You cannot have DO one DO for every DO probe that would be nice to have it sitting in the probe bottle. But unfortunately, we do not have that many do you have to take it out and use it for 100 other samples.

So some of these groups can cost up to I think they can get a cheap probe of 50; 60,000 rupees but it can also go up to maybe a lack or more. So depending on the characteristics of the group. So those are quality control questions, I think so if you think they are important to you have to worry about corrections for that otherwise does not matter. So the volume is not really important

that much here the only way volume is important here is if it creates a headspace then it is a problem because oxygen will now go and equilibrium there.

And available oxygen for us in the BOD is not enough. So then once you do the initial thing you take the BOD bottle and incubated for t or time t is, typically t is 5 days in the standard method t is 5 days. Now, this is not set in stone, this is also not a fixed thing, because it can vary from vase to vase can vary from a comprehension of the vase. So, what happens in t is the microorganisms that are present in the water.

Consume oxygen to degrade whatever is the organic compounds call us for the degradation of organic compounds that are present in the in the water. So you have to what are the possible errors in this? We are talking about quality control, what are the possible so after 5 days you take out and measure the probe do again. So, you are going to get a decrease in the DO from the initial to the final, which means that corresponding to the amount of organic load that is present there is certain amount of degradation that has happened oxygen has been utilized for the integration of organic compounds.

And therefore this is difference is expressed as less BOD is in demand. What else could be there and this so it is not I am not giving you full information this is not standard method means they will tell you everything. Now, whether you should move your hand this much keep it there, keep it here all that everything is invalid. So, a lot of missing information here like I give you a BOD bottle and water sample and DP probe, what will you do?

Will add this take then what do you do where do you incubated the word incubate means it is kept at what are the different things we are doing biodegradation here, we are doing biological degradation. And you have to think about what are the things that affect biodegradation and also things that affect oxygen consumption, the oxygen in the water, we talked about biodegradation one thing that biodegradation is influenced by is temperature.

So, you have to keep it at some temperature. So, this BOD can different biodegradation can happen rates that can happen at different temperatures short 5 days. I can consume 1 level of

oxygen and it can change you fight my temperature changes. So, you have to standardize this you have to tell everybody that all you all people do incubation at some temperature and not do it whichever temperature you want.

So, if I keep it outside temperature in Chennai is 35, 33 degrees 10 temperature in somewhere else, somewhere some other part of the world is different temperature in Delhi is in 20 degrees, 30 degrees difference between Chennai and Delhi all those things are there so, you cannot have that, this is what we call a standardization when you say standardization, it means everybody follows the same method which means you have to do it at some temperature.

And typically the temperature is 25 or 30 degrees, I think the standard method asks for 25 or 30, 20 is 20 degrees centigrade. And one may argue, is 20 degrees and never happens in India often is India temperature and South India is always 20 plus night or day it does not really affect us. Yes, it does not the point the BOD is a surrogate measurement, it is not a real measurement. It is already a surrogate measurement. Because it is measuring oxygen, it is not measuring organic compound.

It is also a reference point at 20 degrees centigrade and for 5 days. So you have to understand it in that context that you have to map this to an actual concentration of some chemical. So the value of BOD is to give you a parameter that will indicate the quality of water that is all so there are standards based on this, to tell you that if the BOD level is above some value, the water is considered to be unhealthy or considered to be polluted.

So, these are all the same references important why everybody follows a standard method. Irrespective of you know, if you do not have the equipment to follow a standard method, you should not call it a standard method, you should say that I measured BOD at 35 degrees centigrade using, you know, some other instrument or you can develop your own method for it is not standard method of BOD.

That is all everything will change a lot of things will change. And the second problem is that you can also get oxygen production using by photosynthesis is something is there in the water

including microorganisms including some biological matter which will contribute to oxygen production during photosynthesis. You do not want that you want oxygen neutral systems in reference to that so, that will cause problems.

So, we are only looking at oxygen demand by for degradation. This entire thing is done in the dark. You put it away from sunlight, dark. So there is another problem here this is a general 2 conditions for very highly polluted water can use one day and all that. So that is that is also possible. But everything else remains the same. But typically when somebody says BOD is BOD 5 days of incubation.

The third another factor that influences the analysis, the results for BOD is what if you do not see any change or you see very small change, does that mean what could it mean to possibilities? If you see a very small change in the oxygen content of the between the 0 and DOt, what does it what does it mean? What could it mean? And there are no microorganisms whether or not right but oxygen is not changing from initial, not going on anywhere, it is not been consumed.

There are no microorganisms that is 1 possibility that there is no microorganisms in the wastewater to do the microbial degradation that is 1 possibility. So what is the second possibility? No, that is it is there is other possibility, no biomedical argument so that covers it. So they could be organic matter, but it is non-biodegradable. So it is quite possible that it is non-biodegradable. It is biodegradable but does not contain no microorganisms.

Here it is non-biodegradable, you cannot do anything about it. You cannot use this method no matter what. But to if there is no microorganisms can be addressed by adding microorganisms from outside so this is called a seeding. We will see the system by adding mechanisms, which are against standard, but this is more, trickier because standard means somebody has to sell you those microorganisms.

To tell this, is even more, trickier because there is a specific species of microorganism and microorganism profiles in different places across the world are very different. It may be very different here compared to what is then 10 kilometers away from you. There is a wide variation

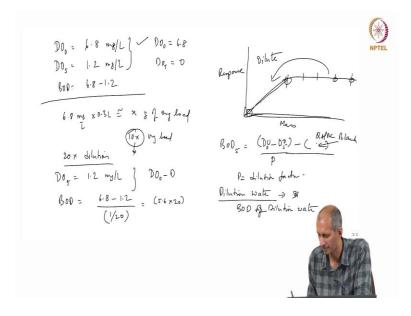
depending on what is the substrate? Available here and all that. So that is a very complex problem to people do not get into that. So instead, they will add whatever is available.

So this is again, not standardized, but I think I would be very careful when you are doing the seeding you are doing with all references must be same in the same seeding. This is a bit of a tricky thing that will not get into that. So that analysis of that requires you to analyze the population all that is generally there is some recommendation of a seed, but people may not have access to that sometimes they will just add whatever is available.

So, what we call a general microbial populations. So, the idea of a serious that natural water have some microbial populations that are there already so, it will degrade. So, the idea behind it is that it will degrade by itself things will biodegrade naturally, which means that you have to rely on whatever is there naturally, it could not it may not be 1 particular bacterial strain or microbial strain it could be multiple family of them depending on what is there in the water they will eat something will survive and will grow metabolic metabolize whatever is there.

So, you see, so, let us say that so, ideally what you would hope is that, at the end of 5 days, all your biodegradation is done, finished. And so, you start so, you take an example like the example in the next page.

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Let us say that the DO initial is the 9.8 milligrams per liter not 9.8, 6.8 and DO 5 is 1.2 is very straight forward. BOD is the difference between these 2 is 6.8 - 1.2 over. Suppose I get instead of this thing. DO0 is 6.8, DO5 is 0. What does this mean? What could this mean? 0 is a problem, because in our case of when we are doing calibration if you recall calibration bonds is response mass or something, if your calibration does this 0 is equivalent to this part.

I have no idea where this is what does 0 mean? 0 means all oxygen is consumed, which means 6.8 grams of oxygen can be consumed by some x grams of organic load, what if there is nx organic load 6.8 milligrams per liter worth for 600 milligrams either multiplied by 0.3 liters is the amount of oxygen you have in there this corresponds to x grams of organic load. What if there are 10 x organic load? You have no idea it will see the 0 you once this x is gone.

The remaining 9 x is still sitting there. Your analysis is incomplete. So what do you do? BOD 5, is there an easier method? Whatever happens in the first 10 minutes? Now, whenever we do is what do we do if you have a signal response like this, if you are in this range, what do we have to do? We have to bring it into the range where it is measurable. So, what do we do in this case? We dilute, so here how much will you dilute your 10 x you are diluting at least 10 times you bring it below 6.8 below the range where it is going happen.

So I will dilute it by a lot large amount. So I can do multiple dilution this is what people do, they have no idea what the concentration is their dilute 10 times, 20 times, 30 times, 100 times somewhere his value will be above 0. Then they take that value as the reference. So, if you dilute this in fact, I will say 20 times and my DO5 is 1.2 milligrams per liter. My actual DO we know we DO0 – DO my BOD will be 6.8 - 1.2.

But this 638 - 1.2 corresponds to 1/20th of the sample. So I have to divide by 1/20th dilution this is the dilution factor that goes in there. So this actual BOD of the sample is 6.8 - 1.2 is what 5.6 multiplied by 20 typically, this is what is done. You do not have very few samples which are BOD of 15 and 10 and all that. It is always more so, dilution is inevitably done. So, the standard method equation or straightaway the standard whether the equation is DO0- DO5 divided by what is called as P is known as the dilution factor in this case is 1/20.

So, you have to good question. So, you use, it is called what do you use for dilution is a dilution water whatever call it now this could be whatever is the purest water that you have now, what is the quality control here? So, there is a matrix effect now, if I am diluting 20 times or 100 times which means that in the 300 ml sample the bottle 3 ml is actual sample rest of it is my dilution water. So, the dilution water is a matrix is the water.

So, now you have to check if what is the BOD of dilution water? This is a matrix blank, you have to do this matrix blank because the water you are adding for dilution can have a BOD of its own. So, you have to calculate that separately you know to calculate that separately. So this must be subtracted, something must be subtracted from this DO is a change in DO of the blank of the dilution water. How much value is that so that you have to divide that and when do the dilution there is another possibility that the seed that you are adding also has its own its own BOD.

You are saying it is microorganism, but it may also accumulate BOD, it may also have some consumption going on. So, along with this blank, if you are writing a seed, you put that seed also report some amount of seed and then calculate the consumption of oxygen for the seed. So, these are the this is a quality control, this is a QA and QC blank the blank term goes here so you do not consider that as part of the sample.

It is part of your method, your sample diversion water or the seed or anything else. Oxygen leaks all this are the way we do with blanks what we do with blanks some false positive and all that you have to do here. Also, if you want to look at leak of oxy and leaking of oxygen across the way we do is we have a standard what is the standard here? What could be a standard there is no standard here what is the standard here oxygen is a standard we are measuring oxygen.

So, you have to calibrate the instrument using oxygen your dissolved oxygen probe is measuring oxygen from 1 point to 1 point the DO probe usually, so we operate at which means you have to make standards at multiple concentrations of oxygen very difficult to do. So what people do is they have maximum what is the maximum concentration of oxygen and water that is highest you can get is this solubility saturation.

So they will bubble oxygen or air through water for a long time, 1 day, 2 days they will measure keep measuring what is a solid solubility of oxygen and what dissolved oxygen solubility roughly 8 surround period 9, 10 around that milligram do not say a ppm is a very I do not I would suggest you use absolute units always because there is a lot of confusion ppm and all that. You know, if you forget you make a mistake. It will cost you 1000 factors. The factor of 1000 milligrams per liter is around 10, 9, 10, 25 degrees certificates is around 10, 8 between 9 and 10 depending on the water. So it is not very high.

So you can make it so whatever you get the highest value you if you know what the value is you keep it at one the other end you come completely remove oxygen from water, which is also not easy. Because you are now exposed to oxygen whenever you expose our water, oxygen, oxygen always wants to go in. So you have to burn the oxygen out of it, there are ways to do it. One way is to add an oxidizing an agent that will react with oxygen, all oxygen can be taken away.

There are methods to do it will not go into it but you have to prepare this and it is not trivial to prepare this. One way is to just purge with nitrogen will remove all oxygen and there is 1 way of doing it. It is a lengthy process it takes time this must transfer of nitrogen using oxygen and then that is 1 end so you have a 2 point calibration is 2.7 very dangerous, but that is the best you can do as of now because any anything else in between is not stable oxygen is by the time.

You prepare everything system open it put the do probe guns, it is not the same different so it is a bit tricky. Any questions is this overview of this? Now, so, sometimes a seed may also have, you are taking seed from somewhere that could have some organic growth that will also consume microorganisms the venue of collecting it practically may have something else in it unless you are using your culture and all that.

So, this is it may or may not give you any value. So, it is the same quality control is done just to ensure any such artifacts. Sometimes it may be important especially because of the dilution factor. So, the reason you are if you are using dilution factor of 100 and all that any small error

there will magnify by 100 times here so that is the reason why worried about small things like otherwise it is trying to be not be a big issue.