

Video « Calibration – Drawing the calibration line »

Time	Text
00 :09	In this video, I am going to present you how to draw a calibration curve. In the sheet related to calibration, you have access to a video about the preparation of a standard range, this video about the analysis of the standards and the drawing of the calibration curve, and the last one about the quantification. So, all standards being already analysed, this video will focus here on the drawing of the calibration curve.
00 :32	I will start by the external calibration, which is the simplest, and I will continue with the internal calibration and finish with the standard additions method.
00 :41	Concerning the external calibration, I am reminding you that a stock solution has been used to make standard solutions at different concentrations. Then, each standard is measured. Therefore, for each concentration we get a measurement.
01 :00	At least five standards are necessary to draw a calibration curve. Once the measures are done for the different concentrations, there are two ways to proceed: <ul style="list-style-type: none"> - The simplest, the most archaic, but which is still used in laboratory, is to get a pencil and a graph paper. The concentrations or the quantities of each analysed compounds are placed on the x-axis and the measured values got for each standard are on the y-axis. Each axis is drawn with the correct scale. Therefore, these points are the measures of each standard. Then, a handmade line is drawn (it is better to draw it with a ruler) to connect as well as possible these points. In another video, I will present you how to quantify the products from this calibration line.
02 :07	<ul style="list-style-type: none"> - The second way to draw this calibration line is to use Excel (with a computer). If I take the same example of the titration of the ammonium in aqueous solutions, I have to make two columns: the first column represents the standard solutions with their concentration and the second column represents the measures, which are, in this example, the absorbance of the titration of ammonium, measured by spectrophotometry. With this table, it is possible to make a graph. Mind to choose the "cloud of dots" mode, otherwise you will get a graph not convenient to use.
02 :58	Do not forget to, first, add an explicit title. On this example, the title is not indicative enough. It is necessary to write that this is a calibration line for the titration of the ammonium in aqueous media.
03 :13	Moreover, mind to add the legends and the units for each axis. It is very important because it allows a different person to understand what you did titrate and what are the units.
03 :30	Once the line is drawn, it is possible to add a tendency curve by right clicking on a point of the line and by selecting « add a tendency curve ». Choose "linear curve" and do not forget to add the line equation as well as the determination coefficient. It allows to add an information on your graph which is useful for the quantification of a given sample, as I will present you in the next video.
04 :00	The principle is the same concerning the internal calibration, which is a calibration mode that could be interesting. Again, there is a standard range but this time, in each standard with increasing concentration, a known concentration of internal standard is added in each standard. Obviously, it is added before the adjustment of the volume of each flask. It is important to avoid any mistake on the concentrations.
04 :28	The analysis of these standards is the same as earlier but, in that case, 2 information are provided for each standard: <ul style="list-style-type: none"> - An information concerning the concentration of the compound of interest - And an information for the internal standard which has been also measured
04 :40	Then, as previously, it is possible to work with Excel or with graph paper to draw the calibration line, using the same method as earlier. But in that case, it is the ratio of the

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	measured concentration and the concentration of the standard (which is constant) that is represented on the x-axis. And it is the ratio of the measure of the compound of interest and the measure of the internal standard that is on the y-axis.
05 :12	Concerning the standard additions method, it is a little bit different. As a reminder, the analysis starts with the first measure of a sample. Then, the compound of interest is adding to this sample, and a second measure is done. And again, and again ...
05 :34	So, a known quantity of the compounds of interest is adding to the same sample again and again, which allows to get several points to draw the calibration line.
05 :40	As previously, the line is drawn by using Excel or graph paper. But this time, the origin won't be on the line. Indeed, the point for x equals 0 corresponds to the measure of the sample, it means without any adding. Then, in the same way as earlier, each adding corresponds to a different concentration, and a point is adding to the graph, which corresponds to the measure performed after the adding.