- What is the purpose of calibration curve?

  Answer: Most analytical instruments generate an electrical output signal such as a current or a voltage. A calibration curve establishes the relationship between the signal generated by an instrument and the concentration of the substance being measured. Different chemical compounds and elements give different signals. When an unknown sample is measured, the signal from the unknown can be converted into concentration using the calibration curve.

- How to make a calibration curve?

  Answer: Prepare a series of "standard solutions" of the substance and measure the signal (e.g. absorbance, if you are doing absorption spectrophotometry); then plot the concentration on the x-axis and the measured signal for each standard on the y-axis. Draw a straight line as close as possible to the points on the calibration curve (or a smooth curve if a straight line won't fit), so that as many points as possible are right on or close to the curve.

- How do you use a calibration curve to predict the concentration of an unknown sample?

  Answer: This can be done in two ways, graphically and mathematically. Graphically, draw a horizontal line from the signal of the unknown on the y axis over to the calibration curve and then straight down to the concentration (x) axis to find the concentration of the unknown. Mathematically, fit an equation to the calibration data, and solve the equation for concentration as a function of signal. Then, for each unknown, just plug its signal into this equation and calculate the concentration. For example, for a set of calibration data, the best fit line equation is Signal = Slope × Concentration + Intercept, where slope and intercept are determined by a linear (first order) least squares curve fit to the calibration data. Solving this equation for Concentration yields Concentration = (Signal - Intercept) / slope, where Signal is the signal reading (e.g. absorbance) of the unknown solution.

- The molar absorption coefficient, molar extinction coefficient, or molar absorptivity, is a measurement of how strongly a chemical species absorbs light at a given wavelength. It is an intrinsic property of the species.