

Video « The Kjeldahl method – Theory »

Time	Text
00 :09	Welcome. In this video, I am going to present you the titration of the total nitrogen thanks to the Kjeldahl method. This method allows to determine the quantity of proteins in a bioproduct.
00 :19	When I say bioproduct, it means that you can use this method either on food (solid or liquid), either on ingredients (liquid extract, pasty extract, powder) or directly on animal or vegetal raw materials as cereals.
00 :35	Just one thing if you work with solids, you have to mind to grind your samples first. So, what is the principle of this method?
00 :50	The organic compounds with nitrogen, which are proteins and, in some matrices, nucleic acids, will be broken down at high temperature with sulfuric acid and a catalyst. This catalyst is made with potassium sulphate, which allows to rise the boiling point of sulfuric acid, but also with copper sulphate which acts directly as a catalyst of the reaction.
01 :13	Therefore, the organic nitrogen will quantitatively form ammonium sulphate, as you can see in this reaction called "mineralisation".
01 :21	During the second step, the ammonia will be taken from its salt by sodium hydroxide and then distilled by steam distillation and stored into a known quantity of hydrochloric acid, used in excess. This step is called "distillation".
01 :41	Finally, the quantity of hydrochloric acid in excess, which has not been used to trap ammonia, will be measured by a titration with sodium hydroxide.
01 :49	How can we get the content of proteins in our sample? In order to get that, we need to analyse two different samples: <ul style="list-style-type: none"> - A blank sample without any bioproduct - One sample in which the bioproduct is added.
02 :06	What will happen? In the sample with bioproduct, the organic nitrogen will be transformed into ammonium and then ammonia. This ammonia will be trapped by HCl. So, the titration will get the quantity of excess HCl in the sample.
02 :24	In the blank sample, there is no organic nitrogen. Therefore, none HCl reacts during the distillation. So, by making the subtraction between blank and bioproduct, we can calculate the difference between total HCl and excess HCl, which will give the quantity of HCl which has reacted, it means the quantity of HCl which has been used to trap the ammonia during the distillation.
02 :47	How can you get the quantity of nitrogen and proteins from that? According to these reactions, one mole of ammonia is formed from one mole of ammonium, which is formed from one mole of organic nitrogen. So, it is possible to know the quantity of organic nitrogen and, with the molar mass of nitrogen, to calculate the mass of nitrogen in a given mass of bioproduct.
03 :11	How can you get the mass of proteins from the mass of nitrogen? You have to consider that the average of nitrogen in a protein is 16%. So, you have to multiply your mass of nitrogen in your mass of bioproduct by 100, and then to divide this result by 16, which is the same as multiply your nitrogen mass by the Kjeldahl factor equal to 6.25.
03 :31	If you know precisely in your bioproduct the proportion of nitrogen in proteins, it is possible to adjust this factor in order to adapt as well as possible to the matrix you are working on.
03 :41	Do you need specific scientific material for this method? Yes.
03 :47	As you will see in the video concerning the demonstration of this method, you will need a mineralizing device and a specific distiller for this method.
03 :55	Last but not least, this method requires to manipulate very concentrated sulfuric acid, so you have to wear the individual protections (lab coat, safety glasses, gloves), but also be aware of the collective protection (work under a hood) to do this method. Let's go for the demonstration.