The raw material for insulin production is the pancreas of cattle and pigs. Due to the presence of the enzyme trypsin in the pancreas, the production of insulin for a long time remained ineffective. IN1921 year. Canadian researchers FG Beiting and Ch. X. Best first isolated insulin from the pancreas when it was treated with acidified strong ethanol. In this case, enzymes are inactivated and lose the ability to destroy insulin. Currently, there are several technologies for the release of insulin, which differ from each other only in details. Fresh or frozen tissues of the gland are ground in a grinder-top and extracted by bismaceration in an enamelled reactor with a stirrer. As an extractant for the first maceration, 80-85% ethanol is used, for the second - 57% ethanol, acidified with orthophosphoric acid (sulfuric, hydrochloric) to a pH value of 2.8-3.0. The extraction is carried out for 1.5-4 hours with stirring. The extracts are separated by filtration or centrifugation, combined and purified.

Preliminary, coarse purification of the extract is the same for all technologies, it comes down to the removal of acid anions. So, the anion of orthophosphoric acid (PO4 "3) is removed with a solution of calcium chloride at a pH of 3.3-3.8. Then, deproteinization is carried out - the sequential separation of ballast protein substances at a pH of 4.5-5.1 and 3.5 at a temperature of O ° C. The precipitates are separated by centrifugation. The transparent extract is concentrated by evaporation in a film-type vacuum distillation unit at a temperature not exceeding 30 ° C to an ethanol content of 10-25% in the shortest possible time, subject to strict adherence to the temperature regime, since prolonged heating leads to insulin inactivation. The concentrated residue is purified from lipids and ballast proteins by settling in the cold (0-4 ° C) at a pH of 3.0-3.3. The extract is separated from the floating layer of lipids and the sediment of ballast proteins by filtration. Raw insulin is isolated from the condensed, purified extract by double salting out with a 25% sodium chloride solution or 40% ammonium sulfate solution. This method is used in some factories with1928 H... Until now.

The most progressive method is the isolation of insulin by ion exchange chromatography, developed and implemented in1970 year... at the Minsk plant of medicines. According to this method, the evaporation operation is excluded, and insulin is sorbed from the extract, partially freed from ballast proteins, on the domestic macroporous sulfo-cation exchanger KU-ZZ-30 / IOO at a pH of 3.0-3.3 in fluidization mode. To remove fat, the cation exchanger is washed with 65-67% ethanol, and to remove ballast proteins -0.3 Msolution of acetate buffer (pH 5.3). Desorption of insulin is carried out 0.01-0.05 Mammonium buffer solution (pH 9.8-10.0). Insulin is unstable in an alkaline environment, therefore, it is desorbed quickly, the eluate is immediately acidified with hydrochloric acid to a pH of 4.1-4.5 and acetone is added. The ballast sediment is removed. Insulin is precipitated in the form of zinc-insulin with a solution of zinc acetate (pH 6.1-6.2), purified by crystallization. Zinc insulin is dissolved in water acidified with citric acid to a pH value of 2.6-2.8. The solution is left to stand for 1 hour and the precipitated ballast proteins are removed by filtration through diatomaceous earth. The filtrate is mixed with acetone, zinc chloride and phenol are added, cooled to 0 ° C, creating conditions for slow crystallization of insulin. The solution is made alkaline to a pH value of 8.0-8.5; leave for 2-3 minutes, then create a pH value of 6.7-6.8 and stir for 1 hour; at pH 6.5 the mixture is stirred for 2 hours; at pH 6.2 and 6.0, stir for 2 hours and stand for 18-20 hours and at pH 5.8 stir for 2 hours and stand for 48-96 hours at 5 ° C. The precipitated insulin crystals are separated by centrifugation, washed on a Buchner funnel with distilled ice water, acetone and ether. Dry insulin in air, in a fume hood and desiccator.

Insulin activity is determined biologically: by the ability to lower blood sugar in healthy rabbits. The activity unit is taken as the activity of 0.04082 mg of crystalline insulin (standard), it should be 24-26 U per 1 mg.

Crystalline insulin preparations. Insulin for injection (Insulinum pro injectionibus) is obtained by dissolving crystalline insulin with an activity of 24-26 U / mg in water acidified with hydrochloric acid to a pH of 3.0-3.5. A solubilizer (1.6-1.8% glycerin) and a preservative (0.25-0.3% phenol) are added to the solution. The solution is sterilized by filtration through sterilizing filters.

Suiisulin (Suinsulinum) - a solution of crystalline insulin obtained from the pancreas of pigs, in acetate buffer. The solution has a pH value of 7.0-7.5, contains a preservative - nipagin.

Insulin for injection and suinsulin are used in the treatment of diabetes, injected under the skin or intramuscularly. In cases of urgent need for a patient, for example, with a diabetic coma, it is administered intravenously. When administered subcutaneously or intramuscularly, the action of the drugs develops after 15-20 minutes and lasts about 6 hours. Swinsulin rarely causes allergic reactions, is used for complications after injections of insulin obtained from the pancreas of cattle, and for insulin resistance.

The disadvantage of insulin solutions is the short duration of action, therefore, a number of prolonged-release drugs are produced.

Suspension and isulin-protamine for injection {Suspensio insulin-protamini pro injection ibus) - the first long-acting insulin preparation, obtained by mixing a solution of crystalline insulin with protamine sulfate (protein from sturgeon milk). Upon contact with tissues, insulin is gradually separated from protamine and lowers blood sugar. The effect occurs after 2-4 hours, reaches a maximum after 8-12 hours and lasts 16-18 hours.

IIrotamine-zinc-insulin for injection (Protamin-zinc-insulinum pro injectionibus) is obtained by adding a small amount of zinc chloride to insulin-protamine, which further prolongs the action of the latter. The effect of the drug occurs 3-6 hours after injection, reaches a maximum after 14-20 hours and lasts for 24-36 hours. The drug is a sterile suspension of white spp, in phosphate buffer, pI value 6.9-7.3, preservative - phenol (0.25-0.3%).

A certain success in nisulnoterosh was caused by the appearance of three prolonged-acting insuli - suspensions of zinc-insulin-a. They are obtained by mixing sterile solutions of crystalline insulin with zinc chloride and a buffer solution. By changing the order of mixing and the duration of stirring, it is possible to precipitate two physical fractions of zinc-insulin: amorphous and crystalline Suspension of amorphous zinc-insulin (the value of the fraction; does not exceed 2 μm) is obtained with short-term stirring of solutions, a suspension of crystalline, with slow stirring (30-40 / min) for 18-20 hours. In this case, crystals of a diamond-shaped shape with a size of 10-40 microns are formed. Zinc insulin suspensions are stable at neutral pH. The composition of preparations for 100 IU of insulin, mg: sodium aietata - 3.4; sodium chloride - 17.5; zinc chloride - 0.20; pH value 7.1-7.5; phenol - 0, 25-0.3%. Suspensions of zinc-insulin administered subcutaneously are gradually absorbed and cause a decrease in blood sugar, the duration of which depends on the physical state of zinc-insulin.

Suspension of zinc-insulin amorphous for injection (Suspensio Zinc-insulini amorphi pro injectionibus) shows a quick action - after 1 - 1.5 hours, but it is relatively short - lasting 10-12 hours.

Suspension of crystalline zinc-insulin for injection (Suspensio Zinc-insulini crystallisatt pro injectionibus is a drug of the longest action, a decrease in blood sugar occurs after 6-8 hours, reaches a maximum after 16-20 hours and lasts 30-36 hours.

Suspension of zinc-insulin for injection (Suspensio Zinc-insulini pro injectionibus) is a drug with an average duration of action. It is obtained by mixing amorphous and crystalline zinc-insulin suspensions in a ratio of 3: 7. A decrease in blood sugar when injected under the skin occurs after 2-4 hours, moderately increases by 5-7 hours, reaches a maximum after 8-10 hours and lasts 20-24 hours. This drug allows many patients to get by with one injection per day.

Insulin preparations are produced in vials with a capacity of 5 or 10 ml, sealed with rubber stoppers, followed by metal rolling. Insulin activity 40 and 80 units in 1 ml. Store according to list B, protected from light, at a temperature of 1 to 10 ° C. Freezing is not allowed.

Insulin M and its preparations. IN 1983 year... VNIITK.GP developed the technology of insulin M, which does not contain proinsulin and ballast proteins, which determine the ability of drugs to increase the body's immune responses. Additional purification of crystalline insulin was carried out by ion-exchange chromatography on a macroporous anion exchanger of domestic production AV-171-40 / 100 and repeated recrystallization.

Crystalline insulin is dissolved in 40% ethanol with the addition of ethylenediamine tetraacetic acid disodium salt, which binds zinc ions and thus increases its solubility. The concentrated insulin solution is centrifuged and sorbed on the AB-171-40 / 100 anion exchanger. Insulin is eluted with an ammonium buffer solution containing 40% ethanol, sodium chloride, pH 7.4-7.6. Collect fractions of the eluate containing chromatographically pure insulin. Insulin is crystallized from citrate buffer containing zinc chloride and 20% ethanol at pH 6.9 and 5.8. Highly purified insulin is obtained with an activity of at least 26 U / mg.

On the basis of suinsulin M, obtained from the pancreas of pigs, highly purified preparations have been created, similar to preparations of crystalline insulin, but characterized by low antigenic activity. A solution of suinsulinum M (Suinsulinum M) in an acetate buffer, in contrast to a solution of crystalline insulin, has not an acidic, but a neutral reaction, a pH value of 7.0-7.5. Extended-release drugs - suspensions: zinc-insulin M amorphous for injection or insulin-semilong (Insulin semilong), zinc-crystalline insulin for injection or insulin-ultralong (Insulin ultralong) and zinc-insulin M for injection or insulin long (Insulin long)