**Lesson 7**

**Analysis of drugs containing phenothiazine derivatives: dosage forms of aminazine and nozepam.**

**PHENOTHIAZINE DERIVATIVES**

The basis of the chemical structure of medicinal substances included in this group is the phenothiazine heterocyclic system with nitrogen and sulfur heteroatoms. Phenothiazine consists of a condensed heterocyclic system consisting of a thiazine heterocycle and two benzene nuclei:



Phenathiazine derivatives belong to the group of drugs of great importance and perspective in modern pharmacy and pharmacology. Starting from the 30s of the 20th century, the first substance with an antipsychotic effect, chlorpromazine, was discovered during the search for antihistamine substances that are phenothiazine derivatives. In the course of research, it was determined that substances with alkyl derivatives of phenothiazine have neuroleptic and antipsychotic effects, and acyl derivatives have antiarrhythmic effects. The general formula of drugs containing phenothiazine derivatives is as follows:

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Phenothiazine derivatives are mainly divided into 2 groups according to their chemical structure and pharmacological action: 10 alkyl derivatives and 10 acyl derivatives:

S

N

C

H

2

R

1

R

2

5

6

7

8

1

2

3

4

9

1

0

S

N

C

R

2

R

1

O

10-alkyl derivative 10-acyl derivative

In medical practice for more than 50 years, more than 40 preparations with neuroleptics or neuroleptic derivatives of phenothiazine have been used for the treatment of diseases accompanied by schizophrenia and psychoses. The pharmacological effect of phenothiazine derivatives is associated with dopamine receptor blockade.

Depending on the nature of the alkyl substituent (R1), located in the 10th position in the phenothiazine molecule, neuroleptics are divided into the following groups:

1) dialkylaminoalkyl derivatives: chlorpromazine (aminazin), promazine (propazine), promethazine (diprazine).

2) alkylpiperazine derivatives: trifluoperazine (triftazine), fluphenazine (fluorophenazine), perphenazine (etaperazine).

3) derivatives of alkylpiperidin: periciazin (neuleptil), thioridazine (sonapaks).

10-acyl derivatives: etacizin, moracizin (ethmosin), fluacizin (fluorazicin).

Phenothiazine derivatives used in medicine differ from each other by natural substituents in the 10th and 2nd positions. The introduction of halogen in the 2nd position enhances the sedative effect of the derivative characteristic of the CNS.

Antiarrhythmic derivatives of phenothiazine (etimosin, etazisin, nonaxalazine) are acyl derivatives.

Medicinal substances containing phenothiazine derivatives have psychotropic and antiarrhythmic effects, as well as antihistaminic, cholinolytic, etc. also has impact properties. The pharmacological effects of these substances mainly depend on the structure of the substituent located in the 10th position in the molecule. Thus, the substituted aliphatic chain of neuroleptics (aminazin, propazine, tryftazine, etc.) has 3 carbon atoms, and diprazine with antihistaminic action has 2 carbon atoms. In the molecule of antiarrhythmic drugs (ethmosin, etazisin, nonaxalazine), the urea group (in the urethane chain) is located in the 10th position (table).

Relationship between structure and activity in derivatives of phenothiazine

Table

|  |  |  |  |
| --- | --- | --- | --- |
| № | Dərmanmaddəsi | Əvəzedicilər | Əsas farmakoloji təsiri |
| R1 | R2 |
| **1.** | Promazin(Propazin) |  | $$-$$ | neyroleptik |
| **2.** | Prometazin(Diprazin) |  | $$-$$ | antihistamin (allergiya əleyhinə)  |
| **3.** | Xlorpromazin(Aminazin) |  | $-$Cl | neyroleptik |
| **4.** | Levopromazin |  | $-$OCH3 | neyroleptik  |
| **5.** | Triflüoperazin (Triftazin) |  | $-$CF3 | neyroleptik  |
| **6.** | Morasizin(Etmozin) |  |  | aritmiya əleyhinə |
| **7.** | Etasizin |  |  | aritmiya əleyhinə |
| **8.** | Nonaxlazin  |  | $$-$$ | aritmiya əleyhinə |

The acquisition of phenothiazine derivatives consists of 3 stages:

1) purchase of phenothiazine core;

2) Synthesis of alkyl or acyl radical;

3) Addition of the radical to the phenothiazine nucleus and obtaining the hydrochloride of the organic base.

The synthesis of phenothiazine derivatives can be shown as follows on the example of chlorpromazine: to synthesize chlorpromazine, 2-chlorophenothiazine is obtained from 2,4-dichlorotoluene at the initial stage:

3

H

C

l

C

C

l

H

N

O

3

C

O

O

H

l

C

C

l

 2,4-dixlortoluol 2,4-dixlorbenzoy turşusu

N

H

2

a

n

i

l

i

n

C

H

O

O

C

l

N

H

6

3

$-$CO2

 3-xlordifenilamin-

 6-karbon turşusu

N

H

C

l

S

2

5

0

C

0

3-xlordifenilamin

N

H

C

l

S

In the 2nd stage, dialkylaminoalkyl compounds are obtained from simple organic substances. For example, 3-dimethylaminopropyl chloride-hydrochloride is obtained according to the following scheme:





3-dimethylaminopropyl chloride-hydrochloride

In the 3rd step, the addition of dialkylamine alkyl chloride is carried out by replacing the hydrogen atom in the 10th position to the phenothiazine core. For example, in step 3 of the synthesis of chlorpromazine hydrochloride, 2-chlorophenothiazine is reacted with 3-dimethylaminopropyl chloride:





*chlorpromazine chlorpromazine-hydrochloride*

*basic (Aminazine)*

The 10-acyl derivatives of phenothiazine are synthesized by a similar scheme, but in step 3 the phenothiazine derivative is treated with chloroanhydride of β-chloropropionic acid:



By replacing the chlorine atom with the corresponding radicals, acyl derivatives are obtained. Thus, moracizyme, etacizin and other derivatives are synthesized by this method.

Alkylamine derivatives have anti-neuroleptic, hypotensive, anti-emetic, and anti-histamine effects, while acyl derivatives are used in cardiovascular diseases. 10-alkyl derivatives that have a neuroleptic effect also enhance the effect of drugs, sleeping pills, analgesics, and anticonvulsant drugs.

The following representatives of phenothiazine derivatives are widely used in medical practice:

**1. Chlorpromazine Hydrochloride – Chlorpromazine Hydrochloride**

**(Aminazine)**



General structure of phenothiazine derivatives

2-chloro-10-(3/-dimethylaminopropyl)

phenothiazine-hydrochloride

M. k. 355.33

It is a white crystalline powder with a white or brown tint. Hygroscopic, dark on the world. Very easily soluble in water, easily soluble in alcohol and chloroform, practically insoluble in ether and benzene. Melting temperature 195-1980С.

This is a neuroleptic and sedative. Chlorpromazine-hydrochloride 0.025; 0.05; 1 in 0.1 g dragee (tablets, coated) and 2.5% solution for injection; Available in 2 and 5 ml.

Analysis of phenothiazine derivatives

Definition of personality

When analyzing derivatives of phenothiazine, their acid-base properties, reducing properties and other reactions are mainly used.

Most medicinal substances containing phenothiazine derivatives are salts of strong mineral acids and organic nitrogenous bases. Organic bases are separated by the action of solutions of alkaline preparations, carbonates and solutions of ammonia.

Derivatives of phenothiazine in the form of nitrogen-containing salts interact with general alkaloid precipitation reagents (Mayer, Dragendorff, Wagner-Bukhara-picric acid, etc.). Since the bases of medicinal substances with phenothiazine derivatives are not crystalline, but amorphous or oil-like substances, the determination of the melting temperature of the complexes formed by them with common alakloid reagents is of great importance in the analysis of these substances.

Crystals of some complexes of drugs of the phenothiazine group with Dragendorff's reagent have a characteristic shape, and this property is used in toxicological chemistry.

When determining the identity of phenothiazine derivatives, oxidation, salt and complexation reactions are also used, as well as reactions related to the determination of nitrogen, sulfur and chlorides. Phenothiazine derivatives are easily oxidized with the formation of colored products and are widely used.

Physico-chemical methods are used for the analysis of phenothiazine derivatives, including UV-spectrophotometry, IR-spectroscopy and YEMX methods.

1) Derivatives of phenothiazine are easily oxidized and give colored products. Bromine water, chloramine solution, HNO3, iron 3-chloride, H2O2, solid H2SO4 (sometimes with methylene blue), KIO3, KBrO3, Ce(SO4)2, etc. are used as oxidizers. in an acidic environment. berutsya solutions of substances.

The formation of colored products is caused by the formation of cationic (paramagnetic) and dicationic (diamagnetic) radicals.



The sulfur atom in the molecule of phenothiazine derivatives is oxidized and forms various colored products. The paramagnetic cation oxidizes to a diamagnetic dication, which in water forms products such as sulfoxides and sulfones:



sulfoxides

The color in the C2 state depends on the nature of the radical and does not depend on the nature of the oxidant. Thus, the final products of oxidation are 9-S-oxide, 9,9-dioxide (sulfone), 3-oxy, 3,7-dioxy-3-one-, 3-oxy-7-one, thioridazine 2,9-S- thio oxide, 2, 2, 9, 9 - tetraoxide can be:

S

N

R

1

9

R

2

5

6

7

8

1

2

3

4

O

S

N

R

1

R

2

O

O

9-S-oxide 9,9-S-dioxide

Bromine water is more typical of the oxidizing reagents for the phenothiazine nucleus. Thus, the reaction with bromine water is used to distinguish phenothiazine derivatives from each other (table ...). Phenothiazine is oxidized with bromine water to form red colored perbromophenothiazonium:



perbromophenothiazonium

Bromine water, nitric acid, 0.15% methylene blue along with sulfuric acid are considered group reagents for phenothiazine derivatives. Instead of bromine water, 1% potassium bromate with 0.15 ml of hydrochloric acid can be used.

Color reactions of phenothiazine derivatives with bromine water and solid nitric acid

Table

|  |  |  |
| --- | --- | --- |
| **Preparatın adı** | **Bromlu su** | **Qatı nitrat turşusu** |
| Aminazin | açıq-moruğu | qırmızı rəng və ağ bulanıq, yenidən reaktiv əlavə etdikdə rəng itir |
| Propazin | bulanıqlı tünd-qırmızı | tünd –çəhrayı |
| Diprazin | narıncı, moruğu rəngə keçir və çöküntü alınır | qızdırdıqda qırmızı |
| Triftazin | qəhvəyi, çəhrayı rəngə keçən  | qırmızı  |
| Etmozin vəEtasizin | əvvəlcə açıq yasəməni, sonra parlaq-bənövşəyi |  |

2) Phenothiazine derivatives precipitate with precipitating reagents belonging to alkaloids (solution of iodine in potassium-iodide, picric acid, Mayer, Marme, Dragendorf).

3) They alkalize the solution of salts of phenothiazine derivatives in water with NaOH; a white precipitate is formed. After 5 minutes, they filter the solution and carry out a specific reaction for chlorides in the filtrate:

Aminazine-base · HCl + NaOH → NaCl + H2O + aminazine-base↓

4) UV-spectrophotometry: Phenothiazine 10-alkyl derivatives give two absorption maxima in the UV region (290-330 nm). In 10-acyl derivatives, a hypsochromic shift is observed in both maxima. A 0.00055% solution of chlorpromazine in 0.01 M hydrochloric acid has a wavelength of 255 (E\_1cm^(1%) 830) and 307 nm, a solution of promazine-hydrochloride at the same concentration in water has a wavelength of 251 nm, fluazicin in 0.1 M hydrochloric acid The 0.002% solution gives maximum absorption at 258 nm wavelength (E\_1cm^(1%) 190-220).

5) Dissolve 1-2 mg of the drug in 3-5 drops of water, add 10 drops of HNO3 and heat it in a hot water bath for 1 minute. After cooling the solution, carefully add 15-20 drops of 25% NH3 solution through the wall of the test tube and observe under UV rays. Violet fluorescence is observed in fluphenazine and trifluoperazine, and green fluorescence is observed in fluazine (fluorescence is not observed in chlorpromazine, promazine, promethazine, diprazine, perphenazine).

6) Phenothiazine derivatives form colored complex compounds with Fe3+, Hg2+, Co2+ and Pt2+ ions.

7) To determine the sulfur in the phenothiazine nucleus, the preparation is heated with Na2CO3 and NaNO3. They dissolve the residue in water, filter it and carry out a sulfate reaction.

8) Designation of phenothiazine derivatives with a piperazine nucleus in the molecule.

Place some of the preparation in a test bottle, soak it with a 10% solution of p-dimethylaminebenzaldehyde in alcohol and put a filter paper on it; They heat the preparation for 1-2 minutes on the stove until it darkens; due to the effect of pyrrole vapors, a red-purple color is obtained on the filter paper (piper nucleus). This reaction is given by trifluoperazine (tryftazine), fluphenazine (fluorphenazine) and perphenazine (etaperazine).

9) Determination of fluorine. 0.025 g of the preparation is melted with sodium metal, cooled, diluted with water and filtered. 0.5 ml of glacial acetic acid is added to the filtrate. 0.1 ml of the obtained solution is treated with a mixture of freshly prepared alizarin red C and 0.2 ml of a 0.1% solution of zirconium nitrate in hydrochloric acid; the red color of the solution changes to light yellow. This reaction is given by trifluoperazine (triphtazine), fluphenazine (fluorphenazine) and fluazizin (fluorazine).

10) Pyrrole is formed when morpholine derivatives are heated with strong acids due to the presence of the CH2 – CH2 – N = group in morasin (morphine residue).

The preparation is dissolved by heating in dilute hydrochloric acid and boiled for 1 minute. The solution darkens and a light lilac color is obtained. After cooling, the mixture is divided into two parts. They add water and bromine water to a part of it; light lilac color changes to violet (phenothiazine derivative). Filter the 2nd part of the solution, add a few drops of sodium-nitrite solution to the filtrate; a green color passing to yellow is obtained (morpholine).

11) As a result of the hydrolysis of the carbethoxy group (-COOC2H5) in the 2nd position of etasin and morasin, ethyl alcohol is formed, which gives an iodoform test in an alkaline medium with an iodine solution:

C

O

O

C

2

H

5

+

2

N

a

O

H

N

H

R

N

a

2

C

O

3

+

C

2

H

5

O

H

+

R

-

N

H

2

C2H5OH + 4I2 + 6NaOH → HCOONa + 5NaI + CHI3 + 5H2O

0.03 g of the drug is dissolved in 1 ml of NaOH solution; add a few drops of 0.05 M iodine solution and heat until it boils; smell of iodoform.

12) Wagner-Buchard reagent (solution of iodine in potassium-iodide) is used to determine nitrogen in phenothiazine derivatives.

14) Phenothiazine derivatives can be distinguished by the NTX method. "Silufol" plates are used, the ethylacetate - ethanol - diethylamine (17:2:0.5) system is used as a solvent. After running the chromatogram with iodine vapors, depending on the nature of the substituent in the 2nd position, the adsorption areas are colored blue-green (promazine, promethazine, chlorpromazine hydrochlorides) or pink-orange (trifluoperazine - hydrochloride, fluorphenazine).

Determination of cleanliness

Foreign impurities are checked by the NTX method. Silufol plates and hexane-acetone-diethylamine (50:30:2) solvent system are used.

Quantitative methods of phenothiazine derivatives:

1) Spectrophotometry method. Quantification of chlorpromazine, promazine and fluazizin can be done by this method. In parallel, an experiment is carried out with a standard sample (see identity determination 5).

2) Iodometry method. The assignment is based on the formation of chlorpromazine periodide (chlorpromazine · 3I2). The excess of iodine is titrated with 0.1 M sodium thiosulfate solution (E=M.k./6; T=0.005922 g/ml).

Quantification of promazine and chlorpromazine is also carried out by bromatometry and iodochlormetry methods.

3) Neutralization (alkalimetry) method. Most phenothiazine derivatives are determined by this method.

Titration is carried out with 0.05 M NaOH solution in chlorpromazine and 0.1 M NaOH solution in promazine (indicator-phenolphthalein; T=0.01775 g/ml in chlorpromazine, T=0.03209 g/ml in promazine). The organic base obtained as a result of the reaction is extracted with chloroform.



4) Aqueous titration method.

Perphenazine n.k. dissolved in a mixture of acetic acid and acetic anhydride (30:20) (20 ml of benzene is additionally taken in moracin, and acetic acid as a solvent in furacin) with the presence of mercury 2-acetate solution (in some cases it is replaced by formic acid) and 0.1 m with perchloric acid (indicator violet crystal). Titration in perphenazine is blue-green (T=0.02385), and in other preparations it is yellow (T=0.0464 g/ml in moracin and t=0.04309 g/ml in fluacin).

It should be noted that the anhydrous titration method is used in different variants during the determination of the amount of phenothiazine derivatives. However, all variants use perchloric acid as a titrant. In the analysis of promazine, promethazine and chlorpromazine hydrochlorides, acetone is used as a solvent, and methyl orange (in acetone) is used as an indicator.

For hydrochlorides of 10-alkyl derivatives of phenothiazine, the titration process in anhydrous medium proceeds as follows:



5) In determining the identity, purity and quantity of phenothiazine derivatives, UV- and IR-spectroscopy, YEMX, thin-layer and paper chromatography (solvent: n-butanol-acetic acid-water (50:1:50), the chromatogram is clarified with Dragendorf reagent and at this time only one spot should be visible) methods are used.

"Silufol" plates are mainly taken in thin layer chromatography. The solvent here is ethylacetate-ethanol-diethylamine (17:2:0.5); hexane-acetone-diethylamine (50:30:2), chloroform-diethylamine (9:1), etc. Is used. Stains are clarified with iodine vapors, Dragendorf's reagent.

Since phenothiazine derivatives are easily oxidized, they are stored in dark glass containers. These drugs enter the body through the respiratory tract, skin and mucous membranes and cause itchiness, depression, and lowering of arterial pressure.

Therefore, when working with those preparations, you should use an absorbent cabinet and gloves. After finishing work, it is necessary to wash hands with cold water without soap (when using soap, phenothiazine derivatives can be transferred to the base form, as a result, they cannot be washed with water).

**Oxazepam-Oxazepam**

**(Nozepam, Tazepam)**



7-chloro-2,3-dihydro-3-oxy-5-phenyl-“H-1,4-benzodiazepine-2-one