MODERN ANALITICAL COLUMNS FOR CHROMATOGRAPHY – APPLICATIONS, PROBLEMS AND PROSPECTS

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Introduction. According to leading experts the segment of chromatographic methods in analytical chemistry has approached almost 50% in the last years. Among all chromatographic techniques High-Performance Liquid Chromatography (HPLC) is considered a leader in all areas – development of new theories and methods of separation, development of adsorbents, chromatographic columns, etc.

In 2004 Waters Corporation presented new chromatographic system, which provided 1000 atm pressure, and analytical columns with 1.7μm particles. This system was called UPLC – Ultra Perfomance Liquid Chromatography and was awarded with R&D 100 Award and Pittcon Editors' Gold Award.

Aim. The main purpose of our work was to study normative documents, latest information sources dedicated to development and application of modern analytical columns for chromatography analysis. We tried to assess the prospects of the development of different variants of «fast» liquid chromatography, progress in the implementation of new technologies by leading manufacturers, discussion of difficulties and challenges in development of these methods.

Materials and methods. The «core» of every chromatographic system is a column which provides fast and efficient separation of a complex compound.

Nowadays the leading manufacturers launch more than 1000 different kinds of columns. Experts estimate an annual growth rate of about 50. Labs Column Selection Database was created to make the work with this variety of choices much easier. It is a database where all the columns are classified by "Tanaka" system that includes such important parameters as hydrophoby, selectivity to homologue, structural selectivity, contribution of hydrogen bond, total ion-exchange capacity at a pH of 7,6 and ion-exchange capacity at a pH of 2,7. ACD/ChromGenius program helps to predict the time of separation of substances and kind of chromatograms on the base of the structural formulas of compounds.

A great amount of attention in the leading Pharmacopoeias is paid to global trends of chromatographic methods. We studied materials devoted to methods of chromatography with columns and such variants as Gas Chromatography (GC), Liquid Chromatography (LC), Size-Exclusion Chromatography (SEC), Supercritical Fluid Chromatography (SFC) by The State Pharmacopoeia of Ukraine (SPhU) 2.0 (2014, 2015), The United States Pharmacopoeia (USP) 38 (2015), The European Pharmacopoeia (Ph. Eur.) 8.0 (2013), The British

Pharmacopoeia (BP) (2013), The Pharmacopoeia of Russian Federation 13 (2015).

Results and discussion. For the evaluation of organic related substances Pharmacopoeias usually recommend to use chromatographic methods. Lately experts have marked the trend of changing Thin-Layer Chromatography (TLC) method for HPLC, which is more specific and accurate.

It should be noted that HPLC became the leader in the USP in the area of quantification of medical substances of organic nature at the same time when Ph. Eur. began to recommend using HPLC in about 20% of assay cases.

There is a complete list of packings (L), phases (G) and support (S) used in USP-NF test and assays in USP 38 in the section of Reagents, Indicators and Solutions-Chromatographic Columns.

Among leading manufacturers of packings for chromatographic columns USP recommends Waters Corp., Tosoh Bioscience, Dionex Corp., Agilent Technologies, Chiral Technologies, Metrohm, Sigma-Aldrich (now a part of Merck), ZirCrom Separations, Thermo Fisher and others.

Reversed-phase HPLC is used to assay drugs by LC method. Hydrophobic (modified) silica gels, for example, sorbing agent with C_{18} phase (octadecyl), or C_8 (octyl) and some other polymers with 2-10 μm particles in LC (less than 2 μm in UPLC) are often used as stationary phases. These phases are stable in the pH range of 2-8 in the mobile phase. Columns with porous graphite or some polymers allow working in a wider range of pH. Water-methanol and water-acetonitrile mixtures are often used as a mobile phase in reversed-phase HPLC.

Merck Corporation developed monoliths for chromatography of the first generation in the beginning of 1990s, monoliths of the second generation in 2012. These columns have a high efficiency of separation, they are significantly less resistance to flow of the mobile phase and they compete with columns for UPLC with particle size of 2 μm.

At Pittcon 2016 Merck, which bought Sigma-Aldrich for $17 billions in the end of 2015, presented such new technologies as chromatographic columns Watercol® – new capillary columns with innovative stationary phases for water assay by gas chromatography. They also presented new columns Ascentis® Express Biphenyl for UPLC/HPLC, which separate pharmaceutical drugs and their metabolites, and SLB®IL (i-series) – new capillary columns for gas chromatography which provide selectivity of separation of polar compounds.

Conclusions. Chromatographic methods of analysis are now leading in different fields and in pharmaceutical analysis in particular. The success of every chromatographic assay in GC, LC, HPLC, UPLC, SEC, SFC, CapLC, μLC depends on the right choice of method, chromatographic column, conditions of separation of complex compounds, detector, etc. Future pharmacy specialists, who study at the National University of Pharmacy, successfully master these methods and knowledge.