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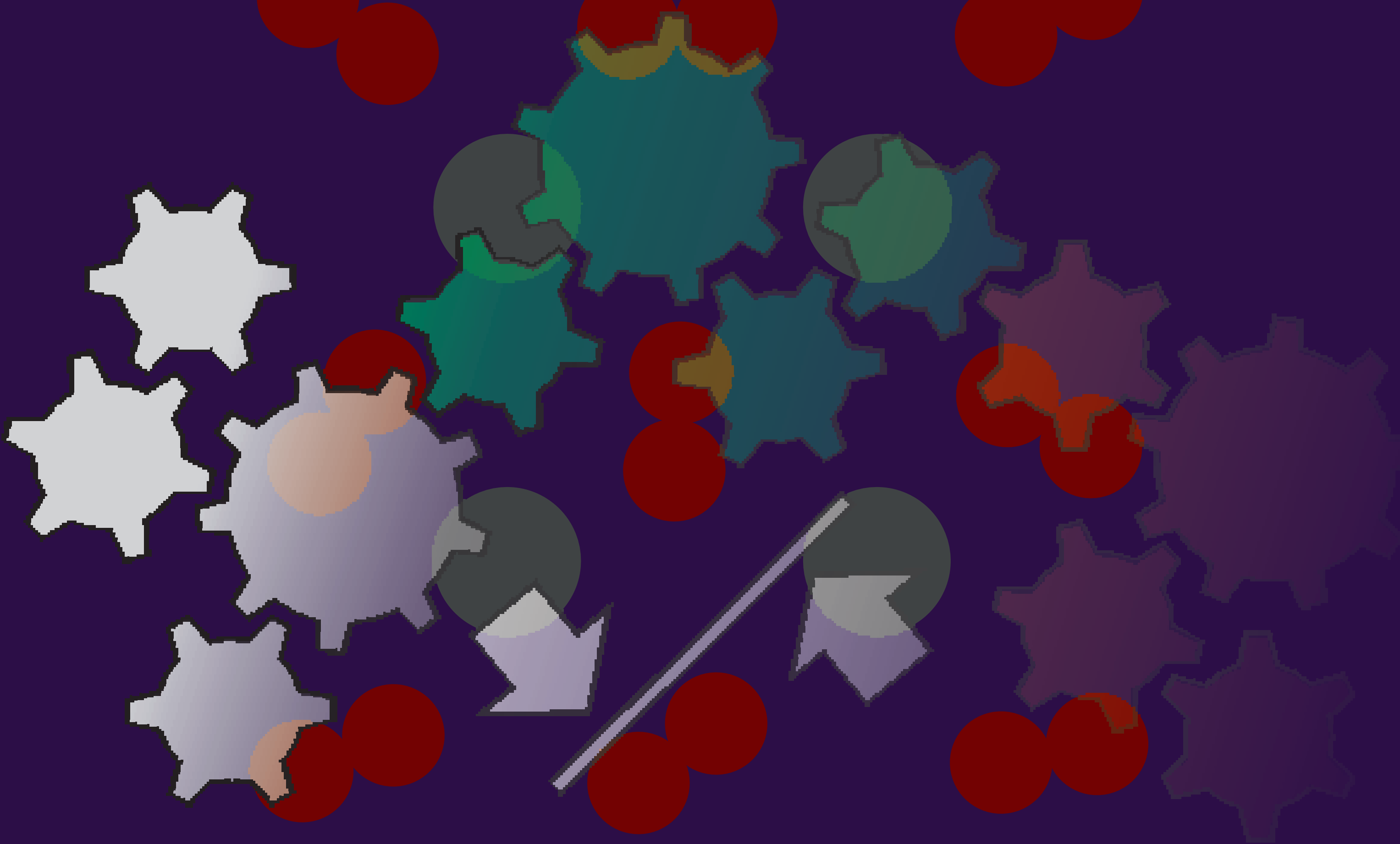

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ANALYSIS OF PROTEINS AND PEPTIDES BY MASS SPECTROMETRY

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Keywords: proteins, peptides, mass spectrometry
Abstract: Analysis of proteins and peptides by mass spectrometry became possible by the end of the 20th century. MS z is a method of analytical chemistry. It works with a *m/Q* ratio where *m* is the mass and *Q* is the fragment charge. It is used to determine the mass of particles, to determine the elemental composition of a sample or molecule, and to elucidate the chemical structure of molecules such as peptides and other chemical compounds. Its principle is based on ionizing chemical compounds and measurements of their mass relative to charge.

1 Introduction, analysis and identification

Analysis of proteins and peptides by mass spectrometry became possible by the end of the 20th century. MS z is a method of analytical chemistry. It works with a *m/Q* ratio where *m* is the mass and *Q* is the fragment charge. It is used to determine the mass of particles, to determine the elemental composition of a sample or molecule, and to elucidate the chemical structure of molecules such as peptides and other chemical compounds. Its principle is based on ionizing chemical compounds and measurements of their mass relative to charge.

1. The sample is placed in the apparatus and undergoesevaporation.
2. The sample components are ionized in one of a number of ways (for example, the impact of an electron beam), resulting in the formation of charged particles - ions.
3. The ions are separated according to the m/Q ratio in the electromagnetic field analyzer.
4. Ions are detected, usually by a quantitative method.
5. Ion is processed by mass spectrometer, Figure 1 [1].

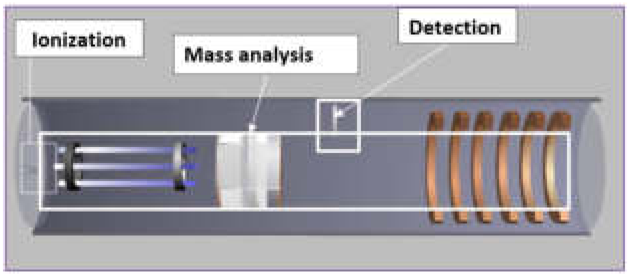

Figure 1 Basic parts of MS [2]

The mass spectrometer consists of three modules:

- The first module is a source of ions that can convert gas molecules to ions
- The second module is a mass analyzer that sorts the ions by their mass using electromagnetic fields
- The third module is a detector that measures the amount of the indicator quantity and thus provides the data to calculate the abundance of each ion in real time.

Technique has qualitative and quantitative use. These include identifying unknown substances, determining the isotopic composition of the elements in the molecule, and determining the structure of the compound by observing its fragmentation. Further use involves detecting the quantitative amount of the mixture in the samples, or studying the gas phase ion bases (ion and neutral molecules in vacuum). Mass spectrometry is commonly used in analytical laboratories that study the physical, chemical or biological properties of various compounds.

1.1 Steps of mass spectrometry

In the Figure 2 you can see the graphical representation of individual steps of mass spectrometry.

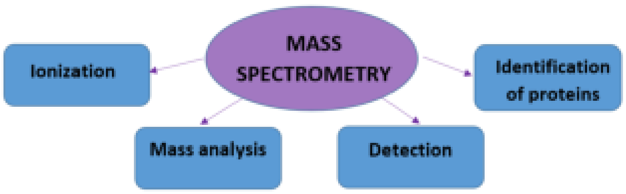

Figure 2 Steps of mass spectrometry

1.1.1 Ionization

The ion source is part of the mass spectrometer. This component ionizes the material by analysis (analyte). The ions are then transported by magnetic or electric fields to a mass analyzer. Ionization techniques have been the key to determining what types of samples can be analyzed by mass spectrometry. Electrone and chemical ionization are used for gases and vapors. In the chemical ionization of sources, the analyte is ionized by chemical ion-reacting molecules during collisions in the source. Examples of ion sources are:

ESI ("electrospray ionization")

For ESI, a sample of proteins dissolved in a solvent (liquid medium), in a volume of about 1 µl, is placed in the capillary. The conically tapered capillary allows a sample flow rate of 20-40 nl / min, the volume of the sample can be analyzed for up to 1 hour. The liquid is drawn into the electrostatic field, the droplets are sucked in by the drying air, the solvent evaporates and the particle breaks up, increasing the repulsive forces. Repeating this process makes it possible to obtain the ions of proteins (charged molecules) that move in the vacuum tube, the process zones and their movement is directed by the electric field. Analyzed molecules can carry multiple charges. This type of ionization and mass analyzers allows the analysis of proteins with molecular weight to 5000 [3,4].

MALDI (matrix-assisted laser desorption/ionization).

For MALDI, the sample is dissolved in a volume of about 1-2 µl, mixed with the same volume of a matrix solution containing an organic substance (e.g., 3,5-dimethoxy-4-OH-cinnamic or 2,5-dihydroxybenzoic acid) about a solid environment. The sample solution is applied to a target and dried at room temperature. The sample crystals are broken down by a small-laser laser and released from the target. The ions are moved in the vacuum tube and the movement is directed by the electric field. The time at which the ions reach the detector is proportional to the second power of the mass / charge ratio (m/Q). Smaller proteins, therefore, make the detector much longer (time of flight) on the basis of which we can determine their mass. This type of MS allows the analysis of proteins with a molecular weight of up to 100,000, Figure 3 [3].

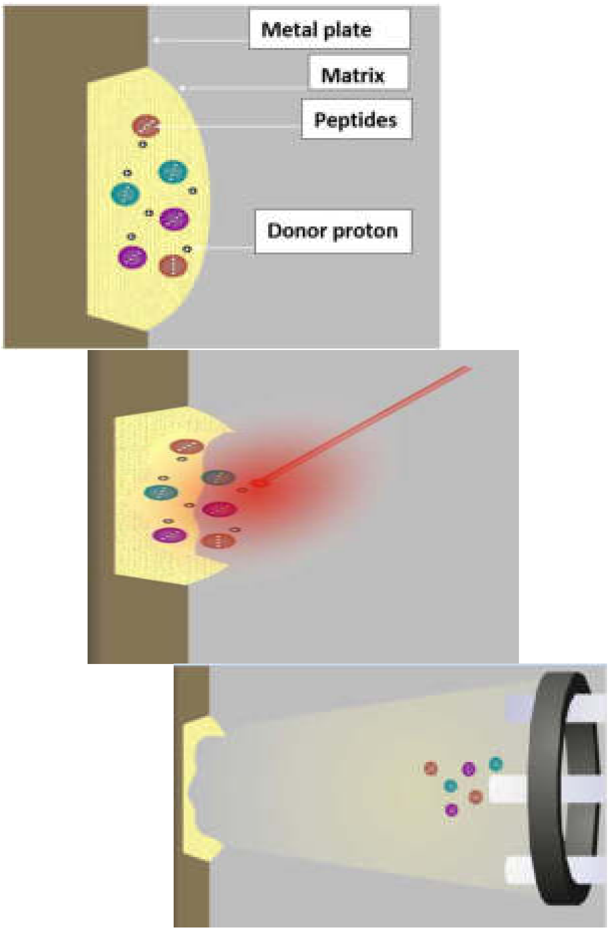

Figure 3 The MALDI principle [2]

1.1.2 Mass analysis

Mass analyzers separate the ions according to their m/Q ratio. The following two laws (1), (2) are governed by the dynamics of charged particles in electric and magnetic fields in vacuum:

$$F = Q(E + v \times B) \quad (1)$$

Lorentz's law of power

$$F = m.a \quad [N] \quad (2)$$

Newton's second movement law in an unrealistic case, i. only applies at an ion rate significantly lower than the speed of light

F – ionic force,
m – the mass of ions,
a - acceleration,
Q – ion charge,
E – electric field,
$v \, x \, B$ - vector product of velocity ions and magnetic field.

The sign of equality between the expressions for the above forces acting on the ion implies (3):

$$(m/Q).a = E + v \times B \qquad (3)$$

This differential equation is the classical motion equation for charged particles. Together with the particles of initial conditions, the particles completely determine the motion in space and time as regards the m/Q ratio [5].

There are many types of mass analyzers: static or dynamic, magnetic or electrical, but all operate according to the above differential equation. Each type of analyzer has its strengths and weaknesses. Many mass spectrometers use two or more mass analyzers for tandem mass spectrometry (MS / MS). There are also other types of analyzers eg:

Quadrupole

The four-pole mass analyzer uses an oscillating electric field to selectively stabilize or destabilize the ion path by radio frequency, the electric field being formed from four parallel electrode rods, alternating at different frequencies. Only ions within a certain range of m/Q ratios are sold through the system at any time. Potential changes on the electrodes allow a wide range of m/Q values. The four-pole filter acts as a selective filter in the mass analyzer and is closely associated with the quad-core ion trap (Figure 4). The other variations of the four-pole are three-quadruples.

Triple quadrupole mass spectrometers have three consecutive quadruples arranged in series. The first quadrupole acts as a mass filter, the second acts as a collision target, where the ions are selected into fragments. The resulting fragments are analyzed by the third quadrupole [5].

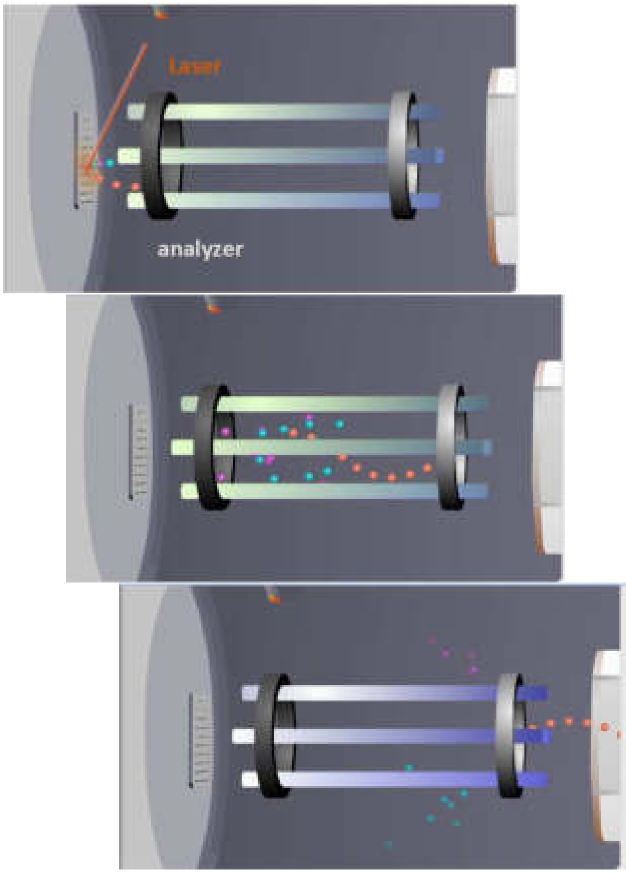

Figure 4 Quadrupol analysis scheme [2]

Quadrupole Ion Trop

The four-pole ionic trap operates with the same physical laws as four-pole mass analyzers, but the ions are trapped and subsequently evaluated. The sample is ionized either internally (e.g., electron or laser beam) or externally, in which case the ions are often introduced through the hole to the so-called “face” of the electrode.

There are many weight separation and isolation methods, but the most unstable is a mass unstable mode in which the radio frequency potential is maintained such that the orbits of the mass “a” > “b” are stable while the ions with the mass "b" < "a” are unstable and are catapulted to the *z-axis* on the detector. Non-destructive methods of analysis are also used there.

The ions can also be evaluated by the resonant excitation method when the additional oscillation excitation voltage is applied to the face of the electrode, and the amplitude capture and / or frequency of the excitation voltage are manifoldly transferred to the resonant state in order of their m/Q ratio. The linear four-pole ionic trap is similar to a four-pole ion trap, but it captures the ions in a two-dimensional four-pole array instead of three-dimensional quadruple fields as in a 3D four-pole ion trap. An example of a linear ion trap is Thermo Fisher LTQ [5].

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Time of Flight Mass Spectrometry (TOF-MS)

TOF - MS uses the differences in times when the ions pass through the separation field, and they are separated by mass. It works in pulse mode, so the ions have to be formed or must be extracted from the ion source in pulse mode. The electric field accelerates all ions to the unbounded area of drifting with kinetic energy $z.V$ (4) where z is ion charge and V is the applied voltage. Since the kinetic energy of the ion is $\frac{1}{2}mv^2$, the more easier the ions have a greater velocity than the heavier ions, and the detector reaches the end of the drift area earlier.

$$E_{kin} = \frac{1}{2}mv^2 = z.V \quad [J] \qquad (4)$$

The speed of the moving ions is proportional to the second power of the ratio of their double kinetic energy and the mass of the ion (5):

$$v = \sqrt{\frac{2.z.V}{m}} \quad [ms^{-1}] \qquad t = \frac{L}{\sqrt{\frac{2V}{m/z}}} \quad [s] \quad (5)$$

The flight time t through the drift tube is L/V, where L is the length of the drifting tube.

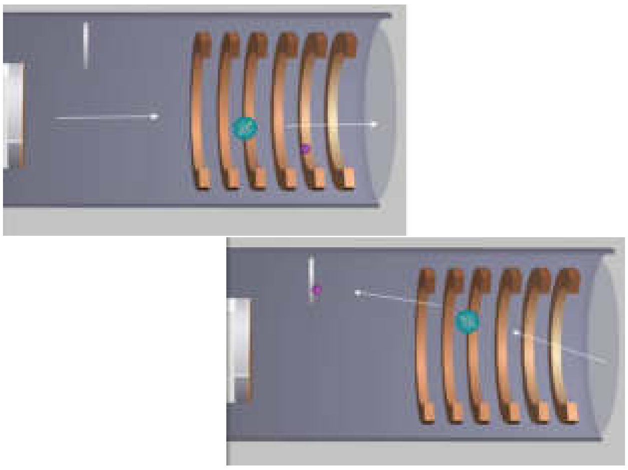

Figure 5 The scheme of TOF analyzer for mass, before and after protein discoloration [7]

The flight time analyzer uses an electric field to accelerate ions with the same potential. If the particles have the same charge, the kinetic energy of the particles will be identical, and their velocity will depend only on their weight. Easier ions reach the first detector (Figure 5) [5].

1.1.3 Detection

The last element of the mass spectrometer is the detector. Recognizes induced or current ion passes or their effects. The detector consists of two metal plates that record the passage of ions. The ion current on the detection slot is very small (approximately 10^{-9}-10^{-18}A). Therefore, a photoelectronic multiplier is used which provides an increase of up to about 10^{-6}.

As a record of mass spectra, fast mirror galvanometers were used in the past to record on photographic paper that was developing itself in the light. Currently PC computing technique is used and spectra are obtained both in tabular form and graphically in standard form [5,6].

1.1.4 Protein identification

In the Figure 6 you can see the diagram of proteins identification.

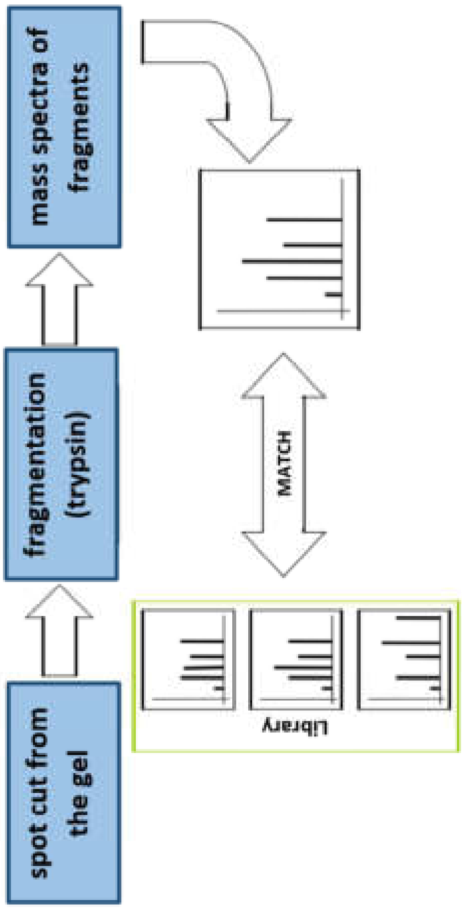

Figure 6 Diagram of proteins identification [1]

There are several ways that MS use to identify proteins. You are here for example:

Peptide mass fingerprinting Maldi Time of Ftight (peptides generated by digestion can be determined by mass spectrometry using peptide mass spectra) utilizes the masses of proteolytic peptides as input for searching in the database of predicted values generated by the digestion of known proteins. If the protein sequence is in the database list, there is evidence that this protein is present in the original sample.

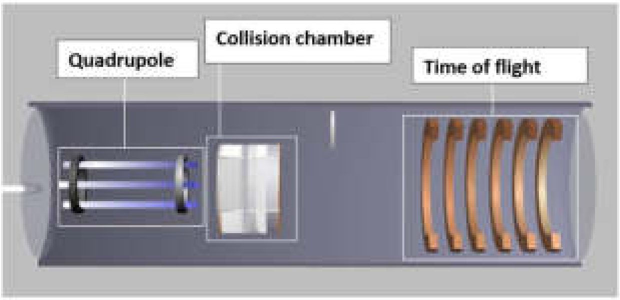

Figure 7 The scheme of Tandem Mass Spectrometry – Quadrupole Time of Flight [7]

Tandem Mass Spectrometry is an increasingly popular experimental method used to identify proteins, characterize their amino acid sequences, and post-translational modifications. Collision-induced dissociation is used in conventional applications to generate a fragment of a particular peptide ion. The fragmentation process results in the cleavage of products by electron capture which decomposes along the peptide bond in the gas phase. With such unnecessary fragmentation, it is possible to use the observed fragments from the mass to match the predicted sequence database to one of the many peptide sequences listed. This technique is referred to as a "top-down or botton-up" approach, because starting with the whole mass and then pulling it from one another, proteolytic fragments are cracked, the protein being assembled together by de novo repeated detection, Figure 7 [3, 7].

2 Conclusions

Today, we are aware that the path from genomics to proteomics would not have been possible without the application of improved special techniques using mass spectrometry principles. There is probably no other current technique that would overcome the mass spectrometer in a variety of applications in basic and applied research as well as in diagnostics.

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5D OF BIM – RESEARCH STUDIES AT HOME AND SLECTED COUNTRIES AROUND THE WORLD

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Keywords: building information modelling, 5D, cost, implementation, cost management tools
Abstract: Currently time is characterized as a stage of dynamic progress and expanding use and implementation of information and communication technology generally. Data and information about cost and budgeting are required in the knowledge society heads grow every day. Several studies suggest that the use of Building information modelling (BIM) and cost management tools contributes on cost reducing of project and enterprise management. However, these studies also indicate that their use in small and medium sized enterprises is low. Several surveys have been carried out abroad on this issue. The exploitation of Building information modelling to support cost management in many enterprises is a priority and one of the main steps and procedures to successfully manage enterprises and construction projects. Building infomration modelling and cost management tools are one of the possible solutions for cost reducing. Article discusses issue of possibility to use cost management tools and Building information modelling in Slovak construction industry and abroad from various point of view. The main objective of this article is to make an overview of using BIM technology and cost management tools at home and abroad. The paper also aims to point to new research opportunities and to identify research issues in this area.

1 Introduction

Building information modelling is the process of creating and data managing about the building during its life cycle. According to National Institute of Building Science (NIBS, 2015), digital model represents a physical and functional object with its characteristics and specifics. It serves as an open database of information about the object for its execution and construction in all lifecycle [1].

BIM model is more than 3D model. BIM is common data environment. The common data environment can be divided on Figureical and non-Figureical information. According to BIM principles there exists 7 established dimensions, which are:

- 3D – geometry,
- 4D – time,
- 5D – costs,
- 6D – sustainability,
- 7D – facility management [2].

Building Information Modelling (BIM) and automated quantities technologies provide both enormous opportunities and challenges for the project cost management profession.

Fifth dimension is used for budget tracking and cost analysis. Creating a construction budget is one of the most important parts of the life cycle of a project. Determining the amount of costs is demanding in terms of the quality of the documents provided and the method of budgeting [3].

The fifth dimension of BIM – costs bring a lot benefits for example better visualization construction, improvement collaboration on projects, increase project quality and BIM Data Quality, increase project conceptualization, increase efficiency of Take-offs, increase efficiency of Cost Planning, earlier risks identification and other [4].

2 5D BIM -tracking and cost management tool

Exploitation of computers and software has increased productivity and speeded up of processes. In fact, more than 55% of estimates fail due to insufficient software solutions in estimating process. The best software solutions may not be complicated or costly in cost management and estimating process. It must meet the following conditions in particular:

- data import tool,

- an integrated cost database (custom pricelists or company data compared to commercially available data),
- a cost estimating tool [5].

The construction market is aware of the merits of eliminating errors caused by incorrectly compiling the report, which will then be transferred to the actual pricing, so there is an increasing supply of software applications for the creation of calculations, budgets, and quotes. The overview of software tools supporting 5D BIM and their use in countries is shown in Table 1.

Table 1 Overview of software application

Software application	Company	Use in countries
Cubit Buildsoft	Australia	Australia, New Zealand, United Kingdom, Ireland
Nevaris Nemetschek	Germany	Germany, Switzerland, Austria
Exactal COST X	United Kingdom	worldwide (Europe - United Kingdom, Ireland)
INNOVAYA Visual Estimating	USA	USA
BIM estiMate	Poland	Poland
Vico Cost Planner	USA	worldwide (Europe - United Kingdom, Sweden, Asia, Australia)
Gala Construction Software	Croatia	Croatia
iTWO 4.0	Germany	Austria, Cyprus, Czech Republic, Denmark, Germany, Slovakia, Spain, Switzerland, Great Britain, Ireland
Synchro Software	USA	worldwide (Europe - United Kingdom)
Calcus	Norway	Norway

Building information modelling brings many benefits. Many countries have implemented BIM in their laws and standards and research about state of implementation BIM was applied. In this chapter is presented overview to realised researches oriented on state of implementation fifth dimension - 5D costs, in selected countries.

2.1 New Zealand and Australia

Organisation Masterspec conducted national BIM (New Zealand) survey found that the proportion of BIM users increased from 34 % (2012) to 57 % (2013), with a year-on-year increase in overall BIM awareness in the construction industry, from 88 % (2012) to 98 % (2013) [6].

New Zealand's BIM maturity level remains fairly rudimentary, and most of the industry is still operating at Stage 1B ('Intelligent 3D') of the Australian Institute of Architects' BIM implementation scale [7].

5D BIM is rarely being implemented in New Zealand, except on a very few large projects, which have recently used a single integrated BIM model, which can be shared with other project participants [8]. A recent survey of 20 quantity surveyors in New Zealand found that almost all respondents had experienced no more than 5 projects which had used 5D BIM [9]. Despite the purported benefits of BIM, the adoption of 5D BIM in New Zealand and Australia is significantly slow-moving due to a number of barriers limiting its implementation within industry, which are ultimately centred around the fragmented nature of the construction industry, suggesting that a shift in current workflows is required [6,10].

In 2014 Dr. Peter Smith conducted a research called BIM and project cost management – implementation issues and creative solutions. Results revealed that there are considerable implementation issues. This research presented these findings:

- the quality of the BIM models (nevertheless, an increasing number of firms are utilizing 5D BIM tools),
- the technology is always evolving and the interviewees commented that a lot of time and expense can be spent on software and training with uncertain outcomes,
- lack of Standards/software incompatibility,
- sharing cost data information [11].

Respondents interviewed used automated quantities software to prepare quantities on their projects. Four of the firms used this software extensively particularly in the cost planning stages whilst the other two firms used such software in a limited capacity. The firms used both proprietary and in-house software with the CostX program the most commonly used program [11].

2.2 Iceland and Scandinavia

The Scandinavian region has a strong BIM development and implementation track record. In publication Implementation of BIM, Elvar Ingi Jóhannesson focused on comparing implementation of BIM on Island and in Scandinavian countries. In this comparison, Jóhannesson focused on using BIM in various fields, for example marketing, facility maintenance, cost estimation, scheduling and other [12].

Jóhannesson focused on comparing implementation of BIM on Iceland and in Scandinavian countries. BIM´s rate of utilization in cost estimating is higher in Scandinavian countries - 14%.So are interesting tendering and quantity take-off data's. Also in this case Scandinavian countries are in the foreground – tendering 9% and quantity take-off 18% (Figure 21)[12].

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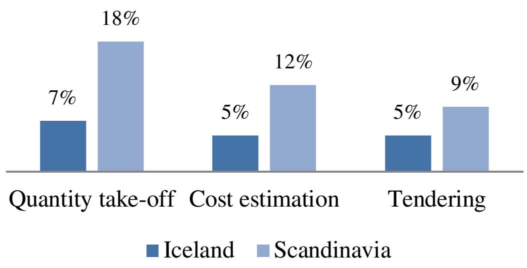

Figure 1 Use of information modeling of buildings – comparison Iceland and Scandinavia [12]

2.3 United Kingdom

Important country on European and world forum is Great Britain. In 2011, British government released a document of labor groups interested in strategies of implementation and using BIM. Also government expressed an intention, that since 2016 it will require using BIM tools for all drafts for public contracts. Great Britain created National BIM Library (NBL), where it offers more than 350 constructing presets (walls, ceilings, windows, doors, etc.), products from different producers. Library is constantly extending, therefore strategy of BIM´s implementation support in British civil engineering industry is supported [13].

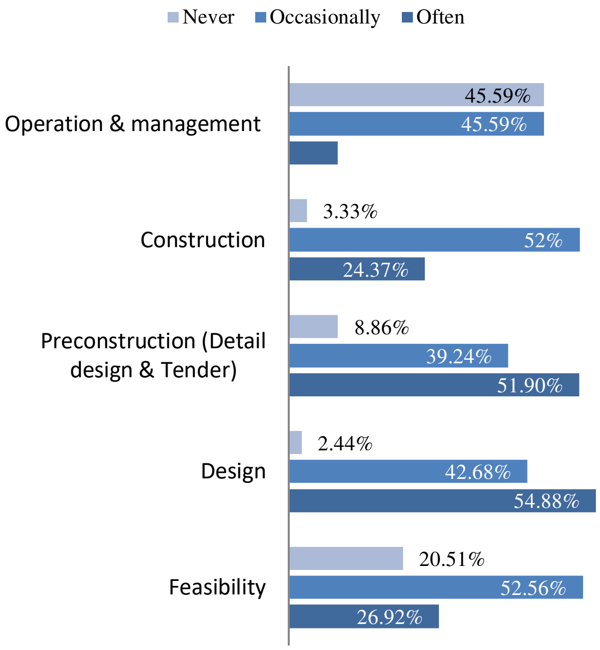

Figure 2 The use of BIM software tools [14].

In 2016, Software Advice analyzed small and medium organizations and their needs. Key discoveries from this survey are follow:

- **60%** seeks in this software options for estimate of construction costs
- **24%** wants to increase project´s transparency and to upgrade its monitoring
- **50%** currently use manual methods for computing costs, offers preparation and managing projects [15].

Robert Eadie´s et. al. conducted research under name of BIM Implementation throughout the UK construction project lifecycle: An analysis. Following the results, software tools supporting information modeling of buildings are used mostly in design (54,88%) and occasionally in realization (Figure 2). In process of pricing and detail design, information modeling is used often, in 51,90% cases, 39,24% occasionally and only 8,84% is not using it [14].

2.4 Slovakia

At present, there are a number of specialized programs for budgeting and cost management in Slovakia. For a example: Cenkros 4, Kalkulus and Odis and so on. These tools can not be considered as full software tools supporting information modeling of buildings.

Implementation of information modeling of buildings in Slovakia is weak because overall there is a great degree of awareness and reluctance to learn with new knowledge. The overall state of implementation in Slovakia is in its early stages. Interesting findings on the state of implementation of BIM in Slovakia brought research carried out by Mesároš, Selín and Mandičák in 2016 under the title Identifying Approaches to Cost Management in Slovak Construction Enterprises. Research has focused primarily on the use of BIM technologies to reduce the total costs of construction enterprises [16].

Research has produced the following findings. It was assumed that there was a direct link between the size of the construction company and the use of BIM technology, but the results rejected this assumption. The survey showed the greater impact of BIM technologies on cost reducing in small enterprises. Accordint to Mesaros, Selin and Mandica, the reason it could be the lower rate of use of BIM technologies in the construction company, or the fact that BIM technology has a significant impact on the reduction of the total cost of construction enterprises, different from the participant in the construction project. BIM technology is mainly used by designers and architects. As part of the research, these participants were smaller enterprises and therefore no significant impact on the size of the construction companies can be confirmed and the use of BIM technologies does not have a significant impact on the reduction of the total costs of the construction enterprises, apart from the size of the construction enterprises [16].

The second research hypothesis examined was to investigate the assumption that the exploitation of BIM technology had a significant impact on the reduction of the total cost in construction enterprises, apart from the majority owner of the construction enterprises. But this hypothesis was also rejected. However, research has confirmed the hypothesis: The exploitation of BIM technology has a significant impact on the total cost reducing in construction projects in a different way from the participant in the construction project. Based on the survey results, the designer and architect have the greatest impact on the use of BIM technology on the total cost of construction projects, followed by major contractors, subcontractors and developers [16]. Even in this area, however, profitability is important, as it say other surveys [17]. That is why we need to look for another link to this.

3 Future research

Knowledge of structure and costs´ dynamic in construction of company is a starting point for effective price making. Constructing itself is characterized by high amount of input data's, information, which has to be taken into account in whole construction process. BIM allows to work with information briefly and effectively. Pricing of civil engineering production does not use information modelling of buildings. Next research goal is to map out and identification current state of civil engineering pricing in Slovakia and define suppositions and barriers of pricing in BIM. Subsequently, based on realized researches and acquired knowledge from Slovakia and foreign countries, suggest and develop exact method of planning economical and costs parameters for civil engineering pricing in information modelling of building.

4 Conclusion

BIM technology offered a lot of functionality and it´s very helpful tool in construction project management. Cost management is very important part of construction project management. Accurate and information based cost management may have an serious impact on the success of construction project. BIM technology has a lot of functionality, includes cost management too. Research tried to make an overview of using BIM technology and cost management tools at home and abroad. Research also point to new research opportunities and to identify research issues in this area. Exploitation of BIM technology has a different level in worldeide countries. The overall state of implementation in Slovakia is in its early stages. Comparing with other countries is quite interesting. Exploitation of BIM technology is better in countries as United Kingdom, New Zealand and so on. In these countries, the issue of BIM is mainly incorporated into legislation. This is often the reason why the use of BIM is to such an extent. The inter-country survey has also highlighted the specific software options and practices that are being used. The survey opened up other issues that need to be addressed.

Acknowledgement

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SPECIFIC FEATURES OF NATURAL GAS SUPPLY IN SOME EU COUNTRIES

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Keywords: seasonal effects, monthly domestic production, monthly net import, monthly gas consumption, gas balance
Abstract: The monthly gas balances for Hungary and for five other EU Member States were investigated between 2008 and 2017. For the analysis, monthly data available in the EUROSTAT database [nrg_103m] was used to ensure that the data for the different countries are comparable. Time charts for each country were used with a three-figure set of figures. For each country studied, the first member of the graph group shows the seasonal changes in the use of natural gas for the period 2008-2017. The timely changes in the liability side of domestic production and net imports, the domestic user's use of the user's page, are time-consuming, and the storage activity is illustrated by the quantities extracted from the containers. In the second graph of the group of figures, the monthly opening and closing stock of the gas stored in the given country / country group, as well as the change in the net import, are shown. Finally, the third figure illustrates the change in inventory variability for the gas year from 1 April to 31 March. The authors conducted the analysis for the EU-27, Hungary, Austria, France, Italy and the Czech Republic, and their findings were formulated for the listed countries.

1 Introduction, seasonality, opportunities and risks of natural gas supply

Natural gas, which is more favorable to GHG emissions than other fossil fuels, is transmitted to end-users via a pipeline power supply network. In most countries, the bulk of this energy carrier is used by the household, industry and service sectors. In many countries there is also a significant amount of natural gas used for electricity generation. Over the past two decades, new perspectives have also emerged, from which the transport sector has to be highlighted.

The European Union's firm commitment to reducing GHG emissions has led to the gradual shift from coal to natural gas in the field of electricity production from the statistical data. There is also evidence of improved efficiency in the use of natural gas in the household and service sectors [1,2].

From the point of view of natural gas use it is favorable that natural gas can be stored in exhausted oil and gas fields and in underground water reservoirs (aquifers). In some European countries, it is possible to store in artificial salt domes. In the past decades, commercial and strategic (supply security) storages have been set up in several European countries. The authors present, through the example of some countries, the seasonal changes in the supply of natural gas and the role of storages in this process [3-7].

The changes in the storage strategy of the EU-27 and other EU Member States during the period 2008-2017 were further examined. EUROSTAT monthly data was used to present and analyze monthly gas balances.

2 Natural gas supply in the EU

Figure 1 shows the change in the EU's monthly gas consumption from January 2008 to March 2018. From the gross domestic consumption curve it can be seen that the lowest monthly consumption of natural gas was 857.9 PJ, the largest being 2666.3 PJ. The figure also shows that the gas demand exceeded 2600 PJ in 2010 (January and December) and 2017 in the coldest winter months. The biggest gas demand was in January 2010 with 2666.3 PJ. The lowest gas demand was registered in August, August, with a value of 857.9 PJ.

Figure 2 shows that net imports between 2009 and 2012 were changing in wider, but after 2013, they were changing in a narrower range. From 2015, a moderate growth trend can be seen. During the period 2008-2013, the monthly opening and closing stock of stored gas changed in the range of 1000-2500 PJ but no relevant growing or decreasing trend occurred. After 2013, there was a clear and significant increase in the amount of gas stored, especially in the maximum values. From a financial point of view, the inventory of winter-end (remaining) stocks is interesting, as this is a "frozen cost" for the traders.

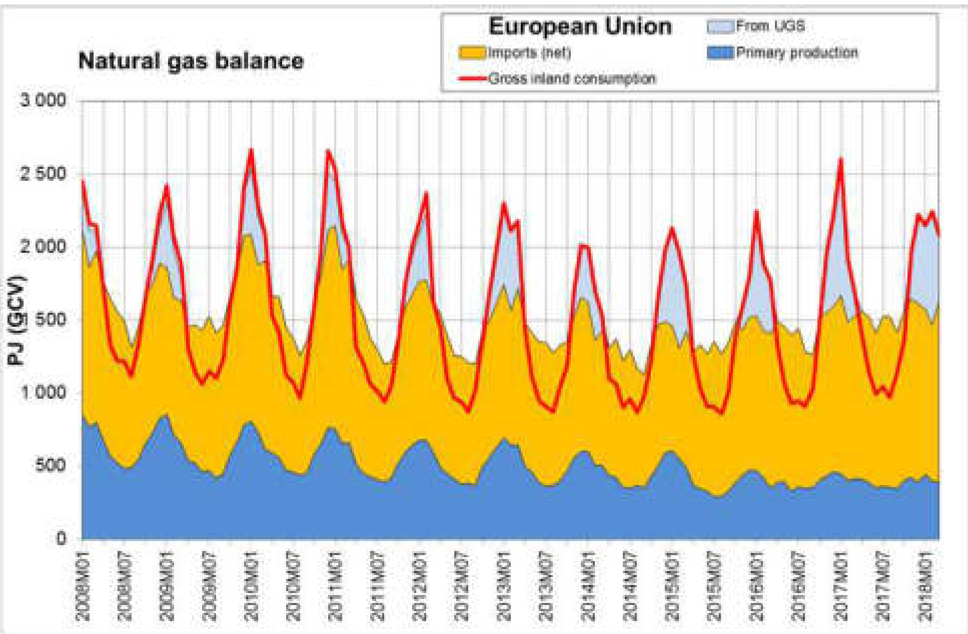

Figure 1 Monthly gas balance for the European Union between 2008-2017 (EUROSTAT, 2018)

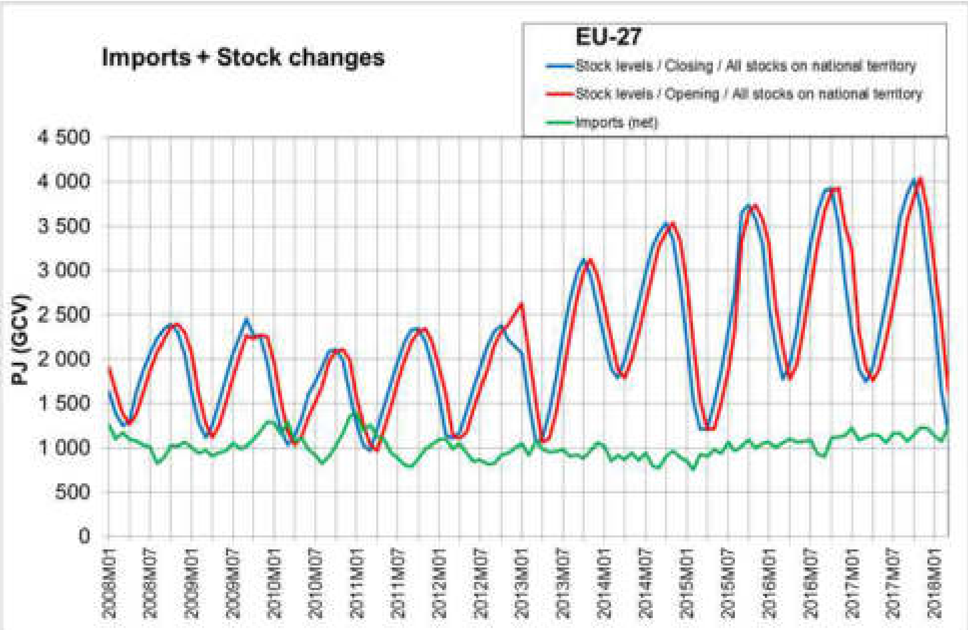

Figure 2 Net import and changes in storage reserve for the European Union between 2008-2017 (EUROSTAT, 2018)

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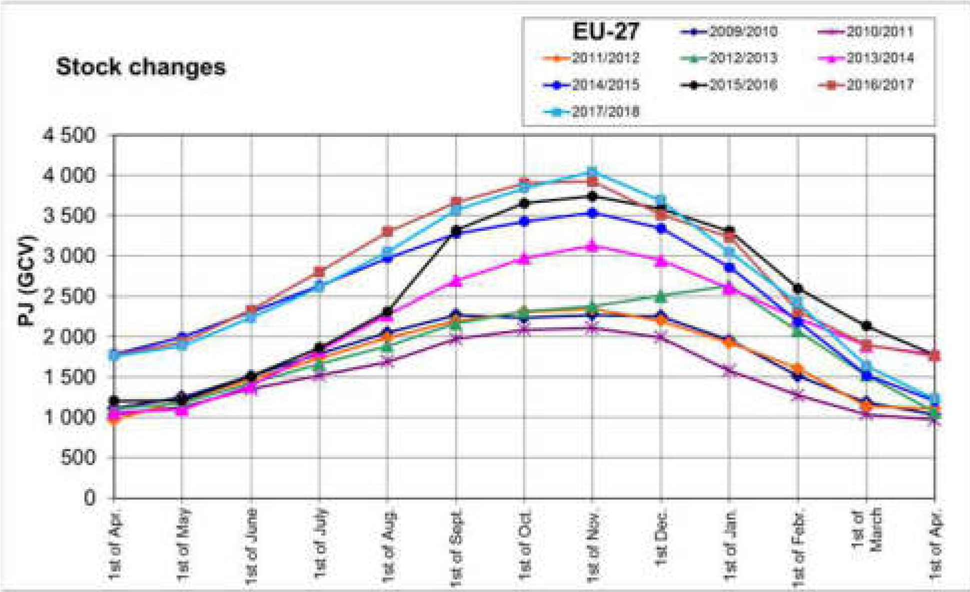

Figure 3 Changes in storage reserve for the European Union between 2008-2017 (EUROSTAT, 2018)

In Figure 3, the inventory changes of gas storage are shown for the “storage” gas year, i.e. from 1 April to 31 March of the following year. In various winters the amount of gas stored varied at different rates during the winter months. The maximums of curves fall at the end of October. Subsequently, the loss of storage stocks can be observed due to the higher gas demand in the cold winter months.

3 Natural gas supply in Hungary

Figure 4 shows Hungary's monthly gas consumption from January 2008 to March 2018. From the gross domestic consumption curve it can be seen that the lowest monthly gas consumption was 12.8 PJ in the month of August 2014 and the highest was 71.7 PJ in January 2008. It can be seen from the figure that in the years 2008-2011, winters with almost the same cold temperatures were successive years with significant natural gas consumption. The figure clearly shows that domestic gas production decreased slightly during the period under review, but its monthly / seasonal fluctuation was small. Monthly net import volumes have changed significantly, sometimes due to international conflicts that are independent of Hungary.

Figure 5 shows the changes in the net import and the underground storage inventory. During the period under review, there have been several years of considerable gas remaining underground storage after mild winters. Based on the figure, it is likely that, after the fall of 2013, the estimation of storage stocks reflects an increasing experience. The net monthly import rate changed in the 6.7 to 51.9 PJ interval.

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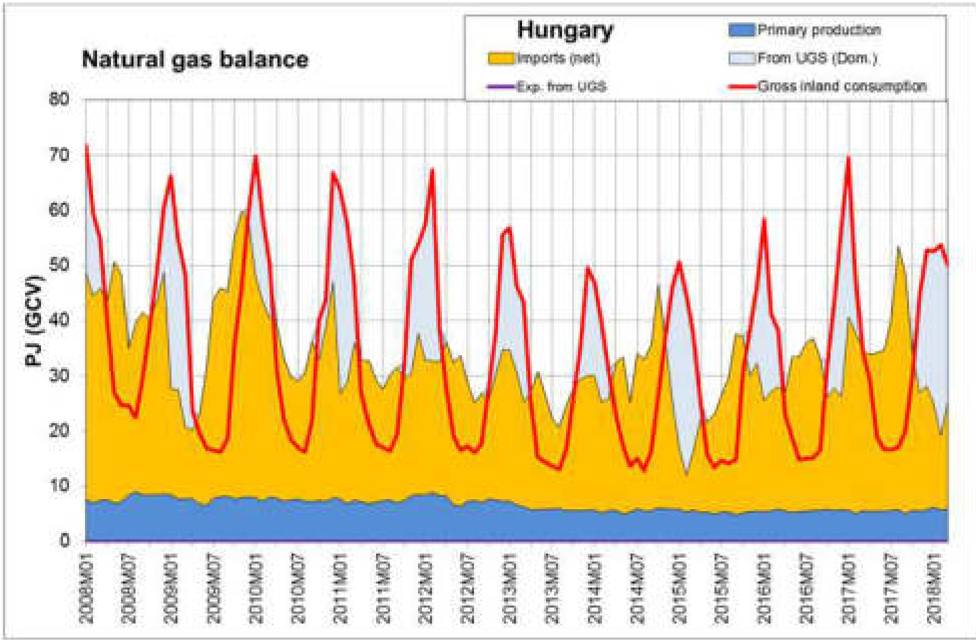

Figure 4 Monthly gas balance for Hungary between 2008-2017 (EUROSTAT, 2018)

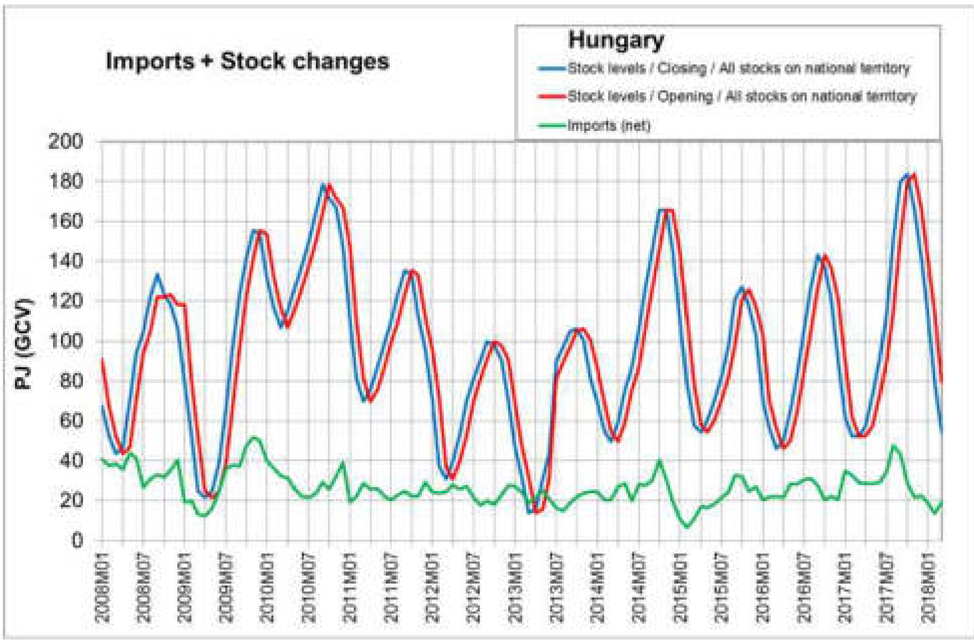

Figure 5 Net import and changes in storage reserve for Hungary between 2008-2017 (EUROSTAT, 2018)

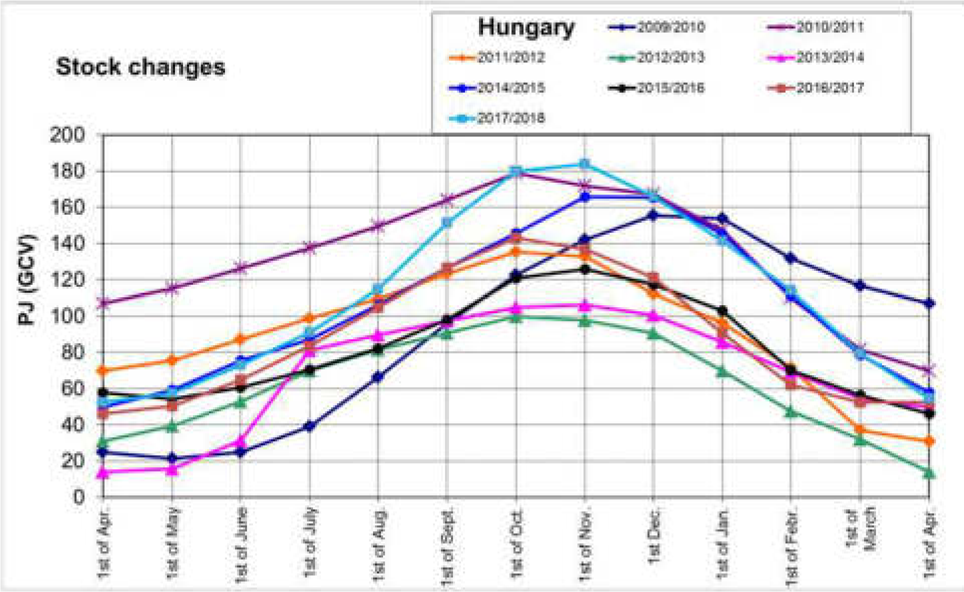

Figure 6 Changes in storage reserve for Hungary between 2008-2017 (EUROSTAT, 2018)

Figure 6 shows the storage reserve changes over the investigated gas years. In differently cold winters the quantities of gas stored were different, these changes are different in magnitude and in their pace in the winter months. It can be seen that the volume of natural gas that was stored began to decrease in early October in some years, and in other years it only occurred only in November or December.

4 Natural gas supply in Austria

Figure 7 shows Austria's monthly gas consumption from January 2008 to March 2018. From the gross domestic consumption curve it can be seen that the lowest monthly gas consumption was 12.8 PJ in July 2013 during the period under review and was highest with 59.5 PJ in January 2017. It can be seen from the figure that domestic production was continuous but its size was considerably lower than gas demand. The amount of natural gas stored in the underground storage facilities played an important role in satisfying gas demand.

The figure shows (violet line) the special shape of the "area" of the stored gas quantities and its passage to the negative range. This phenomenon is related to the activities of the Central European Gas Hub (CEGH). In Austria, the role of the underground gas storage facilities are considerably wider than in other countries in the region. In addition to satisfying the seasonal gas demand of the country, it is necessary to meet the temporary storage needs of CEGH. Figure 7 clearly shows that significant storage capacity is a prerequisite for the operation of an international gas trading center in Austria.

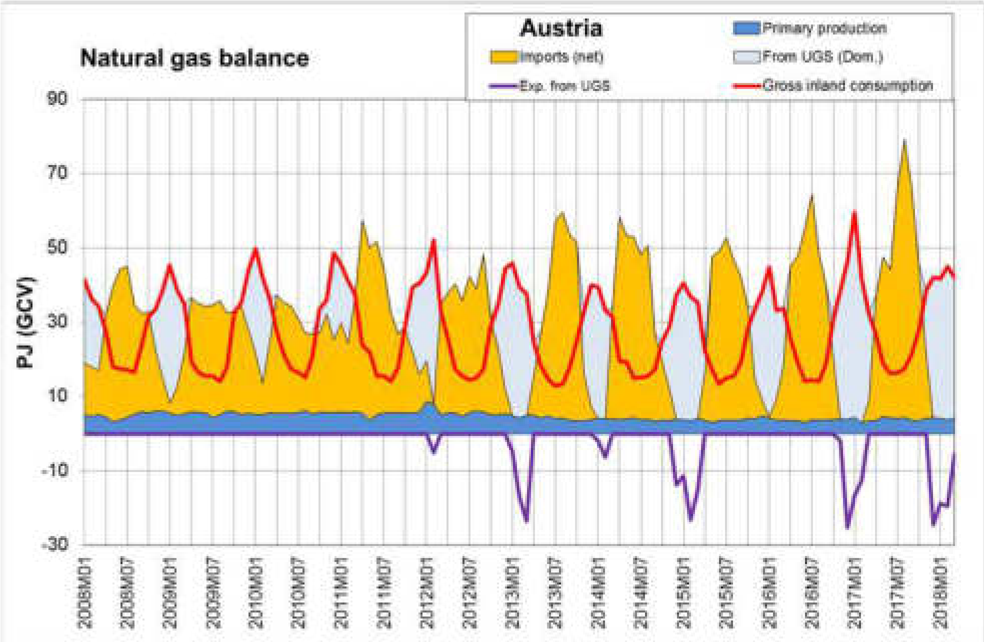

Figure 7 Monthly gas balance for Austria between 2008-2017 (EUROSTAT, 2018)

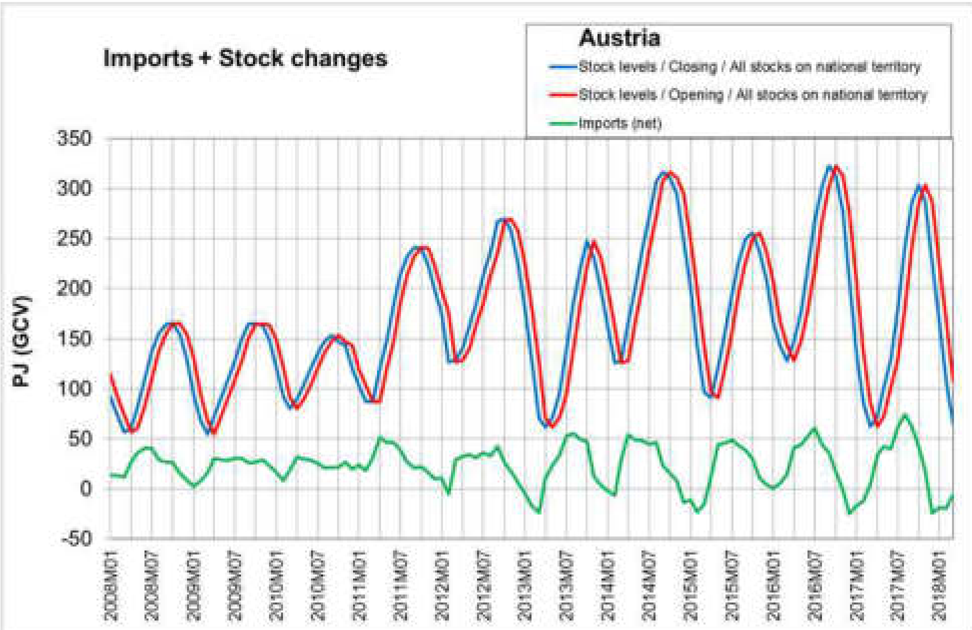

Figure 8 Net import and changes in storage reserve for Austria between 2008-2017 (EUROSTAT, 2018)

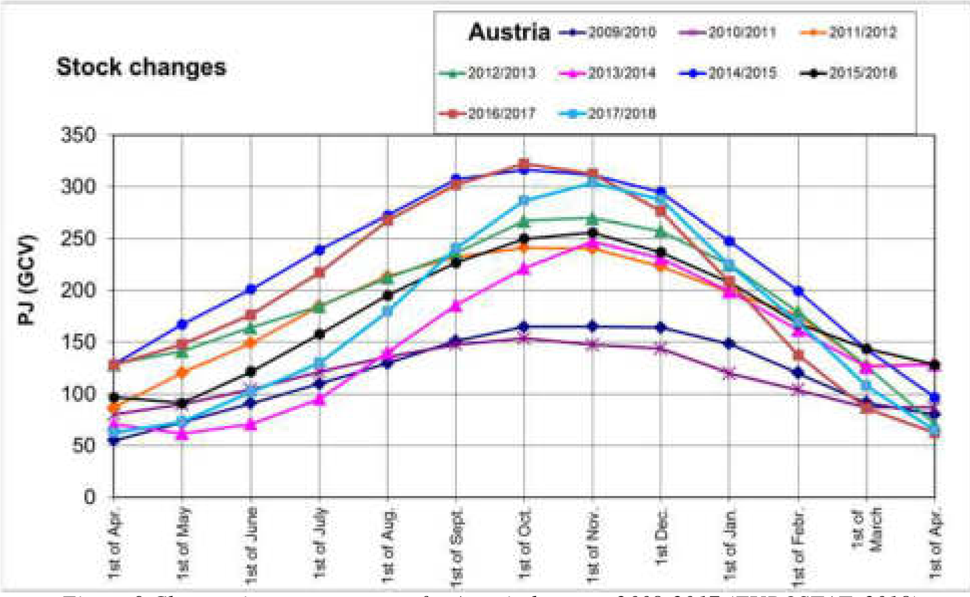

Figure 9 Changes in storage reserve for Austria between 2008-2017 (EUROSTAT, 2018)

Figure 8 shows the changes in the net import and the underground storage reserve. During the period under review, there have been several years since, after mild winters, significant amounts of gas has remained in underground storage, which is unfavorable to traders. The figure suggests that after the fall of 2013 gas traders have become more experienced, the estimation of storage reserves reflects an increasing experience. It can be seen in Figure 9 that the storage reserve in Austria during the period under consideration changed from 67 to 308 PJ. Net imports changed between the 74.8-25.1 PJ interval.

Figure 9 shows the storage inventory changes for the period under review for the years concerned. Comparing Figure 4 and 7 and Figure 6 and 9, it is clear that in Austria, significantly lower storage capacities and storage rates would be sufficient than those shown in Figure 9 to equalize seasonal fluctuations in Austria. However, it can also be seen that the changes of the gas loading and gas removal curves from the storages are very similar to the curves shown in Figure 3 and 6.

5 Natural gas supply in France

Figure 10 shows France's monthly gas consumption from January 2008 to March 2018. From the gross domestic consumption curve it can be seen that during the period under review, the lowest monthly gas consumption was 44.8 PJ in August 2012 and the highest was 307.9 PJ in January 2010. It can be seen from the figure that the size of gas production was not significant in the first half of the examined period, and decreased by 0.1-0.2 PJ after 2013. Because of the high import share and the minimal domestic production, underground storage plays an outstanding role in the country's natural gas supply. It should be noted that in France, most of the underground gas storage facilities are aquifer, i.e. deep gas storages in artificial water reservoirs.

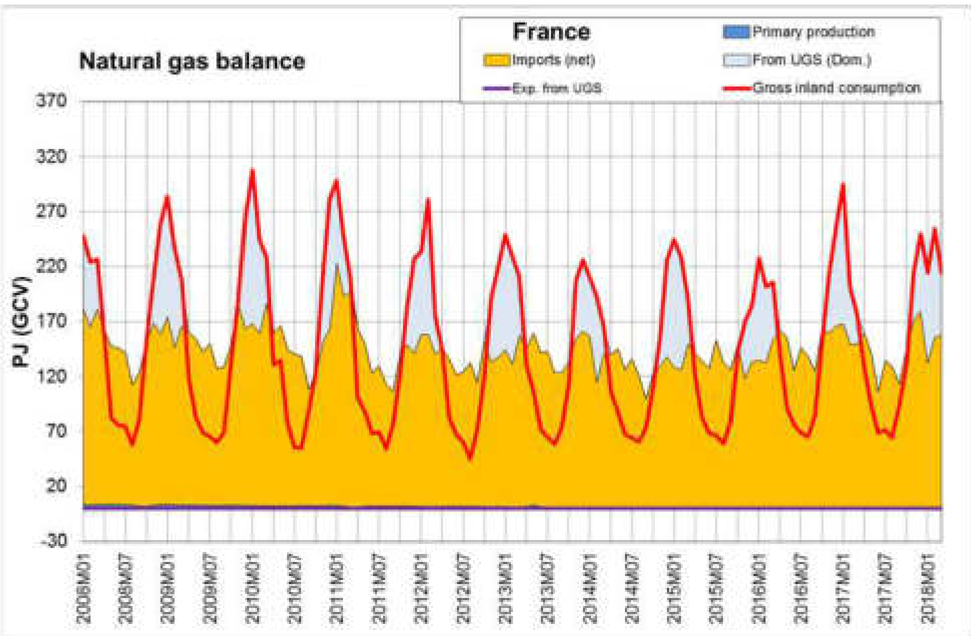

Figure 10 Monthly gas balance for France between 2008-2017 (EUROSTAT, 2018)

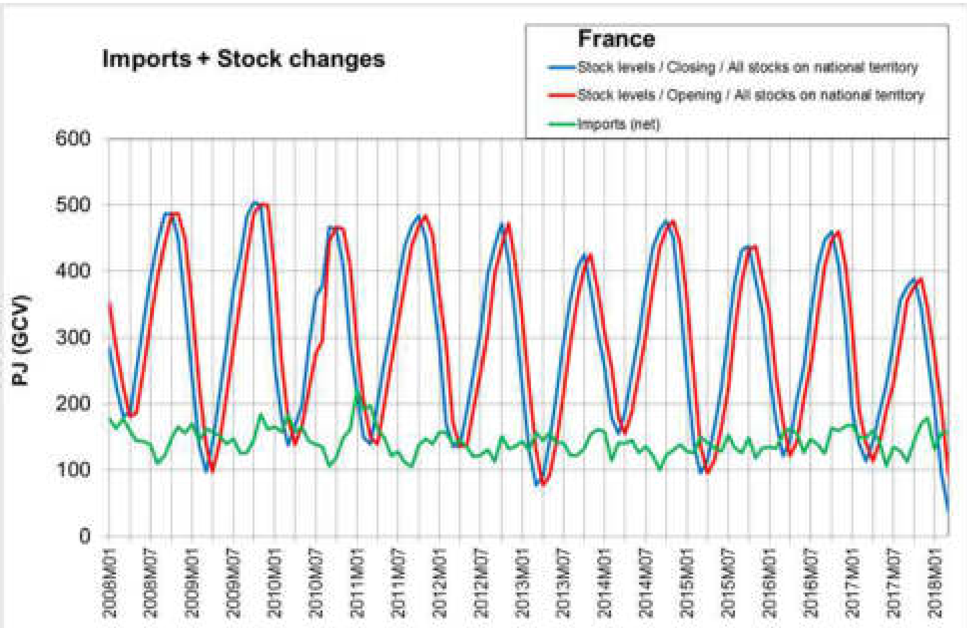

Figure 11 Net import and changes in storage reserve for France between 2008-2017 (EUROSTAT, 2018)

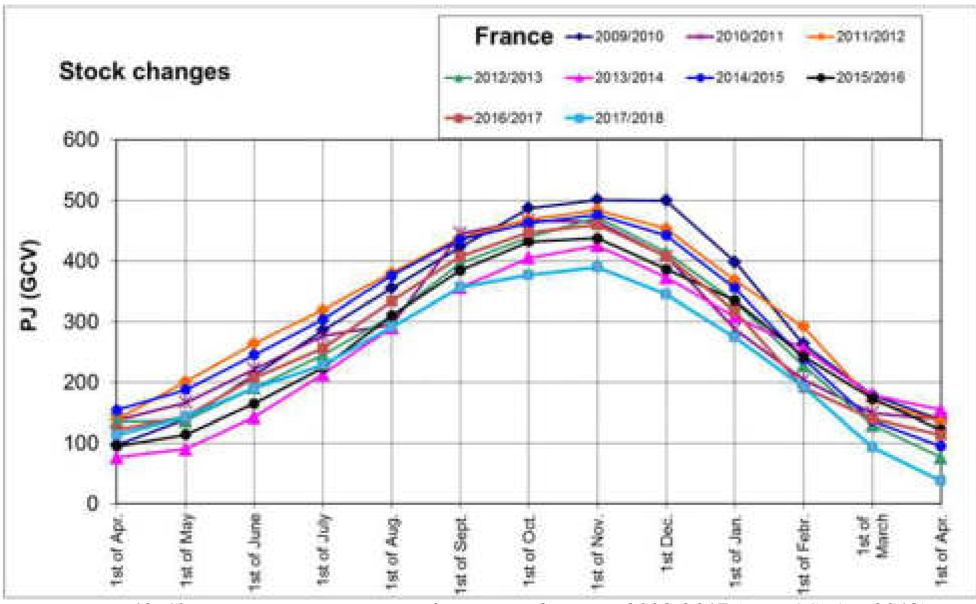

Figure 12 Changes in storage reserve for France between 2008-2017 (EUROSTAT, 2018)

Figure 11 shows that during the period under review, the storage opening volume ranged between 76.7 and 500.9 PJ. Net imports, in particular in the first half of the investigated time interval, changed in a broader range of 99.7 to 221.7 PJ intervals.

Figure 12 shows the storage reserve changes for each gas year. It is conspicuous that in some gas years the storage reserve has changed very narrowly.

6 Natural gas supply in Italy

Figure 13 shows Italy's monthly gas consumption from January 2008 to March 2018. The gross domestic consumption curve shows that the lowest monthly gas consumption was 115.6 PJ in the month of August 2014 and the highest was 424.5 PJ in January 2017. It can be seen from the figure that the volume of domestic production in the months of 2008 has exceeded 30 PJ, but gradually decreased over the following years. Due to the high share of imports, underground storage plays an important role in the country's natural gas supply.

Figures 13 and 14 show that, in the first half of the investigated period, the volume of imports in Italy was significantly higher in the coldest months of the year than in the rest of the year. However, this practice changed after 2014, and the monthly net import became much more even.

Figure 15 shows that during the period under review the storage opening volume in Italy was between 264.1 and 771.5 PJ. Net imports, in particular in the first half of the investigated period, changed in a broader range of 134.7-310.6 PJ.

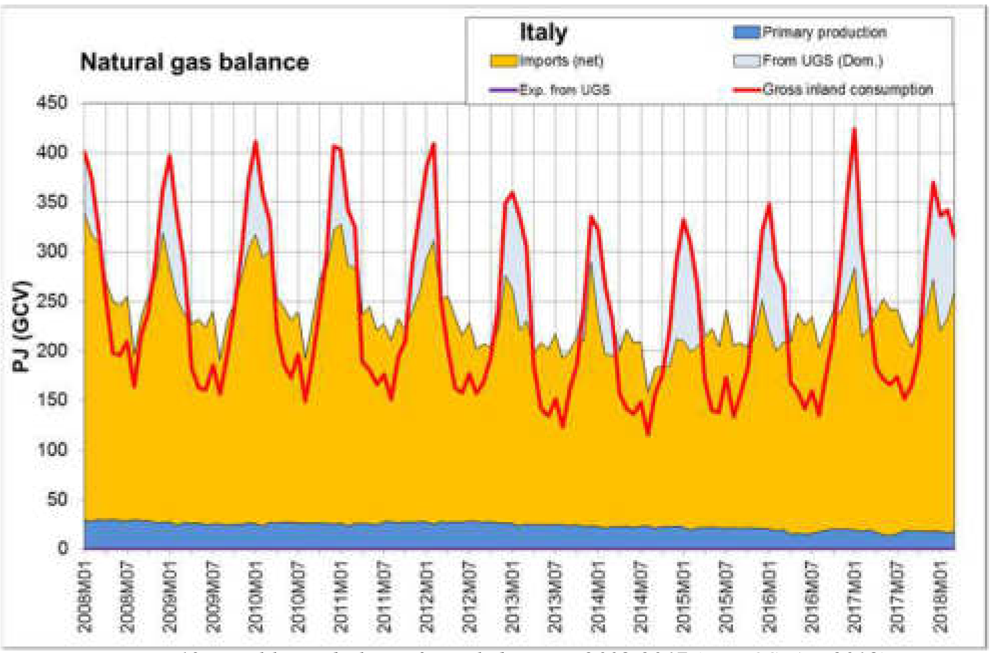

Figure 13 Monthly gas balance for Italy between 2008-2017 (EUROSTAT, 2018)

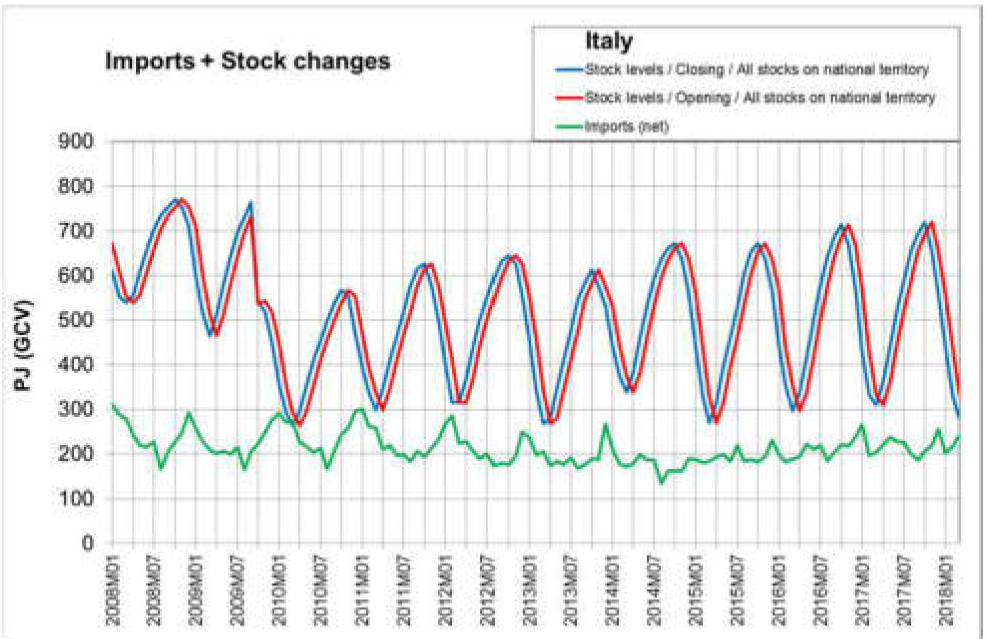

Figure 14 Net import and changes in storage reserve for Italy between 2008-2017 (EUROSTAT, 2018)

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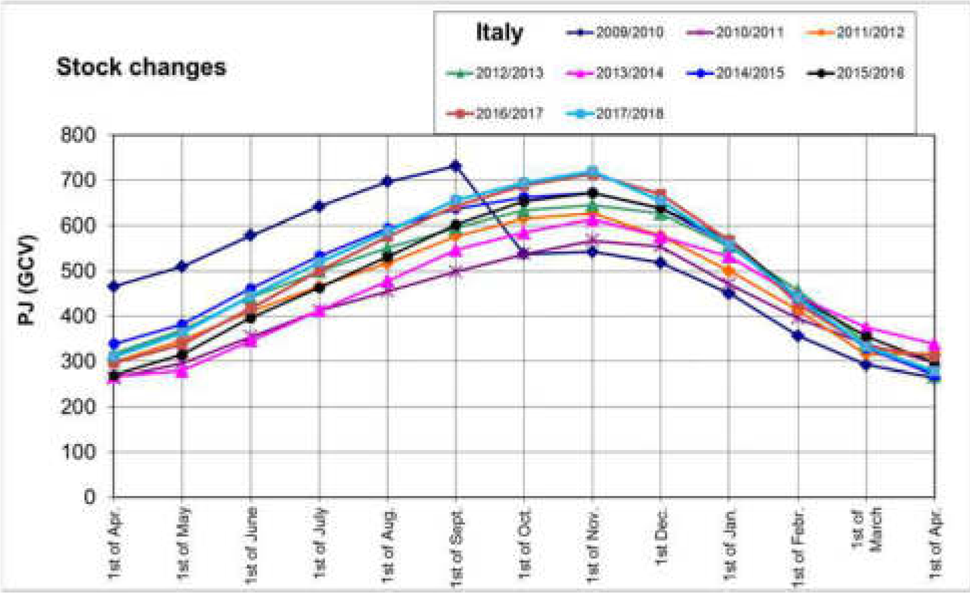

Figure 15 Changes in storage reserve for Italy between 2008-2017 (EUROSTAT, 2018)

7 Natural gas supply in the Czech Republic

Figure 16 shows the change in the monthly gas consumption in the Czech Republic from January 2008 to March 2018. From the gross domestic consumption curve it can be seen that the lowest monthly gas consumption in the period under review was 10.7 PJ in August 2009 and the highest was 56.8 PJ in January 2017. It can be seen from the figure that the size of the domestic production during the period under investigation did not change significantly between 1.0 and 0.4 PJ. Because of the high import share, underground storage has played a prominent role in the country's natural gas supply.

Figure 17 illustrates the changes in the size of the storage opening capacity between 118.4 and 10.5 PJ during the investigated period in the Czech Republic. The net import in varied a broader range between 44.3-10.2 PJ.

Figure 18 shows changes in gas storage capacity for the investigated period. It is conspicuous that in the individual gas years, the storage capacity in charged state varied at a reduced rate compared to the greater changes in months with almost depleted storages.

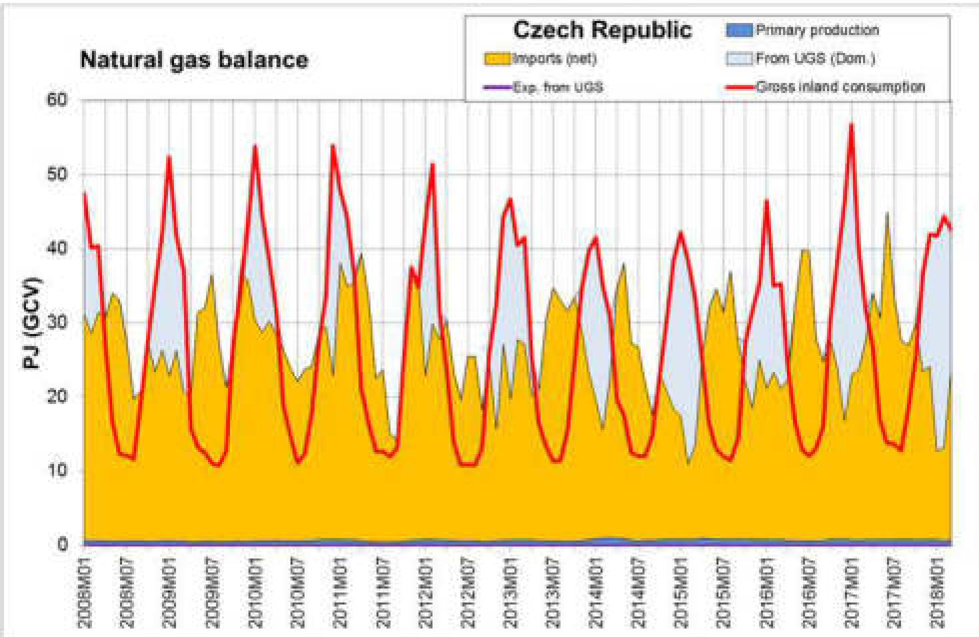

Figure 16 Monthly gas balance for the Czech Republic between 2008-2017 (EUROSTAT, 2018)

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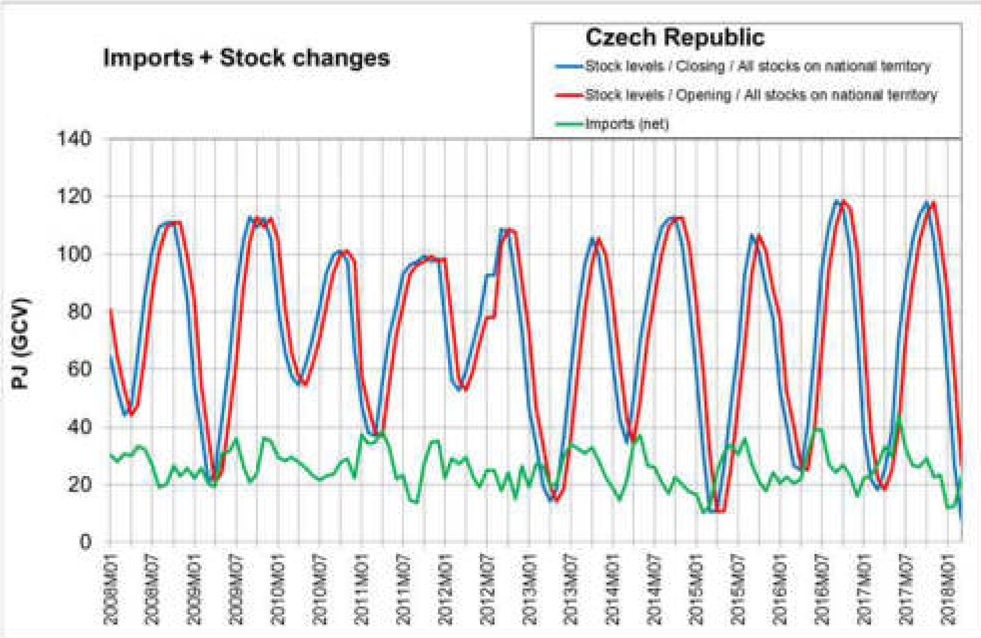

Figure 17 Net import and changes in storage reserve for the Czech Republic between 2008-2017 (EUROSTAT, 2018)

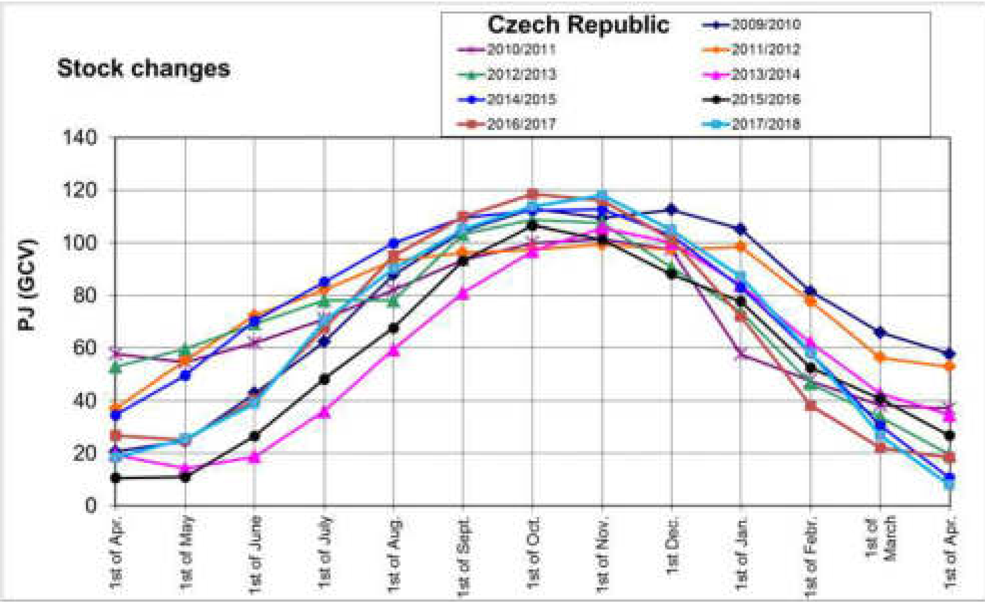

Figure 18 Changes in storage reserve for the Czech Republic between 2008-2017 (EUROSTAT, 2018)

8 Summary, conclusion

The magnitude and timely changes of natural gas consumption and storage reserves were investigated for five EU Member States for 2008-2018 on the basis of the EUROSTAT database monthly data.

From the comparative analysis the following conclusions can be drawn:

- For economic reasons during the investigated period in the countries studied, the monthly volumes of natural gas and imported gas fluctuated within narrow limits;
- Natural gas was used in some countries for end-use purposes and partly for electricity generation;
- It was shown in the surveyed countries that the monthly volume of natural gas consumption showed significant seasonal fluctuations due to the significant gap in heating usage;
- In order to compensate for seasonal fluctuations in the examined countries, natural gas transmission pipeline systems have different gas storage types with different capacities;
- The primary task of storages is the balancing of narrow gas sources and seasonally changing gas

requirements, which are necessary for supply and national security;

- Depleted oil and gas fields are used primarily for the storage of large quantities of natural gas, but aquifers, artificial salt caves are used seldomly;
- In European countries where regional gas trading centers operate, storage facilities are also essential for international gas commerce;
- It can be stated that the storage strategy has changed in the countries under investigation after 2010, that more and more import gas is stored in the summer months and, the volume of imports is reduced in the winter months when possible.

Analysis made to the EU member states surveyed highlighted ways to balance the resource side and the seasonal fluctuations of the user side under significantly different conditions.

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