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Water quality of the river Váh - Ružomberok (Slovakia), experience after 35-year water treatment

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Abstract: The chemistry of the water in the river Váh in the town of Ružomberok was examined. The waste water treatment plant (WWTP) in Ružomberok was built between 1977 and 1982. It was constructed as a joint WWTP for the town sewage as well as for the treatment of industrial waste water. Industrial waste water in the area is primarily a byproduct of the pulp and paper industry near the town, with Mondi SCP producing approximately 600 000 tons of paper and 100 000 tons of pulp for sale each year. Water testing was performed at six sampling sites; four upriver and two downriver from the treatment station. Samples were collected between September 2011 and January 2017. In total 429 samples were analysed, 70 -72 from each site. A significant increase in water pollution was discovered in Hrboltová, downriver from the WWTP. Significantly higher values of total dissolved solids (TDS), conductivity (COND), chemical oxygen demand (COD), salinity, sulphates and other chemical compounds were found downriver from the WWTP. We hypothesize this is a result of insufficient waste water treatment of water used in the production of pulp and paper. Mondi SCP, currently owns the treatment complex, and is also a large industrial contributer in the area. Our data confirms that following 30 years of operation, the plant may require restoration to effectively treat the water going forward.

Key words: water quality, river Váh, water pollution, paper and pulp production

Introduction

The river Váh in Slovakia is a major affluent of the Danube River. At 402 km long and with a basin of 19 696 km^2, Váh is the longest river in the Slovak republic. The region surrounding the river Váh is characterised by the presence of many industrial sources of pollution, including paper, pharmaceutical, automobile, metalworking, and wood and leather processing complexes. Similarly, the environment near the river is also affected by highly developed regional agriculture, well-developed industrial centres and settlements along its riverbanks (Halmo *et al.* 2009).

The pulp and paper industry is the 5th most energy consumptive industries in the world; accounting for more than 4% of worldwide industrial energy consumption. During the pulp and paper production process, a huge amount of waste is produced. It is estimated that about 500 million tons of paper will be produced per year in 2020. Three main raw materials are used in the pulp industry – non-wood fibres, and both hard and soft wood materials. Waste and wastewaters are a byproduct of both the pulp and bleaching processes. Additionally, 100 million kilograms of toxins are released bythis industry into the environment every year (Ince *et al.* 2011). Solid waste from different parts of the pulp and paper production process are listed in Table 1 below. Table 2 includes types of air pollutants from these production processes.

Mondi SCP, a.s. Ružomberok is a part of the Mondi Group, and produces uncoated fine paper. It is an integrated papermaking factory. Mondi is the biggest employer in this region and one of Slovakia's top 10 exporters of paper. Producing 8 million sheets per hour, each year it yields more than 620 000 tons and exports approximately 32 000 trucks of paper. In 2010, Mondi SCP won the PPI Award for Environmental strategy of year (Mondi SCP 2011). Mondi SCP also produces around 13 500 tons of dangerous waste per year which is stored on the Mondi SCP Ružomberok and WWTP Hrboltová grounds. Waste products are comprised of wood waste, dregs, sulphuric acid, hydrochloric acid, nitric acid and many others (Mondi 2016).

The basic function of wastewater treatment is to speed up the natural processes by which water is purified. There are two basic stages in the treatment of wastes - primary and secondary. During the primary stage, solids settle and can be removed from wastewater. The secondary stage uses biological processes to further purify wastewater. Sometimes these mechanical and biological processes are combined (EPA 1998), as is the case with the treatment plant in Hrboltová. 75% of wastewater treated is a byproduct of the pulp and paper mill, and 25% is municipal wastewater from Ružomberok and the surrounding area. The process of sedimentation of dregs, mechanical cleaning, oxygenating in the biological cisterns, filtrating in the filter bearing and the retaining of

Source	Waste Type	Waste Characteristic
Wastewater Treatment Plant	Sludge	Organic fraction consists of wood fibres and biosludge.
		Inorganic fraction consists of clay, calcium carbonate, and other materials
		20 - 60 % solid content
		ph = 7
Caustic Process	Dregs, muds	Green liquid dregs consisting of non – reactive metals and insoluble materials; lime mud
Power Boiler	Ash	Inorganic compounds
Paper Mill	Sludge	Colour waste and fiber clay including slowly biodegradable organics such as cellulose, wood fibers and lignin

Table 1. Solid wastes types and sources from pulp and paper mills (Ince *et al.* 2011).

Source	Major Pollutants
Pulping Process	VOCs (terpenes, alcohols, phenols, methanol, acetone, chloroform, methyl ethyl ketone. (MEK))
	Reduced sulphur compounds (TRS)
	Organo-chlorine compounds
Bleaching	VOCs (acetone, methylene chloride, chloroform, MEK, chloromethane, trichloroethane)
Wastewater Treatment Plant	VOCs (terpenes, alcohols, phenols, methanol, acetone, chloroform, MEK)
Power Boiler	SO2, Nox, fly ash, coarse particulates
Evaporator	Evaporator nonconcondensibles (TRS, volatile organic compounds: alcohols, terpenes, phenols)
Recovery Furnance	Fine particulates, TRS, SO2, Nox
Calcining (Lime Clin)	Fine and Coarse particulates

Table 2. Air pollutant sources and types from pulp and paper mills (Ince *et al.* 2011).

dregs, (which are then energetically evaluated) all takes place at this location (Sika 2013).

Primary treatment, which includes screening and grit removal, is carried out at the start of the treatment process. Primary treatment includes removing solid objects as well as oil and grease, which impede efficient wastewater treatment and are unwanted in the final biosolid product. Primary treatment also reduces the biochemical oxygen demand of the wastewater. Biochemical oxygen demand is a measure of the strength or pollution potential of the wastewater (Watercare 2016).

Secondary treatment is used to convert dissolved and suspended pollutants into a form that can be removed, producing a relatively highly treated effluent. Secondary treatment normally utilizes biological treatment processes followed by settling tanks and removes nearly 85% of the biochemical oxygen demand and TSS in wastewater (www.cctexas.com 2016). Biological wastewater treatmeant began in the early twentieth century and is now foundational to wastewater treatment worldwide. It involves confining naturally occurring bacteria at much higher concentrations in tanks. These bacteria, together with protozoa and other microbes, are collectively referred to as activated sludge. The bacteria remove small organic carbon molecules by consuming them. Then, the bacteria grow, and the wastewater is cleansed. The treated wastewater or effluent can then be discharged to receiving waters (Davies 2005).

While the concept is simple, control of the treatment process can be very complex, because of the large number of variables that can affect it. As a result of variation in the composition of bacterial flora in the treatment tanks, as well as in the sewage passing into the plant, the influent can show variations in chemical composition, flow rate, pH, and temperature. Many municipal plants also have to contend with surge flows of rainwater following storms. Plants treating industrial wastewater must cope with both chemicals that are slowly degradable, as well as more toxic chemicals that inhibit the function of the activated sludge bacteria. High concentrations of toxins can produce a toxic shock that kills the bacteria. When this happens the plant may pass untreated effluent directly into the environment, until the dead bacteria have been removed from the tanks and new bacterial 'seed' is introduced (Davies 2005).

New pollutants have placed additional stress on wastewater treatment systems. Today's pollutants, including heavy metals, chemical compounds, and toxic substances, are more difficult to remove from water. The increasing need to reuse water calls for better wastewater treatment. These challenges are being met through better methods of removing pollutants at treatment plants, or through prevention of pollution at the source. To return more usable water to receiving lakes and streams, new methods for removing pollutants are being developed. Advanced waste treatment techniques in use or

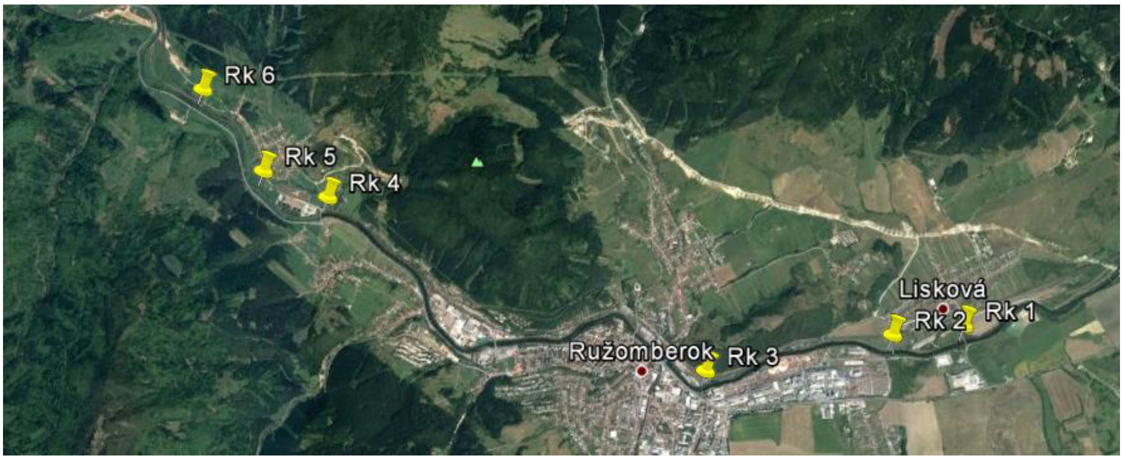

Fig. 1. Sampling sites of the water quality in area of Ružomberok (source: Google Earth 2016).

under development range from biological treatment capable of removing nitrogen and phosphorus to physical-chemical separation techniques such as carbon adsorption, distillation, filtration and reverse osmosis. These wastewater treatment processes, separately or in combination, can achieve almost any degree of pollution control needed. Waste effluents purified by such treatment, can be used for agricultural, industrial, or recreational purposes, or even drinking water (EPA 1998).

The main aim of this study was to describe and evaluate the impact of the paper industry and human activities on the water quality of the river Váh in Ružomberok and well as to evaluate the effectiveness of the WWTP built approximately 30 years ago.

Material and Methods

For a detailed description of location and methods see Gondová *et al.* (2017). Water samples were collected from 6 sampling sites (Fig. 1). The sampling sites were selected in suitable places where we expected different water quality in the Váh river. The first sampling site was at the upstream of Lisková village (RK1). The second sampling site downstream of Lisková village (RK2), where we expected to see the effect of the village on water quality. The third sampling site was in Ružomberok (RK3) near the pulp and paper factory (Mondi SCP), where we expected to see the effect of pulp production on water quality. The fourth sampling site was upstream of the WWTP Hrboltová (RK4). The fifth sampling site was situated downstream from the WWTP Hrboltová but before Hrboltová village (RK5), where we would expected to measure the effect of treatment on water quality. The sixth sampling site downstream of Hrboltová village (RK6). The following number of samples were collected at each site: RK1 – 72, RK2 – 72, RK 3 – 72, RK4 – 72, RK5 – 71 and RK6 – 70. Seasonal variations in water chemistry are presented in Gondová *et al.* (2017). This paper examines and discusses the special diffences between sampling sites.

At each sampling sites physical parameters were measured (in situ), including salinity, temperature of water, pH, conductivity (COND), total dissolved solids (TDS), and oxygen (O_2), using the Multi 3430 device (WTW GmbH, Weilheim, Germany). Water samples were collected for chemical analysis into sterilized 700 ml polyethylene bottles, conserved hermetically and transported to the laboratory. Colorimetry (YSI inc., Ohio, USA, YSI 9500) was used to detect the concentration of chlorides, sulphates, nitrates, phosphates, ammonia iones and $CaCO_3$. Chemical consumption of oxygen was measured by oxidative titration analysis using potassium permanganate. All analysis was completed 24 hours after sampling.

Data was analysed using Statistica 12 software (StatSoft, USA). Principal component analysis was performed to identify the potential relations betwen variables. This analysis uses an orthogonal transformation to convert a set of observations of possibly correlated variables into a set of values of linearly uncorrelated variables called principal components. The differences between categories of row data or among component scores were compared by variance analysis. Levels of TDS, Cl, S amount and conductivity and their difference among sampling sites are presented in Gondová *et al.* (2017).

Results

The results from each sampling site - RK1, RK2, RK3, RK4, RK5, RK6 are presented in Fig. 2 through 6. The quality of water was significantly lower below WWTP Hrboltová (sites RK5 and RK6).

At sites RK5 and RK6, the indicators were significantly different from sites R1 to RK4 (upstream from treatment). These sampling sites are located downstream from the WWTP and the treatment plant likely had a significant impact on water quality. The quality of the hydrological environment deteriorated as evidenced through increasing concentration of TDS, COND, COD, salinity, Cl, NaCl, S, SO_4^{2-} and PO_4^{2-}. This pollution is likely a result of insufficient purification of wastewater by the WWTP. At the RK5 sampling site, the TDS concentration increased from 281.3729 mg/l (RK4) to 364.3263 mg/l and at the RK6, the TDS concentration slightly decreased to 355.9359 mg/l. Conduc-

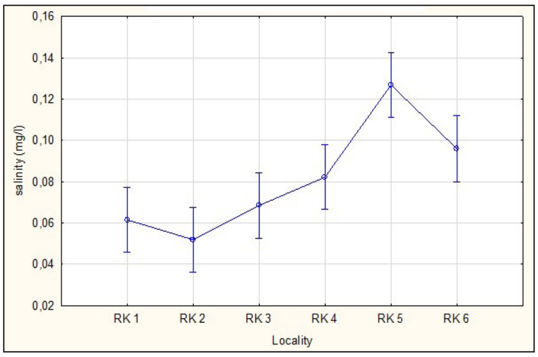

Fig. 2. Differences of measured values of salinity in the Váh river [One – way ANOVA F(5,423)=11.414, p=0.00000]. Salinity raised below WWTP hrboltová (RK5 and RK6).

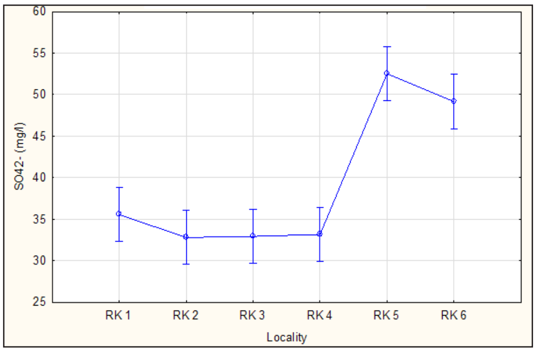

Fig. 3. Differences of measured values of sulphates (SO_4^{2-}) in Váh river [One – way ANOVA F(5,404)=29.467, p=0.0000]. The highest value of sulphate was measured in RK5 – 52.529 mg/l.

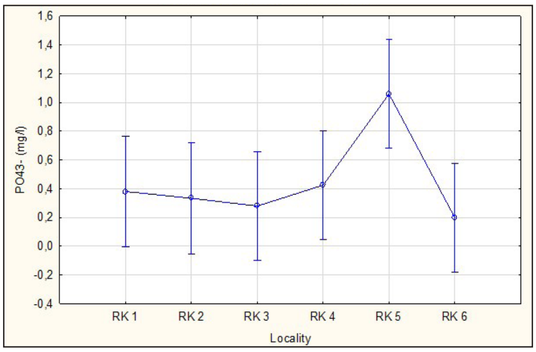

Fig. 4. Differences of measured values of phosphates (PO_4^{3-}) in the Váh river [One – way ANOVA F(5,332)=2.6130, p=0.02461]. Phosphate increased in RK5 – 1.059 mg/l and consequently the value dropped.

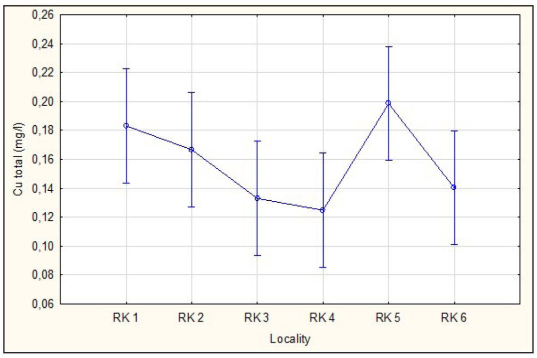

Fig. 5. Differences of measured values of copper (Cu) in Váh river [One – way ANOVA F(5,282)= 2.1898, p=0.05552]. Copper decreased from RK1 to RK4 and than rose in RK5 – 0.199 mg/l.

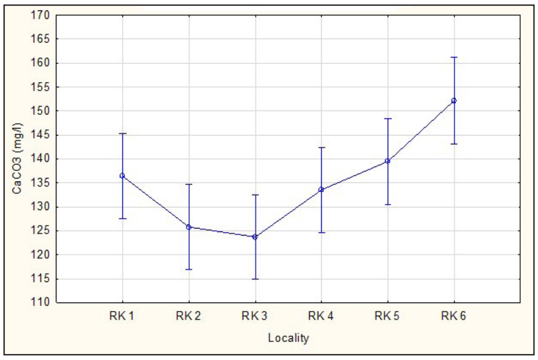

Fig. 6. Differences of measured values of calcium carbonates ($CaCO_3$)in Váh river [One – way ANOVA F(5,405)= 5.1378, p=0.00014]. The highest measured value was in RK6 – 152.164 mg/l.

tivity also increased from 274.4856 µs/cm (RK4) to 362.3083 µs/cm (RK5), COD from 3.977344 (RK4) to 5.778436 (RK5), salinity from 0.082250 mg/l (RK4) to 0.126887 mg/l (RK5), Cl from 6.632356 mg/l (RK4) to 8.364179 mg/l (RK5), NaCl from 11.23220 mg/l (RK4) to 13.97069 mg/l (RK5), S from 11.38333 mg/l (RK4) to 19.52542 mg/l (RK5), SO_4^{2-} from 33.17391 mg/l to 52.52941 mg/l (RK5) and PO_4^{2-} from 0.424386 mg/l (RK4) to 1.058596 mg/l (RK5). We found that highest concentration of pollution at the RK5 sampling place, downstream from the WWTP.

In Table 3 the principal component weights of the original measured variables are presented. The components (factors in the table) indicate mutual interactions among physico–chemical properties of water samples. Highlighted numbers in bold represent the link between the most significant variables for each factor. Seasonal effects on the first three component scores are presented in Figs. 7-9.

The most serious effect of waste in the waters of the Váh is synergy of increased sulphates, carbonates and conductivity. The increased pollution is most significantly evident during summer and autumn (Fig.7).

Increased ammonia content and water temperature in summer is a natural phenomenon and did not differ between localities. The lowest levels of ammonia in water were found during the cold weather in winter, and the highest levels were measured during warmer weather in the spring and summer (May to September) (Fig. 8).

The third factor describes the synergy among sulphates, salinity, TDS and conductivity. The effects were more visible at localities below the treatment plant and increased during summer and autumn (Fig. 9).

Discussion

Our results show a deterioration in water quality of the Váh river, downstream from the WWTP Hrboltová, (sampling sites RK5 and RK6). Indicators such as COD, TDS, COND, S, SO_4^{2-}, Cl, NaCl and PO_4^{3-} significantly increased at these sampling sites. This deterioration is largely a result of high quantities of wastewater from that paper industry, which the WWTP is responsible for treating, but urban ag-

Variables	Factor 1	Factor 2	Factor 3	Factor 4	Factor 5	Factor 6
Temperature (˚ C)	0.200	**0.426**	0.285	0.122	0.225	0.023
pH	-0.129	-0.118	-0.107	-0.081	0.203	-0.147
COND (S/cm)	**0.513**	-0.001	**0.610**	-0.300	-0.160	-0.045
TDS (mg/l)	0.391	0.015	**0.582**	-0.393	-0.192	-0.132
salinity (mg/l)	0.181	0.121	0.425	0.001	-0.255	0.176
O_2 (%)	0.187	0.184	0.211	0.214	0.079	0.382
$CaCO_3$ (mg/l)	**0.910**	-0.106	-0.368	0.027	0.019	-0,117
$CaCO_3$ (mmol/l)	**0.910**	-0.106	-0.368	0.027	0.019	-0.117
$CaCO_3$ (mg Ca^{2+}/l	**0.910**	-0.106	-0.368	0.027	0.019	-0.117
$CaCO_3$ (˚ dH)	**0.910**	-0.106	-0.368	0.027	0.019	-0.117
N (mg/l)	-0.115	0.097	-0.109	0.003	**0.554**	0.140
NO_3 (mg/l)	-0.029	0.357	-0.027	0.095	**0.676**	0.262
N-ammonia (mg/l)	-0.055	**0.895**	-0.234	-0.033	-0.187	-0.123
NH_3 (mg/l)	-0.015	**0.912**	-0.216	-0.016	-0.178	-0.100
NH_4 (mg/l)	0.020	**0.833**	-0.256	0.017	-0.188	-0.040
Cl (mg/l)	0.170	0.064	0.393	**0.820**	-0.018	-0.284
NaCl (mg/l)	0.156	0.001	0.392	**0.833**	0.013	-0.259
SO_4^{2}- (mg/l)	**0.563**	0.285	**0.478**	-0.281	0.316	0.032
S (mg/l)	**0.686**	0.203	**0.418**	-0.201	0.190	0.131
Cu total (mg/l)	-0.133	0.044	0.129	-0.161	-0.316	-0.234
PO_4^{3-} (mg/l)	0.189	-0.077	0.003	0.196	-0.424	**0.627**
P (mg/l)	0.401	0.002	-0.204	0.259	-0.239	**0.554**

Table 3. Principal component (Factors) vectors (loadings), which indicate mutual interaction of physico-chemical properties of water samples. (Factor coordinates of the variables, based on correlations).

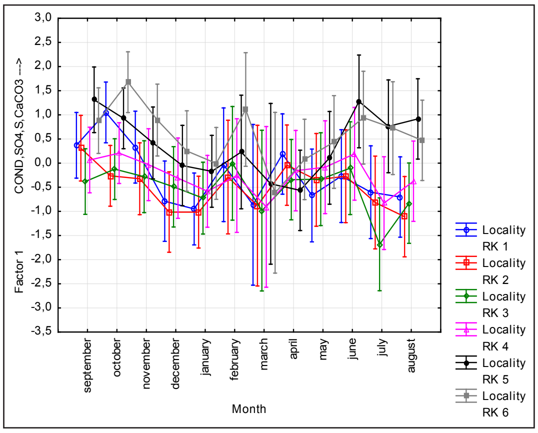

Fig. 7. Comparison of mean monthly COND, SO_4, S, $CaCO_3$ among localities by ANOVA [Locality (F=11.5, p=0.000) * Month (F=5.9, p=0.000) Interactivity: F (55, 210) =0.743, p=0.903].

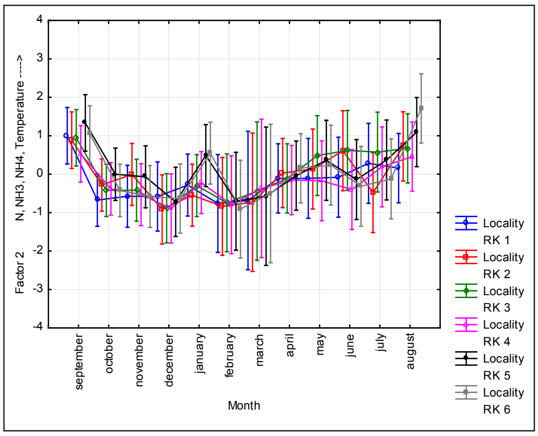

Fig. 8. Comparison of mean monthly N, NH_3, NH_4 among localities by ANOVA [Locality (F=0.801, p=0.550) * Month (F=8.997, p=0.000) Interactions: F (55,210) = 0.383, p=0.999].

glomeration is a secondary factor. To improve water quality, wastewater treatment must be improved.

Primary treatments such as more effective floatation are generally employed prior to biological purificationt. Some smaller enterprises use filtration as the only treatment of waste water, and according to Garcilaso (2001), the removal rate for dissolved solids (TDS) may be between 60-90%. Secondary treatment uses aerobic and anaerobic methods. Aerobic methods are used for sewage water, which contains a large amount of degradable organic substances.The process of separating activated sludge is the most widespread process in secondary treatment. Using activated sludge separation, BOD is reduced by 85-96% and COD by 75-90%. Tertiary treatment processes wastewater that still contains fine particles and nutrients, particularly phosphorus and nitrogen.

Numerous studies exist that discuss the potential improvement of wastewater treatment. Many

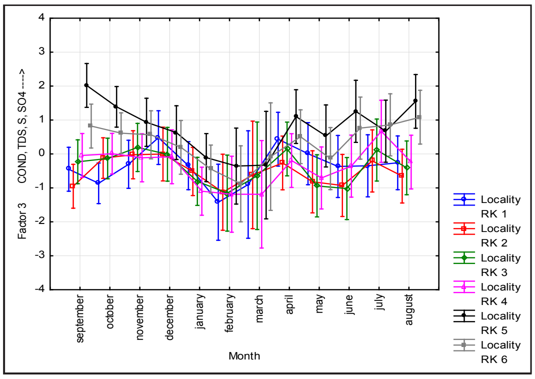

Fig. 9. Comparison of mean monthly COND, TDS, S, SO_4 among localities by ANOVA [Locality (F=15.31, p=0.000) * Month (F=4.97, p=0.000) Interactions: F (55, 210) =0.983, p= 0,515].

researchers are analysing increasing the effect of biological cleaning throught he use of active bilge. Wastewater from the paper industry contains a high volume of solid particles like bark chips, sawdust and other wood byproducts. Haarhoff and Bezuidenhout (1999) suggest the implementation of floatation propr to biological cleaning, which has had a significant impact on the effectiveness of biological cleaning in Great Britain and Sweden. Through this method, the reduction of insoluble substances in wastewater reached 90% (Wenta and Hartmen 2002).

In addition to active bilge, other cleaning methods are emerging, such as utilizing an aeration lagoon and a dosing sequential reactor. Anaerobic biological cleaning produces less biopass, a lower energy output, and requires a smaller physical footprint for the reactor building when compared to aerobic biological cleaning. The combination of both aerobic and anaerobic biological cleaning of wastewaters for the paper industry significantly reduces industrial sulphates (Chen *et al.* 2003). Other common wastewater treatment techniques include ultrafiltration, ozonation, adsorption and membrane technologies. The combination of coagulation, floatation and multimedial filtration could be the most effective tertiary cleaning method for wastewater produced by the pulp and paper industry.

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Orchid diversity of the Súľovské vrchy Mountains and the northern part of Strážovské vrchy Mountains

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Abstract. The aim of this study was the mapping and evaluation of diversity of the orchid family in the Súľovské vrchy Mountains in the northern part of the Strážovské vrchy Mountains and the foothill areas in the Považské podolie and Žilinská kotlina Basin. The field survey was conducted during the spring and summer season in 2017 and 2018. Coordinates of each individual or the population of all orchid species were recorded by a GPS device in various habitats including forests, rocks and scree, water springs, mires and fens, meadows and pastures, xerophilous and thermophilous grasslands, bushes, and ruderal habitats at altitudes ranging from 237 - 1213 m a.s.l. Historical records of orchids in the studied area were verified and changes in species composition were evaluated. The result is a list of 65 species, subspecies and hybrids with a brief description of the locality, a brief commentary, a map of the expansion in the Central European square map and a species diversity map.

Key words: *Orchidaceae*, *Epipactis*, threatened and rare species, Inner Western Carpathians, Slovakia

Introduction

In the past, the mountains on the western edge of the Western Inner Carpathians were called the Strážovská hornatina Mountains. It is currently divided into separate two geomorphological units; the Súľovské vrchy Mountains and the Strážovské vrchy Mountains. These two mountains are one of the best preserved areas in Slovakia and share a rugged terrain, carbonate rock subsoil, diverse habitats in a small area, and the highest species diversity of the *Orchidaceae* family in Central Europe. It is likely that this location represents the highest species representation of the genus *Epipactis* Zinn worldwide in one mountain range.

Orchids have attracted the attention of many botanists because of their diversity, variability, way of life, and the possibility of discovering a new species in Slovakia. These mountains were visited by significant botanists from Austria-Hungary and Czechoslovakia such as J. Ľ. Holuby, K. Brančík, J. Futák and K. Domin. P. Mereďa Jr. is another important botanist who contributed significantly to the description of new species of *Epipactis* in the Strážovské vrchy Mountains. Additionally, found in the the Súľovské skaly Geomorfological Subunit in the Súľovské vrchy Mountains is a veritable orchid paradise (Potůček and Kryška 1975). Although the area studied is visited by many professional and amateur botanists, many professional works have now become outdated because the landscape has changed greatly over the last several decades.

The Strážovské vrchy Mountains and Súľovské vrchy Mountains have been visited by botanists since the 17^{th} century (Mereďa 2006). The most visited areas were the parts of the mountain range most famous for natural beauty, such as the southern parts of the Strážovské vrchy Mountains, including Trenčianske Teplice, Vápeč, Strážov, Ostrá Malenica and Súľovské skaly. One of the first references to orchids occurs in the literary work Tri razy na Malenici by J. Ľ. Holuby (Holuby 1900). This prominent Austrian-Hungarian botanist found *Ophrys insectifera* L. (listed as *Ophrys myodes* Jacq.) on Ostrá Malenica hill for the first time in his life. His next discovery was an intergeneric hybrid ×*Cephalopactis hybrida* (Jáv.) Domin = *Cephalanthera damasonium* (Mill.) Druce × *Epipactis helleborine* (L.) (Feráková 1986).

K. Domin practiced botany in Strážov hill, Manínska tiesňava Gorge, Kostolecká tiesňava Gorge and Trenčianske Teplice town in 1882 and in 1954. J. M. Novacký practiced botany in Strážov hill (Mereďa 2006). J. Futák dealt with xeroterm vegetation. He wrote the Candidate dissertation on the topic of xerothermic vegetation of the southern part of Strážovské vrchy (Futák 1960). He also dealt with the ecology and expansion of several rare plant species in the northern part of Strážovské vrchy Mountains, including species found at Manínská tiesňava Gorge and Súľovské skaly NNR (Šípošová 2016) and in was there that he recorded *Gymnadenia odoratissima* (L.) Rich. (Futák 1932). He found *Ophrys holubyana András.* (listed as *Ophrys fuciflora*) on the southern slope of Stráne hill near Košecké Podhradie village (Štátna ochrana prírody SR 2014), where he proposed the protection of xerothermic habitats (Šipošová 2016).

E. Sajverová and K. Prach recorded eight taxa of orchids in the Strážovská dolina valley, such as *Ophrys holubyana* and *Ophrys insectifera* (Sajverová and Prach 1985).

O. Potůček and F. Kryška reported the results of their survey between 1968 – 1975 in the article Súľov – a paradise of orchids. There in is a list of 33 species and three hybrids, including two new species for Czechoslovakia (Potůček and Kryška 1975).

O. Potůček drew up a complete list of species, subspecies and hybrids in the Súľovské vrchy Mountains and Strážovské vrchy Mountains. This list includes 67 taxa (Potůček, unpublished data), and makes reference to *Gymnadenia conopsea* subsp. *montana* Bisse. The taxonomic value of the identifying signs of this taxon is unclear, so some botanists have not recognized it as a subspecies, but only as a synonym *Gymndadenia conopsea* (L.) R. Br. subsp. *conopsea*.

Materials for mapping of 19 taxa from the Súľov Area were made by Velísek, Potůček, Kryška, Projezdka, and Terková between 1968 – 1992 (Velísek 1992). An inventory and detailed mapping of the 31 orchid species in the Súľovské vrchy Mountains and the northern part of Strážovské vrchy Mountains was made by Velísek (1993).

P. Mereďa Jr. is most well know for his research on the genus *Epipactis* in Slovakia. He described three new species in the Strážovské vrchy Mountains: *Locus classicus* of *Epipactis futakii* Mereďa et Potůček, *E. pseudopuraputa* Mereďa and *E. komoricensis* Mereďa. *E. pseudopurpurata* is found near the town of Trenčianske Teplice (Mereďa 1996a), *E. komoricensis* near the town of Ilava (Mereďa 1996b), and *E. futakii* in the town of Trenčianska Teplá (Mereďa and Potůček 1998). Along with P. Mereďa Sr., he conducted field research between 1990-2002 and compiled a list of species and the extension of the genus *Epipactis* in the territory of the administration of the Strážovské vrchy Mountains PLA (Mereďa 2002). Hallonová collected a herbarium of *Epipactis helleborine* from Súľovske vrchy Mountains in 1978 that was later identified by P. Mereďa Jr. as *Epipactis greuteri* H. Baumann et Künkele, a new species of the Slovak flora (Mereďa 2000).

J. Smatanová has been the most active botanist in Strážovské vrchy Mountains in recent years. She works in the administration of the PLA Strážovské vrchy. The Slovak Republic State Nature Conservancy database includes more than 2500 orchid records from A. Beňová, K. Boublík, A. Cvachová, K. Devánová, V. Dinga, D. Dítě, A. Dobošová, D. Dúbravková, M. Duchoň, J. Dupkala, J. Ďuriga, E. Fajmonová, V. Feráková, T. Figura, J. Futák, M. Garčár, V. Grulich, L. Hrouda, M. Janišová, M. Kolník, J. Koštál, P. Koutecký, Lepší, P. Lustyk, B. Machciník, M. Maňák, P. Mereďa Jr., S. Mertanová, D. Michálková, J. Němec, P. Novosadová, K. Olšavská, D. Pavlišin, Z. Pčolová, M. Pepichová, E. Pietorová, M. Pirchala, Z. Plesková, K. Prach, I. Rizman, G. Runkovič, E. Sajverová, J. Smatanová, V. Stanová, I. Škodová, S. Štefániková, J. Štěpánek, B. Trávníček, S. Uhrin, Urbanová, S. Vačková, J. Vnuk, and M. Vyšinský (Štátna ochrana prírody SR 2014).

In 2003, in the Strážovské vrchy Mountains, the 42th floristic course was conducted (Mertanová and Smatanová 2006). The flora of grassland communities (meadows, pastures, orchards, wetlands) was studied in the central part of Strážovské vrchy Mountains during the 2002 growing season (Janišová *et al.* 2004).

In their bachelor thesis, Z. Dávidíková dealt with orchids found at Terasy za Baranovským krížom near the town of Rajec, where 14 species were recorded (Dávidíková 2006). Management measures have been proposed in this area to strengthen flora populations.

P. Novosadová elaborated on this topic in their master's thesis: Proposal of Part of the Management Plan for the National Nature Reserve of Súľovské skaly, where a detailed description of the area and a list of 23 orchid taxa from the year 2016 is provided (Novosadová 2017).

F. Lajcha conducted a direct floristic survey of the Orchid family in Opatovská dolina valley. 17 taxa were recorded (Lajcha 2008).

The aim of this study is to present a comprehensive picture of the total diversity and distribution of the family *Orchidaceae*. In the growing season of 2017 and 2018, historical findings were confirmed or disproved by direct field survey and new locations of some taxa were found. Qualitative and quantitative location information were identified, such as the number and density of flowering individuals, as well as the inclination and orientation of the terrain.

Material and Methods

Definition of the study area

The monitored area is situated in the northern half of Strážovské vrchy Mountains, Súľovské vrchy Mountains and their foothills in Žilinská kotlina Basin and Považské Podolie. In the west, the border of the study area is Hričov Canal, Nosice Reservoir and the Váh river. In the east it is bordered by the Rajčianka river. The southern boundary is not clearly defined. The border crosses the cadastral municipality of Trenčín, Soblahov, Mníchová Lehota, Trenčianske Teplice, Timoradza, Kšinná, Čavoj, Kľačno and Fačkov.

The issue of taxonomy

Some taxa names are not universally accepted between authors of the database or become accepted very slowly. For example, Reichenbach (1885) regarded *Listera* and *Neottia* as one genus. Szlachetko (1995) included the genus *Listera* in *Neottia*, but his classification was ignored or not accepted by many botanists. Recent studies suggest merging both genera on the basis of nuclear ribosomal DNA (Kotilínel *et al.* 2015). Epipactis pseudopurpurata, which is found in the Strážovské vrchy Mountains, was suggested as a separate species in the scientific work of Jakubska-Busse et al. (2012). However, this work is questioned by the author of the taxon. Issues with the acceptance of some species lie mainly in genera *Dactylorhiza* (Taraška 2014), *Ophrys, Epipactis* and *Gymnadenia*. Subspecies *Gymnadenia conopsea* subsp. *montana* can be found in older records, but today it is not accepted taxonomy in Slovakia and it is only referred to as a synonym for *Gymnadenia conopsea*. *Epipactis leptochila* s. l. includes a rather complicated group of *Epipactis*. Five taxa in this aggregate are known in Slovakia and several others are awaiting description.

Fig. 1. View from the top of Vápeč hill. Rudelar habitats in the basins, beech-oak forests on steep slopes, xerotherm meadows on extreme sites, this is a typical landscape in Strážovské vrchy Moutains (Photo: V. Ruček, 2018).

The most problematic is *Epipactis neglecta* Kümpel. Its taxonomy is perceived differently by different experts (Mereďa 2010; Delforge and Gévaudan 2002).

Material and Methods

Issues specific to orchid species, preferred habitats, flowering, and plant morphology had to be studied prior to the field survey. These topics were discussed with P. Mereďa Jr., M. Kolník and J. Smatanová.

In 2017 and 2018, a field survey was conducted on relevant habitats based on the time of flowering to the for each species. The growing season can not be precisely determined due to climatic influences, but in general, it runs from April to September. Both years throughout which the survey was conducted were very dry and as a result the growing season for some species was recorded as occurring up to two weeks earlier than average. Field surveys in 2017 were conducted for 30 days and field surveys in 2018 were conducted for 34 days.

Field survey took place in various habitats including forests, rocks and scree habitats, water springs, mires and fens, meadows and pastures, xerophilous and thermophilous grasslands, bushes, and ruderal habitats (Fig. 1) at altitudes ranging from 237 - 1213 m a.s.l., (from the lowest point to the highest in the studied area).

Species determination was carried out directly at the site by determining key and detailed description according to: Batoušek and Kežlínek (2012), Baumann *et al.* (2009), Vlčko *et al.* (2003), Potůček and Čačko (1996), Mereďa (1999) and the AHO-Bayern e.V. website (Gügel *et al.* 2010). Genus *Dactylorhiza* and *Epipactis* are known for their high variability and cross-over, and it was difficult to identify some individuals. Therefore, photos of important identifying signs were made and sent to J. Smatanová, P. Mereďa Jr., M. Kolník, J. Šmiták, P. Batoušek and other experts in genus *Dactylorhiza*. Geographical coordinates for WGS84 were determined by the Garmin Etrex 30 navigation device, and were used to identify the sample location of all orchid species. The list of coordinates is in the author's personal database. Rare findings are embedded in a comprehensive information and monitoring database (Štátna ochrana prírody SR 2013).

Floristic data are complemented by a list of earlier findings. The list of taxa is listed in alphabetical order by Latin name. Taxa nomenclature follows Vlčko *et al.* (2003) and Batoušek and Kežlínek (2012). Behind the name is the abbreviation for the category of threats according to Eliáš *et al.* (2015). Localities are arranged from south to north and their names are listed by OpenStreetMap contributors (2018). Locality and taxon information are listed in the following symbols and sequences and inspired by the works of Kolník (2004) and Kučera (2005):

1. Name of cadastral municipality,
2. Precise localization,
3. Altitude (meters above sea level),
4. Locality size: S – small (to 25 m^2), M – middle (to 2500 m^2), H – huge (over 2500 m^2) ,
5. Terrain slope: P – plain, S – slight, M – medium, G – great inclination,
6. Slope orientation: 0 – indefinite , W – west, N – north, E – east, S – south,
7. Number of individuals: O – to 10 pcs, T – tens, H – hundreds, or exact number is given,
8. Density: S– sparse, scattered over a larger area, G– group, several isolated groups, I– isolated, one group,
9. Date,
10. Quadrant Code of the Central European Network Mapping (Niklfeld 1971),
11. Name of the mapper,
12. Author's comments.

Individual information is separated by a symbol „ ; ", if information is missing, in its place it is " / ", records of findings are separated by a symbol „ * ". Other record authors are listed in a separate section. Behind the author's name is the cadastral area, year of record and number of base field and quadrant of Central European Network Mapping.

Results

Orchids occurring in the studied area

Anacamptis morio (L.) R. M. Bateman, Pridgeon et M. W. Chase, VU
Košecké Rovné; Dolná Stredná, cottage area; 460; S; S; N; O; I; 22.5.2015; 7076a; P. Smatanová; verified in 2017 and 2018, unconfirmed, reason: drought, population dynamics.

Records of occurrence according to K. Devánová: Rožňová Neporadza (2015, 7174d), Šípkov (2015, 7175b). According to M. Duchoň: Valaská Belá (2014, 7176a). According to Lajcha (2008): Dobrá (2007, 7074d). According to Čačko (1993): Horná Poruba (to 1993, 7075d), the data has not been confirmed. According to Grulich (Ambros 1996): Timoradza (1994, 7175c). According to J. Smatanová: Dubnica nad Váhom (2017, 7075c), Prejta (2017, 7075c), Veľké Košecké Podhradie (2010, 7076a). According to Velísek (1992): Hlboké nad Váhom (1969, 6777d), Dolný Hričov (1973, 6777d).

Cephalanthera damasonium (Mill.) Druce, NT
Omšenie; Havránovský potok NM, 1.5 km SW of Nad Vyhorencom hill; 425 ; S; P; 0; O; I; 2.8.2017; 7175a; V. Ruček; / * Omšenie; 1.3 km south of the Omšenie village; 440 ; S; S; N; 13; I; 12.6.2018; 7175a; V. Ruček; / * Dubnica nad Váhom; 0.6

km NE of the Ostrý vrch Top; 350 ; /; M; E; /; /; 2.7.2015; 7075c; V. Ruček; / * Malý Kolačín; 150 m SE of Markovica Top; 560; S; M; S; 1; I; 16.7.2018; 7075c; V. Ruček; / * Trenčianske Teplice; 0.1 km from the Trenčianske Teplice town, southern slope of Grófovec hill; 298; S; M; S; 1; I; 16.7.2018; 7075c; V. Ruček; / * Omšenie; surroundings of Omšenská Baba hill; 525 – 575; H; M; S; T; G; 12.6.2017; 7075c; V. Ruček; / * Dubnica nad Váhom; Dúbravy, 0.8 km SE of Kopanica Settlement; 534; S; M; S; 1; I; 12.7.2018; 7075c; V. Ruček; / * Dolná Poruba; 0.7 km east of Slopský vrch hill; /; S; S; E; 4; I; 20.7.2017; 7075d; V. Ruček; / * Dolná Poruba; southern slope of Homôlka; 774 – 817 ; M; S, M; S; T; S; 21.7.2018; 7075d; V. Ruček; the species was registered near the monument in the saddle * Horná Poruba; surroundings of Vápeč hill; 523 – 920 ; H; P, M; W, S; H; G; 18. – 19.5.2018; 7075d; V. Ruček; verified older records * Kopec; Kruhy Locality, 2 km SSW of Kopec village; 692 – 777; H; S,M; N; T; G; 19.5.2018; 7076c; V. Ruček; / * Košecké Rovné; surrounding of Horná Stredná Settlement; 550 – 660; H; /; /; 56; G; 5.6.2018; 7076c; V. Ruček; / * Dolná Stredná; surroundings of Dolná Stredná Settlement; 440 – 580; H; /; /; 56; G; 5.6.2018; 7076c; V. Ruček; / * Košecké Rovné; surrounding of Košecké Rovné village; 570 – 772; H; /; /; 32; G; 2.6.2017, 5.6.2017, 27.7.2017, 20.7.2018; 7076c; V. Ruček; 2.6.2017 a field survey was conducted by J. Smatanová * Zliechov; the area between Javorina and Strážov hill, north of Zliechov village; 730 – 820; H; /; /; 40; G; 30.5.2017.20.6.2017.5.6.2018; 7076b; V. Ruček; / * Pružina; Strážovská dolina valley, 0.3 km below Dolný Strážovký vodopád Waterfall; 705; S; M; N; 11; I; 4.6.2018; 7076b; V. Ruček; in 1999, D. Dítě recorded it about 1 km north * Pružina; Strážovská dolina valley, near Priedhorie village, from the mouth of the valley to the cross in Široké pažite Locality; 425 – 483; H; P; N; S; 25.5.2006, 21.6.2015; 7076b; S. Vačková, J. Smatanová, K. Devánová; verified 5.6.2015 and 4.6.2018 * Pružina; 1.1 km NE of Čierny vrch hill; 619; S; M; N; 1; I; 6.8.2017; 7077a; V. Ruček; / * Vrícko; 0.3 km south of Vrania skala Top; 905; S; S; S; O; I; 13.6.2017; 7078a; V. Ruček; / * Horná Poruba; Smrčkovci Settlement, 0.6 km SE of Vlčinec Top; 475; S; M; E; 1; I; 11.8.2018; 7075b; V. Ruček; in the vicinity of Vlčinec hill was recorded it by V. Feráková in 2003 (Mertanová and Smatanová 2006) * Košeca; Zábystrie Locality, 0.4 – 0.7 km SE-E of Norovica Top; 597 – 618; M; S; 0; O; S; 26.6.2017; 7075b; V. Ruček; 1.6 km SW-W it was recorded by K. Boublík 2003 * Kopec; Jedličná Locality, 1.3 km east of Tomanovci Settlement; 544; S; S; W; 1; I; 1.8.2017; 7075b; V. Ruček; / * Veľké Košecké Podhradie; the forest steppe of Stupičie hill, Pancier, Dúbravy, Veľká and Malá Šimerka Locality; 367 – 810; H; /; S; 357; G; 2017 – 2018; 7076a; V. Ruček; the species was recorded at 127 microlocalities, the first record is by E. Fajmonová from 1995, another by J. Smatanova from 2002 and 2017 * Mojtín; 1.3 – 2.7 km west of Mojtín village; 622-815; H; /; N-E; 59; G; 31.7.2017, 24.5.2018; 7076a; V. Ruček; older records are from M. Pirchala from 2013 and P. Smatanova from 2015 * Beluša; surroundings of the Ostrá Malenica, Rohatín and Rakytník hill; 495 – 725; H; /; /; /; G; 1989 – 2018; 6976c; V. Dinga, D. Michálková; J. Smatanová; V. Ruček; the oldest record is from V. Dinga in 1989 from Ostrá Malenica hill, D. Michálková worked in the vicinity of Rohatín hill on 26.5.2001, 6.6.2001 and 7.6.2001, J. Smatanová on 3.7.2003 and 30.5.2017, V. Ruček 30.5.2017 (3 ex.), 22.7.2017 (1 ex.) and 24.7.2018 (2 ex.) * Veľké Košecké Podhradie; Ladecká Pálkovica Locality, Dehetník hill ; 528; S; S; E; O; I; 26.6.2018; 6976c; V. Ruček; / * Hloža-Podhorie; Pod Tlstou horou Locality, above the Zahorú Cottage area, in front of a quarry; /; H; G; S; O; S; 3.8.2017; 6976c; V. Ruček; / * Beluša; Filipová kopanica, Za Vŕškami Locality; 421; S; S; 0; 2; I; 28.6.2017; 6976c; V. Ruček; / * Beluša; surroundings of Kamenica hill, near the Rybníky Cottage area; 332 – 420; H; /; /; 6; G; 29.7.2017; 6976a; V. Ruček; 4 microlocalities * Visolaje; Zákluky Locality, 0.6 – 1 km west of Jankov Háj Top; 293 – 321; S; M; W; 3; G; 29.7.2017; 6976a; V. Ruček; / * Tŕstie; Jaseňová, about 1 km south of Tupý hrádok Top; 460 – 537; H; /; /; O; S; 24.7.2018; 6976c – 6976d; V. Ruček; / * Podskalie; southern slope of Tŕnie hill; 516 – 622; H; M; S; 8; G; 16.8.2018; 6976d; V. Ruček; 3 micro-localities, the first record is by G. Runkovič from 1983 and 1989, A. Cvachová from 1988 (6976b), E. Fajmonová from 1995, J. Smatanová 2002, D. Pavlišin from 2014 (6976b) * Čelková Lehota; surroundings of the Čelková Lehota village; 499 – 629; H; /; /; 14; G; 30.6.2017, 1.7.2017, 2.7.2017; 6977c; V. Ruček; 5 micro-localities * Ďurďové; Richtárska, 1.6 – 2.3 km north of Ďurďové village; 548 – 678; H; /; S; O; S; 2.7.2017; 6977c; V. Ruček; the first record is by M. Vyšinský from 2014 from Stráne hill * Fačkov; Siguty, 1.1 km NE of Fačkov village, near the first class road; 515; S; M; W; 1 ; I; 13.6.2017; 6977d; V. Ruček; / * Vrchteplá; Pod Vieškami, 1.1 km north of Vrchteplá village; 574; M; S; S; 16; I; 26.5.2018; 6877c; V. Ruček; / * Považská Teplá; surroundings of Veľký Manín and Malý Manín hill; 458 – 506; H; M; S,W; 23; G; 11.6.2017, 23.7.2017, 9.8.2017, 12.8.2017, 14.7.2018; 6876d; V. Ruček; 10 micro-localities, verified Runkovič's record in Manínska tiesňava Gorge from 1990 * Súľov-Hradná; Súľovské skaly NNR; north of Súľov-Hradná village; 413 – 653; H; /; T; G; 26.5.2018, 27.7.2018; 6877b; V. Ruček; verified older findings * Súľov-Hradná; near Súľovský hrádok NM; 423; M; M; E; 6; S; 25.7.2017; 6877a; V. Ruček; verified older findings.

Records of occurrence according to J. Němec (2015, 7174d). According to the list of records from Floristic course Pruské 2003 (Mertanová and Smatanová 2006): Trenčianske Mitice (2003, 7174d), Slatinka nad Bebravou (2003, 7175b). According to Lajcha (2008): Dobrá (2007, 7074d). According to the list of records from Floristical contribution from the central part of Strážovské vrchy Mountains (Janišová *et al.* 2004): Valaská Belá (2002, 7176a). According to J. Smatanová: Kľačno (2017, 7077b), Prečín (2017, 6977a), Malé Lednice (2017, 6977b). According to E. Fajmonová: Čičmany (1995, 7076d), Horné Kočkovce (2003, 6876c), Domaniža (2004, 6977c). According to K. Rejšek: Dobrá (2004, 7074d). According to P. Jánsky (Eliáš Jr. 2010): Trenčín (2009, 7174a), Zamarovce (2009, 7074c). According to Rybáriková *et al.* (1994): Rajec (to 1994, 6977b). According to S. Štefaniková: Dolný Moštenec (2018, 6976b). Accoding to D. Pavlišin (2014, 6877c). According to V. Velísek: Vrchteplá (to 1993, 6877c). According to J. Limánek: Rajecké Teplice (2017, 6878c). According to A. Dobošová: Lietavská Svinná (2004, 6878a), Závodie (2014, 6778c). According to F. Kryška (Potůček, unpublished data): Paština Závada (1968 – 1980, 6777d).

Cephalanthera longifolia (L.) Fritsch, NT
Horná Poruba; 0.9 km S-SE of Hoľazne Top, near the forest path; 625; M; S; S; O; S; 19.5.2016; 7075d; V. Ruček; / * Horná Poruba; 1.2 km NW of Vápeč Top; 571; M; S; W; 7; S; 18.5.2018; 7075d; V.Ruček; /.

Records of occurrence according to the list of records from Floristic course Pruské 2003 (Mertanová and Smatanová 2006): Trenčianske Mitice (2003, 7174d), Trenčianske Teplice, Omšenie (2003, 7075c). According to D. Pavlišin: Košecké Rovné (2013, 7076c). According to M. Pirchala: Košecké Rovné (2014, 7076a) – unconfirmed, reason: drought, population dynamics, incorrect location description; Lietavská Svinná (2013, 6878a), Súľov-Hradná (2014, 6877a). According to M. Vyšinský: Veľké Košecké Podhradie (2013, 7076a), Fačkov (2014, 7077a), Domaniža (2014, 6977c), Kardošová Vieska (2014, 6977d), Považská Teplá (2014, 6877a). According to J. Smatanová: Tužina (2017, 7077d), Hloža-Podhorie (2017, 6976c). According to Vačková (1997): Pružina (1996, 6976d). According to E. Fajmonová: Počarová (1995, 6977a). According to Urbanová: Považská Teplá (1991, 6877c). According to J. Smatanová: Súľov-Hradná (2016, 6877a). According to P. Novosadová: Súľov-Hradná (2016, 6877b).

Cephalanthera rubra (L.) Rich., NT
Omšenie; Omšenská Baba NR; 570 – 660; H; /; S; 74; G; 12.6.2017; 7075c; V. Ruček; 17 micro-localities * Omšenie; Bartošovica Locality, 2.7 km NE of Omšenie village; 670 – 700; M; /; S; 3; S; 20.7.2017; 7075c; V. Ruček; 2 micro-localities * Dolná Poruba; 0.9 km east of Slopský vrch Top, near the red hiking trail; /; M; S; S; 3; S; 20.7.2017; 7075d; V. Ruček; / * Veľké Košecké Podhradie; Mlynovica valley, Stráne hill; 527-535; H; M; S; O; S; 26.6.2017; 7075b; V. Ruček; 3 micro-localities * Košeca; 0.7 km SE of Norovica Top; 618; M; S; W; 5; S; 26.6.2017; 7075b; V. Ruček; / * Košeca; under Močeková skala Rock, Húboč; 432; S; M; N; O; I; 26.6.2017; 7075b; V. Ruček; / * Veľké Košecké Podhradie; the forest steppe of Stupičie hill, Skalica hill, Pancier hill, Veľká and Malá Šimerka hill; 380 – 600; H; M-G; S; 73; S; 1.6., 15.6., 17.7., 31.7., 4.8.2017, 16.6.2018; 7076a; V. Ruček; 41 micro-localities, verified data by J. Smatanová from 2002 and 2006 * Mojtín; the slopes of Suchý vrch hill; 632 – 778; H; G; E; O; S; 31.7.2017, 19.7.2018; 7076a; V. Ruček; / * Košecké Rovné; Zanadlúčie Locality, 1.3 km north of Košecké Rovné, under of Gábrišské vrch Hills; 702 – 735; H; G; S; 5; S; 27.7.2017; 7076a; V. Ruček; 4 micro-localities * Zliechov; 1.8 km NE of Zliechov village, north of red hiking trail, Strážov NNR; 813 – 830; H; M; W; O; S; 5.6.2018; 7076b; V. Ruček; 4 micro-localities * Veľké Košecké Podhradie; Ladecká Pálkovica hill, 1.6 km SW of Dielec Top; 459 – 470; M; M; S; 7; S; 26.6.2018; 7076a; / * Hloža-Podhorie; under Tlstá hora hill, 0.9 km south of Prvé vráta Gorge; 516 – 526; H; S; 0; O; S; 3.8.2017; 6976c; V. Ruček; verified data by Ujházyová *et al.* (2007) * Pružina; Rečica valley; 479 – 535; H; P; 0; O; S; 2.7.2017; 7076b, 6977c; V. Ruček; / * Čelková Lehota; 1.6 km south of Čelková Lehota village; 529; S; S; N; 1; I; 30.6.2017; 6977c; V. Ruček; / * Briestenné; Briestenské skaly NM and close surroundings; 415 – 483; H; /; S; O; S; 30.6.2017; 6976d; V. Ruček; 3 micro-localities * Ďurďové; Podstránčie, 0.6 km east of church of Ďurďové village; 494; S; S; S; 6; I; 2.7.2017; 6977c; V. Ruček; / * Domaniža; Richtárska, 1 km south of Stráne hill; 598 – 639; H; G; S-E; 10; G; 1.7.2017; 6977c; V. Ruček; 3 micro-localities * Ďurďové; Kohilovec Top; 636; S; S; 0; 1; I; 16.8.2018; 6976d; V. Ruček; / * Domaniža; Blatnica valley; 390; M; S; 0; 2; S; 29.6.2018; 6977c; V. Ruček; / * Fačkov; Mackov laz Settlement; 525; S; S; S; 1; I; 13.6.2017; 6977d; V. Ruček; / * Považská Teplá; 0.9 km west of Veľký Manín Top; 554; S; S; W; 3; I; 9.8.2017; 6876d; V. Ruček; / * Vrchteplá; ridge of Havrania skala hill; 551 – 672; H; M; S; O; S; 23.7.2017; 6877c; V. Ruček; / * Súľov-Hradná; Súľovský hrádok NM, Súľovské skaly NNR, near the Súľov Castle; 478 – 548; H; /; /; O; S; 26.5., 19.6.2018; 6877a; V. Ruček; /.

Records of occurrence according to N. Hatala: Trenčianske Mitice (2015, 7174d). According to K. Devánová: Krásna Ves (2015, 7175c), Šípkov (2015, 7175b). According to I. Rizman: Omšenie (2015, 7175a), Valaská Belá (2015, 7176a), Jablonové (2015, 6877a), Porúbka (2015, 6878c). According to the list of records from Floristic course Pruské 2003 (Mertanová and Smatanová 2006): Slatinka nad Bebravou (2003, 7175a), Šípkov, Omšenie (2003, 7175b). According to the list of records from Floristical contribution from the central part of Strážovské vrchy Mountains (Janišová *et al.* 2004): Valaská Belá (2002, 7176a, 7176b), Čavoj (2002, 7176b). According to Lajcha (2008): Dobrá (2007, 7074d). According to A. Cvachová: Podskalie (1988, 6976b). According to J. Limánek: Fačkov (2017, 6977d), Šuja, Rajecká Lesná (2018, 6977b). According to E. Fajmonová: Domaniža (2004, 6977c). According to P. Smatanová: Prečín (2015, 6977a). According to J. Smatanová: Počarová (2006, 6977a), Záskalie (2007, 6877c), Súľov-Hradná (2002, 6877d). According to D. Pavlišin: Bodiná (2014, 6877c). According to M. Ujházyová and K. Ujházy (Ujházyová *et al.* 2007): Kostolec (2005, 6877c), Považská Teplá (2005, 6877c), Plevník-Drienové (2005, 6877a). According to Králik *et al.* (2006): Porúbka (to 2006, 6878c). According to Novosadová (2017): Súľov-Hradná, Paština Závada, Hrabové, Hlboké nad Váhom (2016, 6877b). According to Velísek (1993): Hrabové (to 1993, 6877b). According to F. Kryška (Potůček, unpublished data): Hričovské Podhradie (1968 – 1980, 6777d).

Corallorhiza trifida Châtel., LC
Omšenie; Omšenská Baba NR; 624; S; S; E; 1; I; 12.6.2017; 7075c; V. Ruček; / * Vrícko; 0.6km NEE of Vrania skala Top; 925; S; M; S; 1; I; 13.6.2017; 7078a; V. Ruček; /.

Records of occurrence according to the list of records from Floristic course Pruské 2003 (Mertanová and Smatanová 2006): Slatinka nad Bebravou (2003, 7175b), Omšenie (2003, 7175a). According to the list of records from Floristical contribution from the central part of Strážovské vrchy Mountains (Janišová *et al.* 2004): Valaská Belá (2002, 7176a).

According to Lukáš: Kubrica (1972, 7174b). According to Čačko (1993): Horná Poruba (to 1992, 7075d). According to E. Fajmonová: Zliechov, Pružina (1995, 7076b). According to J. Smatanová: Pružina (1999, 6976d). According to G. Runkovič: Podskalie (1989, 6976d). According to M. Ujházyová and K. Ujházy (Ujházyová *et al.* 2007): Kostolec (2005, 6877c). According to Urbanová: Považská Teplá (1991, 6877c). According to Novosadová (2017): Súľov-Hradná (2016, 6877a, 6877b), Hlboké nad Váhom (2016, 6877b). According to F. Kryška (Potůček, unpublished data): Hrabové (1968 – 1980, 6877b), Hričovské Podhradie (1968 – 1980, 6777d).

Cypripedium calceolus L., NT
Records of occurrence according to the list of records from Floristic course Pruské 2003 (Mertanová and Smatanová 2006): Zliechov (2003, 7076b) - The locality was verified in 30.5., 20.6.2017 and 5.6.2018. The occurrence has not been confirmed, reason: drought, population dynamics, shading of the micro-locality. M. Maňák found one sterile exemplar in 2018. According to B. Machciník: Kľačno (2017, 7077b).

Dactylorhiza fuchsii (Druce) Soó subsp. *fuchsii*, NT
Kopec; Kúty Locality, 1.6 km SSW of Kopec village; 580; M; S; N; 1; I; 19.5.2018; 7076c; V. Ruček; verified older data by E. Fajmonová and J. Smatanová, there is often crossing with D*actylorhiza majalis* * Súľov-Hradná; Čierny potok valley; 395; M; S; N; O; S; 13.6.2017; 6877a; V. Ruček, J. Smatanová; in the vicinity there is *Dactylorhiza fuchsii* subsp. *sooiana*.

Records of occurrence according to J. Smatanová: Horná Poruba (2001, 7075d). According to E. Fajmonová: Kopec (1990, 7076c), Pružina (1995, 7076b).

Dactylorhiza fuchsii subsp. *sooiana* (Borsos) Borsos, NT
Súľov-Hradná; Čierny potok valley; 390; S; S; N; 1; I; 13.6.2017; 6877a; V. Ruček; confirmation of an older finding by J. Smatanová.

Records of occurrence according to J. Smatanová: Kopec (2002, 7076c) – it was not confirmed in 19.5.2018, the reason may be crossing with other species of the genus *Dactylorhiza* and the extinction of parental taxons, incorrect determination in the past; Horná Poruba (2001, 7075d), Plevník-Drienové (2000, 6877a). According to A. Dobošová: Fačkov (2018, 6977d).

Dactylorhiza incarnata (L.) Soó subsp. *incarnata*, NT
Omšenie; Omšenská Baba NR, 0.8 km north of Omšenie village; 460; M; S; S; 23; S; 12.6.2017; 7075c; V. Ruček; / * Čičmany; Hanušová; 720; M; P; E; /; I; 9.6.2017; 7076d; J. Smatanová; in the population there are hybrids with *Dactylorhiza majalis*, so determination of some individuals is difficult * Hloža-Podhorie; 0.6 km north of Dielec Top; 362; M; P; 0; 3; S; 12.5.2018; 6976c; V. Ruček; there is Petasites succession * Prečín; 1 km west of Prečín village; 395; M; /; 0; T; /; 8.6.2017; 6977a; J. Smatanová; /.

Records of occurrence according to the list of records from Floristic course Pruské 2003 (Mertanová and Smatanová 2006): Valaská Belá (2003, 7076c), Zliechov (2003, 7076b), Čičmany (2003, 7077a). According to P. Mereďa Jr.: Prejta (1998, 7075a). According to J. Smatanová: Veľké Košecké Podhradie (2006, 7075b). The locality in Zliechov and Veľké Kosecké Podhradie was verified, the species there was not confirmed.

Dactylorhiza lapponica (Laest. ex Hartm.) Soó, NT
Records of occurrence according to A. Dobošová: Fačkov (2010, 7077b), Šuja, Rajecká Lesná (2010, 6977b). Localities require revision.

Dactylorhiza majalis (Rchb) Hunt & Summerh, NT
Kopec; Kúty Site, 1.6 km SSW of Kopec village; 580; M; S; N; T; I; 19.5.2018; 7076c; V. Ruček; verified older data by J. Smatanová and E. Fajmonová, there is often crossing with *Dactylorhiza fuchsii* subsp. *fuchsii* * Košecké Rovné; Strednianské lúky Meadows, between Dolná Stredná and Horná Stredná Settlements; 575; M; S; E; 12; I; 5.6.2017; 7076c; V. Ruček; / * Kopec; the mouth of Kopčianská dolina valley; 370; M; P; 0; 10; I; 19.5.2018; 7076a; V. Ruček; an individual of varying variability, it may be hybrids, verified older data by E. Fajmonová from 1995 * Zliechov; 0.3 – 1.8 km north of Zliechov village, Plešivé, under Javoriná and Strážov hill, Kopce; 655 – 770; H; /; S; H; G; 30.5.2017; 7076b; V. Ruček; 6 localities, verified older data by J. Smatanová from 2006, J. Košťál, J. Kochjarová, P. Koutecký from 2003 (Mertanová and Smatanová 2006) * Pružina; Priedhorie Settlement, the mouth of Strážovská dolina valley; 440; M; P; 0; T; I; 2016; 7076b; V. Ruček; verified older data by S. Vačková from 1996, there were reported *Dactylorhiza lapponica* and Dactylorhiza maculata subsp. *maculata*, but it is probably incorrectly determined. In the case of D. lapponica, it is probably D. majalis and in the case of *D. maculata* subsp. *maculata*, it is another taxon of the *Dactylorhiza maculata* agg. * Podskalie; Nivy, football field, 1.1 km NNE of Podskalie village; 370; M; P; 0; T; I; 24.5.2016; 6976b; V. Ruček; verified older data by J. Smatanová from 2000 * Sádočné; the stream near the quarry, south between the Čelková Lehota and Sádočné village; 431; M; P; 0; O; I; 1.7.2017; 6977c; V. Ruček; / * Domaniža; Hodoň valley; 405 – 420; H; P; W; T; G; 25.5.201, 14.5.2018; 6977c; V. Ruček, J. Smatanová; verified older data by V. Grulich from 2004, E. Fajmonová from 2004.

Records of occurrence according to K. Devánová: Soblahov (2014, 7174b). According to P. Jánsky: Petrová Lehota (2009, 7174b). According to S. Mertanová: Omšenie (2003, 7175a, 2009, 7075c). According to D. Galvánek: Dolná Poruba (2003, 7175b, 7075d). According to the list of records from Floristical contribution from the central part of the Strážovské vrchy Mountains (Janišová *et al.* 2004): Valaská Belá (2002, 7176b), Čavoj (2002, 7176b). According to the list of records from Floristic course Pruské 2003 (Mertanová and Smatanová 2006): Valaská Belá (2003, 7176a), Čičmany (2003, 7077a, 7077d), Beluša (2003, 6976c). According to J. Smatanová: Horná Poruba (2010, 7075d), Fačkov (2004, 7077b), Pružina (1999, 7076b), Podskalie (2011, 6976d), Prečín (2006, 6977a), Milochov (2005, 6876c), Veľká Čierna (2001, 6977a), Súľov-Hradná (1999, 6877d; 2002, 6877a), Kostolec (1999, 6877c), Záskalie (2007, 6877c), Plevník-Drienové (1999, 6877a). According to E. Fajmonová: Veľké Košecké Podhradie (1995, 7076a), Počarová (2004, 6977a), Horné Kočkovce (2003, 6876c). According to J. Ďuriga: Tužina (1998, 7077a). According to Vačková (1997): Pružina (1996, 6976d). According to P. Smatanová: Malé Lednice (2015, 6977a). According to Rybáriková *et al.* (1994): Rajec (to 1994, 6977b). According to Cvachová et al (1990): Súľov-Hradná (1990, 6877b). According to Velísek (1992; 1993): Jablonové (1992, 6877a), Lietavská Svinná (1993, 6878c). According to F. Kryška (Velísek 1992): Hričovské Podhradie, Paština Závada (1973, 6777d). According to A. Dobošová: Rajecká Lesná, Šuja (2018, 6977b), Fačkov (2008, 6977d).

Dactylorhiza sambucina (L.) Soó, NT
Records of occurrence according to K. Devánová: Omšenie (2002, 7175a). According to K. Devánová: Dolná Poruba (2015, 7075d). According to Čačko (1993): Horná Poruba (to 1993, 7075d). According to J. Smatanová: Dubnica nad Váhom (2017, 7075c), Prejta (2017, 7075c), Košecké Rovné (2015, 7076c) - verified data in 2017 and 2018, not verified, reason: dry and visit outside the growing season, Čičmany (2006, 7076b), Prečín (2006, 6977a), Súľov-Hradná (1999, 6877d), Fačkov (2004, 7077b). According to J. Němec: Zliechov (2015, 7076d). According to M. Duchoň: Čičmany (2015, 7077c). According to G. Runkovič: Podskalie (1989, 6976d), Kostolec (to 1990, 6877c). According to B. Veselský (Velísek 1992): Hričovské Podhradie (1973, 6777d). According to F. Kryška (Potůček, unpublished data): Súľov-Hradná (1968 – 1980, 6877b).

Dactylorhiza viridis (L.) A. M. Bateman, A. M. Pridgeon & M. Chase, NT
Veľké Košecké Podhradie; 2.8 – 3.5 km SW of Suchý vrch Top, Michalová, Klokočová, Dúbravy Forest steppe; 410 – 615; H; M; S; 10; G; 1.6.2017; 7076a; V. Ruček; 2 micro-localities * Hričovské Podhradie; near a hiking trail to the Hričov Castle; /; H; S; 0; 2; /; 2018; 6777d; Milan Jánoš; there is over 8 years, in 2017 there were 8 exemplars.

Records of occurrence according to the list of records from Floristic course Pruské 2003 (Mertanová and Smatanová 2006): Čičmany (2003, 7076b). According to J. Smatanová: Pružina (1999, 6976d), Kostolec (1999, 6877c). According to E. Fajmonová: Vrchteplá (1995, 6877c). According to Cvachová et al (1990): Súľov-Hradná (1990, 6877b). According to F. Kryška (Potůček, unpublished data): Hrabové (1968 – 1980, 6877a, 6777c), Paština Závada (1968 – 1980, 6777d).

Dactylorhiza ×aschersoniana (Hausskn.) Borsos et Soó (*D. incarnata* subsp. *incarata* × *D. majalis*)
Čičmany; Hanušová; 720; M; P; E; /; I; 9.6.2017; 7076d; J. Smatanová; in the population there are *Dactylorhiza majalis* and *D. incarnata* subsp. *incarnata*, so determination of some individuals is difficult.

Dactylorhiza ×braunii (Halácsy) Borsos et Soó (*D. fuchsii* subsp. *fuchsii* × *D. majalis*)
Kopec; Kúty, 1.6 km SSW of Kopec village; 580; M; S; N; 19; I; 19.5.2018; 7076c; V. Ruček; verified older data by E. Fajmonová from 1995 and by J. Smatanová, in this locality is also *Dactylorhiza majalis* (23 ex.) and *Dactylorhiza fuchsii* subsp. *fuchsii* (1 ex.). The mouth of Kopčianská dolina valley (7076a), 4 km north of Kúty Locality are individuals similar to *Dactylorhiza ×braunii*, but for unambiguous determination it is necessary to investigate it in other growing seasons.

Epipactis albensis Nováková et Rydlo, NT
Dubnica nad Váhom; 1.2 km east of Holaňovec Top; 467; M; S; W; 15; I; 12.7.2018; 7075c; P. Mereďa Jr. and P. Mereďa Sr., V. Ruček, J. Smatanová, Z. Pčolová; / * Veľké Košecké Podhradie; Podhradský potok Stream, 1.1 km west of Veľké Košecké Podhradie village; 313; S; P; 0; 1; I; 3.8.2018; 7075b; V. Ruček; / * Košeca; behind the canal, 1.1 km NW of Košeca village; 235; M; P; 0; 14; G; 22.7.2018; 6975c; V. Ruček; / * Ladce; 1.4 km west of Ladce village, near Váh river; 248; S; P; 0; 7; I; 22.7.2018; 6975d; V. Ruček; / * Hloža-Podhorie; near Slatinský potok Stream and the quarry in Beluša; 335; S; P; 0; 1; I; 2018; 6976c; V. Ruček; / * Beluša; Belušské Slatiny, near Slatinský potok Stream, 0.7 km west of Hradište Top; 338; S; P; 0; 6; I; 5.8.2018; 6976c; V. Ruček; / * Streženice; 0.4 km south of football field in Streženice village; 257; S; P; 0; 1; I; 1.8.2018; 6975b; V. Ruček and M. Kolník; / * Udiča; at the junction to Upohlav village; 290; S; P; 0; 1; I; 1.8.2018; 6876a; V. Ruček and M. Kolník; / * Jablonové; near the guarry and Hradnianka Stream, 0.6 km SE of Jablonové village; 340; S; P; 0; 2; I; 27.7.2018; 6877a; V. Ruček; / * Mikšová; in close proximity to Hričovský kanál Canal near Beňov Settlement; 300; M; P; 0; T; S; 1.8.2018; 6876b; V. Ruček and M. Kolník; / * Veľká Bytča; in close proximity to Pšurnovický potok Stream, 1.3 km west of Bytča town; 307; S; G; 0; 1; I; 1.8.2018; 6777c; V. Ruček and M. Kolník; / * Porúbka; near Slnečné skaly Camp on the right bank of Rajčianka river; 410; S; P; 0; 1; I; 25.8.2018; 6878c; V. Ruček; /.

Records of occurrence according to Kolník and Kučera (2002): Kubrica (2000, 7174b). According to M. Kolník: Súľov-Hradná (to 2002, 6877a).

Epipactis atrorubens (Hoffm.) Besser, LC
Omšenie; Lánce NNM; 440; M; S; N; 9; G; 12.6.2017; 7175a; V. Ruček; / * Omšenie; Omšenská Baba NR; 460 – 600; H; M; S; O; G; 12.6.2017; 7075c; V. Ruček; / * Dolná Poruba; south slope of Homôlka hill; 832; S; G; S; 2; I; 21.7.2018; 7075d; V. Ruček; / * Horná Poruba; south slopes of Vápeč hill; 680 – 850; H; G; S; T; S; 29.6.2017, 13.7. and 18.7.2018; 7075d; J. Smatanová, V. Ruček; / * Malé Košecké Podhradie; 1 km NE of Sokol Top; 407; S; M; 0; 1; I; 19.6.2018; 7075b; V. Ruček; / * Veľké Košecké Podhradie; south oak slopes of Stráne hill between Mlynovica and Norovica valley; /; S; G; S; O; I; 26.6.2017; 7075b; V. Ruček; / * Veľké Košecké Podhradie; forest steppe southern slope of Stupičie, Pancier, Skalica, Veľká Šimerka, Malá Šimerka, Klokočová hill; 420 – 705; H; G; S; H; S; 2017 and 2018; 7076a; V. Ruček, J. Smatanová; 56 micro-localities, the color variation lusus lutescens was recorded in the eastern part of the locality * Košecké Rovné; valley below Gábrišské vrchy hill, 0.1 – 0.3 km NE of Košecké Rovné village; 533; M; M; S; O; S; 27.7. and 4.8. 2017; 7076a; V. Ruček; / * Zliechov; 1.3 km SSW of Strážov Top; 773; S; M; S; 1; I; 24.6.2018; 7076d; V. Ruček; / * Zliechov; 1.3

km NW of Strážov Top; 840; S; G; S; 1; I; 20.6.2017; 7076b; V. Ruček; / * Pružina; Strážovská dolina and Rečiča valley; 430 – 480; H; S; 0; 16; S; 20.6., 2.7., 6.8.2017, 4.6.2018; 7076b; V. Ruček; 3 micro-localities * Trstie; Jasenová valley, 1.4 km SEE of Dievča Top; 511; S; G; S; 1; I; 24.7.2018; 6976c; V. Ruček; / * Briestenné; Briestenské skaly NM; /; H; M; S; 25; S; 30.6.2017; 6976d; V. Ruček; / * Čelková Lehota; 0.8 km SW of Čelková Lehota village; /; M; M; E; 5; I; 1.7.2017; 6977c; V. Ruček; / * Sádočné; Jaseňová valley, along the stream, 0.8 – 1.1 km SW of Sádočné village; 444 – 455; H; P; 0; 15; G; 1.7.2017; 6977c; V. Ruček; 4 micro-localities * Domaniža; Hodoň and Blatnica valley; 391 – 426; H; /; 0; T; S; 29.6.2018; 6977c; V. Ruček; / * Fačkov; Siguty, 1.1 km NE of Fačkov village; 575; S; M; S; 1; I; 13.6.2017; 6977d; V. Ruček; / * Ďurďové; rocks in Richtárska Locality and 0.9 km north of Ďurďové village; 472 – 704; H; G; S; O; G; 2.7.2017; 6977c; V. Ruček; 4 micro-localities * Prečín; Protected Area Svarkovica; 385; M; M; S; O; S; 29.6.2018; 6977a; V. Ruček; / * Jasenové; the valley between Dubová and Žibrid hill, along the stream; 510; H; /; 0; 8; G; 16.7.2017; 6877d; V. Ruček; 2 micro-localities * Vrchteplá; southern ridge of Vodičná hill; 693; S; M; S; 1; I; 23.7.2017; 6877c; V. Ruček; / * Porúbka; Slnečné skaly NR; 410; S; G; E; 1; I; 4.9.2018; 6878c; V. Ruček; / * Súľov-Hradná; Súľovske skaly NNR and Čierny potok valley, 0.3 – 0.8 km east of Súľov village; 360 – 407; H; G; W, S; O; G; 26.5., 19.6.2018; 6877a; V. Ruček; 2 micro-localities.

Records of occurrence according to the list of records from Floristic course Pruské 2003 (Mertanová and Smatanová 2006): Mníchová Lehota (2003, 7174d), Krásna Ves (2003, 7175c), Slatinka nad Bebravou (2003, 7175c), Šípkov (2003, 7175b), Veľký Kolačín (2003, 7075c), Prejta (2003, 7075b), Ilava (2003, 7075b). According to the list of records from Floristical contribution from the central part of the Strážovské vrchy Mountains (Janišová *et al.* 2004): Valaská Belá (2002, 7176a, 7176b), Čavoj (2002, 7176b). According to Lajcha (2008): Opatová (2007, 7074d, 7174b). According to M. Pirchala: Tužina (2014, 7077d), Milochov (2014, 6876c), Babkov (2013, 6877b), Lietavská Svinná (2013, 6878a). According to D. Pavlišin: Tužina (2014, 7077a), Košecké Rovné (2013, 7076c), Podskalie (2014, 6976b), Bodiná (2014, 6877c), Plevník-Drienové (2014, 6877a). According to M. Vyšinský: Kardošová Vieska (2014, 6977d), Počarová (2014, 6977a), Považská Bystrica (2014, 6876c). According to P. Mereďa Jr.: Malý Kolačín (1998, 7075c), Praznov (1994, 6877c). According to J. Smatanová: Dubnica nad Váhom (2012, 7075d), Malé Lednice (2002, 6977b), Vrchteplá (1999, 6877c), Záskalie (2016, 6877c), Považská Teplá (2016, 6877c). According to Vačková (1997): Trstie (1996, 6976d). According to T. Figura: Podskalie (2015, 6976d). According to Novosadová (2017): Hrabová (2016, 6877a). According to Rybáriková *et al.* (1994): Rajec (to 1994, 6977b). According to F. Kryška (Potůček, unpublished data): Paština Závada (1968 – 1980, 6777d), Hričovské Podhradie (1968 – 1980, 6777d). According to F. Kryška (Velísek 1992): Hlboké nad Váhom (1971, 6777d), Lietavská Závadka (1992, 6877b). According to Velísek (1993): Lietavská Svinná (1993, 6878a). According to A. Dobošová: Závodie (2003, 6778c). According to Z. Pčolová: Rajecká Lesná (2014, 6977b), Porúbka (2013, 6878a).

Epipactis futakii Mereďa et Potůček, EN
Veľký Kolačín; 0.4 km NE of Grófovec Top; 457; S; M; N; 3; I; 16.7.2018; 7075c; V. Ruček; verified older recorded by P. Mereďa Sr. and P. Mereďa Jr from 1994 – 1996, they also found it on the slopes of Drieňová and Markovica hill in 1994-2002 (Mereďa 2002).

Records of occurrence according to Mereďa Jr. (2010): Timoradza (1998, 7175d), Slatinka nad Bebravou (1998, 7175a), Omšenie (2002, 7175a), Trenčianske Teplice (1999, 7175a). Type locality is on Ostrý vrch hill (7074d) near the Trenčianska Teplá town (Mereďa Jr. and Potůček 1998).

Epipactis futakii was recorded near the studied area in Trenčianske Jastrabie (7174c), Považský Inovec Mountains (Eliáš Jr. 2010).

Epipactis greuteri, EN
Považská Teplá; the western foothills of Veľký Manín hill; 400 – 475; H; S; W; 43; G; 11. – 12.8.2017, 14.7.2018; 6876d; V. Ruček; 7 micro-localities, verified older recorded by P. Mereďa Sr. and P. Mereďa Jr. from 2001.

Records of occurrence according to Mereďa Jr. (2000): Lietavská Svinná (1998, 6878c, 6878a).

Epipactis helleborine (L.) Crantz subsp. *helleborine*, LC
Malý Kolačín; 0.6 km NW of Markovica Top; 501; S; S; N; 1; I; 17.7.2018; 7075c; V. Ruček; / * Omšenie; 0.5 km NE of Omšenská Baba Top; 627; S; M; S; 1; I; 20.7.2017; 7075c; V. Ruček; / * Dolná Poruba, Valaská Belá; SE, SW and NW slopes of Homôlka hill; 800 – 800; H; G; N, S, E; 5; S; 21.7.2018; 7075d; V. Ruček; / * Horná Poruba; Vápeč hill; 660 – 790; H; /; S, W; O; S; 1.8.2017, 13.7.2018; 7075d; J. Smatanová, V. Ruček; 3 micro-localities * Kopec; 1.1 km west of Kopec village; 578; S; S; E; 1; I; 1.8.2017; 7076c; V. Ruček; / * Zliechov; in valley 0.8 – 1.5 km SW of Strážov Top; 731 – 912; H; G; S; O; G; 18.6.2017, 23. – 24.6.2018; 7076b, 7076d; V. Ruček; / * Zliechov; 1.3 km NW of Strážov Top; 872; S; S; S; 1; I; 20.6.2017; 7076b; V. Ruček; / * Košecké Rovné; south slopes and valley under Gábrišské vrchy hill; 531 – 863; H; S, G; S; O; S; 27.7., 4.8. 2017; 7076a, 7076b; V. Ruček; / * Pružina; 0.8 km north of Srážovské vodopády Waterfalls; 653; S; S; N; 1; I; 4.6.2018; 7076b; V. Ruček; / * Košecké Podhradie; Podhradská dolina valley, 2 km SW of Mojtín village; 629 – 723; H; G; S; 6; S; 17.7.2017, 19.7.2018; 7076a; V. Ruček; / * Veľké Košecké Podhradie; Veľká and Malá Šimerka, Klokočova, Pancier Sides, Podhradský potok Stream; 365 – 700; H; /; S, N; T; S; 2017, 2018; 7076a; V. Ruček; / * Mojtín; 0.3 km west of Suchý vrch Top; 803 – 813; H; M; S, N; 8; S; 19.7.2018; 7076a; V. Ruček; / * Mojtín; 1 km south of Mojtín village; 711; S; S; N; 1; I; 20.7.2018; 7076a; flower mutation * Kopec; 1.5 km NW of Mačičie Top; 539; M; S; W; O; S; 1.8.2017; 7075b; V. Ruček; / * Trstie; eastern slopes of Dievča hill; 515 – 691; H; S, M, G; E, N; 12; G; 24.7.2018; 6976c; V. Ruček; 5 micro-localities * Beluša; 0.9 km NW of Rohatín Top; 626; S; G; N; 1; I; 21.7.2017; 6976c; V. Ruček; / * Beluša; 0.6 km north of Pasienok Top; 422; S; S; N; 1; I;

16.9.2018; 6976c; V. Ruček; / * Hloža-Podhorie; under Kavčia skala Rock, Prvé vráta Gorge; 360 – 401; M; G; E; 3; S; 29.6.2018; 6976c; V. Ruček; / * Beluša; surroundings of Belušské Slatiny Settlement; 280 – 349; H; /; E; 4; S; 2017, 2018; 6976c; V. Ruček; / * Pružina; Sekaná hill, 0.2 km east of Priedhorie Settlement; 578; S; S; W; 1; I; 22.8.2018; 6976d; V. Ruček; / * Ďurďové; 1 km NE of Ďurďové village; 452; S; S; S; 6; I; 2.7.2017; 6977c; V. Ruček; / * Súľov-Hradná; the southern border of the village; 565; M; S; N; 10; G; 9.8.2018; 6877d; V. Ruček; / * Vrchteplá; ridge of Vodičná hill; 686; S; M; S; 1; I; 23.7.2017; 6877c; V. Ruček; / * Považská Teplá; the western foothills of Veľký Manín hill; 430 – 490; H; S, M; W; O; G; 14.7.2018; 6876d; V. Ruček; 3 micro-localities * Považská Teplá; slopes of Malý Manín hill; /; H; M, G; W, S, E; 22; G; 2017, 2018; 6876d, 6877c, 6877a; V. Ruček; 9 micro-localities * Mikšová; in close proximity to the Hričovský kanál Canal near Beňov Settlement; 300; S; P; 0; O; S; 1.8.2018; 6876b; V. Ruček and M. Kolník; /.

The botanical records of *Epipactis helleborine* subsp. *helleborine* are from the entire area, but they may be incorrectly determined. This species can be easily interchangeable with *Epipactis leutei* and these two species have not been differentiated in the past. A similar problem is with *Epipactis helleborine* subsp. *orbicularis* subspecies. Occurrence of the species is expected on the entire study area.

Epipactis helleborine subsp. *orbicularis* (K. Richter) E. Klien, NT
One record is from Malý Manín hill (Ujházyová *et al.* 2007), but this was not confirmed.

Epipactis komoricensis Mereďa, NT
Dolná Poruba, Valaská Belá; eastern and southern slopes of Homôlka hill; 792 – 867; H; G; S, E; 4; S; 21.7.2018; 7075d; V. Ruček; / * Horná Poruba; western and southern slopes of Vápeč hill; 739 – 787; H; M; W, S; T; G; 13.7., 18.7.2018; 7075d; V. Ruček, J. Smatanová, P. Mereďa Jr. and P. Mereďa Sr.; 2 micro-localities, verification of the older record from 1991 – 1997 (Mereďa Jr. 1998) * Považská Teplá; 1.4 km NW of Veľký Manín Top; 462; S; S; W; 1; I; 13.7., 14.7.2018; 6876d; V. Ruček, J. Smatanová, P. Mereďa Jr. and Sr.; / * Považská Teplá; 0.9 km west of Malý Manín Top; 468; S; S; W; 1; I; 14.7.2018; 6876d; V. Ruček; /.

Records of occurrence according to Mereďa Jr. (1998, 2002): Timoradza (1997, 7175c), Slatinka nad Bebravou (1996, 7175a), Omšenie (1994 – 1997, 7175a, 7075c), Dolná Poruba (2002, 7075d), Prejta (2002, 7075d), Horná Poruba (1993 – 1996, 7075d), Ilava (1993 – 1997, 7075b) – locus classicus, Trstie (1998, 6976c), Prečín (1997, 6977a), Kostolec (1997, 6877c), Jablonové (1994 – 1995, 6877a), Sádočné (1998, 6977c), Fačkov (1998, 7077a). According to Novosadová (2017): Súľov-Hradná (2016, 6877a).

Type locality is 0.5 km SE of Ilava town (Mereďa Jr. 1996b).

Epipactis leptochila s.l.
The list includes undescribed species in *Epipactis leptochila* aggregate. *Epipactis leptochila* s. str. does not occur in the studied area.

Dolná Poruba, Valaská Belá; Homôlka hill; 810 – 850; H; G; N, E; 2; S; 21.7.2018; 7075d; V. Ruček; 2 micro-localities * Horná Poruba; the southern slopes of Vápeč hill; 730 – 770; H; M, G; S; O; S; 18.7.2018; 7075d; P. Mereďa Jr., V. Ruček, J. Smatanová; 2 micro-locaitons, * Prejta; 1 km SE of Kopanica Settlement; 560 – 600; M; G; S; O; I; 12.7.2018; 7075c, 7075d; P. Mereďa Jr., P. Mereďa Sr., V. Ruček, J. Smatanová, Z. Václavová; / * Košecké Rovné; the southern slopes of Gábrišské vrchy hill; 726 – 777; H; M; W; 7; G; 27.7.2017, 20.7.2018; 7076a; V. Ruček; 2 micro-localities * Mojtín; west and south of Suchý vrch Top; 745 – 815; H; M; S, N; 11; S; 31.7.2017, 19.7.2018; 7076a; V. Ruček; 6 records * Ďurďové; Richtárska Site, 2.2 km NE of Ďurďové village; 627; S; G; S; 1; I; 1.7.2017; 6977c; V. Ruček; / * Jasenové; Hradisko Locality, 0.7 km west of Jasenové village; 564; S; M; E; 1; I; 16.7.2017; 6877d; V. Ruček; / * Plevník-Drienové; 0.8 km north of Malý Manín Top; 630; S; M; W; 1; I; 12.8.2017; 6877a; V. Ruček; / * Súľov-Hradná; 0.6 km NW of Súľov Castle; 403; M; M; N; O; I; 27.7.2018; 6877a; M. Jánoš; /.

Others records are from 1996 when a new Epipactis komoricensis was described (Mereďa Jr. 1996b), since it is distinguished from other taxa in Epipactis leptochila aggregate. Older records were not used because it could be an Epipactis neglecta, Epipactis komoricensis or other undescribed species. According to the list of records from Floristic course Pruské 2003 (Mertanová and Smatanová 2006): Slatinka nad Bebravou (2003, 7175a).

Epipactis leutei Robatsch, EN
Veľký Kolačín; 0.6 km NE of Grófovec Top; 463; S; S; N; 1; I; 26.7.2018; 7075c; V. Ruček; / * Dolná Poruba, Valaská Belá; slopes of Homôlka hill; 819 – 854; H; G; S, E, N; 3; S; 21.7.2018; 7075d; V. Ruček; / * Dubnica nad Váhom, Prejta; Za humnami, Harmanová Locality under Beňová skala Top; 670 – 774; H; S, M; S, W; O; S; 12.7.2018; 7075d; V. Ruček; J. Smatanová; P. Mereďa Jr. and P. Mereďa Sr.; 4 micro-localities * Košecké Rovné; valley under Gábrišské vrchy hill, near the yellow-marked hiking trail from Košecké Rovné to Mojtín village; 545 – 739; H; S; W; 2; S; 27.7., 4.8.2017; 7076a; V. Ruček; 2 micro-localities * Mojtín; Hrianková Locality, 1 km south of Mojtín village, near the yellow-marked hiking trail; 730; S; S; N; 1; I; 20.7.2018; 7076a; V. Ruček; / * Mojtín; the northern slopes of Suchý vrch hill, 2 km east of Mojtín village; 762 – 801; H; M; S, N; 9; G; 19.7.2018; 7076a; V. Ruček; 4 micro-localities * Považská Teplá; 0.4 km east of Malý Manín Top; 616; S; G; W; 1; I; 14.7.2018; 6877c; V. Ruček; /.

Epipactis microphylla (Ehrh.) Swartz, LC
Slatinka nad Bebravou; 0.2 km south of Peršová Top, near the blue-marked hiking trail; 789; S; /; S; 1; I; 1.8.2017; 7175b; V. Ruček; / * Dolná Poruba, Valaská Belá; slopes of Homôlka hill; 767 – 867; H; M, G; S, E; 14; S; 21.7.2018; 7075d; V. Ruček; 10 micro-localities * Dolná Poruba; 1.4 km east of Slopský vrch Top, near the red-marked hiking trail; 602; S; M; S; 1; I; 20.7.2017; 7075d; V. Ruček; / * Prejta; 0.3 – 0.6 km west of Beňová skala Top, near the forest road; 736 – 748; H; S; W; 4; G; 12.7.2018; 7075d; V. Ruček; 2 micro-localities * Horná Poruba; southern slopes of Vápeč hill; 602 – 764; H; M; S; O; G; 13.7., 18.7.2018; 7075d; V. Ruček, J.

Smatanová; 4 micro-localities, verified older record by P. Mereďa Jr. and P. Mereďa Sr. from 1990 – 2002 * Horná Poruba; 0.6 km SSE of Vlčinec Top, east of Smrčkovci Settlement; 504; S; M; E; 1; I; 11.8.2018; 7075b; V. Ruček; / * Kopec; 0.7 km east of Mačičie Top, near the forest road; 560; S; S; S; 3; I; 1.8.2017; 7075b; V. Ruček; / * Zliechov; 0.7 – 1.2 km SSW of Strážov Top in valley under Harvanica Rock; 776 – 910; H; M, G; S; O; S; 23.-24.7.2018; 7076b, 7076d; V. Ruček; 3 micro-localities * Veľké Košecké Podhradie, Košecké Rovné; Podhradská dolina valley, Stupičie hill, southern slopes in the forest steppe, forest edges, 40 – 700 m above the third class road; 400 – 720; H; M, G; S; T; S; 15.6., 17.7., 31.7.-4.8.2017, 19.7.2018; 7075b, 7076a; V. Ruček; 14 records * Mojtín; Suchý vrch hill, 0.9 – 2.5 km east of Mojtín village; 682 – 829; H; M, G; N, E, S; T; S; 31.7.2017, 19.7.2018; 7076a; V. Ruček; 9 records * Veľké Košecké Podhradie, Košeca; slopes of Norovica hill and under the Mončeková skala Rock, 0.8 – 2.4 km NW of Košecké Podhradie village; 521 – 625; H; M, G; S, N; T; S; 26.6.2018; 7075b; V. Ruček; 10 records * Pružina; NE ridge of Sokolie hill, 1.6 km SW of Priedhorie Settlement; 667; S; M; E; 1; I; 23.8.2018; 7076b; V. Ruček; / * Veľké Košecké Podhradie; Dehetník, /; 1.8 km SWW of Dielec Top; M; M; E; O; S; 26.6.2018; 6976c; V. Ruček; 2 records * Trstie; in valley, 0.8 – 1.9 km east of Rakytník Top; 504 – 660; H; M; E; O; S; 24.7.2018; 6976c; V. Ruček; 2 records * Hloža-Podhorie, Beluša; Pod Tlstou hill, Pasienok hill, under Kavčia Rock near Prvé Vráta Gorge, 0.7 km east of Belušské Slatiny Settlement (Nad Martovou Hill); 365 – 498; H; M, G; S, E; O; S; 2017, 2018; 6976c; V. Ruček; 5 micro-localities, verification of the older record Pod Tlstou hill (Ujházyová *et al.* 2007) * Podskalie; 0.3 – 0.5 km north of Kohilovec Top on the ridge; 604 – 631; M; S; /; O; G; 16.8.2018; 6976d; V. Ruček; 2 micro-localities * Považská Teplá; 1.4 km NW of Veľký Manín Top; 483; S; /; W; 1; I; 14.7.2018; 6876d; V. Ruček; / * Považská Teplá; western and eastern slopes of Malý Manín hill; 526 – 621; H; M, G; W, E; 36; G; 2017, 2018; 6877c, 6877a; V. Ruček; 2 localities, 14 records, verification of the older record (Ujházyová *et al.* 2007) and record by Urbanová before 1991.

Records of occurrence according to the list of records from Floristic course Pruské 2003 (Mertanová and Smatanová 2006): Zemianske Mitice (2003, 7174d), Slatinka nad Bebravou (2003, 7175a), Slatina nad Bebrabou (2003, 7175b), Čierna Lehota (2003, 7175b, 7176a), Omšenie (2003, 7175a, 7075c). According to P. Jánsky: Soblahov (2009, 7174b). According to E. Fajmonová: Čičmany (1995, 7076d). According to Ujházyová *et al.* (2007): Kostolec (2005, 6877c), Plevník-Drienové (2005, 6877a). According to Mereďa (2010, 2002): Timoradza (to 1998, 7175c, 7175d), Čierna Lehota (1996, 7176c), Trenčianske Teplice (1993-2002, 7075c), Veľký Kolačín (2002, 7075c), Malý Kolačín (1992 – 2002, 7075c), Dubnica nad Váhom (2000, 7075c), Klobušice (2002, 7075a), Ilava (1993 – 2000, 7075a, 7075b), Košeca (1993 – 2002, 6975d), Rajecké Teplice (1998, 6878c), Porúbka (to 1979, 6878c), Lietavská Závadka (to 1985, 6877b), Zemianská Závada (to 1985, 6977a), Hričovské Podhradie (to 1985, 6777d).

Epipactis muelleri Godfery, NT
Valaská Belá; saddle under Homôlka hill, near the second class road; 760; S; S; S; 1; I; 21.7.2018; 7075d; V. Ruček; / * Malý Kolačín; Huštik Site, 1.6 km NW of Markovica Top; 309; S; M; N; 1; I; 17.7.2018; 7075c; V. Ruček / * Dolná Poruba; 0.8 km east of Slopský vrch Top, near the red-marked hiking trail; 600; M; /; E; 3; I; 20.7.2017; 7075d; V. Ruček; / * Horná Poruba; the southern slope of Vápeč hill and near red-marked hiking trail 1.2 km east of Horná Poruba village; 554 – 800; H; S, M, G; S; O; S; 13.7., 18.7.2018; 7075d; J. Smatanová, V. Ruček; 2 localities, 7 records * Zliechov; 1.3 km SSW of Strážov top in valley under Harvanica Rock; 772; S; M; S; 1; I; 23.6.2018; 7076d; V. Ruček; / * Košecké Rovné; 0.5 km SSW of Gábrišské vrchy Top; 753; S; M; S; 1; I; 20.7.2018; 7076a; V. Ruček; / * Veľké Košecké Podhradie; under Klokočová Locality, side valley of Podhradská dolina valley, 2.5 km NW of Čierny vrch hill; 405 – 459; M; M; S; 6; G; 16.6.2018; 7076a; V. Ruček; 2 micro-localities * Veľké Košecké Podhradie; Stráne hill, 0.2 km north of Košecké Podhradie village; 411; S; G; S; 1; I; 19.6.2018; 7075b; V. Ruček; / * Mojtín; 0.3 – 0.6 km east of Suchý vrch hill; 801 – 837; H; M, G; S; 4; S; 31.7.2017, 19.7.2018; 7076a; V. Ruček; / * Hloža-Podhorie; under Kavčia Rock, Prvé vráta Gorge; 395; S; G; E; 2; I; 29.6.2018; 6976c; V. Ruček; / * Domaniža; Hodoň Settlement; 422; M; S; 0; O; S; 29.6.2018; 6977c; V. Ruček; / * Počarová; near the road 0.2 km from the cemetery in Počarova village; 412; M; M; S; 8; I; 29.6.2018; 6977a; V. Ruček, J. Smatanová; / * Podmanín; 1.1 km west of Veľký Manín Top; 508; S; S; W; 1; I; 14.7.2018; 6876d; V. Ruček; / * Považská Bystrica; 0.7 km west of Malý Manín Top; 538; M; M; W; O; S; 14.7.2018; 6877c; V. Ruček; /.

Records of occurrence according to Mereďa Jr. (1998, 2002): Timoradza (2004, 7175d), Kšinná (to 1996, 7176c), Slatinka nad Bebravou (2002, 7175a), Čierna Lehota (1996, 7176c), Trenčianske Teplice (1993 – 2002, 7075c), Omšenie (1993 – 2002, 7075c), Dubnica nad Váhom (1994-2002, 7075c), Prejta (1999, 7075d), Malé Košecké Podhradie (1992, 7075b), Klobušice (2002, 7075a), Ilava (2002, 7075a), Ladce (1998, 6975d), Praznov (1994, 6877c), Rajecké Teplice (to 1985, 6878c). Súľov-Hradná (until 1985, 6877b). According to the list of records from Floristic course Pruské 2003 (Mertanová and Smatanová 2006): Krásna Ves (2003, 7175a), Slatinka nad Bebravou (2003, 7175a), Veľký Kolačín (2003, 7075c), Zliechov (2003, 7076b). According to Michálková (2003): Hloža-Podhorie (2001, 6976c). According to Ujházyová *et al.* (2007): Hloža-Podhorie (2005, 6976c), Kostolec (2005, 6877c). According to Velísek (1992, 1993): Súľov-Hradná (1992 – 1993, 6877a, 6877b), Hričovské Podhradie (1992, 6777d).

Epipactis neglecta (Kümpel) Kümpel, VU
Records of occurrence according to Mereďa Jr. (2002, 2010): Malý Kolačín (1998, 7075c), Horná Poruba (1990 – 2000, 7075d), Považská Teplá, Plevník-Drienové (1998 – 2002, 6877a). According to P. Novosadová: Súľov-Hradná (2016, 6877b), Hrabové (2016, 6877b).

Epipactis palustris (L.) Crantz, NT
Omšenie; NR Omšenská Baba, under Omšenská Baba hill; 460; M; S, M; S; T; I; 12.6.2017; 7075c;

V. Ruček; verification of older record recorded by S. Mertanová and I. Škodová from 2003 (Mertanová and Smatanová 2006) * Kopec; Kúty Locality, 1.6 km SSW of Kopec village; 590; M; S; N; /; I; 9.6.2017; 7076c; J. Smatanová; / * Kopec; Kopecká dolina valley, 1.8 km NE of Mačičie Top; /; S; P; 0; 1; I; 19.6.2018; 7076a; V. Ruček; / * Zliechov; 0.5 – 0.7km NE of Zliechov village; 660 – 675; H; S; W; T; G; 7.7.2016, 20.6.2017; 7076b; V. Ruček; verification of older record recorded by P. Koutecký and J. Koštál (Mertanová and Smatanová 2006) * Kľačno; 0.4 km SW of Vrania skala Top; 905; S; S; S; 1; I; 13.6.2017; 7078a; V. Ruček; / * Domaniža; Blatnica valley; 400; H; P; 0; T; G; 29.6.2018; 6977c; V. Ruček; 2 micro-localities * Jasenové; valley between Jasnové and Malá Čierna village, around the stream; 475 – 500; H; S; 0; H; G; 16.7.2017; 6877d; V. Ruček; / * Súľov-Hradná; Čierny potok valley; 415; S; P, S; N; O; I; 19.6.2018; 6877a; V. Ruček; According to Novosadová (2017) there were hundreds of exemplars in 2016.

Records of occurrence according to Mereďa Jr. (2002, 2010): Dubnica nad Váhom (1999, 7075d), Lietavská Svinná (to 1979, 6877d). According to the list of records from Floristic course Pruské 2003 (Mertanová and Smatanová 2006): Mníchová Lehota (2003, 7174c, 7174d), Soblahov (2001, 7174b), Petrová Lehota (2009, 7174b), Čierna Lehota (2003, 7175b), Valaská Belá (2003, 7176a), Omšenie (2003, 7175a), Čičmany (2003, 7077a), Trstie (2003, 6976d). According to LT-SES Trenčín of 1998: Opatová (to 1998, 7174b). According to Lajcha (2008): Opatová (2007, 7074d). According to K. Devánová: Dolná Poruba (2001, 7175b). According to the list of records from Floristical contribution from the central part of the Strážovské vrchy Mountains (Janišová *et al.* 2004): Čavoj (2002, 7176b). According to J. Smatanová: Čičmany (2017, 7076d), Košecké Rovné (2001, 7076a), Podskalie (2006, 2010, 6976d, 6976b), Prečín (2001, 6977a), Záskalie (2007, 6877c). According to Z. Plesková: Veľké Košecké Podhradie (2015, 7076a), Domaniža (2015, 6977c). E. Fajmonová: Horná Poruba (2001, 7075b), Horné Kočkovce (2003, 6876c), Bodiná (to 1995, 6877c). According to Rybáriková *et al.* (1994): Rajec, Šuja, Rajecká Lesná (to 1993, 6977b). According to Potůček (unpublished data): Paština Závada (1968 – 1980, 6877b, 6777d). According to A. Dobošová: Šuja (2018, 6977b), Závodie (2015, 6778c). According to J. Limánek: Fačkov (2017, 6977d).

Epipactis placentina Bongiorni & P.Grünanger, EN
Dubnica nad Váhom; Harmanová Locality; 649 – 680; H; S, M, G; S; 10; G; 12.7.2018; 7075d; P. Mereďa Jr., P. Mereďa Sr., V. Ruček, J. Smatanová, Z. Václavová; verification of the locality of occurrence from 1993 – 2002 discovered by P. Mereďa Jr. and P. Mereďa Sr. (Mereďa Jr. 2002).

Epipactis pontica Taubenheim, LC
Hloža-Podhorie; 1.2 km SE of Butkov Top; 434 – 575; H; P, S, G; S; T; G; 3.8.2017, 24.7.2018; 6976c; V. Ruček; verification of older data recorded by Mereďa Jr. (1997) * Považská Teplá; 1 – 1.2 km NWW of Veľký Manín Top; 496 – 518; H; S, M, G; W; 37; G; 14.7.2018; 6876d; V. Ruček, J. Smatanová; 2 micro-localities, verification of older data recorded by P. Mereďa Jr. and P. Mereďa Sr. (Mereďa Jr. 2002).

Records of occurrence according to Mereďa Jr. (2002, 2010): Horné Motešice (1997, 7175c), Slatinka nad Bebravou (2002, 7175a), Čierna Lehota (1997, 7176c), Veľký Kolačín (1997, 7075c), Dubnica nad Váhom (1991 – 2000, 7075a), Prejta (1997, 7075a, 7075b), Horná Poruba, Kopec (1997, 7075d), Kopec (1997, 7075b, 7076a), Klobušice (2002, 7075b), Prečín (1997, 6877c). According to the list of records from Floristic course Pruské 2003 (Mertanová and Smatanová 2006): Veľký Kolačín (2003, 7075c).

Epipactis pseudopurpurata Mereďa, VU
Omšenie, Veľký Kolačín; 1.6 km north of Baračka, Kamenné vráta, Pri altáne Locality; 497 – 515; H; S; S; O; S; 16.7.2018; 7075c; V. Ruček; 2 localities, 2.5 km from the *locus classicus* of this species * Dubnica nad Váhom; 1.7 km SSE of Kopanica Settlement; 520; M; S; S; O; I; 12.7.2018; 7075c; P. Mereďa Jr., P. Mereďa Sr., V. Ruček, J. Smatanová, Z. Václavová; / * Horná Poruba; the southern slopes of Vápeč hill; 689 – 762; H; S, M; S; O; G; 13.7. – 18.7.2018; 7075d; P. Mereďa Jr., P. Mereďa Sr., V. Ruček, J. Smatanová; 2 micro-localities * Košecké Rovné; the souther slopes Gábrišské vrchy, 0.2 – 0.9 km east of the yellow-marked hiking trail; 747 – 793; H; S, M; S; 10; S; 20.7.2018; 7076a; V. Ruček; 7 records, verification of older data recorded by P. Mereďa Jr. and P. Mereďa Sr. from 1998 (Mereďa 2002), in the locality there is an occurrence of Epipactis purpurata and *Epipactis ×merediorum* hybrid * Košecké Rovné; Podhradská dolina valley, 1.9 km SW of Mojtína village; 700 – 720; S; G; S; 2; I; 19.7.2018; 7076a; V. Ruček; / * Trstie; Blatnatá Locality, 1.1 km SEE of Dievča Top; 575; S; S; 0; 2; I; 24.7.2018; 6976c; V. Ruček; nearby there occurs *Epipactis purpurata* * Beluša; the western foothills of Rohatín hill; 475 – 500; H; S; W, S; O; S; 21.7., 22.7.2017; 6976c; V. Ruček; 2 micro-localities spaced 0.8 km; verification of older data recorded by D. Michalková and P. Mereďa Jr. from 2001 (Mereďa 2002) * Súľov-Hradná; 0.7 km NW of Súľov Castle, in valley; 400; S; M; /; 1; I; 2018; 6877a; M. Jánoš; 3 km south E. purpurata was recorded by Novosadová (2017).

Records of occurrence according to Mereďa (2002, 2010): Slatinka nad Bebravou (1996, 7175a), Omšenie (1998, 7175a), Zliechov (1998, 7076b), Veľké Košecké Podhradie (1997 –1998, 7075b), Rajecké Teplice (2001, 6878c), Porúbka (1998, 6878a). According to the list of records from Floristic course Pruské 2003 (Mertanová and Smatanová 2006): Veľký Kolačín (2003, 7075c). *Locus classicus* is near Trenčianske Teplice town (7075c; Mereďa 1996a).

Epipactis purpurata Sm., NT
Opatová, Trenčianske Teplice; near the red-marked hiking trail from Trenčianske Teplice town to Opatovská dolina valley; 411 – 463; H; S, M; /; O; S; 20.7.2017; 7074b, 7174d; V. Ruček; in Opatovská dolina valley it was recorded in 2005 (Lacha 2008) * Veľký Kolačín; 0.6 km SW of Drieňová Top; 540; M; S; S; 4; S; 16.7.2018; 7075c; V. Ruček; / * Dolná Poruba; 0.8 km SE of Slopský vrch Top, near the red-marked hiking trail; 612; M; M; S; O; G; 20.7.2017; 7075d; V. Ruček; / * Malý Kolačín; 1.6 km NW of Markovica Top; 330; M; S; W; 5; G; 17.7.2018; 7075c; V. Ruček; / * Dubnica nad Váhom; Dubnická

dolina valley, 1.6 km NNE of Kopanica Settlement; 485; M; S; S; O; I; 12.7.2018; 7075c; P. Mereďa Jr., P. Mereďa Sr., V. Ruček, J. Smatanová, Z. Václavová; / * Horná Poruba; 0.6 km SWW of Smrčkovci Settlement; 550; S; M; E; 1; I; 11.8.2018; 7075b; V. Ruček; / * Košecké Rovné; the southern slopes of Gábrišské vrchy hill; 760 – 840; H; S, M, G; S; T; G; 27.7.2017, 20.7.2018; 7076a, 7076b; V. Ruček; 3 micro-localities, verification of older data recorded by Mereďa (2002), there is an occurrence of Epipactis pseudopurpurata and hybrid Epipactis × merediorum * Mojtín; 0.6 km SWW of Suchý vrch Top; 795 – 833; M; S, M; S; O; S; 31.7.2017, 19.7.2018; 7076a; V. Ruček; / * Veľké Košecké Podhradie, Hloža-Podhorie; Mraznica valley, 1.3 km west of Dielec Top; 425 – 480; H; P, G; E; 14; G; 26.6.2018; 6976c; V. Ruček; 2 micro-localities * Hloža-Podhorie, Beluša; western foothills of Rohatín hill; 465 – 500; H; P, S, M; W; 70; G; 21.7., 22.7.2017, 6976c; V. Ruček; 2 micro-localities, *Epipactis pseudopurpurata* occurs in the vicinity, verification of older data from 2001 (Mereďa 2002) * Tŕstie; Blatnatá Locality, 1.1 – 1.4 km SE of Dievča Top; 484 – 570; H; P, S, M; S, E, N; 37; G; 24.7.2018; 6976c; V. Ruček; 5 micro-localities, there is an occurrence of *Epipactis pseudopurpurata* in the vicinity * Beluša; Pasienok hill, Pánský háj hill; 400 – 500; H; P, S, M; S, E, N; T; G; 15.9., 16.9.2018; 6976c; V. Ruček; 7 micro-localities * Beluša; Nad Martovou Locality, 0.4 km west of Belušské Slatiny Settlement; 338; S; S; E; 1; I; 29.6.2018; 6976c; V. Ruček; / * Beluša; 0.1 km east of Kamenica Top; 413; S; S; 0; 1; I; 29.7.2018; 6976a; V. Ruček; / * Visolaje; 0.5 km SE of Pápežov laz Settlement; 360; H; P, S; 0; 31; I; 29.7.2017; 6976a; V. Ruček; / * Podskalie; saddle SE of Tŕnie hill; 625; M; P; 0; 4; I; 16.8.2018; 6976d, 6976b; V. Ruček; / * Súľov-Hradná; 0.7 km SE of guarry in Jabloňové; 360; S; M; E; 1; I; 9.8.2018; 6877a; V. Ruček; /.

Records of occurrence according to Mereďa Jr. (2002, 2010): Slatinka nad Bebravou (1998 – 2002, 7175a), Trenčianske Teplice (1928, 7075c, 7175a), Veľký Kolačín (1996 – 2002, 7075c), Dubnica nad Váhom (1992 – 2002, 7075c), Prejta (1990, 7075a), Klobušice (1990, 7075a, 7075b), Ilava (2002, 7075a), Praznov (1994, 6877c), Plevník-Drienové (1999, 6877a), Hričovské Podhradie (1985, 6777d). According to J. Smatanová: Horná Poruba (2018, 7075d).

Others records are from 1996 when a new *Epipactis pseudopurpurata* was described (Mereďa 1996a) and began to differentiate from similar *Epipactis purpurata*. According to the list of records from Floristic course Pruské 2003 (Mertanová and Smatanová 2006): Zemianské Mitice (2003, 7174d). According to P. Jánsky: Soblahov (2009, 7174a). According to Lajcha (2008): Dobrá (2005, 7074d). According to Novosadová (2017): Paština Závada – NNR Súľovské skaly (2016, 6877b).

Epipactis tallosii Molnár et Robatsch, NT
Dubnica nad Váhom, above Kvášovec Locality; 1.3 km NNW of Ostrý vrch hill; 254; M; P; 0; 50; I; 12.7.2018; 7075c; P. Mereďa Jr., P. Mereďa Sr., V. Ruček, J. Smatanová; verification of older data recorded in 1991 – 2002 (Mereďa Jr. 2002).

Records of occurrence according to Mereďa Jr. (2002, 2010): Horné Motešice (2001, 7175c), Borčice (1992, 1999, 7074b) - unsuccessful verification in 2018.

Epipactis ×merediorum Batoušek (*E. purpurata* × *E. pseudopurpurata*)
Košecké *Rovné*; the southern slopes of Gábrišské vrchy hill; 750 – 790; H; S; S; 3; G; 20.7.2018; 7076a; V. Ruček; 2 micro-localities, occurrence of *Epipactis pseudopurpurata* and *Epipactis purpurata*.

Epipactis ×heterogama Bayer (*E. atrorubens* × *E. muelleri)*
Records of occurrence according to Mereďa P. Jr. (2002): Horná Poruba (1992, 1999, 7075d).

Epipactis helleborine × *E. muelleri*
Records of occurrence according to Mereďa Jr. (2002, 2010): Omšenie (1996, 7175a), Horná Poruba (1990 – 1997, 7075d), Zliechov (1996 – 1997, 7076b).

Epipactis helleborine × *E. pseudopurpurata*
Records of occurrence according to Mereďa Jr. (2002): Košecké Rovné (1997 – 1998, 7076b).

Epipogium aphyllum Sw., NT
Records of occurrence according to P. Mereďa Jr. and J. Smatanová (Mereďa 2003): Malý Manín hill (2002, 6877c) – unsuccessful verification in 2017, 2018. According to Mereďa Sr. (unpublished data): Bodiná (1997, 6977a) – unsuccessful verification in 2018, Bodiná / Praznov (1997, 6877c). According to Potůček (unpublished data): Súľov-Hradná (1968 – 1987, 6877b).

Goodyera repens (L.) R.Br., NT
Súľov-Hradná; NM Súľovský hrádok; 450; M; P; 0; T; I; 13.6., 25.7.2017., 9.8.2018; 6877a; V. Ruček, J. Smatanová; verification of older record recorded by V. Velísek from 1979 (Velísek 1992).

Records of occurrence according to the list of records from Floristic course Pruské 2003 (Mertanová and Smatanová 2006): Slatinka nad Bebravou (2003, 7175d). According to J. Smatanová: Ďurďové (2006, 6977c). According to I. Kalafusová: Malé Lednice (2006, 6977a). According to D. Dítě: Porúbka (1998, 6878c). According to Novosadová (2017): Súľov-Hradná (2016, 6877a, 6877b).

Gymnadenia conopsea (L.) R.Br., LC
Omšenie; NNM Lánce and vicinity; 420 – 450; H; S; N; H; I; 12.6.2017; 7175a; V. Ruček; / * Omšenie; NR Omšenská baba; 450 – 470; M; S, M; S; 28; I; 12.6.2017; 7075c; V. Ruček; / * Dubnica nad Váhom; Trštické lúky Locality, 570; M; S; N; O; I; 12.6.2017; 7075c; V. Ruček; verifications older data recorded by I. Škodová from 2014 * Košecké Rovné, Kopec; Horná Stredná and Dolná Stredná Settlement, mouth of Strednianská and Kopčianská dolina valley; 375 – 650; H; P, S, M; N; 64; G; 2017, 2018; 7076a, 7076c; V. Ruček; 5 localities, verifications older data recorded by J. Smatanová from 2014 and 2015, E. Fajmonová from 1995 * Košecké Rovné; above the cemetery in Košecké Rovné village; 650 – 675; H; S, M; N; O; S; 5.6.2017; 7076a; V. Ruček; 2 micro-localities * Zliechov; 0.8 – 2.3 km NE of Zliechov village; 660 – 765; H; S; W; H; G; 30.5., 20.6.2017, 5.6.2018; 7076b; V. Ruček, J. Smatanová; 3 micro-localities, verification of older data recorded by J. Košťál, P. Koutecký from 2003, J. Smatanová from 2006, 2014, M. Duchoň

from 2014 * Pružina; Strážovská dolina valley, the mouth and end of the valley; 430 – 690; H; S; N; H; G; 20.6.2017, 4.6.2018; 7076b; V. Ruček; 3 micro-localities, verification of older data recorded by K. Devánová from 2015, J. Smatanová from 2000, 2002, 2006, D. Dítě from 1999, S. Vačková from 1996 (Vačková 1997), E. Sajverová from 1984 * Kľačno; 0.3 south of Vrania skala Top; 900; S; S; S; O; I; 13.6.2017; 7078a; V. Ruček, J. Smatanová; / * Veľké Košecké Podhradie; forest steppe of Podhradská dolina valley, 2.6 – 6 km east of Košecké Podhradie village, Prevrat Saddle; 390 – 690; H; M, G; S; 136; S; 2017, 2018; 7076a, 7075b; V. Ruček, J. Smatanová; 42 records, verification of older data recorded by J. Smatanová from 2002, 2006, E. Fajmonová from 1988, 1990, 1995 * Mojtín; Javorina hill, 1 km east of Stanovci Settlement; 801; S; M; W; 3; I; 5.6.2018; 7076b; V. Ruček; / * Pružina, Briestenné; vicinity of the NM Briestenské skaly; 405 – 500; H; S; S; O; S; 30.6.2017; 6976d, 6977c; V. Ruček; verification of older data recorded by J. Smatanová from 2002 * Čelková Lehota; 0.7 km east of Čelková Lehota village, near the third class road; 473; S; P; 0; 4; I; 30.6.2017; 6977c; V. Ruček; / * Sádočné; Jaseňová dolina valley 0.5 – 1 km SW of Sádočné village; 430 – 450; H; P, S; N; 54; I; 1.7.2017; 6977c; V. Ruček; verification of older data recorded by E. Fajmonová from 2004 * Ďurďové; 1 km NE of Ďurďové village; 480; S; M; S; 2; I; 2.7.2017; 6977c; V. Ruček; verification of older data recorded by E. Fajmonová from 2004 * Domaniža; Blatnica and Hodoň valley; 397 – 425; H; S; W; T; G; 29.6.2018; 6977c; V. Ruček, J. Smatanová; 2 localities, verification of older data recorded by Z. Plesková from 2015, J. Smatanová from 2014, E. Fajmonová from 1995, 2004 * Fačkov, Siguty Locality, Rajecká Lesná; 0.9 – 2.4 km NE of Fačkov village; 525 – 560; H; S, M; S, W; H; G; 13.6.2017; 6977d; V. Ruček, J. Smatanová; 2 micro-localities * Počarová; behind the cemetery in Počarová village; 475; M; M; E; O; I; 8.6. 2017; 6977a; J. Smatanová; / * Bodiná; 0.6 km NE of Bodiná village; 487; S; S; W; 6; I; 9.8.2018; 6877c; V. Ruček; / * Jasenové; valley between Jasenové and Malá Čierna village; 453 – 505; H; P, S; 0; 186; I; 16.7.2017; 6877d; V. Ruček; / * Vrchteplá; Čierny potok valley, 1.7 km north of Vrchteplá village; 600; M; S; S; 3; S; 26.5.2018; 6877c; V. Ruček; verification of data recorded by P. Novosadová from 2016, by J. Smatanová from 2014, 2015 * Súľov-Hradná; Čierny potok valley, Súľovský hrádok Locality; 372 – 425; H; M; E, N; T; G; 13.6.2017, 19.6., 9.8.2018; 6877a; V. Ruček, J. Smatanová; 3 micro-localities, verification of data recorded by P. Novosadová from 2016, by Z. Pčolová from 2014 * Porúbka; NR Slnečné skaly; 405; M; M; E; 19; I; 4.9.2018; 6878a; V. Ruček; /.

Records of occurrence according to Lajcha (2008): Opatová (2007, 7174b). According to K. Rejšek: Dobrá (2004, 7074d). According to D. Galvánek: Šípkov (2002, 7175b). According to S. Mertanová: Dolná Poruba (2007, 7175b). According to A. Fehér: Valaská Belá (2015, 7176a). According to the list of records from Floristic course Pruské 2003 (Mertanová and Smatanová 2006): Valaská Belá (2003, 7176a). According to the list of records from Floristical contribution from the central part of the Strážovské vrchy Mountains (Janišová *et al.* 2004): Valaská Belá, Čavoj (2002, 7176b). According to M. Duchoň: Kľačno (2015, 7077b), Rajec (2015, 6977b). According to J. Smatanová: Horná Poruba (2017, 7075d), Čičmany (2015, 7077a, 7077c), Hloža-Podhorie (2000 – 2003, 6976c), Horný Moštenec (2000, 6976b), Prečín (2017, 6977a). According to A. Dobošová: Šuja, Rajecká Lesná (2018, 6977b), Lietavská Svinná (2016, 6878a), Lietavská Lúčka (2007, 6878a), Ovčiarsko (2003, 6778c), Závodie (2015, 6778c). According to E. Fajmonová: Podmanín – Manínec hill (1995, 6876d), Malé Lednice (2004, 6977b). According to Novosadová (2017): Súľov-Hradná – NNR Súľovské skaly (2016, 6877b, 6777d). According to Velísek (1993): Lietavská Svinná (1993, 6878c). According to S. Uhrin: Hlboké nad Váhom (2014, 6777c, 6777d).

Gymnadenia densiflora (Wahlenb.) A. Dietr., NT
Veľké Košecké Podhradie; Mraznica Saddle, 1.2 km NNE of Stupičie Top; 557; S; S; 0; 1; I; 19.6.2018; 7076a; V. Ruček; / * border of NM Briestenské skaly; 405; S; P; 0; 1; I; 30.6.2017; 6976d; V. Ruček; / * Čelková Lehota; 0.7 km NWW of Čelková Lehota village, near the third class road; 474; S; S; 0; 3; I; 30.6.2017; 6977c; V. Ruček; / * Sádočné; Jaseňová dolina valley, 0.5 SW of Sádočné village; 430; S; P; 0; 2; I; 1.7.2017; 6977c; V. Ruček; / * Domaniža; Blatnica valley; 400; S; P; 0; 1; I; 29.6.2018; 6977c; V. Ruček; verification of older data recorded by E. Fajmonová from 2004 and V. Grulich from 2003 (Mertanová and Smatanová 2006) * Jasenové; valley between Jasenové and Čierna Lehota village; 480 – 490; H; P, S; 0; 8; G; 16.7.2017; 6877d; V. Ruček, P. Kučera, M. Kolník, E. Štubňová; 3 micro-localities * Súľov-Hradná; Čierna dolina valley; 400; S; S; N; 1; I; 19.6.2018; 6877a; V. Ruček; verification of data recorded by Novosadová (2017) from 2016, by J. Smatanová from 2002.

Some individuals of *Gymnadenia densiflora* are difficult to distinguish from *Gymnadenia conopsea*, so older records may not be reliably determined. Records of occurrence according to the list of records from Floristic course Pruské 2003 (Mertanová and Smatanová 2006): Omšenie (2003, 7175b), Horná Poruba (2003, 7075d), Čičmany (2003, 7076d, 7077a), Zliechov (2003 7076b), Veľké Košecké Podhradie (2003, 7075b). According to D. Galvánek: Čierna Lehota (2001, 7175b). According to M. Duchoň: Čavoj (2014, 7176b). According to K. Devánová: Soblahov (2014, 7174b). According to D. Dítě: Omšenie (2001, 7175a). According to the list of records from Floristical contribution from the central part of the Strážovské vrchy Mountains (Janišová *et al.* 2004): Čavoj (2002, 7176b). According to J. Smatanová: Podskalie (2000, 6976b), Bodiná (2001, 6877c). According to P. Smatanová: Horná Poruba (2014, 7075d), Kopec (2015, 7076c), Zliechov (2015, 7076b), Malé Lednice (2015, 6977a). According to A. Dobošová: Fačkov (2015, 6977d), Šuja, Rajecká Lesná (2018, 6977b), Podhorie (2011, 6877b), Poluvsie nad Rajčankou (2002, 6878c), Bitarová (2003, 6778c). According to Velísek (1993): Lietavská Svinná (1993, 6878c), Hrabové (1993, 6877b).

Gymnadenia odoratissima (L.) Rich., NT
Súľov-Hradná; Čierna dolina valley; 390 – 410; H; P; N; 2; S; 19.6.2018; 6877a; V. Ruček, P. Kučera, M. Kolník, E. Štubňová; 2 micro-localities, verification of data recorded by Novosadová (2017) from 2016,

by F. Kryška, Terková and V. Velísek from 1975, 1979, 1980, 1992 (Velísek 1992).

Records of occurrence according to I. Škodová: Omšenie (2015, 7175a). According to Lajcha (2008): Opatová (2007, 7174b). According to Vačková (1997): Pružina valley (1996, 7076b). According to Michálková (2003): Hloža-Podhorie (2002, 6976c). According to Velísek (1992): Súľov-Hradná (1979, 6877b), Hričovské Podhradie (1979, 1992, 6777d).

Gymnadenia ×intermedia Peterm. (*G. conopsea × G. odoratissima)*
Súľov-Hradná; Čierna dolina valley; 381; S; S; N; O; I; 19.6.2018; 6877a; V. Ruček, P. Kučera, M. Kolník, E. Štubňová; 2 micro-localities, verification of older data recorded by V. Velísek and J. Terková from 1978, 1993 (Velísek 1992, 1993).

Records of occurrence according to Potůček (unpublished data): Súľov-Hradná (1968-1980, 6877b).

Herminium monorchis (L.) R.Br., CR

One locality of occurrence is in Rajecká kotlina Basin between Strážovské vrchy Mountains and Malá Fatra Mountains (6977d). Verification of an older finding by V. Ruček and J. Smatanová in 2017.

Himantoglossum adriaticum H.Baumann, EN
Records of occurrence according to M. Galovičová: Počarová (2014, 6977a) - unsuccessful verification in 2017, 2018 – probably a random flowering exemplar.

Limodorum abortivum (L.) Sw, NT
Records of occurrence according to the list of records from Floristic course Pruské 2003 (Mertanová and Smatanová 2006): Slatinka nad Bebravou (2003, 7175a). Records of occurrence according to P. Mereďa Sr.: Dubnica nad Váhom (2005 – 2013, 7075c) – probably a random flowering exemplar.

Listera ovata (L.) Bluff & Fingerh., LC
Omšenie; NNM Lánce; 410 – 450; H; S, M; N; 24; G; 12.6.2017; 7175a; V. Ruček; 3 micro-localities, verification of data recorded by S. Mertanová and I. Škodová from 2003 (Mertanová and Smatanová 2003) * Omšenie; NR Omšenská baba; 440; S; S; S; 12; I; 12.6.2017; 7075c; V. Ruček; / * Dubnica nad Váhom; Tršticé lúky Meadows, 1 km NE of Omšenská baba Top; 575 – 590; H; S; N; 17; G; 22.5., 12.6.2017; 7075c; V. Ruček, J. Smatanová; verification of data recorded by J. Smatanová from 2015 * Kopec; Kúty Locality, 1.5 km SSW of Kopec village; 590; S; S; N; O; I; 19.5.2018; 7076c; V. Ruček; verification of older data recorded by E. Fajmonová from 1995 * Košecké Rovné; Strednianská dolina valley, Horná and Dolná Stredná Settlement; 460 – 650; H; P, S, M; N; 22; G; 30.5., 5.6.2017; 7076a, 7076c; V. Ruček, J. Smatanová; 4 micro-localities, verification of data recorded by J. Smatanová from 2005 – 2014 * Zliechov; 1.7 km SSW of Strážov Top; 720; S; S; S; 1; I; 18.6.2017; 7076d; V. Ruček; / * Košecké Rovné; behind the cemetery in Košecké Rovné village; 550 – 580; H; S; N; 211; I; 2.6., 5.6.2017; 7076a; V. Ruček, J. Smatanová; / * Zliechov; 1.3 – 1.5 km NE of Zliechov village; 725 – 750; H; S; W; O; G; 30.5., 20.6.2017; 7076b; V. Ruček, J. Smatanová; verification of older data recorded by J. Smatanová from 2006 * Pružina; the end and mouth of the Strážovská dolina valley, NNR Strážov; 426 – 775; H; P, M; N; 15; G; 20.6.2017, 4.6.2018; 7076b; V. Ruček; verification of data recorded by K. Devánová from 2015, by J. Smatanová from 200, 2002, by E. Sajverová from 1984, by S. Vačková from 1996, by D. Dítě from 1999 * Kľačno; south of Vrania skala Top, near the red-marked hiking trail; 910; H; S; S; O; G; 13.6.2017; 7077b, 7078a; 2 micro-localities * Veľké Košecké Podhradie; Podhradská dolina valley and side northern valleys; 360 – 680; H; P, S, M, H; S; 122; G; 2017, 2018; 7076a; V. Ruček, J. Smatanová; 7 micro-localities, verification of older data recorded by J. Smatanová from 2002, 2006, by E. Fajmonová from 1995 * Beluša; 1 km NWW of Dievča Top; 575; S; M; W; 1; I; 2016; 6976c; V. Ruček; verification of older data recorded by Michalková (2003) from 2000 - 2002 * Sádočné; 0.7 km SE of Čelková Lehota village; 443; S; M; N; 1; I; 1.7.2017; 6977c; V. Ruček; / * Fačkov; 1 – 2.3 km NNE of Fačkov village, near the first class road; 530; H; M; W; O; S; 13.6.2017; 6977d; V. Ruček, J. Smatanová; 2 micro-localities * Súľov-Hradná; Čierny potok valley; 375 – 600; H; P, S; N; H; G; 2017, 2018; 6877a, 6877c; V. Ruček; V. Ruček, J. Smatanová, P. Kučera, M. Kolník, E. Štubňová; 6 micro-localities, verification of data recorded by P. Novosadová from 2016 (Novosadová 2017), by J. Smatanová from 2002 – 2015, by Z. Pčolová from 2014, by Velísek (1992).

Records of occurrence according to J. Němec: Kubrica (2015, 7174b). According to K. Devánová: Šípkov (2015, 7175d). According to D. Dítě: Trebichava (1998, 7175d). According to the list of records from Floristic course Pruské 2003 (Mertanová and Smatanová 2006): Čierna Lehota (2003, 7175b), Valaská Belá (2003, 7075d), Ilava (2003, 7075b), Čičmany (2003, 7077a). According to Lajcha (2008): Opatová nad Váhom (2007, 7074d, 7174b). According to the list of records from Floristical contribution from the central part of Strážovské vrchy Mountains (Janišová *et al.* 2004): Valaská Belá, Čavoj (2002, 7076c, 7176a, 7176b). According to T. Figura: Valaská Belá (2015, 7176a). According to M. Duchoň: Dolná Poruba (2014, 7075d). According to J. Smatanová: Horná Poruba (2017, 7075d), Podskalie (2006, 6976d), Milochov (2005, 6876c), Horný Moštenec (2000, 6976b), Počarová (2017, 6976b, 6977a), Prečín (2002 – 2017, 6977a), Malé Lednice (2017, 6977b), Bodiná (2018, 6877c). According to S. Uhrin: Rajec (2015, 6977b), Zbýňov (2015, 6877d). According to J. Limánek: Poluvsie nad Rajčankou (2017, 6878c). According to A. Dobošová: Porúbka (2012, 6878a, 6878c). According to Novosadová (2017): Súľov-Hradná (2016, 6877b), Hlboké nad Váhom (2016, 6777d). According to Z. Pčolová: Závodie (2014, 6778c).

Malaxis monophyllos (L.) Sw., NT
Súľov-Hradná; Čierny potok valley; 382; M; M; W; O; I; 19.6.2018; 6877a; V. Ruček, P. Kučera, M. Kolník, E. Štubňová; verification of older data recorded by M. Kolník from 1998.

Records of occurrence according to G. Runkovič: Považská Teplá (to 1990, 6877c).

Neotinea ustulata R.M.Bateman, A. M. Pridgeon & M. W. Chase subsp. *aestivalis* (Kümpel) Jacquet et Scappat., EN

Prečín; PA Svarkovica; 380; S; M; S; 1; O; 29.6.2018; 6977a; V. Ruček, J. Smatanová; verification of older data recorded by J. Smatanová from 2010.

In the past, subspecies was not distinguished from *Neotinea ustulata* subsp. *ustulata* (L.) R. M. Bateman, A. M. Pridgeon *et* M. W. Chase. Late-flowering *Neotinea ustulata* was raised to subspecies status by Kümpel and Mrkvicka (1990). Therefore older records may not be correctly determined.

Records of occurrence according to Lajcha (2008): Opatová nad Váhom (2007, 7174b). According to V. Grulich: Timoradza (1994, 7175c). According to J. Smatanová: Pružina (1999, 6976d), Veľké Košecké Podhradie (2007, 7076a). According to Velísek (1993): Dolná Poruba (to 1993, 7075d). According to A. Dobošová: Fačkov (2015, 6977d).

Neottia nidus-avis (L.) Rich.
Omšenie; NR Omšenská baba; 450 – 525; H; M; S; 4; S; 12.6.2017; 7075c; V. Ruček; / * Omšenie; Kamenné vráta and Závratie Locality; 566 – 600; H; M; S; O; S; 20.7.2017, 16.7.2018; 7075c; V. Ruček; 2 localities, verification of older data recorded by J. Štěpánek from 2003 (Mertanová and Smatanová 2006) * Valaská Belá; Homôlka hill; 843 – 900; H; S, G; E; 3; S; 21.7.2018; 7075d; V. Ruček; / * Horná Poruba; Vápeč hill; 630 – 750; H; M; W; 35; G; 18.5., 19.5.2018; 7075d; V. Ruček; 3 micro-localities, verification of older data recorded by J. Smatanová from 2002, by L. Hrouda, J. Smatanová, P. Koutecký, V. Grulich from 2003 (Mertanová and Smatanová 2006) * Košecké Rovné; Horná Stredná Settlement; 625; S; S; E; 1; I; 5.6.2017; 7076c; V. Ruček; verification of data recorded by D. Pavlišin from 2013 * Zliechov; 1.3 – 2 km NE of Zliechov village; 740 – 870; H; S, M, G; W; 10; G; 30.5., 20.6.2017, 5.6.2018; 7076b; V. Ruček; 3 micro-localities * Pružina; Strážovská dolina valley; 535 – 775; H; P, M; N; 8; G; 20.6.2017, 4.6.2018; 7076b; V. Ruček; 3 micro-localities * Košecké Rovné; Pusté Locality, 0.5 north of Košecké Rovné village; 600; S; S; S; 1; I; 4.8.2017; 7076a; V. Ruček; / * Veľké Košecké Podhradie; Podhradská dolina valley, side northern valleys, southern slopes; 375 – 700; H; M, G; S, E; 140; S; 2017, 2018; 7075b, 7076a; V. Ruček; 61 records * Veľké Košecké Podhradie; Stráne hill, 1 km NW of Veľké Košecké Podhradie village; 535; H; G; S; O; G; 26.6.2017; 7075b; V. Ruček; 2 micro-localities * Košeca; Kraví vrch Locality, 1.7 km NWW of Norovica Top; 422; S; S; W; 1; I; 26.6.2017; 7075b; V. Ruček; / * Veľké Košecké Podhradie; Mraznica valley, 0.7 km SWW of Dielec Top; 380; S; S; N; 10; I; 26.6.2018; 6976c; V. Ruček; / * Mojtín; 0.8 km NE of Rakytník Top; 665; S; M; E; 1; I; 24.7.2018; 6976c; V. Ruček; / * Hloža-Podhorie, Beluša; southern and western slopes of Rohatín hill; /; H; G; S, W; T; G; 28.5.2015, 21.7.2017; 6976c; V. Ruček; 2 micro-localities, verification of older data recorded by D. Michalková from 2001 (Michalková 2003) * Beluša; Nad Martovou Locality, 0.4 km west of Belušské Slatiny Settlement; 335; H; S; E; 11; G; 29.6.2018; 6976c; V. Ruček; 2 micro-localities * Beluša; 0.4 km west of Kamenica Top; 405; S; M; W; 1; I; 29.7.2017; 6976a; V. Ruček; / * Briestenné; Bukovina hill, 1.8 km SSE of Briestenné village, near the green-marked hiking trail; 630; S; S; S; 1; I; 30.6.2017; 6977c; V. Ruček; / * Čelková Lehota; 1.2 km SSW of Čelková Lehota village; /; S; M; E; 3; I; 1.7.2017; 6977c; V. Ruček; / * Ďurďové; 0.9 – 1.8 km NE of Ďurďové village; 475 – 510; H; S, M; S; 23; G; 1.7.2017; 6977c; V. Ruček; 2 micro-localities * Jasenové; Hradisko Locality, 0.8 km west of Jasenové village; /; S; G; N; 1; I; 16.7.2017; 6877d; V. Ruček; / * Považská Teplá; 0.9 km NW of Malý Manín Top; 412; S; M; S; 1; I; 12.8.2017; 6876d; V. Ruček; / * Súľov-Hradná; NNR Súľovské skaly; 444 – 650; H; S, M; S, E; T; S; 26.5.2018; 6877a, 6877b; V. Ruček; verification of data recorded by T. Figura from 2015, by P. Smatanová from 2015, by I. Rizman from 2015, by J. Smatanová from 2002, 2005, by D. Dítě from 1999, by A. Cvachová from 1990, by F. Kryška from 1968 – 1987 (Potůček, unpublished data).

Records of occurrence according to the list of records from Floristic course Pruské 2003 (Mertanová and Smatanová 2006): Trenčianske Mitice, Mníchova Lehota (2003, 7174d), Krásna Ves (2003, 7175c), Omšenie (2003, 7175a), Slatinka nad Bebravou, Šípkov, Dolná Poruba (2003, 7175b), Trenčianske Teplice (2003, 7075c), Košeca (2003, 6975d). According to D. Pavlišin: Čavoj (2013, 7176b), Podskalie (2014, 6976b). According to I. Rizman: Dobrá (2015, 7074d), Bánová (2015, 6878a). According to Lajcha (2008): Opatová nad Váhom (2007, 7074d, 7174b). According to M. Garčár: Dubnica nad Váhom (2015, 7075c). According to the list of records from Floristical contribution from the central part of Strážovské vrchy Mountains (Janišová *et al.* 2004): Valaská Belá (2002, 7176a, 7176b). According to E. Fajmonová: Čičmany (1995, 7076d), Podmanín (1995, 6876d). According to M. Juříček: Fačkov (2015, 6977d). According to J. Smatanová: Kľačno (2017, 7077b), Podskalie (2017, 6976d), Bodiná (2018, 6877c). According to Rybáriková *et al.* (1994): Šuja, Rajecká Lesná (to 1994, 6977b). According to A. Dobošová: Rajec (2004, 6977b), Lietava (2004, 6878a). According to D. Dítě: Porúbka (1998, 6878c). According to M. Pirchala: Lietavská Svinná (2013, 6878a). According to Velísek (1993): Hričovské Podhradie (1993, 677d).

Neotinea tridentata (Scop.) R.M.Bateman, Pridgeon & M.W.Chase, NT
Records of occurrence according to M. Ďurček: Soblahov (2016, 7174b). According to Velísek (1993) and J. Smatanová: Malé Lednice (1993, 6977b) – frequent crossing with Orchis militaris. According to A. Dobošová: Porúbka (2012, 6878c), Turie (2012, 6878a).

Ophrys apifera Huds., VU
Omšenie; NR Omšenská baba; 455 – 470; M; S; S; 9; I; 12.6.2017; 7075c; V. Ruček; verification of older data recorded by Mihálová, J. Vlčko (Dítě 1998).

Records of occurrence according to M. Duchoň (Eliáš Jr. 2015): Slatinka nad Bebravou (2014, 7175d), Šípkov (2014, 7175b).

Ophrys holubyana András., VU
Veľké Košecké Podhradie; side northern valley of Podhradská dolina valley; 430; M; S; S; 5; I; 15.6.2017; 7076a; V. Ruček; verification of older data recorded by J. Smatanová from 2002, 2006, a flowering exemplar was not found in 2018 – probable reason: population dynamics * Veľké Košecké Podhradie; forest steppe of southern slope of

Stupičie hill; 610; S; M; S; 1; I; 22.5.2017; 7076a; V. Ruček; a flowering exemplar was not found in 2018 – probable reason : random flowering exemplar * Pružina; Strážovská dolina valley; 480; M; P; 0; 3; I; 2017, 4.6.2018; 7076b; V. Ruček, J. Smatanová; verification of older data recorded by K. Devánová from 2015, by J. Smatanová from 1999, 2006, by E. Sajverová from 1984 (recorded syn. Ophrys fuciflora), by Vačková (1997) from 1996 (recorded syn. Ophrys holosericea) * Súľov-Hradná; protection zone of NNR Súľovské skaly; 392; M; S; N; 1; I; 19.6.2018; 6877a; V. Ruček, P. Kučera, M. Kolník, E. Štubňová; verification of data recorded by Z. Pčolová from 2017, by J. Smatanová from 1999, 2002, by D. Dítě from 2001, by F. Kryška from 1968 – 1987 (Potůček, unpublished data), by J. Terková, F. Procházka, V. Velísek from 1978 - 1992 (Velísek 1992).

Records of occurrence according to K. Rajcová: Peťovka, Petrova Lehota (to 2018, 7174b). According to K. Devánová: Slatinka nad Bebravou (2015, 7175c). According to D. Dítě: Šípkov (1997, 7175b), Čierna Lehota (1999, 7175b, 7176a). According to I. Škodová: Omšenie (2015, 7175a). According to the list of records from Floristic course Pruské 2003 (Mertanová and Smatanová 2006): Omšenie (2003, 7175b). According to M. Maňák and J. Smatanová: Horná Poruba (2017, 7075d). According to J. Futák: Veľké Košecké Podhradie (1965, 7075b) – recorded as Ophrys fuciflora. According to J. Smatanová: Pružina (1999, 6976d), Podskalie (2010, 6976d), Prečín (1999, 2017, 6977a).

Ophrys insectifera L., NT
Omšenie; NR Omšenská Baba; 460; S; M; S; 2; I; 12.6.2017; 7075c; V. Ruček; / * Horná Poruba; NNR Vápeč, below the peak of Vápeč hill; 903; S; G; W; 1; I; 18.5.2018; 7075d; V. Ruček; verification of older data recorded by J. Smatanová from 2002 * Veľké Košecké Podhradie; forest steppe of Podhradská dolina valley; 360 – 825; H; M, G; S; >202; S, G; 2017, 2018; 7076a; V. Ruček, J. Smatanová; 20 micro-localities * Hloža-Podhorie; southern slope of Rohatín hill; 625; S; G; S; 1; I; 28.5.2015; 6976c; V. Ruček; verification of older data recorded by Michálková (2003) from 2000 – 2002 * Fačkov; 2 km NNE of Fačkov village; 525; S; M; N; 1; I; 13.6.2017; 6977d; V. Ruček, J. Smatanová; / * Počarová; behind the cemetery in Počarová village; 413; S; M; S; 1; I; 24.6.2017; 6977a; V. Ruček; / * Súľov-Hradná; protection zone of NNR Súľovské skaly; 392; M; S; N; O; I; 13.6.2017, 19.6.2018; 6877a; V. Ruček, J. Smatanová, P. Kučera, M. Kolník, E. Štubňová; verification of data recorded by Z. Pčolová from 2014, by J. Smatanová from 1999, 2002, 2006, 2016, by F. Kryška from 1968 – 1987 (Potůček, unpublished data), by V. Velísek from 1993.

Records of occurrence according to K. Devánová: Rožňová Neporadza (2015, 7174d). According to V. Grulich: Timoradza (1996, 7175c). According to D. Dítě: Krásna Ves (1998, 7175c), Trebichava (1998, 7175d), Porúbka (1999, 6878c). According to the list of records from Floristic course Pruské 2003 (Mertanová and Smatanová 2006): Omšenie (2003, 7175a, 7175b). According to I. Škodová: Omšenie (2015, 7175a). According to E. Fajmonová: Veľké Košecké Podhradie (1995, 7075b), Domaniža (2004, 6977c). According to P. Smatanová: Košecké Rovné (2015, 7076a). According to Vačková (1997), E. Sajverová, D. Dítě: Pružina (1984, 1996, 1999, 7076b). According to J. Smatanová: Podskalie (2000, 2002, 2006, 6976d), Malé Lednice (2002, 6977b), Prečín (2017, 6977a), Bodiná (1999, 2011, 6877c), Záskalie (2007, 6877c), Vrchteplá (1999, 6877c). According to Z. Pčolová: Šuja (2014, 6977b). According to A. Dobošová: Rajec (2012, 6977b), Lietavská Svinná (2016, 6877b). According to Novosadová (2017): Súľov-Hradná (2016, 6877b). According to F. Kryška (Potůček, unpublished data): Hlboké nad Váhom, Hričovské Podhradie (1968 – 1987, 6777d).

Ophrys ×devenensis Rchb. f. (*O. holoserica* × *O. insectifera)*
Records of occurrence according to P. Potůček (Velísek 1992): Súľov-Hradná (1968, 6877a) – a Locality destroyed by landslide after rain in 1969. it was probably a hybrid Ophrys holubyana × *insectifera*.

Orchis ×canuti Richter (*O. militaris* × *O. tridentata*)
Records of occurrence according to Velísek (1993), J. Smatanová: Malé Lednice (1993, 2002, 2017, 6977b).

Orchis mascula subsp. *signifera* (Vest) Soó, NT
Dolná Poruba; between Dolná Poruba village and Homôlka hill; 550; M; S; W; 60; I; 20.5.2016; 7075d; V. Ruček; / * Horná Poruba; Kotliny Locality, 1.1 km east of Horná Poruba village; 535 – 571; H; S, M; W; 73; I; 18.5.2018; 7075d; V. Ruček; verification of older data recorded by J. Smatanová from 2006 * Košecké Rovné; Strednianská dolina valley, Horná and Dolná Stredná Settlement, Kržlenica Locality; 455 – 630; H; S, M; E, S; 26; G; 30.5., 5.6.2017; 7076a, 7076c; V. Ruček; 3 localities, verification of data recorded by P. Smatanová from 2015 and by J. Smatanová from 2005, 2007 * Košecké Rovné; behind the cemetery in Košecké Rovné village; 560; S; S; N; 2; I; 2.6. – 5.6.2017; 7076a; J. Smatanová, V. Ruček; / * Zliechov, Čičmany; NNR Strážov, a peak of Strážov hill; 1185 – 1205; H; S; E; 22; G; 20.6.2017, 4.6.2018; 7076b; V. Ruček; verification of older data recorded by J. Smatanová from 2002 * Veľké Košecké Podhradie; Tuchyňa valley; 400; S; M; E; 1; I; 7.5.2017; 7076a; V. Ruček; / * Pružina; Hrubá Kačka hill, Samostrel Meadow; 900; M; S; N; 8; I; 4.6.2015; 7077a; V. Ruček; / * Hloža-Podhorie; 0.9 km east of Dielec Top; 400; M; S, M; N; 20; I; 12.5.2018; 6976c; V. Ruček; / * Čelková Lehota; 0.8 km NEE of Briestenné village, near the third class road; 500; M; S; E; T; I; 25.5.2016; 6977c; V. Ruček; / * Podmanín; 0.5 km SSW of Manínec Top; 455; S; S; S; 1; I; 11.6.2017; 6876d; V. Ruček; / * Vrchteplá; north of Vrchteplá village, near the red-marked hiking trail; 575 – 605; H; S; S; 16; G; 26.5.2018; 6877c; V. Ruček; 2 micro-localities, verification of data recorded by P. Smatanová from 2015 * Súľov-Hradná; Čierny potok valley, near the red-marked hiking trail; 385; M; S; N; 3; S; 13.6.2017; 6877a; V. Ruček, J. Smatanová; verification of data recorded by P. Smatanová from 2015, by Z. Pčolová from 2014, by J. Smatanová from 2002, 2006, by Velísek (1993) from 1992 * Súľov-Hradná; NNR Súľovské skaly, near the yellow-marked hiking trail; 450; S; P; S; 1; I; 26.5.2018; 6877b; V. Ruček; /.

Records of occurrence according to Lajcha (2008): Opatová nad Váhom (2007, 7074d). According to P. Smatanová: Dubnica nad Váhom (2015, 7075c). According to D. Galvánek: Dolná Poruba (2003, 7175b). According to J. Smatanová: Čičmany (2017, 2018, 7077c), Kľačno (2004, 7077b), Fačkov (2004, 7077b), Pružina, Podskalie (2000, 6976d), Zemianský Kvášov, Horná Moštenec (2000, 6976b), Milochov (2005, 6876c), Počarová, Prečín (2017, 6977a), Bodiná (2018, 6877c), Súľov-Hradná (1999, 6877d). According to S. Uhrin: Malá Čierna (2015, 6877d). According to Rybáriková *et al.* (1994): Rajec (to 1993, 6977b). According to A. Dobošová: Lietavská Svinná (2004, 6877b, 6878a), Porúbka (2012, 6878c), Závodie (2003, 6778c). According to A. Beňová: Hlboké nad Váhom (2004, 6777c, 6777d). According to Velísek (1993): Lietavská Svinná (1993, 6878c).

Orchis militaris L., NT
Omšenie; 1.1 km south of Omšenie village; 425; M; S; N; O; S; 12.6.2017; 7175a; V. Ruček; / * Zliechov; 1.8 km NNE of Zliechov village, near the green-marked hiking trail; 745; M; S, M; S; 97; I; 30.5.2017; 7076b; V. Ruček, J. Smatanová; / * Veľké Košecké Podhradie; Podhradská dolina valley, side northern valleys, Tuchyňa valley and under Malá Šimerka Locality; 381 – 445; H; S; S; 165; G; 5.6.2017, 11.5.2018; 7076a; V. Ruček; 2 localities, verification of older data recorded by J. Smatanová from 2002, 2006, by E. Fajmonová from 1995 * Hloža-Podhorie; 0.8 km east of Dielec Top; 390; M; M; N; 15; I; 12.5.2018; 6976c; V. Ruček; / * Podskalie; 0.3 km NE of Podskalie village, above the green-marked hiking trail; 375; M; M; E; T; I; 24.5.2016; 6976d; V. Ruček; / * Prečín; PA Svarkovica; 375; M; M; S; O; I; 25.5.2016, 2017; 6977a; V. Ruček, J. Smatanová; verification of data recorded by T. Figura from 2015, by Z. Pčolová from 2014, by J. Smatanová from 2002, 2006, by E. Fajmonová from 1995 * Vrchteplá; north of Vrchteplá village, near the red-marked hiking trail; 575; S; S; S; 1; I; 26.5.2018; 6877c; V. Ruček; / * Súľov-Hradná; protection zone NNR Súľovské skaly; 392; M; S; N; O; I; 13.6.2017; 6877a; V. Ruček, J. Smatanová; verification of data recorded by J. Smatanová from 2002, 2006, 2016, by Velísek (1992) from 1992, 1984.

Records of occurrence according to V. Grulich: Timoradza (1994, 7175c). According to K. Devánová: Rožňová Neporadza (2014, 7174d), Krásna Ves (2015, 7175c). According to M. Duchoň: Slatina nad Bebravou (2014, 7175d). According to D. Dítě: Trebichava (1998, 7175d). According to Local Territorial System of Ecological Stability of Trenčín town from 1998: Trenčín (to 1998, 7174a). According to D. Galvánek: Dolná Poruba (2003, 7175b). According to Lajcha (2008): Opatová nad Váhom (2007, 7074d). According to the list of records from Floristical contribution from the central part of Strážovské vrchy Moutains (Janišová *et al.* 2004): Valaská Belá (2002, 7176b). According to S. Mertanová: Omšenie (2009, 7075c). According to J. Smatanová: Horná Poruba (2006, 7075d), Počarová, Prečín (2017, 6977a), Malé Lednice (2017, 6977b), Hrabové – NNR Súľovské skaly (2002, 6877b). According to E. Fajmonová: Čelková Lehota, Sádočné, Domaniža (2004, 6977c). According to A. Dobošová: Rajecká Lesná (2015, 6977d), Lietavská Svinná (2004, 6877b). According to S. Uhrin: Rajec, Šuja (2015, 6977b), Zbyňov (2015, 6877d). According to F. Kryška (Velísek 1992): Hlboké nad Váhom, Hričovské Podhradie (1969, 6777d). According to Velísek (1993): Lietavská Svinná (1993, 6878c).

Orchis pallens L., NT
Horná Poruba; 1.3 km NW of Vápeč Top; 540 – 555; M; M; W; 20; I; 18.5.2018; 7075d; V. Ruček; / * Veľké Košecké Podhradie; Podhradská dolina valley, southern slopes of Stupičie, Pancier, Skalica hill and Dúbravy Locality; 375 – 825; H; S, M, G; W, S, E; H; G; 2017, 2018; 7076a; V. Ruček, J. Smatanová; 7 micro-localities, verification of older data recorded by J. Smatanová from 2002 and by E. Fajmonová from 1995 * Beluša; Belušské Slatiny Settlement, under Jelenia skala Rock; 340 – 375; H; S, M; S; H; G; 16.4.2019; 6976c; V. Ruček, O. Roučka; verification of data recorded by O. Roučka * Súľov-Hradná; Čierny potok valley, NNR Súľovské skaly; 395; S; M; N; 2; I; 13.6.2017; 6877a; V. Ruček, J. Smatanová; /.

Records of occurrence according to I. Škodová: Trenčianske Mitice (2014, 7174d), Kubrica (2014, 7174b). According to Lajcha (2008): Opatová nad Váhom (2007, 7074d). According to J. Smatanová: Prejta (2017, 7075c), Košecké Rovné (2007, 2005, 7076c), Trstie (2000, 6976c), Milochov (2005, 6876c), Počarová (2000, 6976b, 6977a), Prečín (2017, 2000, 6977a), Záskalie (2017, 6877c), Súľov-Hradná (2017, 6877b). According to M. Dudáš (Eliáš 2015): Košecké Rovné (2013, 7076a). According to E. Fajmonová: Veľké Košecké Podhradie (1995, 7075b). According to H. Kubišová: Podskalie (2019, 6976d). According to Rybáriková *et al.* (1994): Rajec (to 1994, 6977b). According to M. Pepichová: Podmanín (2006, 6876d). According to P. Smatanová: Vrchteplá (2015, 6877c). According to A. Dobošová: Rajecké Teplice (2003 6878c). According to Novosadová (2017): Súľov-Hradná (2016, 6877b). According to F. Kryška (Potůček, unpublished data): Hlboké nad Váhom, Hričovské Podhradie, Dolný Hričov (1968 – 1980, 6777d).

Orchis spitzelii Saut. ex W.D.J.Koch., EN
Orchis spitzelii was first found in Malá Fatra Moutains in 1988 (Čačko 1990) and in 2006 it was also found by B. Machciník in Strážovské vrchy Mounatins. Records of occurrence according to V. Ruček, J. Smatanová: Malá Fatra (2017, 7078a). According to J. Smatanová, B. Machciník: Kľačno (2017, 7077b).

Orchis ×loreziana Brügger (*Orchis mascula subsp. signifera × O. pallens*)
Records of occurrence according to F. Kryška (Potůček, unpublished data): Súľov-Hradná (1968 – 1980, 6877a, 6877b). According to Velísek (1993): Kostolec (1993, 6877c). According to M. Kolník: Súľov-Hradná (1999 – 2002, 6877b).

Platanthera bifolia (L.) Rich, LC
Dolná Poruba; 1 km SSE of Česaná hora Top, near the blue-marked hiking trail; 750; S; G; E; 1; I; 1.8.2017; 7175b; V. Ruček; / * Omšenie; NR Žihľavník, 0.8 km NNW of Žihľavník Top; 820; S; G; N; 1; I; 1.8.2017; 7175a; V. Ruček; / * Omšenie; NNM Lánce; 430; S; M; N; 2; I; 12.6.2017; 7175a; V. Ruček; verification of older data recorded by S.

Mertanová and I. Škodová from 2003 (Mertanová and Smatanová 2006) * Omšenie; above Baračka near the educational trail; 325 – 475; H; G; E; O; S; 16.7.2018; 7075c; V. Ruček; / * Omšenie; NR Omšenská Baba and the peak of Michalová hill; 450 – 610; H; S, M; S; 28; G; 12.6.2017; 7075c; V. Ruček; 3 micro-localities * Dolná Poruba; 0.7 km east of Slopský vrch Top, near the red-marked hiking trail; 595 – 625; M; S; E; 3; S; 20.7.2017; 7075d; V. Ruček; / * Horná Poruba; NNR Vápeč, the western foothills of Vápeč hill, near red-blue-marked hiking trail on NE ridge; 480 – 900; H; S, M, G; W; T; S; 1.8.2018, 18.5.2018; 7075d, 7076c; V. Ruček; verification of data recorded by J. Smatanová from 2014, 2006, 2002, 2001, by V. Grulich from 2003, by A. Cvachová from 1983 * Košecké Rovné; Strednianska dolina valley; 430 – 575; H; P, M; S; 10; G; 9.6.2017; 7076a, 7076c; V. Ruček; 3 localities * Košecké Rovné; behind the cemetery in Košecké Rovné village; 575; S; M; N; 2; I; 5.6.2017; 7076a; V. Ruček; / * Zliechov; NNR Strážov, 1.3 km NNW of Strážov Top; 775; M; M; S; 3; S; 18.6.2017; 7076d; V. Ruček; verification of data recorded by J. Němec from 2015 * Zliechov; 1.7 km NE of Zliechov village, NNR Strážov; 790 – 825; H; M; W; 17; G; 2017, 2018; 7076b; V. Ruček; verification of older data recorded by J. Smatanová from 2006, by J. Košťál, P. Koutecký, J. Kochjarová from 2003 (Mertanová and Smatanová 2006) * Čičmany; NNR Strážov, the peak of Strážov hill; 1201; S; S; E; 1; I; 4.6.2018; 7076b; V. Ruček; / * Pružina; Strážovská dolina valley; 475 – 1062; H; P, S, G; N; 63; G; 20.6.2017, 4.6., 5.6.2018; 7076b; V. Ruček; 6 micro-localities, verification of data recorded by K. Devánová from 2015, by J. Smatanová from 2014, 2006, 2002, by Vačková (1997) from 1996, by E. Fajmonová from 1995 * Košecké Rovné; the southern slopes of Gábrišské vrch hill; 550 – 860; H; G; S; O; S; 27.7.2017; 7076a, 7076b; V. Ruček; / * Veľké Košecké Podhradie; Podhradská dolina valley, southern slopes and side valley; 370 – 830; H; S, M G; S; 126; S; 2017, 2018; 7076a; V. Ruček; verification of older data recorded by M. Pirchala from 2014, by J. Smatanová from 2002, by E. Fajmonová from 1995 * Mojtín; near the Stará Mojtínska jaskyňa Cave; 570; S; G; W; 1; I; 5.6.2018; 7076b; V. Ruček; / * Mojtín; west of Mojtín village, Tretie vráta Gorge – Smetlička, Vŕšky Locality, Suchý vrch hill; 591 – 800; H; M, G; S; T; G; 31.7.2017, 24.5., 19.7.2018; 7076a; V. Ruček; 3 localities * Veľké Košecké Podhradie; Mraznica valley, 1.7 km NW of Dielec Top; 450 – 520; H; G; S; O; S; 26.6.2018; 7076a, 6976c; V. Ruček; / * Hloža-Podhorie; southern slopes of Rohatín hill; 430 – 650; H; M, G; S; T; G; 2015, 2017; 6976c; V. Ruček; verification of data recorded by Michalková (2003) from 2002 * Pružina; Rečica valley; 472; S; P; W; 1; I; 2.7.2017; 7076b; V. Ruček; / * Čelková Lehota; south and west of Čelková Lehota village; 500 – 630; H; S, M; N, E; T; G; 2015, 2017; 6977c; V. Ruček; scattered throughout the territory, verification of older data recorded by E. Fajmonová from 2004 * Briestenné; NM Briestenské skaly; 440; M; S, G; S; 30; I; 30.6.2017; 6976d; V. Ruček * Hloža-Podhorie; under Tlstá hora hill, Kavčia Rock; 375 – 540; H; M; S, E; 25; S; 1.6.2014, 3.8.2017; 6976c; V. Ruček; 2 localities * Beluša; Filipová kopanica Locality, 0.9 km east of church in Belušské Slatiny; 420; S; P; 0; 4; I; 28.6.2017; 6976c; V. Ruček; / * Ďurďové; the eastern half of the cadastral area; 490 – 675; H; S; /; O; S; 1.7., 2.7.2017; 6 977c; V. Ruček; 5 micro-localities * Beluša; Kamenica hill; 375 – 420; H; S; E; O; S; 29.7.2017; 6976a; V. Ruček; verification of older data recorded by D. Michálková from 2004 * Fačkov, Rajecká Lesná; 1.9 km NNE of Fačkov village, near the first class road; 425; M; S; W; O; S; 13.6.2017; 6977d; V. Ruček; / * Bodiná; 1.2 km SWW of Vysoký vrch Top; 480; S; M; W; 1; I; 9.8.2018; 6877c; V. Ruček; verification of older data recorded by J. Smatanová from 1999, by E. Fajmonová from 1995 * Jasenové; valley between Jasenové and Malá Čierna village; 470 – 500; H; S; E; O; S; 16.7.2017; 6877d; V. Ruček; / * Považská Teplá; the western foothills of Veľký Manín hill; 392 – 525; H; S, M; W; 9; S; 14.7.2018; 6876d; V. Ruček; 4 micro-localities * Vrchteplá; the ridge of Vodičná hill and Čierny potok valley; 595 – 765; H; S, M; S; O; S; 23.7.2017, 26.5.2018; 6877c; V. Ruček; 3 micro-localities, verification of data recorded by P. Smatanová from 2015 * Súľov-Hradná; NNR Súľovské skaly, protection zone, Čierny potok valley; 366 – 534; H; P, S, M, G; N, E, S; 40; S; 2017, 2018; 6877a; V. Ruček; verification of data recorded by Novosadová (2017) from 2016, by J. Smatanová from 2016, 2006, 2002, by P. Smatanová from 2015, by M. Pirchala, by Z. Pčolová from 2014, by V. Velísek from 1993, by F. Kryška from 1968 – 1980 (Potůček, unpublished data) * Lietavská Lúčka; 1.2 km east of Lietava village; 388; S; P; 0; 1; I; 1.9.2018; 6878a; V. Ruček; / * Mikšová; in close proximity to the Hričovský kanál Canal near Beňov Settlement; 290; M; P; 0; T; I; 1.8.2018; 6876b; V. Ruček and M. Kolník; /.

Records of occurrence according to the list of records from Floristic course Pruské 2003 (Mertanová and Smatanová 2006): Mníchová Lehota, Trenčianske Mitice (2003, 7174c, 7174d), Opatová nad Váhom (2003, 7174b), Prejta, Ilava (2003, 7075a, 7075b), Košeca (2003, 6975d). According to D. Dítě: Krásna Ves (1998, 7175c), Trebichava (1998, 7175d). According to K. Rejšek: Dobrá (2004, 7074d), Trenčianska Teplá (2004, 7074d). According to K. Devánová: Soblahov (2014, 7174b). According to Lajcha (2008): Opatová nad Váhom (2007, 7074d). According to the list of records from Floristical contribution from the central part of Strážovské vrchy Mts (Janišová *et al.* 2004): Valaská Belá (2002, 7176a), Temeš, Čavoj (2002, 7176b), Prejta (2003, 7075a). According to A. Dobošová: Fačkov (2018, 7077a, 7077b), Rajecká Lesná (2015, 6977d), Zbyňov (2002, 6977b). According to D. Pavlišin: Malé Košecké Podhradie (2014, 7075b), Podskalie (2014, 6976b). According to J. Smatanová: Čičmany (2018, 7077c), Počarová, Prečín (2016, 2017, 6977a). According to S. Štefaniková: Dolný Moštenec (2018, 6976b). According to Z. Pčolová: Rajec (2014, 6977b). According to M. Duchoň: Poluvsie nad Rajčankou (2014, 6878c). According to Novosadová (2017): Súľov-Hradná (2016, 6877b). According to F. Kryška (Potůček, unpublished data): Hričovské Podhradie (1968 – 1980, 6777d).

Platanthera chlorantha (Custer) Rchb., NT
Omšenie; NM Havránkovský potok; 395; S; P; 0; 1; I; 2.8.2017; 7175a; V. Ruček; / * Omšenie; NNM Lánce; 430; S; S; N; 1; I; 12.6.2017; 7175a; V. Ruček; / * Omšenie; NR Omšenská baba; 450 - 475;

M; S, M; S; 13; I; 12.6.2017; 7075c; V. Ruček; / * Veľký Kolačín; 0.8 km south of Markovica Top; 440; S; S; S; 1; I; 16.7.2018; 7075c; V. Ruček; / * Omšenie; NP Havránkovský potok; 395; S; P; 0; 1; I; 2.8.2017; 7175a; V. Ruček; / * Dolná Poruba; 1 km east of Slopský vrch Top, near the red-marked hiking trail; 595; S; S; S; 1; I; 20.7.2017; 7075d; V. Ruček; / * Horná Poruba; 1.2 km SW of Vápeč Top, near the red-marked hiking trail; 512; S; S; W; 1; I; 18.7.2018; 7075d; V. Ruček; / * Košecké Rovné; Dolná Stredná Settlement; 472; S; S; N; 1; I; 19.6.2018; 7076a; V. Ruček, P. Kučera, M. Kolník, E. Štubňová; verification of data recorded by P. Smatanová from 2014, by J. Smatanová from 2000 * Zliechov; 1.2 – 1.8 km NNW of Strážov Top, NNR Strážov; 715 – 760; H; S, M; S; 7; S; 18.6.2017, 23.6.2018; 7076d; V. Ruček; / * Zliechov; 1.4 – 2.2 km NE of Zliechov village; 760 – 800; H; S; W, S; 34; G; 20.6.2017, 5.6.2018; 7076b; V. Ruček; 3 micro-localities, verification of data recorded by J. Smatanová from 2014 * Pružina; Strážovská dolina valley; 440 – 625; H; P, S; N; 12; S; 20.6.2017, 4.6.2018; 7076b; V. Ruček; 5 micro-localities, verification of data recorded by K. Devánová from 2015, by E. Sajverová from 1984 * Veľké Košecké Podhradie; Podhradská dolina valley, southern slopes and side valley; 355 – 690; H; P, S, M; S; 17; S; 2017, 2018; 7076a; V. Ruček; 4 localities, verification of older data recorded by E. Fajmonová from 1997 * Košeca; Beňová Localities, 2.3 km NE of Košeca church; 320; M; S; W; 6; S; 26.6.2017; 7075b; V. Ruček; verification of older data recorded by K. Boublík (Mertanová and Smatanová 2006) * Hloža-Podhorie; 0.7 km north of Dielec Top; 355; S; P; 0; 1; I; 26.6.2018; 6976c; V. Ruček; / * Beluša; Filipová kopanica Locality, 0.9 km east of Belušské Slatiny church; 420; S; P; 0; 11; I; 28.6.2017; 6976c; V. Ruček; 98 cm high exemplars * Trístie; Jaseňová valley, 0.9 km NNE of Tupý hrádok Top; 450; S; M; E; 1; I; 24.7.2018; 6976d; V. Ruček; / * Briestenné; 1.2 km NW of Briestenné village; 495; S; P; 0; 1; I; 30.6.2017; 6976d; V. Ruček; / * Ďurďové; 0.8 km NNE of Briestenné village; 520; S; S; W; 5; I; 30.6.2017; 6977c; V. Ruček; / * Sádočné; 1 km NE of Čelková Lehota village; 440; S; S; N; 1; I; 1.7.2017; 6977c; V. Ruček; / * Fačkov; 2.1 km NE of Fačkov village; 560; S; M; N; 1; I; 13.6.2017; 6977d; V. Ruček, J. Smatanová; / * Prečín; PA Svarkovica; 380 – 390; M; M; S; O; S; 29.6.2018; 6977a; / * Súľov-Hradná; 1.5 km SSW of Hradná village; 560; S; S; N; 1; I; 9.8.2018; 6877d; V. Ruček; / * Vrchteplá; 1.6 km north of Vrchteplá Top, near the red-marked hiking trail; 600; S; S; S; 1; I; 26.5.2018; 6877c; V. Ruček; verification of data recorded by P. Novosadová from 2016 * Súľov-Hradná; protection zone NNR Súľovské skaly, Čierny potok valley; 370 – 410; H; P, S; N; O; S; 13.6.2017, 19.6.2018; 6877a; V. Ruček, J. Smatanová, P. Kučera, M. Kolník, E. Štubňová; verification of data recorded by Novosadová (2017) from 2016, by P. Smatanová from 2015, 2014, by J. Smatanová from 2006, 2002.

Records of occurrence according to K. Olšavská: Trenčianske Mitice (2003, 7174d). According to S. Mertanová: Peťovka, Petrová Lehota (2003, 7174b). According to V. Grulich: Timoradza (1994, 7175d). According to Štepánková: Slatina nad Bebravou (1994, 7175d). According to D. Galvánek: Šípkov (2002, 7175b). According to the list of records from Floristical contribution from the central part of Strážovské vrchy Moutains (Janišová *et al.* 2004): Valaská Belá (2002, 7176a, 7176b, 7076c). According to P. Smatanová: Horná Poruba 2014, 7075d), Kopec (2015, 7076c). According to K. Prach (Mertanová and Smatanová 2006): Ladce (2003, 6975d). According to J. Smatanová: Visolaje (2001, 6976a), Podskalie (2006, 6976b). According to J. Limánek: Fačkov (2017, 6977d). According to A. Dobošová: Šuja, Rajecká Lesná (2018, 6977b). According to Novosadová (2017): Súľov-Hradná (2016, 6877b), Hlboké nad Váhom (2016, 6777d). According to Velísek (1993): Lietavská Svinná (1993, 6878a). According to Z. Pčolová: Závodie (2014, 6778c).

Platanthera ×hybrida Brügger (*P. chlorantha × P. bifolia*)
Košecké Rovné; Dolná Stredná Settlement; 470; S; S; N; 1; I; 19.6.2018; 7076a; V. Ruček, P. Kučera, M. Kolník, E. Štubňová; / * Beluša; Filipová kopanica Locality, 0.9 km east of Belušské Slatiny church; 420; S; P; 0; 4; I; 28.6.2017; 6976c; V. Ruček; /.

Records of occurrence according to B. Trávníček (Mertanová and Smatanová 2006): Veľký Kolačín (2003, 7075c). According to J. Smatanová: Horná Poruba (2006, 7075d), Záskalie, Kostolec (1999, 6877c), Súľov-Hradná (2002, 6877a). According to E. Fajmonová: Veľké Košecké Podhradie (1997, 7076a).

Traunsteinera globosa (L.) Rchb., NT
Košecké Rovné; Stredniansk á dolina valley, Strednianské lúky Meadows, Kržlenica Locality, Horná and Dolná Stredná Settlement; 460 – 655; H; S, M; N; 48; G; 30.5., 5.6.2017; 7076a, 7076c, 7076c; V. Ruček, J. Smatanová; 4 localities, by P. Smatanová from 2015 * Košecké Rovné; behind the cemetery in Košecké Rovné village; 565; M; M; N; 5; I; 2.6., 5.6.2017; 7076a; V. Ruček, J. Smatanová; / * Zliechov, Čičmany; NNR Strážov, near the hiking trail between the upper and lower meadows below the peak of Strážov hill; 1135 – 1200; H; P, M; W, E; 9; G; 20.6.2017, 5.6.2018; 7076b; V. Ruček; verification of data recorded by J. Smatanová from 2010, by J. Košťál and J. Kochjarová from 2003 (Mertanová and Smatanová 2006), by E. Fajmonová from 1995.

Records of occurrence according to D. Galvánek: Dolná Poruba (2003, 7175b, 7075d). According to K. Rejšek: Dobrá (2004, 7074d). According to Lajcha (2008): Opatová nad Váhom (2007, 7174b). According to the list of records from Floristical contribution from the central part of Strážovské vrchy Mountains (Janišová *et al.* 2004): Čavoj (2002, 7176b). According to M. Duchoň: Valaská Belá (2014, 7075d). According to P. Smatanová: Horná Poruba (2015, 7075d). According to J. Smatanová: Dubnica nad Váhom (2017, 7075c), Horná Poruba (2001, 7075d), Podskalie (2010 6976d), Prečín (2006, 6977a). According to P. Mereďa Jr.: Dubnica nad Váhom (1998, 7075d). According to A. Dobošová: Fačkov (2015, 6977d), Šuja, Rajec (2012, 6977b). According to I. Kalafusová: Vrchteplá (2005, 6877c). According to Velísek (1993): Jablonové (1993, 6877a).

Other Orchids from the south part of Strážovské vrchy Mountains

Orchis purpurea Huds., NT
The species is reported in thermophilous forest communities of the southern part of Strážovské

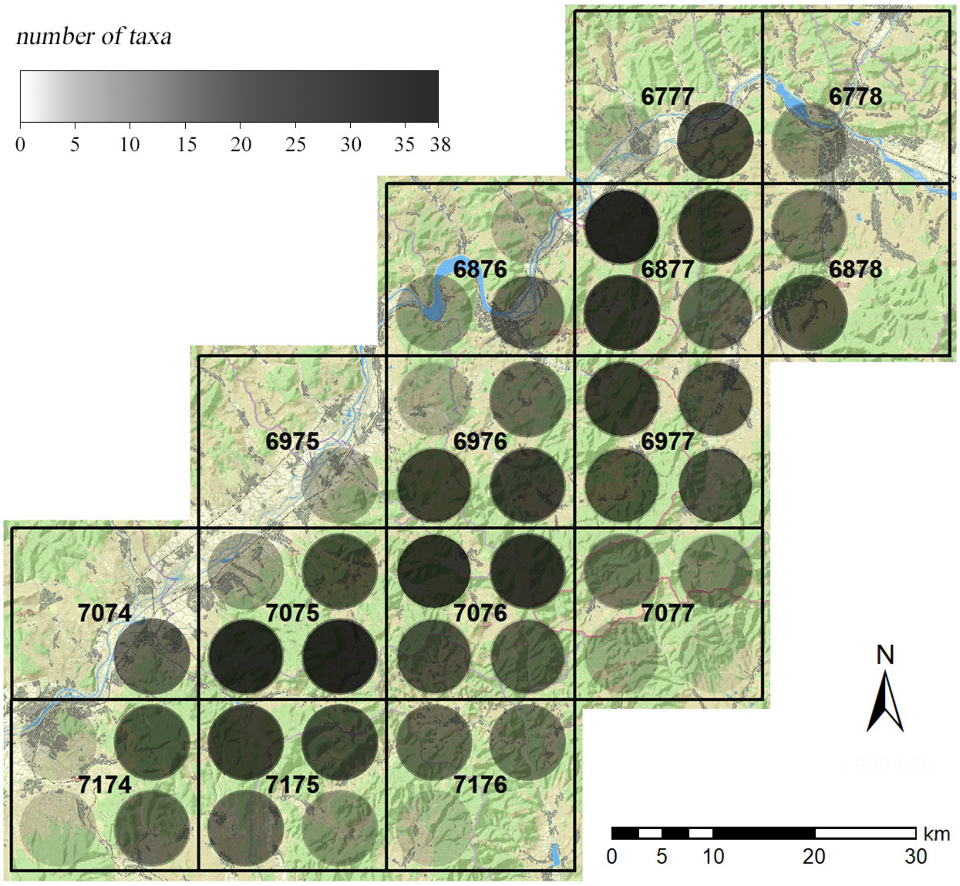

Fig. 2. Species diversity of the family *Orchidaceae* by map quadrants. Includes current and historical findings of 65 taxa. The largest number of taxa is in the darkest dots 7075c, 7075d, and 6877a. there are 35 - 38 taxa / quadrant.

vrchy Moutains (Skořepa 2005), specifically in the Kňaží stôl and Malá Magura Subunits (Štátna ochrana prírody SR 2014).

Pseudorchis albida (L.) Á.Löve & D.Löve, NT
Leucorchis albida (syn.) was recorded in the area of Rokoš hill (Futák 1960). The species is considered to be missing in this area.

Anacamptis pyramidalis (L.) Rich., VU
According to S. Benedikt and J. Raisová (Eliáš 2012): Uhrovské Podhradie (2011, 7276b).

Discussion

A total of 65 taxa of the family *Orchidaceae* (54 species and 11 hybrids) were recorded in the study area, taking into consideration historical and present botanical occurrence. Of these, 57 were verified and found in 2010 – 2018. Eight taxa were not confirmed: *Limodorum abortivum*, *Epipogium aphyllum* and rare hybrids of the genus *Epipactis*, *Ophrys* and *Orchis*. This may be due to climatic conditions or population dynamics.

Súľovské skaly NNR has been called an orchid paradise since the second half of the 20th century (Potůček and Kryška 1975). A systematic survey of orchids has been undertaken in this area since 1968 and therefore the evolution of the number of species is best demonstrated in this area. According to the list of species and hybrids found by F. Kryška between 1968 – 1987, there were 34 taxa in the area of Súľovské skaly (Potůček, unpublished data). There are reported findings of *Epipactis helleborine* subsp. viridis and *Gymnadenia conopsea* subsp. *montana*, whose taxonomic value is questionable (Mereďa Jr. 2010). These are not currently on the list of orchids found in Slovakia. It is likely that *Epipactis leptochila* (Godfery) Godfery was also incorrectly reported. According to Mereďa Jr. (2002), this species does not occur in the studied area, and was likely *Epipactis komoricensis* (Mereďa 1996b), *E. neglecta* or another species not yet described from the *Epipactis leptochila* aggregate. Some localities of occurrence no longer exist, and may have disappeared due to afforestation, intensive grazing or absence of extensive grazing and subsequent natural succession (Potůček, unpublished data). *Dactylorhiza sambucina*, *Ophrys ×devensis*, *Orchis morio*, *Ophrys cornuta* (artificially planted) have been absent for several years, and it is like they they are extinct or are present in a new locality. Following taxonomic revision of some species, the number of species increased in the Súľovské vrchy Mountains. *Dactylorhiza fuchsii* subsp. *fuchsii* was separated from *Dactylorhiza maculata* agg. *Epipactis pseudopurpurata* began to differ from *Epactis purpurata*. Súľovské skaly drew many botanists who produced new findings for the area, such as *Dactylorhiza fuchsii* subsp. *sooana*, *Epipactis albensis*, *E. komoricensis*, *E. neglecta*, *E.*

pseudopurpurata, *Gymnadenia densiflora* and *Malaxis monophyllos*. According to the current list of orchids (Vlčko *et al.* 2003), we can say that in the years 1968 – 1987 there were 32 species in the area of Súľovské skaly. Between 1987 and 2018, nine new taxa were found. In the last century, 4 taxa became extinct (Potůček, unpublished data) and in 2010 – 2018 *Orchis* ×*loreziana*, *Platanthera* ×*hybrida*, *Dactylorhiza majalis*, *Epipogium aphyllum*, *Traunsteinera globosa* were no longer found.

During the growing season in 2017 and 2018, about 8000 predominantly flowering examples of 48 taxa were recorded in 2021 localities/ micro-localities by our own field survey. New localities of some species of orchid were also discovered during this period. Among the most interesting are the presence of *Cephalanthera longifolia* at two locations in the Strážovské vrchy Mountains; *Dactylorhiza incarnata* at one locality in Strážovské vrchy Mountains; *Epipactis albensis* at 11 localities in all studied geomorphological units, which is a new species for the Žilinská kotlina Basin; and *Epipactis komoricensis* at 2 localities in Strážovské and Súľovské vrchy Mountains. *Epipactis leptochila* agg. was present at 5 surveyed localities near both mountains; *Epipactis leutei* at 5 localities near both Mountains. Although this species has not yet been published in the territory, P. Mereďa Jr. mentions its occurrence. *Epipactis microphylla* was found at 17 localities at both Mountains; *Epipactis muelleri* at 14 localities in both Mountains; *Epipactis pseudopurpurata* at two localities in the Strážovské vrchy Mountains; and *Epipactis* ×*crorum* was found in one location in the Strážovské vrchy Mountains. Information on the occurrence of this hybrid in the territory of Slovakia has not yet been published. *Gymnadenia densiflora* was present in 5 localities including both Mountains and the Žilinská kotlina Basin. *Ophrys holubyana* was found in one location in the Strážovské vrchy Mountains. *Ophrys insectifera* was present at 3 localities in both Mountains, and *Platanthera* ×*hybrida* at two localities in Strážovské vrchy Mountains. The occurrence of *Dactylorhiza fuchsii* subsp. *fuchsii*, *D. fuchsii* subsp. *sooiana*, *D. incarnata* subsp. *incarnata*, *D. viridis*, *Dactylorhiza* ×*braunii*, *Epipactis futakii*, *E. greuteri*, *E. komoricensis*, *E. placentina*, *E. pontica*, *E. pseudopurpurata*, *E. tallosii*, *Goodyera repens*, *Gymnadenia odoratissima*, *Gymnadenia* ×*intermedia*, *Herminium monorchis*, *Malaxis monophyllos*, *Neotinea ustulata* subsp. *aestivalis*, *Ophrys apifera*, *Ophrys holubyana*, *Traunsteinera globosa* was confirmed at known localities.

Significant population dynamics of some taxa could be observed even during the two growing seasons in which intensive exploration took place. For example, in Podhradská dolina valley in Strážovské vrchy Mountains there were 30 occurences. *Ophrys insectifera* on one microlocalite during the 2017 growing season, but non observed the following year. Similarly, there were five examples of *Ophrys holubyana* observed in 2017 in two micro-localities in Podhradská dolina valley, but none in 2018. We also recorded a decrease in the number of *Traunsteinera globosa* in Strednianska dolina valley and at the top of Strážov hill in the Strážovske vrchy Mountains. The most likely cause of a small number of flowering exemplars is the extremely dry spring season in 2018, which accelerated the growing season including the time of flowering by approximately two weeks. In the highest parts of Strážov hill in 2017, the flowering of *Traunsteinera globosa* occurred in the third week of July, while in 2018 it occurred in the first week of July. Due to this shift, it was necessary to adapt summer field survey dates. The drought was reflected in the condition of flowers, imperfect development of flower organs, more frequent kleistogamous flowering, as well as the drying of buds, and thus, it was more difficult to determine species in the troubled *Epipactis* genus. Conversely, 2018 was characterized by successful „floodplain orchids", which corresponds to a number of new localities.

The number of orchid taxa in each geomorphological unit and in close proximity is as follows: Strážovské vrchy Mountains (945 km^2) – 52 species (including three species from the southern part of the mountain range outside the studied area) and eight hybrids. Súľovské vrchy Mountains (200 km^2) – 45 species and four hybrids, Považské Podolie Basin - east part of the river Váh (240 km^2) – 12 species, Žilinská kotlina Basin - west part of the river Rajčianka (125 km2) – 23 species. For comparison, the Nízke Tatry National Park with an area of 1830 km^2 has 49 taxa on the list (Dítě and Jasík 2009), Malé Karpaty Mountains has 48 taxa, Biele Karpaty Mountains has 55 taxa (Mráz 2003; Somlyay 2010; Figura 2013; Figura 2014; Dítě 2005; Wild *et al.* 2019).

According to the diversity map (Fig. 2), map quadrants to the west with lowland relief exhibit 1 – 6 species. The southern edge shows a lack of data, so the diversity map may not be accurately expressed. The greatest diversity is concentrated in quadrants 7075c, 7075d and 6877a where 35 – 38 taxa occur. This is a very rugged conservation area with diverse habitats (forest and non-forest) in the vicinity of Omšenie, Horná Poruba, Súľov-Hradná Village, Velký and Malý Manín Hill.

Species recorded

List of species occurring in the study area sorted by threat level (Eliáš *et al.* 2015):

Critically Endangered (CR)
Herminium monorchis.

Endangered (EN)
Epipactis futakii, *E. greuteri*, *E. leutei*, *E. placentina*, *Himantoglossum adriaticum*, *Orchis spitzelii*.

Vulnerable (VU)
Anacamptis pyramidalis, *Ophrys apifera*, *O. holubyana*, *Epipactis neglecta*, *E. pseudopurpurata*.

Near Threatened (NT)
Anacamptis morio, *Cephalanthera damasonium*, *C. longifolia*, *C. rubra*, *Cypripedium calceolus*, *Dactylorhiza fuchsii* subsp. *fuchsii*, *D. fuchsii* subsp. *sooiana*, *D. incarnata* subsp. *incarnata*, *D. lapponica*, *D. majalis*, *D. sambucina*, *D. viridis*, *Epipactis albensis*, *Epipactis distans*, *E. komoricensis*, *E. muelleri*, *E. palustris*, *E. purpurata*, *E. tallosii*, *Epipogium aphyllum*, *Goodyera repens*, *Gymnadenia densiflora*, *G. odoratissima*, *Limodorum abortivum*,

Malaxis monophyllos, Neotinea tridentata, N. ustulata subsp. *aestivalis, Ophrys insectifera, Orchis mascula* subsp. *signifera, O. militaris, O. pallens, Platanthera chlorantha, Traunsteinera globosa.*

Least Concern (LC)
Corallorhiza trifida, Epipactis atrorubens, E. helleborine subsp. *helleborine, E. microphylla, E. pontica, Gymnadenia conopsea, Listera ovata, Platanthera bifolia.*

Unclassified
Dactylorhiza ×aschersoniana, D. ×braunii, Epipactis ×meprediorum, E. atrorubens × E. muelleri, E. helleborine × E. muelleri, E. helleborine × E. pseudopurpurata, E. leptochila s.l. (incl. undescribed species), *Gymnadenia conopsea × G. odoratissima, Neottia nidus-avis, Ophrys ×devensis, Orchis ×canuti, O. ×loreziana, Platanthera ×hybrida.*

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Seasonal variability of physical and chemical properties of the water in lake Kolové pleso, the West Carpathians

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Abstract. This study examines the physical and chemical limnology of lake Kolové pleso in order to understand the response of the high mountain lake ecosystem to seasonal, climatic and anthropogenic impacts. We have combined regular monthly monitoring of physical properties on site and analyzed the chemical composition of the lake, using nutrient photometry, X-ray analysis of elements and COD methods. Fluctuations and concentrations of these variables confirmed that the lake is affected by seasonal fluctuations, subsoil, bioaccumulation of elements, vegetation and atmospheric deposition and climate variables. No potential pollution was detected by these indicators. The GC-MS method was used to qualitatively identify organic substances in the lake that were potentially harmful. PAHs (phenanthrene and pyrene) were found during the heating period (winter months). Due to the extensive rainfall in July 2018, during the experiment, we had the opportunity to observe the effect of flooding on the lake. Flooding has a significant impact on the organic composition of the lake (COD) as well as on some chemical elements (S, K, Rb, Mo, Cd), whose values dropped significantly following the flood. The decrease in COD values influenced the acidity of the lake negatively, as seen in the past.

Key words: high mountain lake, physical and chemical limnology, flood impact, organic pollutants

Introduction

Water is one of the most widespread compounds on Earth. It creates the conditions for the existence of all forms of life, and is a basic biological component and technological raw material (Hanušin 2009). Standing water is characterized by the absence of flow predetermining some of its physicochemical properties, which are significantly different from flowing water. Standing water includes lakes, and this study focuses on glacial lakes. Glacial lakes in the Tatras are called "plesá". They represent 90% of all lakes in Slovakia (Hrabě 1939, 1942; Bitušík *et al.* 2006) and are mostly oligotrophic standing waters with specific characteristics (Psenner 1989; Drever and Zobrist 1992; Beracko *et al.* 2014).

These ecosystems have been identified as key sites for studying global environmental change (Pienitz *et al.* 1997a). Despite their geographical isolation, without direct anthropogenic influences, due to their specific characteristics, alpine lakes are excellent indicators of seasonal changes, as well as increasingly frequent (IPCC 2001) climate change (Wathn *et al.* 1995).

That is why several studies conducted in Arctic region, as well as in Tatra high mountain lakes (e.g. Henriksen *et al.* 1992; Fott *et al.* 1994; Kopáček and Stuchlík 1994; Kopáček *et al.* 2000, Štefková and Šporka 2001; Grimalt *et al.* 2004; Kopáček *et al.* 2004; van Drooge *et al.* 2004; Stuchlík *et al.* 2006; Šporka *et al.* 2006; Kopáček *et al.* 2006; van Drooge *et al.* 2011, 2013), have been devoted to the limnology of such isolated lakes and to this issue.

These studies have shown that aquatic ecosystems are influenced by extensive global environmental changes (Wograth and Psenner 1995; Duff 1999). Research has indicated that global warming is likely to lead to an increase in water temperatures in lakes, a particularly important factor in aquatic ecology (Robertson and Ragotzkie 1990; Hondzo and Stefan 1993; De Stasio *et al.* 1996; Stefan *et al.* 1996; Šporka *et al.* 2006).

Because of fluctuating water levels, floods are notable biochemical events for lake ecosystems (McClain *et al.* 2003). Most lakes are heterotrophic, and dependent on organic inputs from their basins and subsoil (Sobek *et al.* 2007). Floods have the effect of rapidly mobilizing and accumulating organic carbon and nutrients (Nogueira *et al.* 2002; Wantzen *et al.* 2008), which can lead to "wash out" and also cause leaching of heavy metals into the environment from lake bottom sediments; posing a potential risk to water quality (Chrastný *et al.* 2005).

Many studies address the potential negative impact of organic substances accumulating in natural environments (Wania and Mackay 1995) as a result of their extensive use in industrial applications and combustion processes (Douglas and Smol 1994; Antoniades *et al.* 2003; Bruzzoniti *et al.* 2009). Pollutants tend to be transported in the atmosphere over long distances, (van Drooge *et al.* 2004; Landlová 2006; Morales-Baquero *et al.* 2013) and accumulate in isolated regions (Blais *et al.* 1998; Grimalt *et al.* 2001; Zennegg *et al.* 2003; Fernández *et al.* 2005; Meijer *et al.* 2006) such as the Tatra lakes (e.g. Grimalt *et al.* 2004; van

Drooge *et al.* 2004, 2011, 2013). In the High Tatras, which are part of the National Park, there are no industrial or agricultural activities. The closest industrial area is Košice in the south, Ostrava to the west and Polish factories (Krakow, Novy Targ, Zakopane) to the north; almost all of these are located dozens of kilometers from the mountains (van Drooge *et al.* 2004). Despite this, research has found mild PCB and PAH contamination (Grimalt *et al.* 2001; van Drooge *et al.* 2011).

These ecosystems represent the least disturbed inland water environments (Grimalt *et al.* 2001; Meijer *et al.* 2006). They are sensitive environmental indicators for determining changes in air quality and for the long-range transmission of pollutants (van Drooge *et al.* 2013).

Alpine lakes are likely to experience dramatic future physical, chemical and biological changes (Antoniades *et al.* 2003). Therefore, understanding the impact of past, present, and likely future anthropogenic influences and climate change depends on understanding the basic state of lakes (Hamilton *et al.* 2001; Michelutti *et al.* 2002a).

The aim of this study is, therefore, to collect data that will define the natural, present state of lake Kolové pleso, describe the impact of the flood on the lake, and identify and describe potential organic pollution. The data will serve as a reference for future programs for monitoring anthropogenic impacts and global environmental changes (Pienitz *et al.* 1997a).

Material and Methods

Study area

The Tatra Mountains are the highest mountain range in Slovakia. They are about 60 km long and 17 km wide and one-fifth of the range is situated in Poland. They are divided into two geomorphological units, the Western and Eastern Tatra Mts. The Eastern Tatras a further divided into the High Tatras and the Belianske Tatras (Štefková and Šporka 2001).

The valley of Kolová dolina, from the delta of the Kolový potok stream to the foot of Kolový štít peak, is about 2.5 km long. This area is between Jahnence and Bortky. The dominant mountain peaks within this valley are Kolový štít (2418 m a.s.l) and Jahňací štít (2229 m a.s.l) (www.spravatanap.sk 2012).

Fig. 1. Lake Kolové pleso – June 2018 (Photo: V. Ruček).

Lake Kolové pleso (Fig. 1) is situated in Kolová dolina valley at an altitude of 1565 m. It has an area of 18 280 m^2, a perimeter of 735 m, water volume of 10 846 m^3, length of 225 m, width of 123 m, a maximum depth of 1.2 m, and an average depth 0.59 m (Marček 1996).

Climatic conditions of the area

Many specific features (Table 2) that fundamentally determine the existence and development of natural ecosystems, but also environmental phenomena characterize the climate of the High Tatras. Sampling frequency was determined to capture the seasonal variability of the physical and chemical variables of the monitored lake.

Fieldwork

Samples were taken monthly between August 2017 and December 2018 (Table 1). This sampling frequency was determined to capture the seasonal variability of the physical and chemical variables of the monitored lake.

To measure physical parameters of the water samples including water temperature, salinity, pH, U (voltage), conductivity, TDS (total dissolved solids) (mg/l), soluble oxygen/(oxygen level) and saturation of the water, we used a portable multimeter WTW 3430 (GEOTECH, Weilheim, Germany). Along with the Multi 3430 we used compatible probes: IDS pH electrode Sen TixR 940-3, conductivity electrode TetraCon 925-3 and an optic oxygen electrode FDO 925-3.

Water samples were taken at 0.5 meters from the left shore of the lake. Samples were processed in the laboratory using gas chromatography. We used a glass container with one liter of content to

Month	Time	Conditions
August 2017	9:30	cloudy, cold, fog
September 2017	13:00	rain, cold
October 2017	12:30	cold, freeze
November 2017	11:30	ice, fog, snow
December 2017	13:00	ice, cloudly
January 2018	13:00	ice cloudly
February 2018	13:30	ice, pathy cloud, -25
March 2018	10:00	ice, sunny, warm
April 2018	8:30	without snow and ice, sunny
May 2018	13:00	sunny, without clouds
June 2018	12:00	cloudly, rain
July 2018	11:30	floods
August 2018	9:30	rain
September 2018	12:30	sunny, warm
October 2018	10:35	ice, first snow, sunny
November 2018	12:30	ice, sunny, without snow
December 2018	10:00	ice, snow

Table 1. Sampling tapping (August 2017-December 2018).

Month	Global radiation [W/m²]	Air temperature [deg. C]	Air humidity [%]	Wind direction	Precipitation [mm]	Wind speed (m/s)
August 2017	142.5	13.03	78.58	161.36	0.14	0.53
September 2017	84.7	7.03	86.54	163.67	0.3	0.7
October 2017	52.32	2.99	82.04	165.56	0.06	0.48
November 2017	20.0	-0.87	79.23	159.92	0.05	0.7
December 2017	9.35	-4.2	76.72	169.34	0	1.54
January 2018	20.57	-3.16	72.21	150.36	0	1.01
February 2018	18.13	-7.02	89.53	119.76	0	0.35
March 2018	102.56	-4.36	75.86	149.58	0	0.71
April 2018	153.78	7.07	63.12	150.94	0.14	1.31
May 2018	148.39	9.35	78.14	157.15	0.11	0.68
June 2018	100.84	10.18	92.29	159.96	0.22	0.41
July 2018	114.95	11.63	89.78	160.21	0.17	0.37

Table 2. Monthly measurements from a weather station in valley Kolová dolina (August 2017 - July 2018). Note: It should be taken into account that the weather station is not heated, therefore the precipitation totals are not registered exactly in the winter. It is because snow and ice accumulate in the rain gauge through all winter. Data of measurements from the weather station in valley Kolová dolina were kindly provided by the National Forest Center - Forest Research Institute Zvolen.

Fig. 2. Valley Zadné Meďodoly in the day of flood – July 18th, 2018 (Photo: V. Kapusta).

avoid sample contamination because of medium-flowing organic substances in the water. For the purposes of the subsequent analysis, other samples were placed into plastic containers containing 0.7 l. Before the sampling, all containers were properly labeled and disinfected. We were really careful about the proper transportation of the samples and their preservation, trying to keep intervals between sampling and analysis as short as possible. During sampling in the summer (18th of July 2018), the Tatra Mountains were hit by floods (Fig. 2).

Chemical oxygen demand (COD)

We used three boiling flasks with the same sample (100 ml sample of water from lake) to average results for more accurate measurements.

The method is based on the oxidation of organic substances with 20 ml potassium permanganate (K_2MnO_4) solution (0.002 mol/l), in 5 ml acidic sulfuric acid (H_2SO_4, 96%) in a dilution of (1:2) at 10 min boiling. We used 4-5 cooking stones for each boiling flask and a small clock glass was placed on the flask's throat. Oxidation occurs with excess permanganate. After completion of the oxidation, the unreacted $KMnO_4$ is reduced by the excess of standard oxalic acid ($(COOH)_2$) solution (0.005 mol/l), added in the exact quantity (20 ml) to the sample: $2MnO^-_4 + 5 (COOH)_2 + 6H^+ \rightarrow 2Mn^{2+} + 10CO_2 + 8H_2O$.

The solution in the boiling flask was completely decolorized to a clear solution. The clear solution was titrated back with potassium permanganate (0.002 mol/l) to $KMnO_4$ until it was stained a faintly pink color. Consumption at titration indicates the consumption of manganese to oxidize organic matter.

Photometry

We used photometry (optical - analytical method) to determine other chemical parameters of water quality (chlorides, sulfates, nitrates, ammonia, phosphorus, and contents of total hardness). The YSI EcoSense 9500 photometer and accessories compatible with this water analyzer determined concentrations of ions in our samples.

X-ray analysis

The x-ray method determines values of some chemical elements (trace elements). We used a handle ED-XRF spectrometer DELTA (Bas, Rudice, CZECH) and analyzed the water sample in a plastic vial. Samples were analyzed with x-ray beams for 80 seconds. Analytical methods and calibration practices used in the laboratory correspond with internationally accepted standards (Spectrapure Standards, Norway). Detections limits (X-ray) differ for different elements and fulfill the criteria described in the manual to Delta XRF.

Gas chromatography/mass spectrometry (GC-MS)

We used LLE: liquid-liquid extraction, a method commonly used for aqueous samples. In this extraction technique we mixed 1l water samples from the tarn with 3 ml of hexane. The sample was shaken for 2 minutes. The hexane layer was isolated on the top of the solution. The isolated extract was evaporated and re-filled with hexane (300 ml). Thus isolated, the sample can be analyzed using a gas chromatograph.

The separation of the components is based on analyte retention between the stationary - liquid phase and the mobile - gas phase. To identify the components of the mixture, the measured retention times are compared with the already known retention times obtained under the same conditions (Agilent Technologies 2013).

Statistics

For statistical analysis (Pearson´s correlation matrix, one way ANOVA and graphs) we used STATISTICA 8 software.

Results

Physical and chemical variables

Physical and chemical values from 12 samples collected between August 2017 and July 2018 are reported in Appendix 1 and Table 3.

Physical variables

Water temperature values are seasonally dependent (Comparison between two half a year samples - summer versus winter: $F(1.10) = 7.9815$, $p=0.01800$), falling in the summer months and increasing during winter. Based on our research, the water temperature was low all year round (Appendix 1) with a mean value of 5˚C. A maximum temperature of 14.1˚C was recorded in May and a minimum temperature of -0.1˚C was measured in December (Table 3). Water temperature also affects oxygen ratios (Appendix 2), which can be seen in the annual seasonal fluctuations when oxygen levels fall in the summer months and increases at lower temperatures in winter (Appendix 1). The dissolved oxygen values in the lake were high - mean 10.12 mg/l and 98.36% (Table 3). These values correlate negatively with those of titanium and potassium (Appendix 2). The mean pH of the lake was 7.5 with a range of 5.8 to 9.4 (Table 3). The pH also influences the concentration and solubility of some elements (Appendix 2) such as chlorine, potassium, and titanium. The highest pH values were measured in June - July, while the electric voltage, which is inversely proportional to pH (Appendix 2), decreases with temperature rise (Appendix 1). Positive correlation (Appendix 2) of molybdenum with electrical voltage is an interesting result. Electrical voltage had higher values in August, January, and February than in the other months, as did molybdenum, indicating this correlation (Appendix 2). Conductivity is directly proportional to ion concentration (TDS) and

Parameter	Mean	Median	SD	Minimum	Maximum	Detection limit & units
t	5.008	3.85	3.620	-0.1	14.1	**˚C**
pH	7.460	7.548	1.087	5.705	9.437	
U	-53.409	-40.65	35.867	-131.96	-0.33	**mV**
Concent. O_2	10.123	10.26	1.026	8.31	11.99	**mg/l**
Sat. O_2	98.359	99.7	5.777	88.33	110.47	**%**
O_2	172.521	172.7	8.821	155.10	193.10	**mbar**
Conductivity	15.148	13.38	6.563	7.70	27.23	**μS/cm**
TDS	15.066	13	6.675	8	28	**mg/l**
p	*78.926*	79.23	28.401	37.23	130.20	**kΩ*cm**
COD	3.375	2.82	2.664	0.82	10.59	**ml**
Cl^-	7.5	3.7	7.996	0.5	22.2	**0-50 mg/l**
NaCl	13.183	8.55	12.990	0.8	36.7	**0-50,000 mg/l**
tot. hardness $CaCO_3$	12.567	10	11.679	Det limit	40	**0-500 mg/l**
chlorides $CaCO_3$	11.958	7.3	11.026	0.7	31.3	**mg/l**
SO_4^{2-}	12.517	5.35	21.903	0.0	83.3	**0-200 mg/l**
S	4.217	1.85	7.449	0.0	28.3	**0- 200 mg/l**
ammonia N	0.209	0.095	0.256	Det limit	0.72	**0-1.0 mg/l**
PO_4^{3-}	0.107	0.075	0.103	Det limit	0.34	**0-4.0 mg/l**
P	0.035	0.025	0.034	Det limit	0.11	**0-1.3 mg/l**

Table 3. Measured limnological variables from lake Kolové pleso including mean, median, standard deviation (SD), minimum value, maximum value, and analytical detection limit. Det limit - measurement under the detection limit.

Parameter	Climatic variables									
	Global radiation [W/m²]		Air temperature [deg. C]		Air humidity [%]		Wind direction		Precipitation [mm]	
	r	p	r	p	r	p	r	p	r	p
t (° C)	0.5931	0.0421	0.611	0.0348						
U (mV)					-0.5771	0.0494				
O_2 (mbar)	0.5788	0.0487								
Conductivity (µS/cm)			-0.7356	0.0064			-0.6825	0.0145	-0.6824	0.0145
TDS (mg/l)			-0.7592	0.0042			-0.7193	0.0084	-0.6757	0.0159
p (kohm*cm)			0.7147	0.009					0.6617	0.0191
Cl (ppm)	-0.7139	0.0091	-0.695	0.0121					-0.591	0.043
K (ppm)	-0.7488	0.0051	-0.6445	0.0237						
Ti (ppm)	-0.6225	0.0307								
Sn (ppm)									0.5796	0.0483
Sb (ppm)			0.5766	0.0497						

Table 4. Correlation coefficients of selected climatic variables. Only significant values ($p<0.05$) are listed. Climatic variables were kindly provided by the National Forest Center - Forest Research Institute Zvolen.

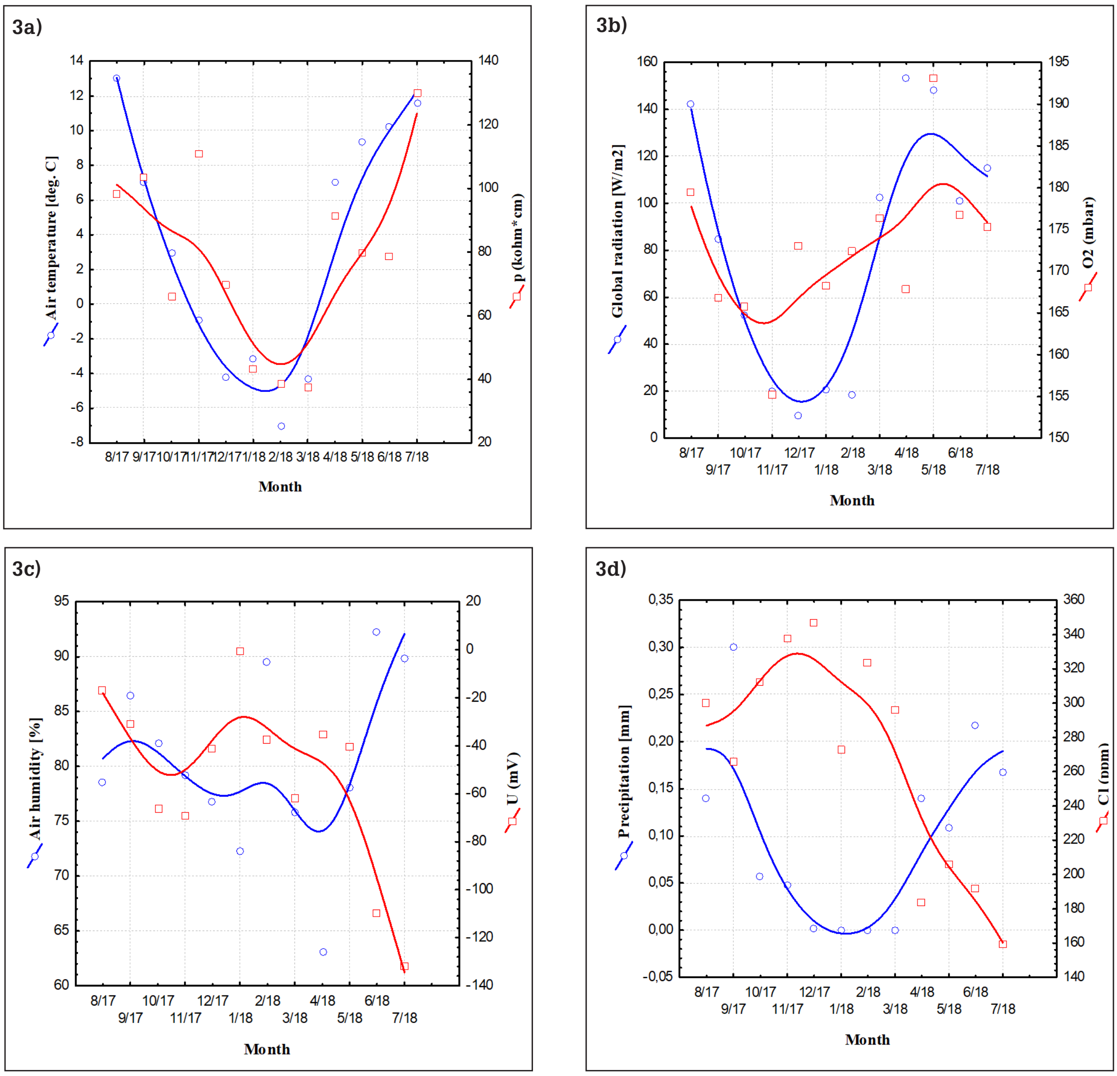

Fig. 3. Fluctuating of multiple variables (negative exponential smoothing) of the lake with climatic variables during the year. The fluctuating of global radiation in conjunction with O_2 (mbar) (3a), air temperature in conjunction with resistance (3b), the air humidity in conjunction electrical voltage (3c), precipitation in conjunction with chlorine (3d), and wind direction in conjunction with conductivity values with respect to the month.

is the inverted value of the electrical resistance of the water (Appendix 2 - high values of the same sign). Seasonally interdependent variables (Comparison between two half a year samples - summer versus winter: Resistance: One way ANOVA: $F(1.10) = 6.6758$, $p=0.02724$, Conductivity: One way ANOVA: $F(1.10) = 6.4953$, $p=0.02893$, TDS: One way ANOVA: $F(1.10) = 7.2196$, $p=0.02282$). Conductivity and TDS values were higher in the winter months (January-March), and dropped during summer months (July- September) (Appendix 1). Resistance negatively correlated with these values during the year (Appendix 2). Salinity showed zero values throughout the experiment.

Nutrients

Four of the 13 original environmental variables (N-NH_4, N-NH_3, NO_3^- and N) were not included in the statistical analysis because they were commonly found outside the detection boundaries. Nutrient concentrations were lower and in some months they were below the detection limit (Appendix 1). Chloride values gradually increased in winter (November - April) and decreased between spring and winter (May - October) (Appendix 1). The most widespread chloride (NaCl) values ranged from 0.8 - 36.7 mg/l (Table 3). Total hardness has a fluctuating tendency (Appendix 1). Peaks occurred in November, January and May and the maximum value was 40 mg/l (Table 3). The lowest values were observed in September, December and March. In Appendix 1, we can see that the sulfates gradually increased in direct proportion to the total hardness during the year. With the exception of one peak in August, when sulfates reached the highest value of 83.3 mg/l (Appendix 1). Phosphate values were higher in the months of October, November, January, and June, and decreased during summer (July-September) (Appendix 1). The highest value was measured in June at 0.34 mg/l (Table 3). Ammonia (N) values gradually increased in direct proportion to the sulfur values during the year. Ammonia (N) had higher values in April, June, November, and December (Appendix 1) than in other months, as did sulfur, suggesting a positive correlation between them (Appendix 2). The highest COD values were observed in the autumn (August-November), and decreased in the following months (Appendix 1). During the year, COD values were low, with the exception of one maximum value of 10.59 ml (Table 3) in November 2017.

Chemical elements

Potassium is seasonally affected (Comparison between two half a year samples - summer versus winter: One way ANOVA: $F(1.10) = 12.999$, $p=0.00480$) and had higher values in the winter months (October - February) (Table 3) than in the summer months (April - August). It has a similar fluctuating tendency to titanium, suggesting a positive correlation between them (Appendix 2). Appendix 2 shows the positive relationship between titanium, NaCl and chlorides ($CaCO_3$). The decrease in the second alkali metal measured in the lake - rubidium (Appendix 1) was recorded at the beginning of spring and summer (March - April, and July) and the highest values were observed from August to February. A smaller decrease in molybdenum values was recorded at the beginning of winter and a more pronounced decrease was recorded in spring (March - July). The highest values were in August, January, and February (Appendix 1). Tin values peaked in September, January, and June (Appendix 1) though in other months, the values were lower. Antimony is affected by temperature (Appendix 2) and thus by season (Comparison between two half a year samples - summer versus winter: One way ANOVA: $F(1.10) = 6.6568$, $p=0.02741$). In summer it gradually increased (Appendix 1). During the winter season it declined between September and April. Chlorine had higher values in winter (October - March) (Appendix 1) and lower values in summer months (April - July), similar to potassium, suggesting a positive correlation between them (Appendix 2) as well as a seasonal dependence (Comparison between two half a year samples - summer versus winter: One way ANOVA: $F(1.10) = 15.518$, $p=0.00278$). Sulfur has a fluctuating tendency (Appendix 1); peaking in November and December as well as between April and June, while in recent months the values were lower.

Direct climate variables also affect the lake, which can be seen in many correlations with lake properties (Table 4). Global radiation positively correlates (Table 4) with water temperature and O_2 (mbar), as seen in the same fluctuations during the year (Fig. 3b). This climate indicator negatively correlates (Table 4) with the chemical composition of the lake, namely the values of chlorine, titanium, and potassium. The temperature as a major climate variable affects the negative conductivity, TDS and chlorine and potassium correlations (Table 4). However, the correlation with resistance is positive, which can be seen in the same fluctuations during the year (Fig. 3a). As shown in Figure 3c, air humidity negatively correlates with electrical voltage. The amount of precipitation correlates favorably with resistance and tin and negative with conductivity, TDS (Table 4) and chlorine in water (Fig. 3d). The dependence of conductivity and TDS on the weather is shown in Table 4.

Effects of flooding

In July of 2018 the High Tatras experienced an extensive flood and significant decrease in concentration was observed in sulfur (S), molybdenum (Mo), potassium (K) and rubidium (Rb) values (Table 5 and Fig. 4a-d). The other mentioned measured elements were unchanged.

The second impact of the flood was on the organic composition of the lake (COD). As we saw with element concentration, COD values (Fig. 5a) also decreases following the flood. The difference is that the decrease in COD was also observed on the day of the flood and thus in the sample from July 18, unlike the other elements whose values fell during the following month, as can be seen in Table 5. However, with the decrease in COD during the summer, pH values (Fig. 5c) are increasing. In July, 2018, values between 7.7 to 9.4 were measured. The highest pH values over the entirety of the sampling pe-

Month	COD (ml)	S (ppm)	K (ppm)	Rb (ppm)	Mo (ppm)	Cd (ppm)
August 2017	6.25	76 ± 64	187 ± 12	1.6 ± 0.6	1.3 ± 0.5	Det limit ± 6
September 2017	4.7	Det limit ± 62	196 ± 12	1.6 ± 1.1	1.1 ± 1	Det limit ± 6
October 2017	2.79	59 ± 48	210 ± 10	1.2 ± 0.8	1.1 ± 0.8	Det limit ± 4
November 2017	10.59	96 ± 67	252 ± 15	1.5 ± 0.5	1.1 ± 1	Det limit ± 6
December 2017	3.27	93 ± 34	193 ± 13	1.5 ± 0.6	Det limit ± 1	Det limit ± 6
January 2018	0.93	Det limit ± 56	203 ± 11	1 ± 1	1.3 ± 0.9	Det limit ± 5
February 2018	0.92	Det limit ± 57	215 ± 12	1.2 ± 0.5	1.3 ± 0.4	Det limit ± 5
March 2018	2.18	Det limit ± 63	187 ± 13	Det limit ± 1.1	Det limit ± 1	Det limit ± 6
April 2018	2.85	196 ± 145	134 ± 9	Det limit ± 0.4	Det limit ± 0.1	Det limit ± 6
May 2018	1.8	Det limit ± 138	152 ± 9	1.3 ± 0.2	Det limit ± 0.1	Det limit ± 6
June 2018	3.41	187 + 144	133 ± 8	1 ± 0.4	Det limit ± 0.1	Det limit ± 6
18th of July 2018	**0.82**	Det limit ± 137	137 ± 8	Det limit ± 0.4	Det limit ± 0.1	Det limit ± 6
August 2018	**0.8**	16 ± 12	1 ± 0.1	Det limit ± 0	Det limit ± 0	Det limit ± 0
September 2018	**0.91**	Det limit ± 11	1 ± 0.1	Det limit ± 0	Det limit ± 0	Det limit ± 0
October 2018	**0.66**	Det limit ± 12	1.1 ± 0.1	Det limit ± 0	Det limit ± 0	Det limit ± 0
November 2018	**0.98**	Det limit ± 12	1.1 ± 0.1	Det limit ± 0	Det limit ± 0	Det limit ± 0
December 2018	**0.77**	13 ± 6	1 ± 0.1	Det limit ± 0	Det limit ± 0	Det limit ± 0

Table 5. Average amount of chemical elements (and COD amount) ± displayed error (deviation from measurement) during a one and half year experiment. Det limit - measurement under the detection limit.

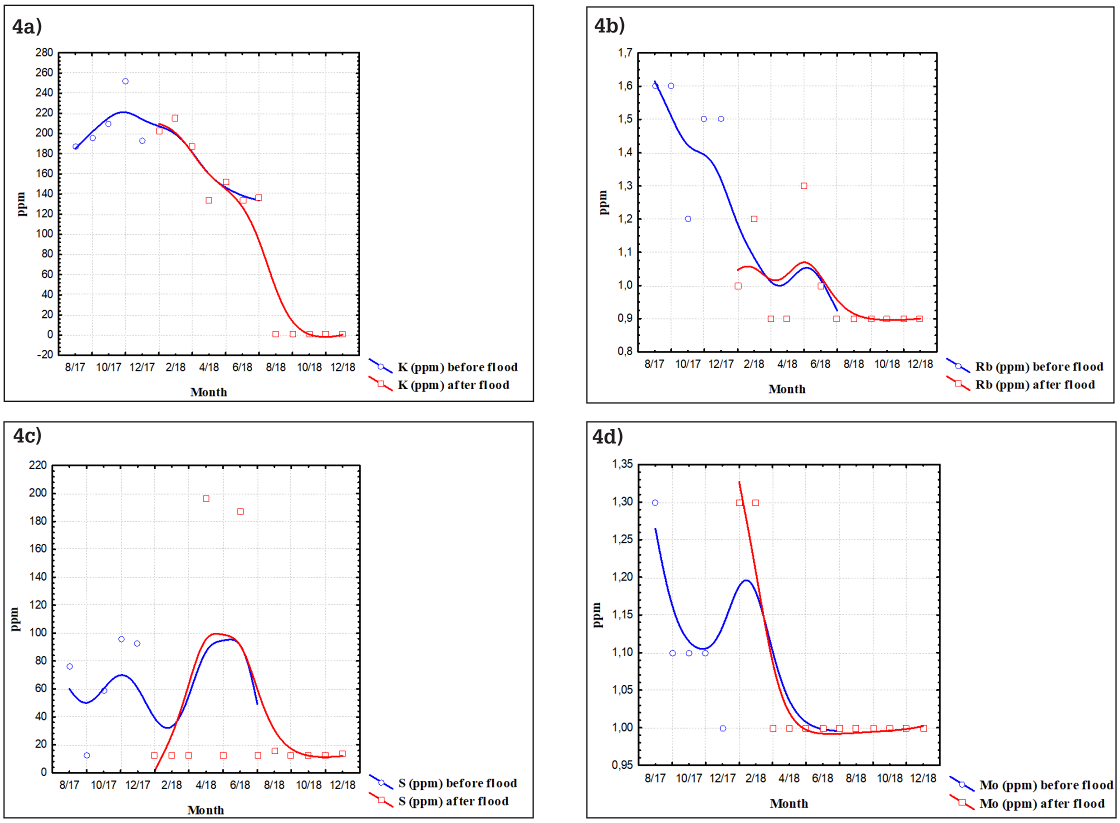

Fig. 4. The fluctuation of elements (K, Rb, S, Mo) prior to the flood (blue line) and post-flood change (red line) in July 2018. A decrease in the amount of potassium (4a), rubidium (4b), sulfur (4c) and molybdenum (4d) after July of 2018.

riod was also on the day of the flood. This negative correlation (pH: COD (ml): r = -0.6003; p = 0.0108) indicates that pH (Fig. 5b) is affected by flood and can be seen in Fig.5c and Fig. 5d. For other measured physical and chemical variables, no changes due to floods were observed.

Organic pollutants

All organic compounds found in the lake and the months in which they were measured are presented in Table 6, except for the phthalates that originated from the laboratory contamination and are therefore

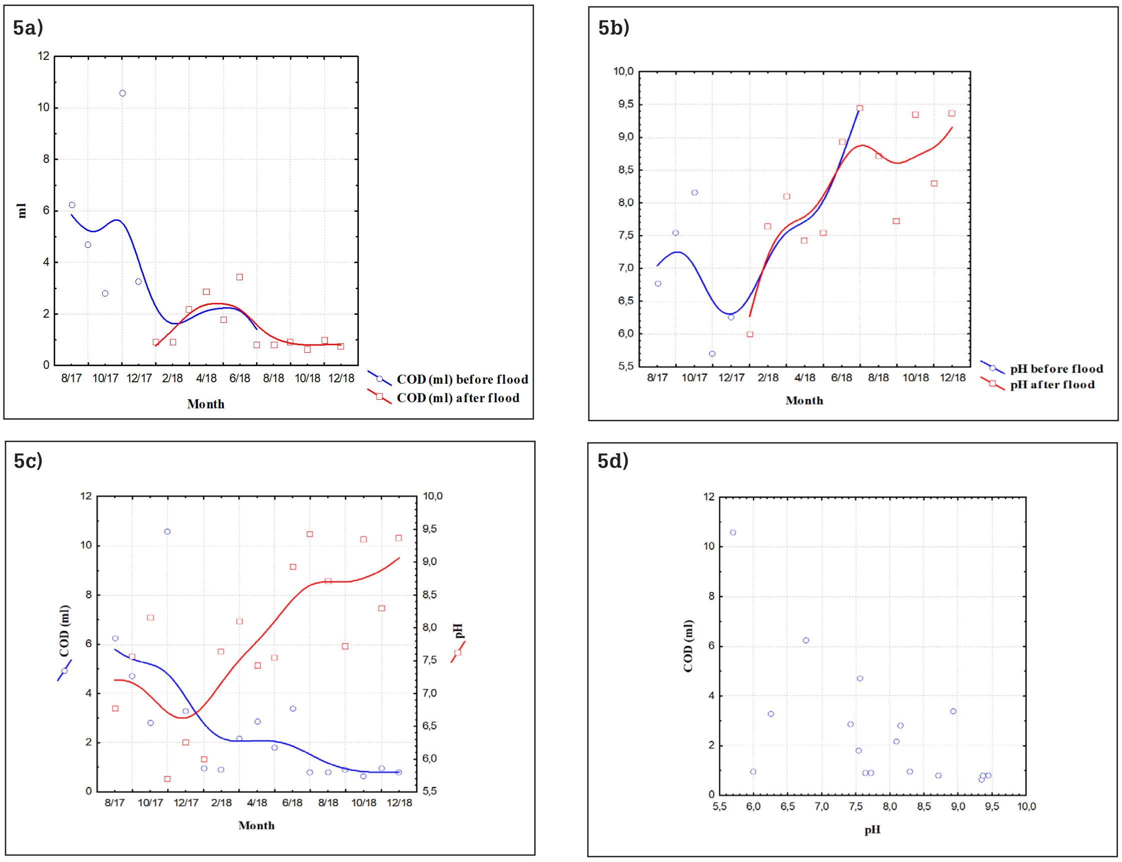

Fig. 5. Changes in COD and pH values due to floods and their interdependence. The fluctuation of values of COD and pH prior to the flood (blue line) and post-flood change (red line) in July 2018 is shown in figures 5a and 5b. Figures 5c and 5d show the correlation (pH: COD (ml): $r = -0.6003$; $p = 0.0108$) of pH values against the COD, during an experimental period.

Organic compounds	Formula	RT	Month 2017	Month 2018
1-Decanol, 2-hexyl-	$C_{16}H_{34}O$	13.6	August	February
1-Dodecanol	$C_{12}H_{26}O$	16.5	-	May
4-Methyl-2,4-bis(p-hydroxyphenyl) pent-1-ene, 2TMS d.	$C_{24}H_{36}O_2Si_2$	18.3	-	November
6-Hepten-3-one, 5-hydroxy-4-methyl-	$C_8H_{14}O_2$	3.1	October	February, June, October
Borinic acid, diethyl-	$C_4H_{11}BO$	4.6	October	January, June, October
Carbonic acid, eicosyl vinyl ester	$C_{23}H_{44}O_3$	12.5	August, December	January, February
Dotriacontane, 1-iodo-	$C_{32}H_{65}I$	13.5	August	February
Eicosane, 1-iodo-	$C_{20}H_{41}I$	12.5	-	January
Eicosyl octyl ether	$C_{28}H_{58}O$	19.3	-	February
Fumaronitrile	$C_4H_2N_2$	3.3	-	November
Hexadecane, 2,6,11,15-tetramethyl-	$C_{20}H_{42}$	13.5	-	January
i-Propyl 14-methyl-pentadecanoate	$C_{19}H_{38}O_2$	20.4	-	January, March
Isopropyl palmitate	$C_{19}H_{38}O_2$	20.4	-	May
Phenanthrene	$C_{14}H_{10}$	14.2	December	January, February
Pyrene	$C_{16}H_{10}$	21.1	December	-
Sebacic acid, but-2-enyl propyl ester	$C_{17}H_{30}O_4$	15.8	-	January
Sulfurous acid, 2-ethylhexyl tetra-decyl ester	$C_{22}H_{46}O_3S$	13.9	-	January

Table 6. List of detected organic compounds in analyzed samples of water during August 2017 – December 2018 from lake Kolové pleso. RT – retention time.

not reported in the results. In September of 2017, November of 2017, April of 2018, July of 2018, August of 2018, September of 2018, December of 2019 no organic compounds were found.

Compounds from all analyzed samples belong to the chemical groups of carboxylic acids, acid esters, ethers of various compounds, alcohols, isoprenoids, PAH and others. In Table 6, we can see that some potentially polluting organic substances were measured in several months during the experimental period. Most of the substances were found in the winter months, when the lake is covered with snow, namely substances from the group PAHs (Phenanthrene; Pyrene), esters (Sebacic acid, but-2-enyl propyl ester; Sulfurous acid, 2-ethylhexyl tetradecyl ester), isoprenoids (Hexadecane, 2,6,11,15-tetramethyl; i-propyl 14-methylpentadecanoate), ethers (Eicosyl octyl ether; Fumaronitrile; Eicosyl octyl ether), but also Eicosane, 1-iodo. Substances analyzed in both summer and winter are from the group of fatty alcohols (1-Decanol, 2-hexyl-) and acids (Dotriacontane, 1-iodo; Carbonic acid, eicosyl vinyl ester). Isopropyl palmitate was measured only in May as well as 1-Dodecanol. Interestingly, acids (6-Hepten-3-one, 5-hydroxy-4-methyl-; Borinic acid, diethyl) were detected in the same months (October, January, and February).

Discussion

Physical and chemical variables

In alpine lakes such as Kolové pleso, water temperatures have very low values throughout the year (Appendix 1), even during periods when the lake is not frozen (Juriš *et al.* 1965; Šporka *et al.* 2006). Although water temperature is dependent on the season, even in summer months the water temperature does not significantly exceed 10° C (Juriš *et al.* 1965). The average water temperature in Kolové pleso was 5° C (Table 3) and during the measurement period, it only rose above 10° C once, and reached a maximum measured value of 14.1° C in May (Table 3). The thermal balance of the lake depends on global radiation absorption (Table 4) and heat exchange with air (Edinger *et al.* 1968; Sweers 1976). Water temperature of the lake has often followed the air temperature closely (Table 4), according to several studies (e.g., McCombie 1959; Edinger *et al.* 1968; Webb 1974; Sweers 1976; Shuter *et al.* 1983; Marti and Imboden 1986; Livingstone and Imboden 1989; Douglas and Smol 1994; Lister *et al.* 1998; Livingstone and Lotter 1998; Kettle *et al.* 2004; Šporka *et al.* 2006). Surface water temperature of the lake more faithfully reflects air temperature during the warmer months (Livingstone *et al.* 1999, 2005; Šporka *et al.* 2006). The highest temperature value (14.1° C) (Table 3) was measured on the warmest day (13.22° C) according to weather station data (weather station data APVV-16-0325 - NFC). Water temperature is also important for the assessment of oxygen ratios (Doláková and Janýšková 2012), as oxygen ratios depend on water temperature (Appendix 2). Saturation in the lake increases in winter and its decrease is caused by the temperature increase in summer (Appendix 1) (Sedláková and Halabuk 2003; Judová *et al.* 2015). The intensity of global radiation (Table 4 and Fig. 3b) also affects dissolved oxygen, which in turn affects photosynthesis in water, as the processes are interrelated. Oxygen concentration in Kolové pleso was around 10.12 mg/l (Table 3) and the average saturation was 98%, which is characteristic for chemically pure waters (Diviš 2008) such as Tatra Mountain lakes (Juriš *et al.* 1965). Based on these parameters, particularly pH values, we can classify the Kolové pleso among oligotrophic lakes (Beracko *et al.* 2014). pH during the analyzed annual cycle (Appendix 1) ranged from 5.7-9.4 (Table 3), which are typical values for this type of lake (Douglas and Smol 1994; Antoniades *et al.* 2000; Hamilton *et al.* 2001; Lim *et al.* 2001; Michelutti *et al.* 2002a, b; Kopáček *et al.* 2006). In these circumstances, pH is most influenced by rock composition (Fyles 1963; McNeely *et al.* 1979; Michelutti *et al.* 2002a) and its values affect ion concentration (Faure 1991; Hamilton *et al.* 2001), which is likely why pH correlates with a number of elements (Appendix 2) measured in the lake. The electrical voltage as an inverted value of pH has a negative effect in the lake (Appendix 2), specifically on the bioaccumulation capacity (Orolínová 2009) of molybdenum in winter months. Other seasonally dependent physical parameters of the lake include conductivity, TDS and resistance. These values are inverted, (Appendix 2) (Doláková and Janýšková 2012; Judová *et al.* 2015) and were also influenced by climatic variables such as air temperature (Table 4 and Fig. 3a) (Tölgyessy *et al.* 1984; Doláková and Janýšková 2012), precipitation and wind conditions (Table 4). Values are (Appendix 1) below the limit (The law 296/2005 Coll. Requirements for surface water quality and water pollution limits), because of the oligotrophy of the lake.

Similarly to studies by Douglas and Smol (1994); Antoniades *et al.* (2000); Lim *et al.* (2001); and Michelutti *et al.* (2002a, b), measurements showed nutrients in the lower range of recorded values. COD (Table 3) concentrations in the alpine environment range from 0.6 to 10 ml (Pienitz *et al.* 1997b). Low values (Appendix 1) and limited range are related to an almost complete absence of vegetation and poor drainage in the area (Antoniades *et al.* 2003). Sulfate concentrations in alpine lakes are generally influenced by sedimentary subsoil (Oswald and Senyk 1977; Pienitz *et al.* 1997b). Due to the oxidation of minerals, they increased from October to March (Appendix 1) and subsequently decreased as a result of reduced conditions (Wograth and Psenner 1995). Sulfate values were low (Appendix 1), implying nearly complete regeneration from events taking place in the 80's (Stuchlík *et al.* 1985; Kopáček and Stuchlík 1994; Kopáček *et al.* 2001; Evans *et al.* 2001; Veselý *et al.* 2002). The solubility of sulfuric minerals is a property of water hardness (Howard-Williams and Vincent 1989; Howard-Williams *et al.* 1989; Hamilton *et al.* 2001) and can be influenced by the trend that sulfates in the lake Kolové pleso grow in direct proportion to total water hardness (Appendix 1). Phosphate concentration (Appendix 1) (Wetzel 1983; Hobbie 1984; Pienitz *et al.* 1997a) is also related to the catchment area (Kopáček *et al.* 2006) of the lake. These nutrients are in the range typical for oligotrophic lakes (Wetzel 2001) and values were near to of below the detection limit (Appendix 1 and Table 3), (Juriš *et al.* 1965; Kopáček *et al.* (2006). Nitrate levels in the lake are also at or near the detection level (Rühland and Smol 1998; Hamilton *et al.* 2001; Kopáček *et al.* 2006). Ammonia nitrogen, as an important indicator of water pollution (Doláková and Janýšková 2012), is below or near the analytical value in high-altitude clean lakes (Appendix 1) (Pienitz *et al.* 1997b). The content of ammonia nitrogen (Table 3) is in the range of 0.01-0.72 mg/l for Kolové pleso and is similar to other Tatra lakes (Juriš *et al.* 1965; Kopáček *et al.* 2006).

Ionic composition of Kolové pleso depends on several factors. Most ions and especially trace elements are in very small concentrations in the lake, or are below the detection limit (Appendix 1), which is typical. (Hamilton *et al.* 2001). Lake chemistry is clearly related to climatic variables. As a result of precipitation (Welch and Legault 1986; Pienitz *et al.* 1997a; Kopáček *et al.* 2006), but also due to wind influence (Marchetto *et al.* 1995; Kamenik *et al.* 2001), concentration of elements in the lake (Table 4) and their conductivity were changed. Precipitation (Kerekes 1975) can reduce the content of chloride compounds in the lake (Table 4, Fig. 3d). Another factor that influences the chemical composition of the lake is the geochemistry of the bedrock (Hutchinson 1957; Wetzel 1983) and thus mineralogy (Kamenik *et al.* 2001), which is related to the concentration of sulfate anions and ammonia (Kopáček *et al.* 2006). This also explains the correlation between them (Appendix 2). High continuity (Appendix 1 and Appendix 2) and concentration of potassium and chlorine elements indicate that they are the most commonly (Kerekes 1973, 1975) found in the lake in the form of chloride minerals (chloride salts) such as sylvite, KCl, or similarly to other water bodies, common rock salt (NaCl) (Muck 2006). Dependence of these major ions (K, Cl) both seasonal (Vondrka *et al.* 2013), and due to temperature can be explained by less salt measured in winter than in warmer months. Chlorine levels (Appendix 1) rise in autumn and winter due to chloride leakage from degrading vegetation (Psenner and Catalan 1994; Kamenik *et al.* 2001; Mikuš 2012). The same effect is also observed with potassium (Prentiki *et al.* 1980; Michelluti *et al.* 2002a). However, other water-soluble minerals (Petránek 1993) also accompany major elements dissolved in water. Common chlorides include titanium tetrachloride ($TiCl_4$) and titanium chloride ($TiCl_3$) (www.britannica.com 2019) as seen in the correlation of Titanium with chlorides (Appendix 2). Other elements are also subject to seasonal effects. Molybdenum dependence is due to its significant bioaccumulation capacity (Orolínová 2009), which in winter, due to weaker current (Appendix 1,2) has better conditions for accumulation and has higher values.

Effects of flooding on the lake

Increasingly frequent phenomena (IPCC 2001) such as extreme rainfall and consequently flooding, affected lake Kolové pleso in July 2018, giving the opportunity to observe the way water level fluctuations and flooding affect the alpine lake. The first effect was observed in the decrease of COD values (Fig. 5a) - total organic matter immediately on the day of flood events (Table 5). At the present stage of knowledge and based on studies of water fluctuations and its effects (Junk *et al.* 1989; Tockner *et al.* 2000; Nogueira *et al.* 2002; Junk and Wantzen 2004), we can only speculate that such a fluctuation is caused by a higher density of dissolved sediments. Due to the flood, these substances were precipitated on land and stored in ATTZ (Grossart and Simon 1998). The behavior of the lake, and its COD levels has thus shown that floods mobilize and accumulate organic nutrients in the lake (Keddy and Fraser 2000; Nogueira *et al.* 2002; Coops *et al.* 2003; Mooij *et al.* 2005; Wantzen *et al.* 2008) which are probably washed out and stored on land. We believe that this effect also caused the observed decrease (Table 5, Fig. 4a-d) in some element values and their displayed errors (measurement deviation) (explained in Materials and Methods). The decrease in element values and their deviations likely occurred due to washout following the flood, as more elements were left behind as part of the mineralization process (inorganic compounds). The instrument measures more accurately for values that bind to inorganic compounds than organic, which affects the resulting measurement error. As already mentioned in the results, the deviation for cadmium was also reduced after the flood, although it had values below the detection limit at all times. This finding is also confirmed by the work of Chrastný *et al.* (2005) whose study of the impact of floods on heavy metals revealed that floods may in particular cause the release of cadmium into the environment, as well as other elements (especially their organic compounds). In connection with the decline in COD values, we observed another interesting fact in the results after the flood situation. With decreasing COD values, pH (Fig. 5c) values increased (pH: COD (ml): $r = -0.6003$; $p = 0.0108$, Fig. 5d), which suggests the importance of organic substances for acidic lakes. This negative correlation has been observed in the past by acidification (Donahue *et al.* 1998; Evans and Monteith 2001; Kopáček *et al.* 2003; Kopáček *et al.* 2006). In such poorly polluted surface waters, the pH is most influenced by sedimentation (Fyles 1963; McNeely *et al.* 1979; Michelluti *et al.* 2002b) and precipitation (Judová *et al.* 2015). The highest pH value was measured on the day of the flood when extreme rainfall occured. Harriman and Taylor (1999) point out in their work that, regardless of the cause, the rising values of organic composition will have a significant impact on the lake's acidity (Fig. 5b), as floods and precipitation have also affected the acidity of lake Kolové pleso, with the opposite effect.

Potential organic pollution in the lake

Due to the low solubility of POPs and their extremely low concentration, it is difficult to determine these substances in aqueous samples (Bruzzoniti *et al.* 2009), which is why most studies around the world focus on the analysis of these substances in sediments (Borghini *et al.* 2005; Appleby and Piliposian 2006; Meijer *et al.* 2006; Evenset *et al.* 2007; Pozo *et al.* 2007; Schmidt *et* *al.* 2011). For this reason, our analysis in water samples was only qualitative. Analysis of samples from the lake detected several organic compounds from different chemical groups (Table 6). The main external factor affecting the amount of measured organic substances entering the lake systems is precipitation (Pozo *et al.* 2007). Snow effectively traps organic pollutants, making them easier to detect in the winter months in areas such as the Tatra lakes that experience a high volume of precipitation in the form of snow (Wania *et al.* 1998, 1999; Arellano *et al.* 2011). In most cases, these substances are not included in the lists of priority pollutants (Wania and Mackay 1995) because of their low levels or lack of toxicity and small spread around the world. However, PAHs classified as mutagenic, carcinogenic and teratogenic compounds have also been qualitatively analyzed (Pérez-Cadahía *et al.* 2004; Oliveira *et al.* 2012) and can be rapidly absorbed by both organic matter and water (Mitra *et al.* 1999). The first PAH substance detected is phenanthrene, a substance that is highly toxic to the environment, especially for water, because it accumulates in aquatic organisms. The second is pyrene, which is one of the most widespread pollutants in aquatic environments (Oliveira *et al.* 2012). These substances arise as a result of pyrolytic processes such as imperfect combustion (they are part of coal tar) and industrial activity (WHO 1987). A potential source for this pollution are industrial areas such as Košice in the south, Ostrava in the west and Polish factories (Krakow, Nowy Targ, Zakopane) (van Drooge *et al.* 2004). These industrial areas are a substantial distance from the Tatra mountains, but due to the properties of these substances and their ability to be transported over long distances atmosphereically (van Drooge *et al.* 2004; Morales-Baquero *et al.* 2013), they still pollute these areas (Landlová 2006). The lake Kolové pleso is located in the most remote area of the High Tatras, but due to its altitude it has a similar effect to the global distillation effect (Grimalt *et al.* 2001; Meijer *et al.* 2006). Additionally, the High Tatras mountain range constitutes a natural barrier to air flow due to its orientation. The Northwestern sides, where the lake is located, have higher rainfall loads due to exposure to North Atlantic air (Zasadni and Klapyta 2009). These substances in the form of wet deposition enter the mountain ecosystems and thus the lake (www.scientica.sk 2012). PAHs substances were measured in the months when the heating effect and wind is strongest (December 2017, January 2018, February 2018) (www.shmu.sk 2019). Other substances that have been measured and may cause a potential risk of contamination are Dodecanol, which is harmful to aquatic organisms (Noweck and Grafahrend 2006); toxic fumaronitrile - LT2300000 ($C_4H_2N_2$); and isopropyl palmitate ($C_{19}H_{38}O_2$) (Table 6). However, we do not know their sources. Our measurements, although only qualitative, have been confirmed by recent studies on contamination in European alpine regions (Blais *et al.* 1998; Grimalt *et al.* 2001; Zennegg *et al.* 2003; Fernández *et al.* 2005; Meijer *et al.* 2006) as well as in the High Tatras specifically (Grimalt *et al.* 2004; van Drooge *et al.* 2004, 2011, 2013).

Conclusions

Our research has confirmed that monitoring water quality and the impact of seasonal and climate change on isolated high mountain lakes is important in the context of global climate change. Based on the physical properties and chemical composition of the lake, we have been able to observe its seasonal changes, confirming some well-known ideas about how alpine lakes function during the year. These changes will be very important for the aquatic environment in the future. Due to the rise in water temperature from global warming or water level fluctuations, and the impact of floods, we can see an effect on the organic composition of the lake, and measure concentrations of heavy metals leached into the environment. The effects of industy on the environment, including the deposition of compounds like PAH (pyrene and phenanthrene) are also becoming an increasingly serious issue. This study defines the natural present state of lake Kolové pleso, which will serve as a reference for future programs for monitoring anthropogenic impacts and global environmental changes.

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Appendix 1. Measured limnological variables from lake Kolové pleso during from year period (August 2017 – July 2018). Det limit - measurement under the detection limit.

	Aug.17	**Sep.17**	**Oct.17**	**Nov.17**	**Dec.17**	**Jan.2018**	**Feb.18**	**Mar.18**	**Apr.18**	**May 18**	**Jun.18**	**Jul.18**
t (° C)	6.5	7.7	2	2.3	-0.1	3.8	3.9	3.7	2.03	14.1	6.6	7.6
pH	6.766	7.553	8.164	5.705	6.246	6.000	7.648	8.101	7.427	7.543	8.933	9.437
U (mV)	-16.87	-30.65	-66.27	-69.5	-41.07	-0,333	-37.433	-61.47	-35.20	-40.23	-109.93	-109.93
Concent. O_2 (mg/l)	10.22	8.31	8.59	9.25	11.99	10.54	10.81	10.93	11.05	9.38	10.30	10.11
O_2 (mbar)	179.37	166.90	165.67	155.10	172.97	168.233	172.43	176.40	167.93	193.10	176.86	175.30
Sat. O_2 (%)	102.50	94.80	89.63	88.33	99.27	96.66	100.13	101.63	94.60	110.47	101.06	101.23
Conductivity (μS/cm)	10.23	9.55	15.27	9.03	14.23	23.267	26.567	27.23	10.87	15.30	12.53	7.70
TDS (mg/l)	9.5	9.5	15.33	9	14.33	23	28	26.67	11	13	13	8
p (kΩ*cm)	98	103.55	65.77	111.03	69.97	43.233	38.467	37.27	91.17	79.6	78.86	130.2
COD (ml)	6.25	4.7	2.79	10.59	3.27	0.93	0.92	2.18	2.85	1.8	3.41	0.82
Cl - (mg/l)	6.4	0.63	0.9	22.2	0.9	17	3.2	21.5	11.16	1.43	4.2	0.5
NaCl (mg/l)	10.2	1.23	1.5	36.7	1.5	28	15.8	35	18.5	2.1	6.9	0.8
chlorides $CaCO_3$ (mg/l)	8.7	18.33	1.3	31.3	1.3	24	4.53	30	15.66	1.8	5.9	0.7
tot. hardness $CaCO_3$ (mg/l)	10	0.9	6.6	40	1.7	30	10	Det limit	11.6	20	15	5
SO_4^{2-} (mg/l)	83.3	6.66	4	18.6	8	4	9	10.6	3	0	3	0
S (mg/l)	28.3	2.66	1	6	3	1	3	3.6	1	0	1	0
ammonia N (mg/l)	Det limit	Det limit	Det limit	0.49	0.23	0.15	0,02	0.01	0.66	0.04	0.72	0.15
PO_4^{3-} (mg/l)	Det limit	Det limit	0.16	0.17	0.05	0.26	0.09	0.11	0.06	0.03	0.34	0.01
P (mg/l)	Det limit	Det limit	0.05	0.05	0.02	0.09	0.03	0.04	0.02	0.01	0.11	0.0
S (ppm)	76 ± 64	Det limit ±62	59 ± 48	96 ±67	93 ± 34	Det limit ±56	Det limit ±57	Det limit ±63	196 ±145	Det limit ±138	187 ±144	Det limit ±137
Cl (ppm)	300 ± 31	266 ± 29	312 ± 24	338 ±33	347 ± 32	273 ±27	324 ±28	296 ± 31	184 ± 19	206 ±19	192 ±19	159 ±18
K (ppm)	187 ± 12	196 ± 12	210 ± 10	252 ±15	193 ± 13	203 ±11	215 ±12	187 ± 13	134 ± 9	152 ±9	133 ±8	137 ±8
Ti (ppm)	Det limit ±10	Det limit ±10	Det limit ±7	10 ±10	Det limit ±10	9 ± 9	9 ±9	Det limit ±10	Det limit ±10	Det limit ±10	Det limit ±10	Det limit ±10
Rb (ppm)	1.6 ± 0.6	1.6 ±1.1	1.2 ± 0.8	1.5 ±0.5	1.5 ± 0.6	1 ± 1	1.2 ±0.5	Det limit ±1.1	Det limit ±0.4	1.3 ±0.2	1 ± 0.4	Det limit ±0.4
Mo (ppm)	1.3 ± 0.5	1.1 ±1	1.1 ± 0.8	1.1 ±1	Det limit ±1	1.3 ±0.9	1.3 ±0.4	Det limit ±1	Det limit ±0.1	Det limit ±0.1	Det limit ±0.1	Det limit ±0.1
Sn (ppm)	Det limit ±9	12 ±9	Det limit ±7	9 ±4	Det limit ±9	11 ±4	Det limit ±8	Det limit ±9	9 ±9	9 ±9	12 ± 4	Det limit ±9
Sb (ppm)	9 ± 3	11 ±3	6 ± 3	7 ±3	7 ± 3	9 ± 6	Det limit ±6	9 ± 3	7 ±3	12 ±3	9 ± 3	10 ±3

Appendix 2. Correlation coefficients of selected physical and chemical variables. Only significant values (P<0.05) are listed.

	Physical variables										Chemical variables							
	t (° C)		pH		U (mV)		O_2 (mbar)		p (kohm*cm)		S (ppm)		Cl (ppm)		K (ppm)		Ti (ppm)	
	r	p	r	p	r	p	r	p	r	p	r	p	r	p	r	p	r	p
U (mV)			-0.719	0.0084														
O_2 (mbar)	0.7197	0.0083																
Sat.O_2 (%)	0.6945	0.0122					0.9637	0										
Conductivity (µS/cm)									-0.9437	0								
TDS (mg/l)									-0.9363	0.00001								
NaCl (mg/l)																	0.6632	0.0187
tot. hardness $CaCO_3$ (mg/l)																	0.7978	0.0019
ammonia N											0.884	0.0001						
Cl (ppm)			-0.638	0.0256														
K (ppm)			-0.650	0.0221			-0.5998	0.0392					0.8936	0.00009				
Ti (ppm)			-0.588	0.0446			-0.5942	0.0416					0.7098	0.0097	0.7098	0.0097		
Mo (ppm)					0.5892	0.0438												
Sb (ppm)	0.819	0.0011																

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Ecology of *Proctophyllodes megaphyllus* and *Analges* sp. of the *Prunella modularis* in the West Carpathian region

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Abstract. This study describes the ecology of the dominant feather mite species found on the dunnock (*Prunella modularis*). By comparing mean abundance, prevalence, and intensity during different seasons throughout the annual life cycle of the dunnock, the life cycle of *Proctophyllodes megaphyllus* and *Analges* sp. was examined. Thanks to the collection of feather mites from dead host specimens, it was possible to compare differing abundance on certain parts of the body and thus better understand the population dynamics of both species. Each species have been shown to have different life strategies. In *Analges* sp., population dynamics are adapted to vertical transmission, while *Proctophyllodes megaphyllus* has adapted its to horizontal transmission despite the unusual social behavior of the dunnock. According to our results, feather mites are likely to adapt their lives to their environment, respectively of their host. This may also provide some insights in response to the question of whether feather mites should be called parasitic or ectosymbionic organisms.

Key words: dunnock, ectoparasites, ectosymbionts, acari, ecology, population dynamics

Introduction

This research studies ecology and population dynamics of the two most common feather mite species of dunnock (*Prunella modularis* Linnaeus 1758) - *Analges* sp. and *Proctophyllodes megaphyllus* (Trouessart 1885). These species live in friable layer of feathers, primary flight feathers and basal body parts (Trouessart 1885). The species are symbionts and there is no clear evidence of competitive behaviour between them. Feather mites cannot live on dropped feathers. Their inability to move rarely allows them to survive outside the body of the host (Dubinin 1951). Increasing preening in the spring and autumn influences the behaviour, movement of population, and ecology of each of these ectoparasites (Janiga and Romanová 1996).

Feather mites occupy four main types of microhabitats on the body of birds: plumulaceous down feathers; vane surfaces of contour feathers; the interior of the quills of flight and tail feathers; and the surface of the skin. The dominant species found on *P. modularis* are *P. megaphyllus* and *Analges* sp. (Dabert and Mironov 1999). *Analges* sp. mostly resides in the friable layer of down feathers, while *P. megaphyllus* generally live in primary feathers. It is therefore assumed that there is no spatial or resource competition between the species. The relationship between feather mites and avian hosts like *P. modularis* is still very unclear. Opinions on whether feather mites are considered parasites of ectosymbionts vary considerable. Ecology of feather mites is generally under-studied compared to species such as chewing lice, which also live on bird hosts (Behnke *et al.* 1995). However, with the chewing lice there is a proven detrimental impact on fitness of the host, because of their consumption of blood and paria. Thus, they are considered parasites.

Factors such as seasonal variation, reproduction, social behavior, and patterns related to yearly cycles of dunnock are an important variable in life and population dynamics of ectosymbionts. The reproduction period is referred to as the period with the most load for feather mites. Studies suggest that the life of the host is affected by the ecology of feather mites in large measure. There is no research yet dedicated to the population dynamics and ecology of feather mite species and *P. modularis*. Similar research has been conducted on *Prunella collaris* Scopoli 1786. (Janiga and Kubašková 2000; Kašlík and Janiga 2016), which inhabits exclusively alpine environments. Parasite populations are typically aggregated among their host individuals, but the degree of aggregation varies greatly over time and among populations and species of parasites.

The nature of ecological interactions between mites and their bird hosts is still very controversial today. Many studies consider all symbiotic organisms to be parasites. According to this hypothesis, symbionts are also associated with host characteristics and they are factor in the choice of sexual partner (Blanco *et al.* 1999). While no way yet been explained by which feather mites can harm their host, some authors assume that feather mites are clearly parasites and they can hurt their host and they also provide correlation and experimental evidence for this hypothesis (Pérez-Tris *et al.* 2002; Figuerola *et al.* 2003). According to some research, feather mites can have a detrimental impact on the host. It is likely that some host species are selectively adapting to this relationship by decreasing the size of their uropygial gland. This leads to a reduction in intensity of feather mite species (Galván *et al.* 2008).

On the contrary, other studies suggest that feather mites bring benefits to their hosts (Campos *et al.* 2011; Dona *et al.* 2018) or have no impact (Dowling *et al.* 2001). Feather mites consume uropygial oil and maintain its quantity at optimal levels required for function. Old uropygial oil acuumulates on plumage and caused it to lose its insulating ability (Blanco and Frías 2001). Feather mites can increase the effectiveness of preening by removing excess uropygial oil (Hubálek 1994; Burtt and Ichida 1999; Blanco and Frías 2001). In addition to uropygial oil mites feed on fungal spores, algae, bacteria that damages feathers and in some cases, on pollen (O'Conor 1982). Feather mites can also control the number of pathogenic microoganisms (O'Conor 1982; Blanco and Frías 2001). Therefore, the nature of the interaction between mites and their hosts remains an unanswered question, the resolution of which would have diverse evolutionary implications, given that mites are present in the feathers of almost all bird species (Proctor 2013).

Prevalence and intensity of feather mites on Passeriformes

Differences in feather mite prevalence are related to seasonal changes in the host's physiological state associated with migration. Changes in dispersion and methods of aquisition are related to an increase in host social activity prior to migration. During this period, the hormonal activity that affects the formation of uropygial oil increases. Conversely, the physiological condition of the host after breeding season is stagnating or decreasing, which means there is less sustenance in the form of uropygial oil for mites. Thus, most species have adapted their reproduction to the season, which will provide them with the greatest amount of food and increase the chances of wider dispersion and horizontal transmission (Blanco and Frías 2001).

Research on the prevalence and intensity of feather mites in Passerines (119 bird species) found that these values differ between species (Diaz-Real *et al.* 2014). Differences between habitats were negligible, which means that local factors (breeding season, weather, habitat, spatial autocorrelation and researcher identity) play a secondary role. Almost 100% prevalence was found in Linnet, *Linnaria cannabina* (Linnaeus 1758) with no differences between gender or age groups (Blanco *et al.* 1999). Repeated prevalence and lower intensity values were found in Passeriformes (Diaz-Real *et al.* 2014).

Feather mites living on wings were examined on Portuguese Passeriformes in 1995. A correlation between body mass and abundance values was found. Intensity levels positively correlated with host body size - more body mass provides a larger habitat (Behnke *et al.* 1995). On the other hand, research from 2018 statesvthat there is no relationship between feather mite abundance and body size or fitness, independent of host species and sex (Matthews *et al.* 2018).

In 2013 and 2014 Passerines were studied in the Azores. 19 feather mite species belonging to the superfamily Analgoidea, including the Analgidae and Proctophyllodidae families were detected. In most of the bird host species the prevalence of Analgoidea was very similiar to their prevalence in european passerine species, including *Turdus merula, Pyrrhula murina* and *Fringilla* sp. Prevalence of this species reached 100% in both Analgoideae and Proctophyllodidae families (Rodrigues *et al.* 2015).

Cooperative breeding means that one or more individuals of a social group take care of offspring regardless of lineage. These helpers or auxiliaries are non-breeding adults that help to care for offspring. This care includes feeding, nest construction, and even incubation, which may influence the vertical transmission of mites (Stacey and Koenig 1990). Transmission of mites from parent to child, during or after birth, is called vertical transmission. Horizontal transmission is caused by physical contact between two or more individuals (Biosci 2000). Cooperative breeding is also assumed to result in higher ectoparasitosis levels. Poinani's (1992) comparative analysis investigated this hypothesis in Australian Passerines. This research has shown that in non-migrating species, cooperative breeding increases the number of parasites per host. Conversely, migrating non-cooperative breeding species are characterized by less dense transferable ectoparasites per host. Generally, the number of ectoparasites increases in proportion to the host's weight and relative abundance.

Factors shaping the community and population structure

In symbiosis ecology, understanding why host species vary greatly in ectoparasite or symbiont counts and how this may depend on ecological host and symbiotic characteristics is a major question. Some symbiotic taxa may be specialized in tracking changes in the quantity and quality of food sources that the host provides to improve reproduction and dispersal. Some species can therefore adapt their lifestyle strategy, activity and conditions of reproduction to specific stages in the host's life, such as retching or nesting (Blanco and Frías 2001).

Recently, there has been a hypothesis that mites are ectosymbionts of birds living on the skin, in feathers or on the surface of feathers (Campos *et al.* 2011). Depending on the taxon, they feed on uropygium oil, dead skin, fungi, bacteria and, to a lesser extent, the feathers themselves. Feather mites are a diverse group of ectosymbionts that occur on most bird species. We know of more than 2000 described species (Mironov and Proctor 2011). The size of the individual has been shown to correlate with the size of the mite population on its body (Proctor 2013). Thus, the body weight of the host may also affect mite diversity. Larger hosts provide more resources and therefore support larger ectosymbiont populations (Poulin 2007).

Another factor affecting abundance is the size of the uropygial gland. Since uropygial oil is an important source of food, its production directly correlates with infestation values. This correlation is also associated with mating seasonality. The relationship of infestation and gland size varies between migrating and residential species (Galván and Sanz 2006; Galván *et al.* 2008).

For nearly every bird species there is a specific species of parasite. These can be endoparasites or ectoparasites. Endoparasites, and in this case haemoparasites that inhabit a host's bloodstream, can be one of the factors affecting the life of mites in a bird host. In 2004, it was found that many bird species which were free of haemoparasites, were highly infested by ectoparasites. Forbexample, some Procellariiformes and alpine swifts, were highly infested with ectoparasites but free of haemoparasites. Often, haemoparasites kill their host, or adversely affect them, thereby creating undesireable conditions for feather mite life (Gonzalez-Solis and Abella 1997; Merino and Minguez 1998; Tella *et al.* 1998; Martinez-Abrain *et al.* 2004).

One important factor that is often overlooked in investigating ectosymbionic diversity is the influence of the host's abiotic environment (Malenke *et al.* 2011). In particular, the diversity of arthropods on the body of birds can be affected by many climatic factors (Møller 2009). Kruskal – Wallis tests were used to support the feather mite migration hypothesis of *Proctophyllodes stylifer* species in blue tits (*Cyanistes caeruleus*). The findings revealed that during cold environmental conditions, feather mites actually aggregate on tertiary remiges. In addition, *P. stylifer* not only spread to remiges of blue tits during warm weather conditions, but statistical data revealed that feather mites prefer to aggregate on the host's primary remiges. Thermal imaging supports the hypothesis, that tertiary remiges are actually warmer or better insulated than primary and secondary remiges (Schmit 2011). This may also apply to the migration of the feather mites throughout the body from colder to warmer body parts.

One of the aims of this study is track how the incidence and social behaviour of *P. modularis* affects the population dynamics and the life strategy of its dominant feather mite species. We will compare population dynamics of the feather mite species sharing the host - *P. megaphyllus* and *Analges* sp. – similar to *Analges pollicipatus* which commonly infests Alpine accentors (*P. collaris*). They are morphologically close to the dunnock, due to joint membership of the family Prunellidae (Haller 1882). One of the goals of our study is to describe the ecology of feather mites compared to seasonal activities of the host species, including preening, breeding, nesting and migration, as well as to find out how mite aggregations are related to environmental conditions.

Material and Methods

Study area

All bird samples were collected between 1998 – 2016. The birds were found dead – either as roadkill, or in the ornithological nets used during previous research. Hosts were collected in characteristic habitats for *P. modularis*; mountain and submountain zones of the Western Carpathians, located in Slovakia. This included localities in: The High Tatra mountains (Veľká Studená dolina, Velická dolina, Dolina Bielych plies); Low Tatra mountains (Demänovská dolina, Chopok, Stredná hoľa, Veľký Choč); Western Tatras (Červenec), Belianske Tatras (Tatranská Javorina, Podspády, Ždiar); Great Fatra mountains (Suchý vrch); and the Oravské Beskydy mountains (Oravská priehrada, Babia hora). All locations are between 665 – 1719 m a.s.l.

Collecting the ectoparasites

All bird samples were stored in a deep-freezer, so it was neccesary to melt them before the mites were extracted. Frozen individuals thawed for a minimum of half an hour at room temperature in a Petri dish. Maturity and sex were determined after complete thawing. Gender was determined by examining cloacal protuberancies and surrounding feathers (Janiga, pers. observation). In the case of young individuals or unclear prominences, the sex was determined by autopsy. For the extraction of feather mites, birds were moved to a soft polystyrene pad for the extraction of parasites. Using pins and preparation needles, different areas of plumage were scanned. Mites were collected using feather forceps and transferred to an Eppendorf with 90% ethanol. Species, gender, and the area of the host's body it inhabited were recorded for each mite (Mironov 2012). Mites found in the Petri dish or on the polystyrene pad may not have been included because of the inability to determine the original body part they came from. Mite sites were divided as follows: head, right wing, left wing, chest, back and tail. Mites were stored in an Eppendorf filled with ethanol and placed in the refrigerator for next use (Balát 1959; Zlotorzycka 1972).

Statistical processing of the data

The matrix of data consists of a feather mite identifier (numbers were used), site, feather mite species, sex, maturity (whether feather mite was adult or juvenile), identifier of a dunnock (PMx), date and location of sampling, sex, maturity, altitude and mercury level. The intensity, abundance and prevalence during the periods April-May, June-July, and August-September were evaluated using Quantitative Parasitology 3.0 (Rózsa *et al.* 2000). To understand this study, it is important to be familiar with the terminology of QP3.0 software. This terminology is often used in connection with study of the parasitic relationship. Site refers to the exact location of feather mites on the body of a bird host. Locality is the region or geographic location where the host was found. Prevalence is a very common term used in the field of parasitology. It shows the proportion of hosts infected with a particular species of parasite and the number of individuals examined for this species. It is expressed as a percentage, but in mathematical operations it is used in the form of a ratio, or respectively, a fraction. QP3.0 uses two methods to determine the confidence level for prevalence. It is recommended to use 95% confidence limits for prevalence in most cases. Mean intensity is the average number of parasites found in all hosts excluding the uninfected specimens, which are described as zero values. The median number of examined parasites, therefore, represents a typical level of infestation. Unlike mean intensity, this quantity is not affected by extremely infected hosts. The mean number of parasites found in all host specimens is called the mean abundance. This measure includes uninfected subjects as well (Rózsa *et al.* 2000).

Results

Parasite quantification

Out 30 hosts, 364 specimens of *Analges* sp. and 257 *P. megaphyllus* were found. The total prevalence was 100% (Table 1,2) and was the smallest in males of the symbiont species (93.3%). *Analges* sp. had a mean intensity level of 2.29 in males and 6.83 in females, while the general mean intensity level was 12.10. Females were the most numerous group in our samples, accounting for 56% of the total count. Nymphs and males were responsible for the remaining 26% and 18%, respectively. *P. megaphyllus* mean intensity was 8.57. The lowest mean intensity was 2.0 in nymphal individuals and the variance-to-mean ratio was 0.63 (Table 2). 47% of all specimens were females, 32% males and the smallest group was nymphal and represented 21%.

Seasonality

448% of the feather mites (*Analges* sp.) were collected from hosts found in June - July, 32% in April-May, and 20% in August and September. Mean intensity was significantly higher in August and September compared to April – May and June - July. In June and July prevalence showed a significant increase when compared with other months (Table 3, Table 4). The prevalence values of both feather mite species reached 100% in all months. Mean intensity of *P. megaphyllus* was significantly higher on hosts collected in spring, respectively in April and May. Confidence limits for prevalence were higher in June and July compared to other months (Table 5).

Comparison of sex-relatedgroups

When it comes to sexual variation of *Analges* sp., only male x nymph (Table 6, Table 7) mean intensity was significantly different (p=0.0055). The

	No. of hosts	Infected hosts	Prevalence	Mean intensity	Median intensity	Variance to mean ratio
***Analges* sp. sum**	30	30	100%	12.10	12.0	0.59
***Analges* sp. female**	30	30	100%	6.83	7.0	0.58
***Analges* sp. male**	30	28	93.3%	2.29	2.0	0.67
***Analges* sp. nymphal**	30	30	100%	3.17	3.0	0.46

Table 1. Sumary of *Analges* sp.Collected *Analges* sp. specimens were divided into nymphs, females and males, and a summary of all hosts is included. Prevalence, mean intensity (MI) and median intensity are included. Exact confidence limit levels of confidence range from 95% to 99%.

	No. of hosts	Infected hosts	Prevalence	Mean intensity	Median intensity	Variance to mean ratio
***P. megaphyllus* sum**	30	30	100%	8.57	9.0	0.32
***P. megaphyllus* female**	30	30	100%	4	4.0	0.48
***P. megaphyllus* male**	30	28	93.3%	2.96	1.2	1.14
***P. megaphyllus* nymphal**	30	27	90%	2	2.0	0.63

Table 2. Summary of *P. megaphyllus*. Collected *P. megaphyllus* specimens were divided into males, females and nymphs, and a summary of all of the hosts is included. Prevalence, mean intensity and median intensity are included. Exact levels of confidence range from 97% to 99%.

	Species	MI Bootstrap p-value (two-sided)	MA Bootstrap p-value (two-sided)
Apr-May x June-July	*P. megaphyllus*	0.4165	0.4135
	Analges sp.	0.8085	0.7955
June-July x Aug-Sep	*P. megaphyllus*	0.7740	0.7595
	Analges sp.	0.0870	0.0905
Aug-Sep x Apr-May	*P. megaphyllus*	0.3215	0.3190
	Analges sp.	0.1005	0.1000

Table 3. Tabulated summary of p-values of significantly differing variables in seasons and species. Only statistically significant results displayed. MI = Mean Intensity; MA = Mean Abundance.

	No of hosts	Prevalence	95% Confidence limits for prevalence	Mean intensity	95% (Bca) Bootstrap CL for MI
Apr-May	10	100%	0.7092 - 1.0000	11.7	10.40 - 13.10
June-July	15	100%	0.7778 - 1.0000	11.5	10.27 - 12.40
Aug-Sep	5	100%	0.5000 - 1.0000	15	11.40 - 16.80

Table 4. Mean intensity and prevalence values of *Analges* sp. displayed in different months within the year: April - May, June – July and August – September. Lower and upper confidence limits of prevalences and bootstrap confidence limits (CL) of mean intensities (MI) are included, both at the 95% confidence level.

	No of hosts	Prevalence	95% Confidence limits for prevalence	Mean Intensity	95% (Bca) Bootstrap CL for MI
Apr-May	10	100%	0.7092 - 1.0000	9	8.30 - 9.60
June-July	15	100%	0.7778 - 1.0000	8.5	7.27 - 9.27
Aug-Sep	5	100%	0.5000 - 1.0000	8.2	6.60 - 9.00

Table 5. Prevalence and mean intensity values of *Proctophyllodes megaphyllus* displayed in different months within the year: April - May, June – July and August – September. Lower and upper 95% confidence limits of prevalence and bootstrap confidence limits of mean intensity (MI) are included.

Samples compared	Species	MI Bootstrap p-value (two-sided)	Exact P-value (two-sided)
Female x Male	*P. megaphyllus*	0.0150	0.492
	Analges sp.	0.0000	0.492
Male x Nymph	*P. megaphyllus*	0.0120	1.000
	Analges sp.	0.0055	0.492
Nymph x Female	*P. megaphyllus*	0.0000	1.000
	Analges sp.	0.0000	0.237

Table 6. Tabulated summary of p-values of significantly differing variables in sex and maturity of each species. Only statistically significant results displayed. MI = Mean Intensity; MA = Mean Abundance.

	No of hosts	Prevalence	95% Confidence limits for prevalence	Mean intensity	95% (Bca) Bootstrap CL for MI
Female	10	100%	0.7092 - 1.0000	11.0	10.00 - 13.00
Male	10	100%	0.7092 - 1.0000	13.6	11.80 - 15.20
Juvenile	10	100%	0.7092 - 1.0000	11.7	9.90 - 12.80

Table 7. Prevalences and mean intensities of *Analges* sp. on female, male and juvenile hosts. Lower and upper confidence limits fo prevalence and bootstrap confidence limints of mean intensity (MI) are included.

	No of hosts	Prevalence	95% Confidence limits for prevalence	Mean intensity	95% (Bca) Bootstrap CL for MI
Female	10	100%	0.7092 - 1.0000	9.40	8.60 - 10.00
Male	10	100%	0.7092 - 1.0000	8.10	7.00 - 9.10
Juvenile	10	100%	0.7092 - 1.0000	8.20	6.70 - 9.00

Table 8. Prevalences and mean intensities of *P. megaphyllus* on female, male and juvenile hosts. Lower and upper confidence limits fo prevalence and bootstrap confidence limints of mean intensity (MI) are included.

lowest P-value for Fisher's test was between nymphs and females (p=0.237). The population structure of *P. megaphyllus* shows a statistically significant increase in mean intensity in favor of females (Table 6, Table 8).

Relationship between sex, maturity and site

For *Analges* sp. females there is a visible difference in mean abundance, especially on the chest (2.10), while they have the lowest abundance on the back (0.70). Overall, the highest abundances occured on wings (2.20 – 2.80) (Table 9). Most *Analges* sp. males are located on juvenile wings (1.20) and none on female chests (Table 10). Nymphs of *Analges* sp. are the most represented on female chests, (1.70) and juvenile backs (1.80) (Table 11).

	Head	Wings	Chest	Back	Tail
Juvenile	1.1	2.2	0.7	2.0	0.5
Female	0.4	2.8	2.1	0.7	0.6
Male	0.5	2.6	1.7	2.0	0.5

Table 9. Comparison of mean abundance of *Analges* sp. females depending on host maturity, gender and site.

	Head	Wings	Chest	Back	Tail
Juvenile	0.3	1.2	0.6	0.3	0.3
Female	0.2	0.8	0.0	0.2	0.2
Male	0.2	1.0	0.6	0.6	0.1

Table 10. Comparison of mean abundance of *Analges* sp. males depending on host maturity, gender and site.

	Head	Wings	Chest	Back	Tail
Juvenile	0.2	0.4	0.2	1.8	0.6
Female	0.2	0.6	1.7	0.0	0.4
Male	0.8	1.0	1.1	0.6	0.0

Table 11. Comparison of mean abundance of *Analges* sp. nymphs depending on host maturity, gender and site.

	Head	Wings	Chest	Back	Tail
Juvenile	0.5	1.4	0.7	0.7	0.2
Female	0.3	1.7	1.4	0.3	0.4
Male	0.2	1.8	0.7	0.7	0.5

Table 12. Comparison of mean abundance of *P. megaphyllus* females depending on host maturity, gender and site.

	Head	Wings	Chest	Back	Tail
Juvenile	0.1	1.1	0.4	0.7	0.2
Female	0.2	1.3	1.0	0.6	0.3
Male	0.3	1.1	0.5	0.4	0.1

Table 13. Comparison of mean abundance of *P. megaphyllus* males depending on host maturity, gender and site.

	Head	Wings	Chest	Back	Tail
Juvenile	0.1	0.7	0.0	1.1	0.4
Female	0.2	0.5	0.6	0.5	0.0
Male	0.2	0.5	0.2	0.4	0.1

Table 14. Comparison of mean abundance of *P. megaphyllus* nymphs depending on host maturity, gender and site.

	No of hosts	Prevalence	95% Confidence limits for prevalence	Mean intensity	95% (Bca) Bootstrap CL for MI
<1000	4	100%	0.4729 - 1.0000	11.75	10.00 - 13.75
1000-1500	19	100%	0.8245 - 1.0000	12.21	10.95 - 13.42
1500<	7	100%	0.6229 - 1.0000	12.00	10.29 - 14.00

Table 15. Comparison of prevalence and mean intensity of *Analges* sp. specimens depending on high above sea levels.

	No of hosts	Prevalence	95% Confidence limits for prevalence	Mean intensity	95% (Bca) Bootstrap CL for MI
<1000	4	100%	0.4729 - 1.0000	8.75	7.00 – 9.50
1000-1500	19	100%	0.8245 - 1.0000	8.32	7.37 – 8.89
1500<	7	100%	0.6229 - 1.0000	9.14	7.43 – 10.14

Table 16. Comparison of prevalence and mean intensity of *P. megaphyllus* specimens depending on high above sea levels.

P. megaphyllus females are mainly located on wings. On backs there is 1.40 mean abundance of females, which is twice the value of juveniles and males (Table 12). The highest mean abundance of *P. megaphyllus* males is on wings (1.10 – 1.30) and on female chests (Table 13). Most nymphs were found on juvenile backs (1.10) and none on chests, while female chests had the highest mean abundance of all specimens (Table 14).

Changes in prevalence by altitude

Prevalences of both symbiont species was 100% at each altitude interval. Highest mean intensity of *Analges* sp. occured at an altitude of 1000 – 1500 metres (12.21) (Table 15) and for *P. megaphyllus* this value was 9.14 at altitudes higher than 1500 metres (Table 16).

Discussion

The sexual variation of feather mites depending on site

Mean intensity and mean abundance of *Analges* sp. was highest on males. On females and juveniles similar abundance was recorded. *P. megaphyllus* had the highest mean intensity on females. Matthews *et al.* (2018) showed that mean abundance does not depend on the gender of the host (Marini *et al.* 1996; Hamstra and Badyaev 2009; Carleton and Proctor 2010). Our results are inconsistent with these this. It is unknown how interaction between age and sex influence abundance of feather mites. This could be due to the uropygial gland in males, and thus the quantity of uropygial oil available for mites (Lafferty *et al.* 2006; Matthews *et al.* 2018). The differences in bird sexes can also be reflected in the host´ s body condition and measurements (Rózsa 1997; Galván *et al.* 2008). Despite this, it has been proven that older specimens carry more ectosymbionts than younger ones. In the case of *P. megaphyllus*, this is mostly due to their preferencefor horizontal transmission. Althought *Analges* sp. uses vertical transmission more than horizontal, it is mainly fertilized females and nymphs that are transmitted this way, and thus it takes longer for the population to increase (Dabert and Mironov 1999; Dabert *et al.* 2015).

Females of *Analges* sp. mostly aggregated on wings and chests of female hosts. On the other hand, there was a visible decrease in individual counts found on female backs. High values of mean abundance were also present in nymphs on backs and tails of juveniles. Nymphs of *Analges* sp. aggregated mostly on chests of females and on juvenile backs. Mean abundance of *P. megaphyllus* females on chests and tails of female hosts exceeded mean abundance of nymphs on female host chests. On the contrary *P. megaphyllus* mean abundance reaches highest levels on juvenile backs. This population distribution suggests that feather mites synchronize their aggregation and reproduction with the host species in terms of horizontal transmission (Figuerola 2000; Proctor and Owens 2000). Our results show that *P. megaphyllus* abundace on heads is higher than abundace of *Analges* sp. on heads, and deviate from results found by Lyra-Neves *et al.* (2003), which showed the opposite. This could be proof that *P. megaphyllus* on dunnocks has adapted to special horizontal transmission during cloacal pecking.

This population distribution also seems to depend on the reproductive behaviour of the host and transmission vectors of feather mites. We know that in vertical transmission, predominantly nymphs and fertilized females are transmitted (Dabert and Mironov 1999; Mironov 2012). Our results show that the aggregation of females on lower parts of the host (chest), and nymphs on the back may be the result of vertical transmission, as female chests and juvenile backs are the contact surfaces while nesting. In this period is also possible that nestlings do not form enough uropygial oil, and instead receive oil produced by adult females on their back.

The seasonality of feather mites

The results clearly show that the mean intensities are significantly different between species, but there are no big differences in mean intensity between the seasons, except for during August and September in *Analges* sp., and April and May in *P. megaphyllus*. In *Analges* sp., the mean intensity was significantly higher in August and September than in April – May and June – July, when values were very similiar. This may be related to moulting, which starts following the nesting season. Body condition of bird hosts is usually weak during this period (Blanco and Frías 2001) and it can disrupt the life cycle of mites (Dubinin 1951; Jovani and Serrano 2001). Body condition affects production of uropygial gland vaxes, which affects feather mite abundance (Behnke *et al.* 1995; Haribal 2011). On the other hand, 48% of all host specimens were collected in June and July. Between August and November, hosts migrate to Southern localities (Sol *et al.* 2005). The intensity may be higher precisely because of the increase in the average ambient temperature. Schmit (2011) proved that *Proctophyllodes stylifer* migrate to tertiary feathers in cold conditions, while during the warm season they remain predominantly in primary feathers. Abiotic conditions in the host's environment are important not only for choosing microhabitats, but also because temperature and humidity significantly affect life and reproduction of feather mites (Matthews *et al.* 2018; Melendez *et al.* 2014; Wiles *et al.* 2000). It is logical that feather mites migrate to different areas of the host's body, depending on the temperature of different parts of the body. The warmest places are on the head and the lower parts of bird body. The mean intensity of *P. megaphyllus* was similar throughout the year, but highest in April and May. During these months, spring migration to nesting sites takes place (Ferianc 1979), which could lead to an increase in the intensity of the species. Nesting ecology is a very important factor for understanding feather mite abundance (Matthews *et al.* 2018). During nesting season, the nest represents the host environment, particularly for female and juvenile hosts (Dona *et al.* 2017; Matthews *et al.* 2018). This means that feather mites are affected by the nest environment during nesting season and during transmission from female to juveniles (Galván and Sanz 2006; Matthews *et al.* 2018). During this period, the nest is a very suitable environment for feather mites, because the temperature is higher than 20˚ C and the humidity is significantly higher than during other periods. This leads to an increase in abundance (Marini and Couto 1997; Wiles *et al.* 2000; Moyer *et al.* 2002; Matthews *et al.* 2018).

These results support the theory that feather mites can adapt to the host's life strategy and for vertical transmission during breeding and nesting season (Galván and Sanz 2006; Kašlík and Janiga 2016). Confidence limits for prevalence of both species were highest in June and July; however, they were very similar to those reached in April and May. On the contrary, the lowest confidence levels occurred in August and September. This may be influenced either by the ambient temperature in the summer months or by the fact that the reproduction of *P. modularis* takes place twice a year and lasts from April to July (Ferianc 1979; Sol *et al.* 2005). At the end of breeding season, along with the start of moulting season, decrease in fitness and production of uropygial oil occurs (Neves *et al.* 2000; Lyra-Neves *et al.* 2003; Pap *et al.* 2010), and thus the food source available to mites decreases, impacting abundance in September through August. Feather mites commonly deviate from their regular distribution pattern during this period (Jovani and Serrano 2001).

Feather mites are photosensitive organisms. Another possible factor influencing the prevalence rate and particularly their intensity during the summer months is the length of day and hence the singig, which is a manifestation of hormonal changes similar to *P. collaris* (Proctor 2003; Kašlík and Janiga 2016; Janiga pers. observation). These hormonal changes (as well as mating) can lead to increased formation of uropygial oil and thus an increase in level of infestation and variations in mean abundances (Galván *et al.* 2008; Blanco and Frías 2001; Diaz-Real *et al.* 2014). Increased confidence limits for prevalence, mean intensity and mean abundance may also be the result of autumn moulting, when feathers are lost along with many ectosymbionts (Markov 1940; Baum 1968; Burtt and Ichida 1999). Some studies claim that feather mites can move between primary, secondary and tertiary feathers in order to avoid feathers that are about fall out in the near future, to prevent population decline during moulting (Dubinin 1951; Burtt and Ichida 1999; Jovani and Serrano 2001). However, the mechanism of moult escape has not yet been explained. For example, when *Analges* sp. inhabit down plumage, there is more loss recorded during this period, because of the smaller magnitude of vibrations of loosened feathers (Dubinin 1951; Kašlík and Janiga 2016). In addition to feather loss, moulting season also brings a decreas in uropygial secretion at the time of migration to winter habitats (Glutz von Blotzheim 1985; Sol *et al.* 2005).

In August, the nesting season ends (Ferianc 1979; Sol *et al.* 2005) and host fitness is low. Their feathers are significantly worn out and body condition is poor. Hormonal levels are also reduced, which means less food resources for feather mites and thus decreasing mean abundance (Dubinin 1951; Blanco and Frías 2001; Galván and Sanz 2006; Blanco *et al.* 1997). During August ambient temperatures are also high, creating suitable conditions for feather mite reproduction (Wiles *et al.* 2000; Matthews *et al.* 2018). However, this is also moulting season and mites are forced to migrate to tertiary feathers (Neves *et al.* 2000; Schmit 2011; Diaz-Real 2014), the natural microhabitat and territory of *Analges* sp., where competition for food sources can occur (Malenke *et al.* 2011). Thus, the mites do not focus on reproduction during this period, and instead focus on synchronizing reproduction with the breeding season of their host (Diaz-Real *et al.* 2014). Between August and November is also the beginning of pre-winter migration to southern locations (Sol *et al.* 2005). Preening takes place during this time and may be an important factor in feather mite loads as preening involves the removal of parasites. During breeding season, almost no preening occurs, however, in spring and autumn there is a significant increase in preening and bathing because of winter aggregation and nesting aggregation in spring (Ferianc 1979; Janiga and Romanová 1996; Sol *et al.* 2005).

Life strategies of feathermites

Differing strategies of transmision and population dynamics can be impacted by different host sites of each species. *Analges* sp. occupy down feathers, while *P. megaphyllus* live in primary flight feathers. This means that mites may employ both vertical and horizontal transmission (O´ Conor 1982; Kašlík and Janiga 2016). Horizonal transmission can be dangerous for feather mites even though it takes place during breeding season, when the chance of successful transmission is highest. This is confirmed by our results, as most feather mites were found during the nesting period, when birds are in physical contact in the nest temperature and humidity conditions in the nest are favourable (Wiles *et al.* 2000; Moyer *et al.* 2002; Galván and Sanz 2006; Dona *et al.* 2017; Matthews *et al.* 2018).

It is very likely that for *Analges* sp., it is more difficult to switch hosts during mating, due to its short duration. Morphology is adapted to the particular type of feather, in which each feather mite species lives (Dubinin 1951, 1953; Dabert and Mironov 1999). Feather mites have a strong sexual dimorphism. *Analges* sp. females have small legs and males have hypertrophic legs (Nakamura 1990; Dabert and Mironov 1999). This may be one of the reasons, that *Analges* sp. have a predisposition to vertical transmission as opposed to horizontal. Our results have shown high mean abundances of females and nymphs on host female chests and host juvenile backs during breeding and nesting season. It is much easier to switch hosts during nesting and incubation. *P. megaphyllus* seems more actively mobile and is able to practice horizontal transmission as well (Dabert and Mironov 1999; Kašlík and Janiga 2016). Based on the high mean abundance of *Analges* sp. nymphs it is possible that feather mites common to dunnock have adapted so much, that they are able to use cloaca pecking to achieve horizontal transmission in addition to mating.

As mentioned previously, feather mites are photosensitive and it is possible that solar radiation has a harmful effect on feather mite lifespan, and may even have a lethal effect in high enough quantities (Moyer and Wagenbach 1995). To protect themselves from radiation during the summer season, mites migrate into down feathers or onto the skin (Jovani and Serrano 2001). Solar radiation, in addition to moulting, may cause migration of *P. megaphyllus* from primary feathers to the friable layer of plummage. They are able to sense vibrations in feathers prior to moulting, and as a result, migrate to down feathers. These plumage layers are already inhabited by *Analges* sp. There may be increased competition between these species following breeding season, considering the increased amount of food resources. In this case, *Analges* sp. has the advantage of being in its natural environment. This leads to an increase in mean intensity and confidence limits for prevalence of *P. megaphyllus* in late summer and autumn (August and September).

Most host specimens were found at altitudes 1000 - 1500 m a.s.l. Despite expectations, the highest mean intensity occured in specimens collected at 1500 m and higher. We expected that abundance and intensity would decrease proportionally with increasing altitude. This hypothesis was based on the assumption that feather mites are sensitive to low temperatures (Poulin 2006; Schmit 2011; Møller *et al.* 2013) and solar radiation, which is more intense at higher altitudes (Moyer and Wagenbach 1995). Kašlík and Janiga (2016) found much higher mean intensity on *P. collaris*, which inhabits exclusively alpine ecosystems. Thus, we can say that mites are sensitive to temperature and solar radiation but the only result appears to be is migration to other plumage layers and body parts (Schmit 2011). On the other hand, the level of infestation increases with higher altitude. This could be a result of longer lifespans for dunnock in alpine zones due to fewer predators at lower altitudes, and a reduction in people, traffic and environmental pollution. We cannot say for sure whether feather mites will be able to adapt to such conditions over time. The most likely reason for a decrease in feather mites at lower altitudes is air pollution. Air conditions affect the host´ s fitness, as well as the lifespan of feather mites. If we perceive feather mites as ectosymbionts (Behnke *et al.* 1995; Evans *et al.* 1961), we can say that they are impacted by their host´ s life as it acts as their natural environment.

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One hundred years of nature conservancy in Slovakia

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"One Hundred Years of Nature Conservancy in Slovakia", was the topic of a recent national conference with international attendance organised by The State Nature Conservancy of the Slovak Republic (SNC SR) and the Ministry of Environment of the Slovak Republic (ME SR). The conference took place in Tale, Slovakia on October 5-16, 2019. Its aim was to commemorate the history of state nature conservancy in Slovakia, and to evaluate current developments, as well as the current state of, and the future vision for nature and landscape protection in Slovakia.

Nature conservation in Slovakia has deep social and cultural roots. Initially, utilitarian and world-view motives prevailed, however, thanks to the Hungarian nature conservationist Karol Kaán (1867 - 1940), and framed by the Hungarian Law Act 39/1881 of monuments and the Hungarian Law Act 31/1879 of forestry, the foundations for current nature conservation practices were laid. We consider 1919 (following establishment of the First Czechoslovak Republic) to be the year that institutionalized nature conservancy was established in Slovakia and it marks the beginning of efforts toward systematic protection of nature throughout Central Europe.

On October 20, 1919, Vavro Šrobár, the Minister of the Government of the Czechoslovak Republic, signed a document entitled "Order of the Minister - Plenipotentiary of the Government of the Czechoslovak Republic for the Administration of Slovakia no. 155/1919 on the competence of the Government Commissioner for the Monuments Conservation in Slovakia" (Nariadenie ministra – plnomocníka vlády Československej republiky pre správu Slovenska č. 155/1919 o právomoci Vládneho komisariátu na ochranu pamiatok na Slovensku). As a result, a Government Commission was established in Bratislava as a part of the Ministry of Education and National Enlightenment in Prague, and named the "State Office for Monuments Conservation in Slovakia" (Štátny referát na ochranu pamiatok na Slovensku). This meant that state nature conservation became inherently linked to the conservation of monuments in Slovakia, until 1981 when the state nature conservancy became independent. Over this period of 62 years, conservation developed in parallel with the conservation of monuments, before eventually becoming an independent professional organization - the State Centre of Nature Conservation (Ústredie štátnej ochrany prírody) in Liptovský Mikuláš.

The development of nature conservancy had several important milestones. The most important of these was adoption of the Act No. 1/1955 on State Nature Protection. The foundation of the Slovak Heritage Institute (Slovenský pamiatkový ústav) occurred in 1951, and subsequently became the "Slovak Institute of Monument Preservation and Nature Conservation" in 1958 (Slovenský ústav pamiatkovej starostlivosti a ochrany prírody). Under these institutions, the same level of protection was implemented to safeguard nature as those legislated for species and territorial protection in Slovakia.

The conference was divided into five blocks and two sections, including excursions. In the first block Martin Lakanda (general director of SNC SR), Boris Susko (State Secretary of the ME SR), Ladislav Miko (Head of the Representation of the European Commission in the SR) and Vladimír Dolejský (Deputy for Management of the Nature Protection Department of the Ministry of the Environment of the Czech Republic) spoke to the guests, where they all emphasized not only historical milestones in the area of state nature protection in Slovakia, resp. in Czechoslovakia, but also presented goals and plans for the future. In the first section, "Historical Consequences of Nature Conservancy in Slovakia", Viliam Stockmann (Slovak Museum of Nature protection and Speleology - SMNPS) presented on the origins and development of state nature conservancy during the 1st Czechoslovak Republic (1918-1938) and the 1st Slovak state (1939-1945). Next, Eva Greschová (SMNPS) spoke to the audience regarding the development of nature conservancy in Slovakia between 1945 to 1989, and at the end of this section, László Miklós (Institute of Landscape Ecology of the Slovak Academy of Sciences) focused on modern history of nature protection (since 1989). The second section was focused on "The State of Nature Protection in Slovakia, Forecasts and Visions". Marta Mútňanová (Section Director of the Nature and Landscape Protection of SNC SR), spoke about State nature conservancy at present. Peter Baláž (Deputy Director General for SNC SR) presented, "Steps for future: Envirostrategy, the Conception of Nature Protection and legislative changes". Milan Chrenko (ME SR), Radoslav Považan, (Slovak Environment Agency – SEA) and Richard Filčák (Institute for Forecasting - Centre of

Social and Psychological Sciences) presented "The Scenarios for Nature of Slovakia until 2050". After professional lectures, an interesting and stimulating discussion was moderated by Zuzana Gabrižová (Euractiv), in which the representatives of interested groups spoke about nature and landscape protection. During the third block, which ran in parallel with the first and second blocks, the conference participants had access to: poster presentations; films on nature and landscape conservation; Exhibition Ecoposters (selection of the winning works of Exhibition "Ekoplagát" - organised by SNC SR); and the exhibition 100 Years of State Nature Protection in Slovakia (SMNPS).

The fourth block was organised as a gala evening including award presentation to personalities and organizations in the field of nature and landscape protection in Slovakia. The prizes were presented by the Minister of the Environment László Solymós together with the General Director of SNC SR, Martin Lakanda. Juraj Galvánek, László Miklós, Dušan Slávik, Viliam Stockmann, Ján Terray, Miroslav Fulín, Rudolf Soltés, Jozef Klinda, Jaroslav Halaš, Milan Janík, Anna Jusková, Jozef Kramárik and Štefan Mihálik were awarded honorable mentions. Representatives of the following organizations or associations received honorary awards: National Zoo Bojnice, Slovak Union of Nature and Landscape Conservationists, Carpathian Protectionist Association of Altruists (KOZA Trenčín) and Civic Association - For Nature (OZ Pre prírodu). Thank you letters in memoriam were presented to relatives of the following personalities: Ján Futák, Jozef Šteffek, Dezider Magic, Peter Straka and Ľudovít Dostál. Commemorative letters and commemorative medals of the General Director of SNC SR were given to all departmental organizations of the Ministry of the Environment of the Slovak Republic, as well as other organizations that significantly influence and help the development of nature protection in Slovakia.

During the second day of the conference, excursions were planned for the fifth block. Members had the opportunity to visit the Demänovská cave of Freedom, Dobročský prales (primeval fir-beech forest) and the Čierny Váh forestry railway (Čiernohorská železnička) or the Primeval forest of Bystrá valley (development after natural disturbances).

The expert guarantors of the conference were Prof. László Miklos from Technical University Zvolen, Katarína Butkovská from the ME SR and Martin Lakanda from SNC SR.