CORYLUS AVELLANA L.

POLLEN AND CATKINS CHARACTERIZATION ON GENOTYPES FROM SELECTED LOCALITIES OF WEST SLOVAKIA

Nataliia NIKOLAIEVA, Janka NÔŽKOVÁ, Ján GAŽO

NITRA 2020

DOI: https://doi.org/10.15414/2020.9788055222622

Title: *CORYLUS AVELLANA* L. POLLEN AND CATKINS CHARACTERIZATION ON GENOTYPES FROM SELECTED LOCALITIES OF WEST SLOVAKIA

Authors: Mgr. Nataliia Nikolaieva, PhD. (2,01 AH)

doc. Ing. Janka Nôžková, PhD. (1,30 AH)

Ing. Ján Gažo, PhD. (1,30 AH)

Slovak University of Agriculture in Nitra

Faculty of Agrobiology and Food Resources

Department of Genetics and Plant Breeding

Reviewers: doc. Ing. Pavol Eliáš, PhD.
Slovak University of Agriculture in Nitra

Ing. Ľubomír Mendel, PhD.
The Research Institute of Plant Production Piešťany

Approved by the Rector of the Slovak University of Agriculture in Nitra on 21th August 2020 as an online scientific monograph.

Nitra 2020

ISBN 978-80-552-2262-2

DOI: https://doi.org/10.15414/2020.9788055222622

Table of content

DOI: https://doi.org/10.15414/2020.9788055222622

DOI: https://doi.org/10.15414/2020.9788055222622

Introduction

Hazelnut (*Corylus avellana* L.) has unusual ability to adapt to extreme conditions and is widely spread throughout Slovakia. Under the influence of various environmental factors shape and size of habitus, leaves, fruits, and catkins change. That is the reason why hazelnut became the subject of research in many countries. The important stage in the study of hazelnut plants was the use for cultivation purposes. The traits from cultivated genotypes are transported also on wild genotypes. Genetic diversity of hazelnuts confirmed survival adaptability.

The application research and experimental development in conservation of environment and agriculture are interconnected by different approaches. The biological objectives are native organisms, and they are constantly in inseparable unity with the environment, but environment conditions are unstable. Organisms adapt to new living conditions through the activation the synthesis of nucleic acids and proteins. This factor influences on variability evaluation of genetic resources.

In our study we tried to find the most distinguishing sample by combined empiric knowledge with innovative methods for morphological characterization. We worked with pollen grains and catkins (male inflorescences). We proposed to combine the results of different experimental works for identifying the suitable genotypes for further using.

DOI: https://doi.org/10.15414/2020.9788055222622

DOI: https://doi.org/10.15414/2020.9788055222622

1 Current condition of the field at home and abroad

1.1 General characteristics of *Corylus avellana* L.

1.1.1 Taxonomy and distribution of *Corylus avellana* L.

Corylus avellana L. is an angiosperm dicotyledonous plant from the *Betulaceae* family originated from Eurasia. It is a monoecious plant, self-incompatible and wind pollinated. Some authors place *Corylus* as a separate family, *Corylaceae*, between *Abedulaceae* and *Betulaceae* (Blackmore et al., 2003; Palacios, 2015) (Fig. 1), but recent molecular analysis reveals that *Corylus* belongs to the *Betulaceae* family (APG III system, 2009).

Division: *Angiospermae*

Class: *Magnoliopsida*

Order: *Fagales*

Family: *Betulaceae*

Subfamily: *Coryloideae*

Genus: *Corylus*

Species: *Corylus avellana* L.

Fig. 1

Hazelnut *(Corylus avellana* L.)
(Drawing: Otto Wilhelm Thome, 1885)

According to the U.S. National Plant Germplasm System (2003) *Corylus avellana* L. has synonyms: *Corylus avellana* f. *aurea* (G. Kirchn.) C. K. Schneid., *Corylus avellana* f. *contorta* (Bean) Rehder, *Corylus avellana* f. *fuscorubra* Dippel, *Corylus avellana* f. *heterophylla* (Lodd. ex Loudon) Rehder, *Corylus avellana* f. *pendula* (H. Jaeger) Dippel, *Corylus avellana* var. *aurea* G. Kirchn., *Corylus avellana* var. *contorta* Bean, *Corylus avellana* var. *fusco-rubra* ined., *Corylus avellana* var. *heterophylla* (Lodd. ex Loudon) Loudon, *Corylus avellana* var. *pendula* H. Jaeger, *Corylus colchica* Albov, *Corylus imeretica* Kem.-Nath., *Corylus maxima* Mill., *Corylus pontica* K. Koch, *Corylus tubulosa* Willd.

DOI: https://doi.org/10.15414/2020.9788055222622

According to the Euro+Med PlantBase (Uotila, 2009) *Corylus avellana* L. has synonyms: *Corylus glandulosa* (Godet) Godet, *Corylus imeretica* Kem.-Nath., *Corylus pontica* K. Koch.

Corylus avellena L. is a widespread species in the world and has o lot of common names in different languages (Tab. 1).

Tab. 1

***Corylus avellana* L. common names in different languages**

Language	Name	Source
English	Barcelona-nuts	Duke, 2000
	cobnut	Miller, 1908
	European filbert	Euro+Med PlantBase (Uotila, 2009)
	European hazel	Euro Med Plantbase, 2017
	filbert	Hanelt, 2001
	giant filbert	Euro Med Plantbase, 2017
	hazel	Stace, 1997
French	avelinier	Rehm, 1994
	coudrier	Rehm, 1994
	noisetier	Rehm, 1994
	Noisetier franc	Rehm, 1994
	Noisetier tubuleux	Rehm, 1994
German	gewöhnliche Hasel	Erhardt et al., 2000
	Haselnuß	Encke et al., 1993
	Haselstrauch	Rehm, 1994
	Lambertnuß	Rehm, 1994
	Lambertshasel	Rehm, 1994
Italiano	avellano	Hanelt, 2001
	nocciuola	Hanelt, 2001
Portuguese	aveleira	Rehm, 1994
Spanish	avellano	Rehm, 1994
	avellano de Lambert	Rehm, 1994
Swedish	filberthassel	Aldén et al., 2012
	hassel	Aldén et al., 2012
Chinese (transcribed)	ou zhen	Hanelt, 2001
Russian (transcribed)	leščina	Hanelt, 2001
	orešnik obyknovennaja	Hanelt, 2001

Distribution of *Corylus avellana* L. in the world: Albania, Armenia, Austria, Azerbaijan, Belarus, Belgium, Bosnia and Herzegovina, Bulgaria, Croatia, Czech Republic, Denmark, Estonia, Finland, France (incl. Corsica), Georgia, Germany, Greece, Hungary, Islamic Republic of Iran, Italy

DOI: https://doi.org/10.15414/2020.9788055222622

(incl. Sardegna, Sicilia), Latvia, Liechtenstein, Lithuania, Luxembourg, Moldova, Montenegro, Netherlands, Norway, Poland, Portugal, Romania, Russian Federation (European Russia: Central European Russia, Chechnya, Dagestan, East European Russia, Ingushetiya, Kabardino-Balkariya, Karachaevo-Cherkessiya, Krasnodar, North European Russia, Northwest European Russia, Severo-Osetiya, South European Russia, Stavropol), Serbia, Slovakia, Slovenia, Spain, Sweden, Switzerland, Republic of Macedonia, Turkey in Asia, Ukraine (incl. Krym), United Kingdom (Great Britain, Northern Ireland) (the U.S. National Plant Germplasm System, 2003; Uotila, 2009).

In Slovakia the *C. avellana* L. is distributed in every phytogeographical district. It has abundant occurrence in Western Carpathians, typical populations are part of the flora of Central Carpathians, especially in Veľká Fatra, Nízke Tatry and Chočské vrchy. Its distribution has been significantly influenced by man and by secondary extension to pastures and meadows (Mercel, 2006).

1.1.2 World production of *Corylus avellana* L.

Wild hazelnuts (*Corylus avellana* L.) are widespread throughout Europe, they are only grown commercially in specific locations. Total world production in the years 2017 – 2018 was estimated about 490,000 tons in-shell uncracked nuts. Turkey was the leading hazelnut producer, with approximately 73 % of the world share (International nut and dried fruit, 2017 – 2018). Italy is the next most important producer, with about 70 000 ha of orchards producing about 110 000 tons, almost 15 % of the world's hazelnut crop (Tombesi, 2005). There are 4 main regions of production, Piedmont in the north-west (13 % of Italy's production), Latium, north of Rome (30 % of production), Campania, near Naples (42 % of production) and Sicily (12 % of production).

Spain is also very important producer in Europe with about 22 000 tons of nuts in shell; 3 % of world production. The key center for production is in the coastal district of the "Camp de Tarragona" (Mehlenbacher, 1991).

France production is 5300 tons of nuts in-shell (Sarraquigne, 2005). The main center of production is Agen in the Aquitaine district, average orchard size is 14 – 20 ha.

Small areas of hazelnuts are grown in some other European countries such as Slovenia (Solar and Stampar, 1997) and Romania (Turcu and Botu, 1997), in locations where conditions are favorable for the crop.

Outside Europe, hazelnuts are cultivated in the USA, mainly in the Willamette Valley in Oregon, south of the city of Portland. The total area of production is about 11 000 ha with an average annual crop of about 28 500 tons of nuts in shell and about 5 % of world production (Mehlenbacher, 1991).

DOI: https://doi.org/10.15414/2020.9788055222622

A hazelnut industry is being developed in Chile (Grau et al., 2001). There are also areas in New Zealand (McNeil, 1999; Hunt, 2001).

1.1.3 Conditions of *Corylus avellana* L. growth

The main areas of commercial hazelnut production are in maritime, Mediterranean-type climates in the latitude range 40 – 45° N, with mild, humid winters and cool summers (Mehlenbacher, 1991). All the northern hemisphere centers have more than 50 mm of rainfall in May, when trees would be making active leaf and shoot growth.

The most critical period is the time from fertilization to kernel fill, which is from the end of May to mid-July in the Northern Hemisphere (Mingeau et al., 1994). The rainfall should ideally be well-distributed throughout the year, but dry conditions are desirable for the period of nut fall and harvest, from mid-September to mid-October. The critical temperatures are considered for winter – with a minimum temperature no lower than -15 °C – to avoid damage of buds and dormant inflorescences; with a minimum of -7 °C – to avoid damage of dehiscing catkins, and with a -3 °C in spring – to avoid damage of young shoots after bud break. Districts where the relative humidity was less than 70 % were considered undesirable (Mehlenbacher, 1991).

Mehlenbacher (1991) considered that a soil type has the significant effect on the growth and production of *Corylus avellana* L. On shallow soils, the hazelnut trees may grow well in their early years but subsequently do poorly and are often stunted (Thompson et al., 1996).

Hazelnut trees can make extensive root growth in winter when temperatures are above 5 °C (Thompson et al., 1996). This presumably enables them to develop a good root structure in spring that can obtain moisture for growth and kernel fill during the relatively dry summer months on July and August (Mehlenbacher, 1991).

There appears to be a relationship between soil type and rainfall. High yields, 1700 – 2000 $kg.ha^{-1}$, are achieved on deep, well drained soils with an annual rainfall of about 1100 mm (Mehlenbacher, 2005).

Germain and Sarraquigne (2004) recommended that in France, soils in the texture range sandy loam to clay loam with a clay content of no more than 20 – 40 %, should be selected. Botu and Turcu (2001) reported in their studies on soils in Romania, that soils with a clay content exceeding 35 – 40 % are unsuitable for hazelnut growing. Soils considered suitable are alluvial, brown forest and chernozems (black prairie soils). Germain and Sarraquigne (2004) also considered that very sandy soils are also unsuitable and should be avoided.

DOI: https://doi.org/10.15414/2020.9788055222622

Khokhlov (2001) concluded in studies on the southern coast of Crimea that the most suitable soils are those that are moist, slightly stony, with a light clay texture and are slightly alkaline.

A slightly acid to neutral soil reaction with a pH in water (pH) of 6 – 7 is considered desirable. However, some cultivars seem well-adapted to mildly alkaline conditions. In France, soil pH is considered to be relatively important (pH range of 6.2 - 7.8) is recommended by Germain and Sarraquigne, (2004).

1.1.4 Phenological phases and development of *Corylus avellana* L. flowers

In different European countries the hazelnut pollen season longs from 2 weeks to 1.5 months (Myszkowska et al., 2010). Pollen season depends from meteorological conditions such as temperature, humidity, precipitation, hours of sunshine, wind speed. Also climatic differences and geographic location has influence on pollination period (Kasprzyk et al., 2004).

Researchers as Myszkowska et al. (2010), Puc and Kasprzyk (2013) have admitted that the temperature is the most important factor, which influences a pollen concentration. Mimet et al. (2009) proposed that higher minimum of air temperatures in urban areas causes flowering period earlier than in rural areas. Črepinšek et al. (2012) pointed out that the base temperature for *Corylus avellana* L. flowering is 2 °C and T_{max} >10 °C (Stępalska et al., 2016). Increasing the air temperature on 1 °C shows a high sensitivity of male and female inflorescences of *C. avellana*.

Puc and Kasprzyk (2013) have noted the average start of pollination period for hazelnut from 15th February in Szczecin (Poland) and 12th March in Rzeszów (Poland). In Lithuania European hazel flowering period begins from 27th of March (Romanovskaja et al., 2012), because country belongs to the zone with cool temperate climate. Ianovici et al. (2013) has studied the pollen season in Timişoara (Romania) during 2000 – 2007 and for genus *Corylus* admitted the dates of start (28.02.; 19.02.; 11.02.; 17.02.; 16.02.; 07.03.; 27.02.; 13.01.) and dates of end (26.03.; 18.03.; 10.03.; 10.04.; 04.04.; 14.04.; 11.04.; 09.03.) of the pollen season. Length of the season is from 28 days (min) to 56 days (max). On the north-eastern Slovenia pollen shedding for some *Corylus* cultivars appears at 31st of January and from 1st to 15th of February. The first female flowers are noted in the first 10-day period of February (Črepinšek et al., 2012). Remišová and Vinceová (2007) obtained results about begging of *C. avellana* flowering in Slovakia and the average start date of European flowering is 15th of March. The average start day for phase of European hazel flowering in Zvolen (Slovakia), in period 2007 – 2012, is the 3rd of March (Zverko et al., 2014). Hájková et al. (2009) researched the earliest phenophases of *C. avellana* in Czech over the period 1992 – 2007. They investigated the pollen

DOI: https://doi.org/10.15414/2020.9788055222622

production in lowlands and mountains. For phase 'flower buttons visible' is determined the average start day date on 10th of January, and for phase 'beginning of flowering' is on 12th of January.

Hazelnut establishes male inflorescences in late summer. Chilling period (preceding pollination and period of dormancy) is required to enter a new growing cycle (Puc and Kasprzyk, 2013). Hazelnut dormancy period lasts until the day when cumulative temperature is stable (Suszka, 1980). Period with low temperatures is unfavorable for hazelnut growth (Puc and Kasprzyk, 2013). The accumulation of cooling at 0 – 7 °C is from November (15.11.) until February (28.02.), and then the increasing day requirements for the beginning of hazelnut flowering is due to 2 °C (Črepinšek et al., 2012).

During *C. avellana* flowering it is possible to observe female and male inflorescences (Germain, 1994). In hazelnut the developmental of male inflorescences begins 7 – 8 months before flowering (Frenguelli et al., 1994). The dormancy period begins in autumn, when the temperature is below 12.4 °C. Also, Frenguelli and Tedeschini (1997) reported that the temperature above 4.5 °C induces catkin axis elongation and release of first pollen grains. Young catkins grow about 0.15 mm per day during period from June until mid-October and in mid-October catkins reached a length around 22 mm. From November to the beginning of December catkins grow slowly. In January catkins stop growth and begin to release pollen (Frenguelli et al., 1994).

After four or five months of pollination fertilization occurs by the end of May or during first three weeks of June. The formation of ovules begins in March (Germain, 1994).

In April and May flower cluster drop is observed according to the apical dominance and along peduncle of catkins (Germain, 1994).

In phenological observation of hazelnut some authors (Kruczek and Puc, 2012; Puc and Kasprzyk, 2013) used the Łukasiewicz method for phenological classification of the generative plant development stages:

F_1 – appearance of the first flowers;

F_2 – beginning of full flowering (25 % of flowers open);

F_3 – first flowers being shed (withered);

F_4 – end of full flowering (75 % of flowers open);

F_5 – last flower buds;

F_6 – end of flowering (from the day when the last flowers finished blooming).

The full pollination period of hazelnut is determined from F_3 to F_5 periods (Puc and Kasprzyk, 2013). And the highest hazelnut pollen concentration falls within the phenophases F_3 and F_4 of the full pollination period.

DOI: https://doi.org/10.15414/2020.9788055222622

Silvestri (2015) in her work describes alternative *C. avellana* phenophases classification:

1. male flowering at stage Fm1 (start);
2. male flowering at stage Fm2 (peak);
3. male flowering at stage Fm3 (end);
4. female flowering at stage Ff1 (start);
5. female flowering at stage Ff2 (peak);
6. female flowering at stage Ff3 (end).

All these phenological phases occur in the period from the 4th week of February to the 3rd week of April.

1.1.5 Characteristic of *Corylus avellana* L. male flowers

The European hazelnut (*Corylus avellana* L.) is a monoecious plant with separate male and female flowers on the same tree. These flowers are borne on one-year-old shoots (Germain 1994). Earlier published work by Mercel (1988) does not include the information about morphological variability of male flowers of *C. avellana* L. The male inflorescences (catkins) are comprised of 130 – 290 flowers (Germain, 1994). Each flower contains 4 bifurcated stamens, each terminating in 2 anthers (Mehlenbacher, 2005; Piotrowska, 2008).

Fig. 2
Male flowers of *Corylus avellana* L. (Oravec A., 2017)

Male flowers (Fig. 2) are initiated before female flowers. In France, Germain (1994) reported the first signs of catkin differentiation occurred in buds from mid-May (Mehlenbacher, 2005). The stigmas can be receptive for at least 2 months from the time of exertion (the red dot stage). In time, the stigmas darken and wither. Stigmatic receptivity is at its optimum about 15 days from the beginning of anthesis (Germain, 1994). Only a few of the pollen grains caught by the stigmas produce a pollen tube that reaches the base of the style after germination. Pollen growth within the style is rapid, the pollen tube reaches the base of the style 4 – 10 days, where its growth ceases at the apex of

DOI: https://doi.org/10.15414/2020.9788055222622

the ovary. The tip broadens, develops a callose coat and goes into a quiescent phase (Germain, 1994). In the case of incompatible pollen, the pollen germinates but fails to penetrate between the cells of the stigma (Germain, 1994; Mehlenbacher, 2005). The pollen tubes grow and fertilisation occurs about the end of May to late June, that is, early summer in the northern hemisphere, depending on the cultivar (Germain, 1994).

1.2 Pollen development

Pollen is produced in microsporangia (contained in the flower anther of angiosperms). The internal layer of anther forms the archeosporial cells. After mitosis the archeosporial cells form diploid cells. Then diploid cell forms haploid cell of microspore at the meiosis. The anther has a four-sided structure and consists of four microsporangia located at the corners, two in each lobe. Microspores begin to divide at the meiosis without cytokinesis. Consequence of such division appears sprouted microspore called a pollen grain. Pollen grain has two cells of different sizes. The lager one is a cell of pollen tube (vegetative cell) and the smaller is called generative cell (Tokarev, 2002; Hesse et al., 2009).

Pollen grain develops from microspore, which is located in microsporangia and pollen development consists from two processes – microsporogenesis and microgametogenesis (Hesse et al., 2009; Ďurišová, 2018).

Microsporogenesis begins with differentiation of pollen mother cell (microsporocytes or microspore mother cell) and continues with microspores development from pollen mother cell though meiosis (Tiyayon, 2008; Borg and Twell, 2011).

Goldberg et al. (1993) characterized anther development by two phases. At the 1st phase anther morphology is established, microspore mother cells differentiation undergoes meiosis. After this phase most of specialized cells and tissues present at the anther. Microspores add up to tetrads within the pollen sacs. The cellular processes of anther cells' differentiation are regulated by histospecification program. At the 2nd phase pollen grains differentiate and increased the anther are pushed upward in the flower by filament extension. Degeneration of tissues and pollen grain release occur.

Microsporogenesis begins with differentiation microspore parent cell and it is covered by callose polysaccharide shell. These diploid cells undergo to meiotic or mitotic division and form four haploid microspores that form tetrads. The walls' forming of individual pollen grains occurs when

DOI: https://doi.org/10.15414/2020.9788055222622

they are under the cover. At the same time sporopollenin (biopolymer) forms a resistant outer shell of pollen grains (Tokarev, 2002; Hesse et al., 2009).

Groups of archesporial cells divide periclinary and form an outer, primary parietal and inner sporogenous layers. The primary parietal layer undergoes periclinal and anticlinal divisions to gives rise several concentric layers that differentiate into the endothecium, middle layers and the innermost tapetum. The primary sporogenous layer gives rise to the microsporocytes (meiocytes) directly or after further limited miotic divisions (Tiyayon, 2008).

Tapetum is the most compicuous anther cell layer, because this layer has metabolically active cells, which have short lifespan (Bedinger, 1992). There are two types of tapetum: the secretory (grandular or parietal) and the amoeboid (periplasmoidal) in the pollen wall (Hesse et al., 2009). Tapetal cells finish their physiological functions in the secretory type and in the amoeboid type cells tapetal cells lose their individuality by degeneration of the cell walls.

The granular tapetum builds up endoplasmic reticulum and dictyosome-derived vesicles while pollen mother cells are in prophase (Bedinger, 1992). Tapetum begins to break down after meiosis, releasing lyced cell walls and disintegrate cytoplasm into the locute (Tiyayon, 2009). The amoeboid tapetum is adjacent to specific areas of the microspore surface (exine) and provide regular nutrition for unusual pollen grains (Pacini, 1990). The genus *Corylus* has secretory (glandular) tapetun (Tiyayon, 2008).

Meiosis occurs in microspore mother cells. They are produced by mitotic divisions (Blackmore and Barnes, 1990). Microspore mother cells divide a lot of times and form diploid cells. These cells divide by meiosis (meiotically) and each cell forms four haploid microspores (pollen grains). There are two types of division in angiosperms, such as simultaneous and successive types. In simultaneous division pollen wall formation takes place after Meiosis-I and Meiosis-II. In simultaneous type of division cytokinesis occurs after Meiosis-I and Meiosis-II as well. The duration of meiosis can be as short for one day as long for three months (Tiyayon, 2008).

Microgametogenesis is the development of microspores into pollen grains. Hesse et al. (2009) admitted that microgametogenesis in angiosperms consists from two pollen mitosis. This process begins with the formation of the central vacuole inside mononuclear microscope. Gradually vacuole pushes the nucleus to the cell wall and formed pollen grain. The first mitosis leads to the formation of small generative cell and a large vegetative cell. During the second mitotic division, which takes place in pollen tube, generative cell is divided into two sperm cells. The process, when produce two sperm cells, is called spermatogenesis (D'Cruz et al., 2010).

DOI: https://doi.org/10.15414/2020.9788055222622

Microgametogenesis in angiosperms consists from two mitotic divisions that lead to the formation of male gametes. Gametogenesis begins with the formation of the central vacuole inside mononuclear microspore. Gradually vacuole pushes the nucleus to the cell wall and formed pollen grain. First mitosis leads to the formation of small generative cell and a large vegetative. Subsequently generative cell is separated from the cell wall and covered its own shell. During the second mitotic division, generative cell is divided into two, turns into a vegetative pollen tube, then the germination of pollen grains starts (Tokarev, 2002; Hesse et al., 2009).

Pollen mitosis of the bicellular pollen grain stage, pollenkitt, composed of lipids and proteins, is located at the interface of the tapetal plasma membrane and locus (Clément et al., 1998). Pollen grain is three-celled at the time of anthesis in 25 % of flowering plants (Blackmore and Barnes, 1990; Hesse et al., 2009), and in 75 % of flowering plants investigated that the pollen grain is two-celled (Hesse et al., 2009).

***Corylus avellana* L. pollen development**

In July the hazelnut sporogenic tissues begin intense mitotic activity (Frenguelli et al., 1994). Frenguelli et al. (1994) reported that in *C. avellana* pollen mother cell is vacuolated and wrapped in a thick callose layer in August. In August and mid-September, the first meiotic divisions occur and continue to the end of October (Frenguelli et al., 1994). The tetrad stage lasts throughout September. At that time callose with microspores is dissolved and the formation of the primary exine is stopped. So, *Corylus avellana* L. pollen grains development continues to mid-October, at the end of October binuclear pollen grains appear (Frenguelli and Tedeschini, 1997). During this period the first haploid mitosis occur. Also, Frenguelli et al. (1994) makes a point that polar axis of *C. avelana* pollen grain grow more than equatorial axis and the pollen shape change from oblate to oblate-spheroidal. So, pollen development continues to the end of October. At the beginning of the November pollen grains begin to decrease in sizes (Frenguelli et al., 1994).

1.3 Pollen morphological characterization

Pollen consists from individual pollen grains, which are formed in the anther and they are responsible for the function of reproduction. It is the male gametophyte of higher plants (Tokarev, 2002; Hesse et al., 2009).

Pollen grain has protoplast and two layered walls: exine and intine. Exine is robust and resistant to environmental influences. The exine function is as a protector of pollen from various

DOI: https://doi.org/10.15414/2020.9788055222622

environmental stresses and bacterial attack when pollen moves from anther to stigma (Scott, 1994; Zinkl et al., 1999; Scott et al., 2004). It contains a biopolymer sporopollenin. Exine consists from two parts: ectexine and endexine. Intine consists from cellulose and pectin. As usually in exine thin areas and apertures are present. Spinaperture, leptomate, colpus, pore, sulcate, extoaperture, endoaperture, margo, anullus, operculum, vestibulum, costa are types of apertures and help to characterize the structure of pollen grain (Tokarev, 2002; Hesse et al., 2009).

Exine (Fig. 3) is covered by bulges in the form outgrowths hill, knobs, mesh, and has a thinner seats and small holes – pores. The form of it can be spherical, oval, triangular, rod-like. The shape, size and surface features of pollen grains are very diverse. The color of pollen is often yellowish or orange, but sometimes brown, reddish, bluish like. In some plants flower generates thousands and even millions of pollen grains (Tokarev, 2002; Hesse et al., 2009).

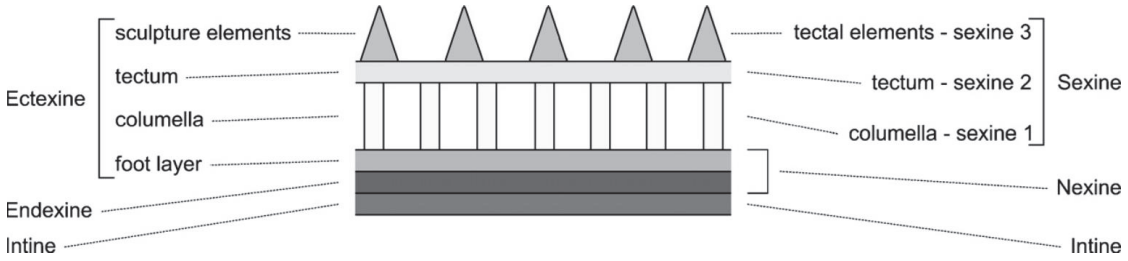


Fig.3
Exine stratification (by Punt et al., 2007)

Intine (Fig. 3) is a thin and delicate layer, composed mainly from pectin. Exine, compared with intine is relatively thick and layered. It contains extremely resistant carbohydrates, sporopollenin, which are practically insoluble in acids and alkalis. Exine has two layers: the outer – sexine (sculpture layer) and internal – nexine. That structure of sexine has many varieties and has a great systemic importance. Generally, in exine thin places are present and these holes serve to release the pollen tube. They are called apertures. Accommodation and form apertures are characterized by considerable variety. By placing they can be polar or zonal, and by form divide into furrows and pores. They provide pollen sculptures characteristic that is important to determine the type of pollen that has systematic basis (Tokarev, 2002; Hesse et al., 2009).

The shape, size, surface, structure, sculpture, structure apertures of exine, their location are varied, but they are constant in the same species. The size of pollen grains ranges from 7 – 10 to 250 μm.

DOI: https://doi.org/10.15414/2020.9788055222622

The polar axis is an imaginary line that runs through the body of pollen grains on its outer (distal) to external (proximal) surface. Pollen can be in solitary form or in groups of two or more pollen grains. Geometrical axis of each pollen grain meets at the geometric center of the complex of pollen particles, in the size of pollen grains are divided into classes defined by the maximum length of the polar axis, or equatorial axis. There are very small (smaller than 10 μm), small (10 – 25 μm), medium (25 – 50 μm), large (50 – 100 μm) and very large (100 – 200 μm), huge (more than 200 μm) pollen grains. Small and medium sizes (20 to 50 μm) pollen causes polynosis. In relation to the length of the polar axis, equatorial diameter distinguished flattened, spherical, oblong form of pollen grains (Tokarev, 2002; Hesse et al., 2009).

Corylus avellana L. pollen morphology

Pollen grain of *C. avellana* (18 × 26 μm) is tripolar with deep structures (lenticular spherical structures, which are unstable to acetolysis and placed at apertures) in each pore (Hofman and Michalik, 1998). They are characterized as angle-apertures (Erdtman, 1952), equatorial apertures and apertures which are located in every corner of the pollen grain. According to previous studies, the average diameter of hazelnut pollen grains is 24.20 μm (Dyakowska, 1959).

Pollen grain of *C. avellana* is smooth, 3-pored grain with a sub-triangular polar view and sub-oblate equatorial view. The surface sculpturing is scabrate (Moore et al., 1991), but usually appears smooth after preparation. Accordingly, to Polleninfo.org database (Pollenatlas, 2013), pollen grains of *C. avellana* were characterized by following features – shape (round, triangular in polar view, and oval in equatorial view), size (polar axis 27.3 μm, equatorial axis 28.9 μm), appertures (tripolar pollen grains with pores of 2 – 3 μm in diameter), pollen shell (rough exine, thin intine with very large bulge regions – lenticular structures); additional attributes – granular cytoplasm.

DOI: https://doi.org/10.15414/2020.9788055222622

2 Goals of monograph

The main aim

Morphological characterization and evaluation of *C. avellana* L. genotypes from different localities of Slovakia (natural, urban, industrial), focused on male reproduction organs (catkins and pollen grains), combined with innovative approaches of image analysis tools can provide a comprehensive and more precise tool for identifying suitable genotypes for further use in the field of plant breeding, and ecology.

Particular aims

1. The characterization of ecological features selected locations in Slovakia, and its connection with data obtained by others experimental methods.
2. Morphological characterization of *C. avellana* catkins and pollen grains.
3. Using the image analysis software as a new and precise approach of phenotype characterization.
4. Determination the pollen production from *C. avellana* catkins.
5. Combining the data obtained by different experimental approaches, and to find the new relationships among genotypes.

Hypothesis

We assume, morphological traits are appropriate for identification, evaluation and classification of intraspecific variability of *C. avellana* genotypes. The assumption of this hypothesis will be the significant differences between genotypes, which grew in different environmental conditions (natural, urban and industrial).

Also, we watched the influence of different environmental conditions (natural, urban and industrial) on phenotypic expression in selected morphological traits.

DOI: https://doi.org/10.15414/2020.9788055222622

3 Materials and methods

3.1 Localities and samples characterization

Hazelnut (*Corylus avellana* L.) is an object of the study. In our experiment we evaluated 14 genotypes from different territories of Slovakia. Table 2 and figure 4 show the selected samples, their geographic location. The closer description of selected localities is described in the Annexes. *Corylus avellana* L. branches with inflorescence were taken. Male inflorescence (catkins) were in stage of dormice (January 2017). From the lowest level of one tree 15 – 20 branches were taken. There were from 2 – 4 to 8 – 12 catkins on one branch. Temperature of outside was 0 – 6 °C. Branches with catkins were put to glass with water at room temperature.

Fig. 4

Map of *Corylus avellana* L. pollen samples preparation in the Slovak Republic

DOI: https://doi.org/10.15414/2020.9788055222622

Tab. 2

Localities of *Corylus avellana* L. samples

Mark	Location	Geographical coordinates		Type of territories	Type of growing
		Latitude	Longitude		
CA-S-01	Čertova pec	48°33'29.1276" N	17°54'57.3912" E	forest (natural – N)	wild (w)
CA-S-02	Banka-1	48°34'45.624" N	17°51'13.9788" E	village (urban – U)	wild (w)
CA-S-03	Banka-2	48°34'43.6332" N	17°51'14.0184" E	village (urban – U)	wild (w)
CA-S-04	Zemianske Podhradie	48°50'24.2304" N	17°49'42.5172" E	village (urban – U)	wild (w)
CA-S-05	Moravské Lieskové	48°49'1.2108" N	17°47'28.5684" E	village (urban – U)	wild (w)
CA-S-06	Ivanka pri Nitre	48°14'42.8928" N	18°6'44.7732" E	village (urban – U)	wild (w)
CA-S-07	Jelenec-1	48°23'51.937" N	18°12'39.591" E	village (natural – N)	wild (w)
CA-S-08	Jelenec-2	48°23'52.014" N	18°12'38.973" E	village (natural – N)	wild (w)
CA-S-09	Žirany	48°22' 52.887" N	18°10'36.864" E	village (urban – U)	wild (w)
CA-S-10	Nitra-1 – Hallská obrovská	48°18'8.646" N	18°6'5.473" E	city (industrial – I)	cultivar (c)
CA-S-11	Nitra-2 – Lombardská biela	48°18'8.363" N	18°6'5.647" E	city (industrial – I)	cultivar (c)
CA-S-12	Nitra-3 – Webbova	48°18'8.222" N	18°6'5.724" E	city (industrial – I)	cultivar (c)
CA-S-13	Nitra (Zobor-1)	48°20'45.137" N	18°5'31.929" E	forest (natural – N)	wild (w)
CA-S-14	Nitra (Zobor-2)	48° 20' 46.241" N	18° 5' 25.576" E	forest (natural – N)	wild (w)

3.2 Biological objects and sampling

In our research we worked with objects:

a. Pollen from selected plant species. In the experiments, the samples abbreviated as “CA” – in full name – CA-P-01;
b. Catkins – hazelnut male inflorescences, obtained by mechanical harvesting from the same genotype as a pollen. In the experiments, the samples abbreviated as “CA” – in full name – CA-P-01.

DOI: https://doi.org/10.15414/2020.9788055222622

For evaluation length, weight and diameter of inflorescence 50 catkins were taken. For samples evaluation of *C. avellana* pollen we used inflorescences, which were prepared by mechanic method – vacuuming or shaking down from the flowers from different genotypes, which grew in different agro-ecological conditions.

3.3 Image documentation

The analyzed biological objects – pollen grains, and catkins were shot by different macroscopic and microscopic systems. The samples for image documentation were prepared according to used facilities (Tab. 3).

Tab. 3

Characteristics of technical equipment for image documentation

Facility	**MIAS 'AeroScope' (NL)**	**Sony (No DSC-HX1)**	**SEM Carl Zeiss Evo LS 15**
Camera	-	Optical lens G (2.8-5.2/5.0-100.0)	Electron optical column
Image acquiring	2048 × 1536 pixel, 24 bit RGB	3456 × 2304 pixel, 24 bit sRGB color	1024 × 768 pixel, 24 bit RGB
Software	PC – Program for the Identification and Classification of stained biological Particles in Microscopic Images – PCS	Digital camera	Smart SEM
Image format	(.jpg), 1056 images from every sample	(.jpg), 12 images from every sample	(.tif), 15 images from every sample

Samples preparation for microscopic analysis:

1. **Microscope image analysis system AeroIScope®** – The pollen for this analysis was gathered in January 2017. Labels and numbers were written on every microscope slide. A drop of water was put on slide glasses. Pollen sample was put on the drop of water. Fixing mixture (glycerin + fuchsine ($C_{20}H_{19}N_3 \cdot HCl$)) was used for sample fixation. After that a cover glass closed pollen sample. Samples were dried in the laboratory at room temperature (Tab. 4);
2. **Sony camera** – catkins were obtained in January 2017 (Tab. 4);

DOI: https://doi.org/10.15414/2020.9788055222622

3. **Scan Electron microscope Carl Zeiss Evo LS 15** – pollen was gathered from catkins in January 2015. The dried pollen was put on the one side of double-sided adhesive tape and the second side tape was attached to the metallic table (diameter 10 mm) (Tab. 4);

Tab. 4

Number of obtained images

Microscope	Number of samples	Number of images per sample	Total number of images
MIAS 'AeroScope' (NL)	14	1056	14784
Sony camera (No DSC-HX1)	14	12	168
SEM Carl Zeiss Evo LS 15	14	17	238

Images of different types of biological objects, which were obtained by using camera, light and scan electron microscopes, are presented at the Fig. 5 – 7.

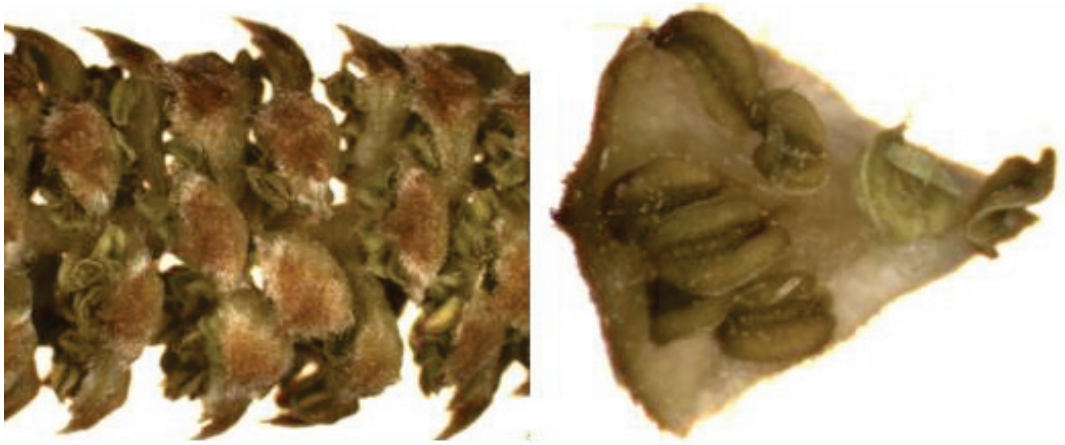


Fig. 5

***Corylus avellana* L. male flowers (Light microscope SteREO Discovery V20)**

DOI: https://doi.org/10.15414/2020.9788055222622

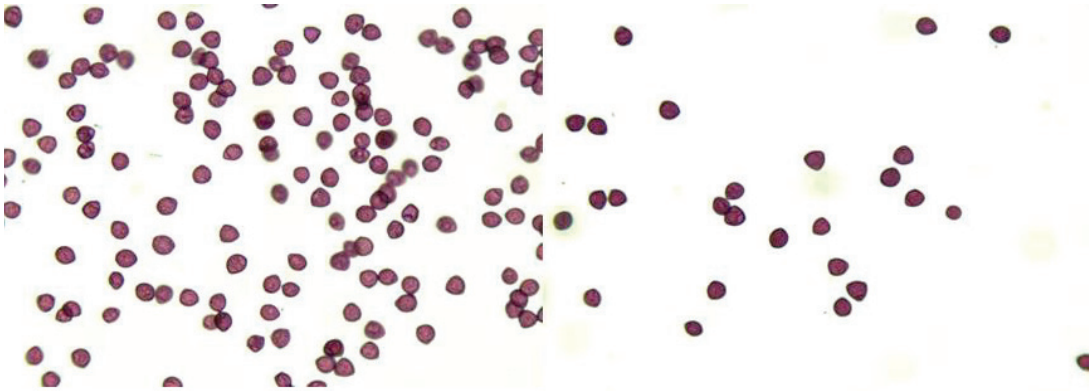


Fig. 6

***Corylus avellana* L. pollen (Microscope image analysis system AeroIScope®)**

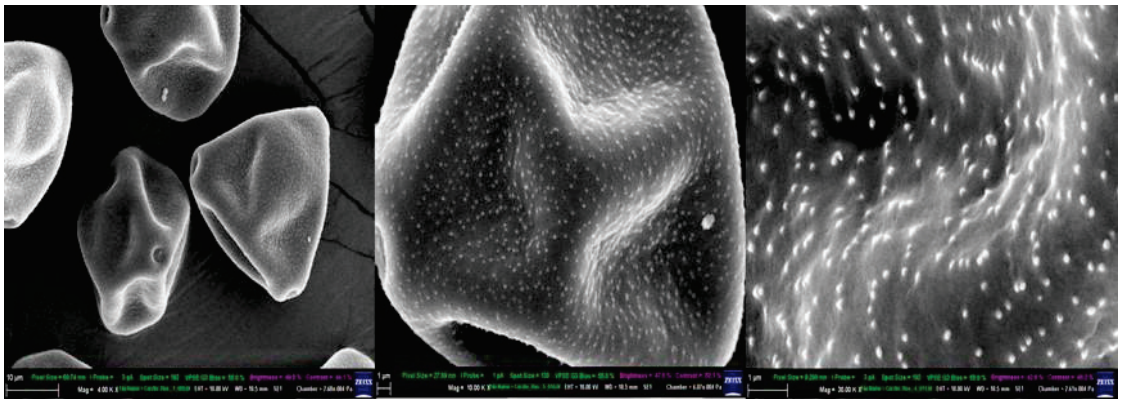


Fig. 7

***Corylus avellana* L. pollen (Electron scanning microscope SEM Carl Zeiss LS 15 EM Evo)**

3.4 Characterization and morphological analysis of catkins and pollen grains

3.4.1 Characterization of *Corylus avellana* L. male flowers

On 50 catkins, taken from every *Corylus avellana* genotypes, were measured length, diameter (mm) and weight (g). These catkins were harvested in January 2017.

Also, the pollen yield (g) was calculated. It was based on the weight difference of 50 catkins before pollen shedding and after pollen release. The methodology was adapted according to Piotrowska (2008).

The number of flowers per inflorescence was counted on 25 inflorescences. The catkins for these analyses were gather in January 2017.

DOI: https://doi.org/10.15414/2020.9788055222622

3.4.2 Morphology of *Corylus avellana* L. pollen grains

Pollen grains morphology was studied on images obtained by scanning electron microscope ZEISS Evo LS 15. Measurement of the morphometric parameters was carried out on 100 pollen grains per genotype by using software Smart SEM. The analysis was realized on pollen which was gathered from catkins harvested in January 2015.

Pollen grains were characterized by parameters: polar axis (P – line which connects proximal pole with distal pole), equatorial axis (E – line locates in equatorial plane and is perpendicular to the polar axis) and aperture diameter (this is a round aperture in which the ratio of length to width is less than two) (Fig. 8), form of pollen grains (elongation factor) was defined by the ratio of the polar axis to the value of the equatorial diameter (P/E).

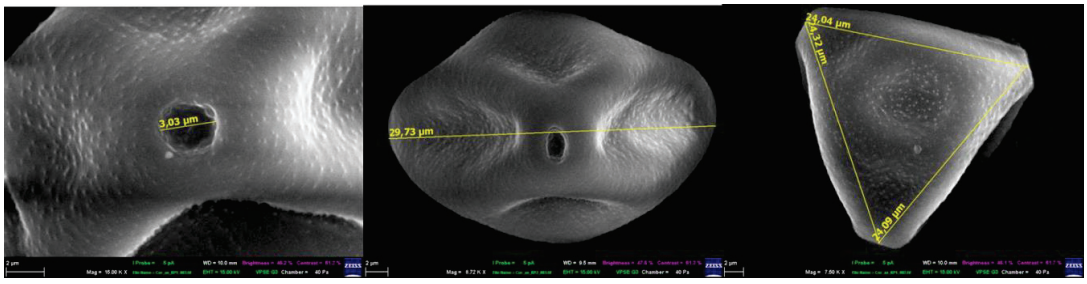


Fig. 8

Morphological parameters of *Corylus avellana* L. pollen: a) diameter of aperture (µm), b) equatorial axis (µm), c) polar axis (µm)

For describing the morphological features of pollen grains was used standard terminology (Kremp 1967; Meyer-Melykyan 1987). For the analysis of the forms of pollen grains the classifier "The forms of pollen grains" (Meyer-Melykyan et al., 1999), and for analysis of sizes – classifier by Tokarev (2002) were used.

Image analysis

The other method we used for analysis of morphology of pollen grains was the image analysis. Nowadays, it is rapidly developed area of research together with development of microscopic devices and related software. We used it for morphology characterization of *C. avellana* L. pollen grains. The pollen was obtained from catkins harvested in January 2017. The main reason of using this method was automatization of pollen grains selection (as region of interest – ROI) and their quantification.

DOI: https://doi.org/10.15414/2020.9788055222622

For analysis we used the software CellProfiler (open source), which is cited in many highly evaluated journals (Carpenter et al., 2006; Lamprecht et al., 2007; Bray and Carpenter, 2018; McQuin et al., 2018). The images we used for this purpose were prepared by automated microscope AeroIScope®.

Firstly, we selected only well focused images. The total number of images used for analysis was 760. The genotypes we selected for analysis are in the Tab. 5.

Tab. 5

The genotypes used for pollen grains image analysis

Sample	Location	Number of images
CA-S-01	Čertova pec	92
CA-S-04	Zemianske Podhradie	79
CA-S-05	Moravské Lieskové	47
CA-S-07	Jelenec-1	158
CA-S-09	Žirany	52
CA-S-10	Nitra-1 (Hallská obrovská)	106
CA-S-12	Nitra-3 (Webbova)	40
CA-S-14	Nitra (Zobor-2)	186
TOTAL number of images		**760**

The different number of selected images per genotype depended on image quality.

The second step was building the pipeline. The pipeline is a sequential set of image analysis modules. The algorithm of pollen grains analysis is presented in the Fig. 9 and in the following steps:

1. To begin creating the project the images were uploaded.
2. The **NamesAndTypes** module allows to assign a meaningful name to each image by which other modules will refer to it.
3. **ColorToGray** module convert the color images to grayscale and then use ApplyThreshold to generate a binary image.
4. **ImageMath** module is used if the objects are black and the background is white, we must invert the intensity using this module.
5. Module **IdentifyPrimaryObjects** identifies objects in an image. This module identifies primary objects in grayscale images containing bright objects on a dark background. Primary

DOI: https://doi.org/10.15414/2020.9788055222622

objects (e.g., cells) are those that can be found in an image without using any corresponding reference image.

6. Then we set different measurement parameters for – object intensity, object size and shape, and texture. **Object Intensity** measures several intensity features for identified objects and measurements are recorded for each object. **Object Size Shape** measures several area and shape features of identified objects, and **texture** measures the degree and nature of textures within objects.
7. The module **ExportToSpreadsheet** exports measurements into one or more files that can be opened in Excel or other spreadsheet programs.
8. **ExportToDatabase** exports data directly to a database, or in database readable format, including an imported file with column names and **a CellProfiler Analyst properties file**, which we used for further pollen grains classification.

For description of selected modules, we used the CellProfiler manual (Carpenter and Jones, 2019). CellProfiler is designed to analyze images in a high-throughput manner. Once a pipeline has been established for a set of images, CellProfiler can export batches of images to be analyzed on a computing cluster with the pipeline (Fig. 9). Totally was analyzed 760 images, 27 286 ROI's (pollen grains) was selected and they were measured by 126 characters for intensity, size, shape, and texture.

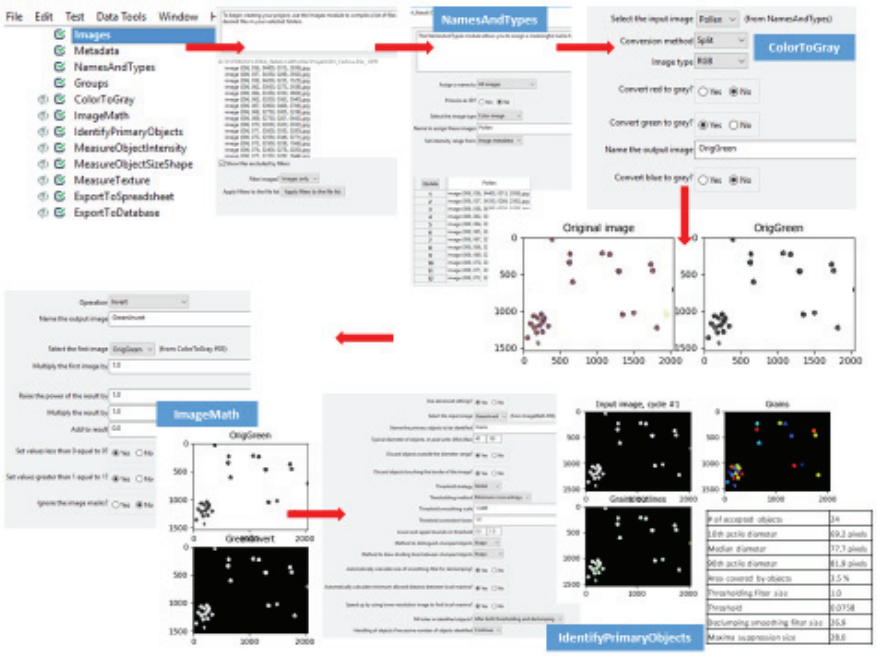


Fig. 9

Scheme of pipeline for automatic selection and analysis of *C. avellana* pollen grains

DOI: https://doi.org/10.15414/2020.9788055222622

The classification of all segmented objects from uploaded images was the other step of image analysis. For this reason, we used the software CellProfiler Analyst (Open Source). CellProfiler Analyst (CPA) allows interactive exploration and analysis of data, particularly from high-throughput, image-based experiments. Included is a supervised machine learning system which can be trained to recognize complicated and subtle phenotypes, for automatic scoring of millions of cells. CPA provides tools for exploring and analyzing multidimensional data, particularly data from high-throughput, image-based experiments analyzed by its companion image analysis software, CellProfiler (Jones et al., 2008; Jones et al., 2009; Dao et al., 2016).

In our experiment we set up simple way how to use this software. The steps of analysis are displayed on pipeline (Fig. 10). We selected the module Classifier. From prepared database we selected images randomly and manually sorted the unclassified objects (pollen grains) into classification bins (positive, negative). In our case positive bin meant pollen grains viewed in polar axis (Fig. 10). The negative bin meant all other pollen grains and objects in images which were not displayed in polar axis view (Fig. 10). This way we prepared the training set which consisted from 100 pollen grains for positive and 100 pollen grains form negative bin. During this process we were also preparing the rules set. Final set consisted from 15 rules. When we were satisfied with classification accuracy of software (Fig. 10) and we corrected errors, then we pressed the Score All button.

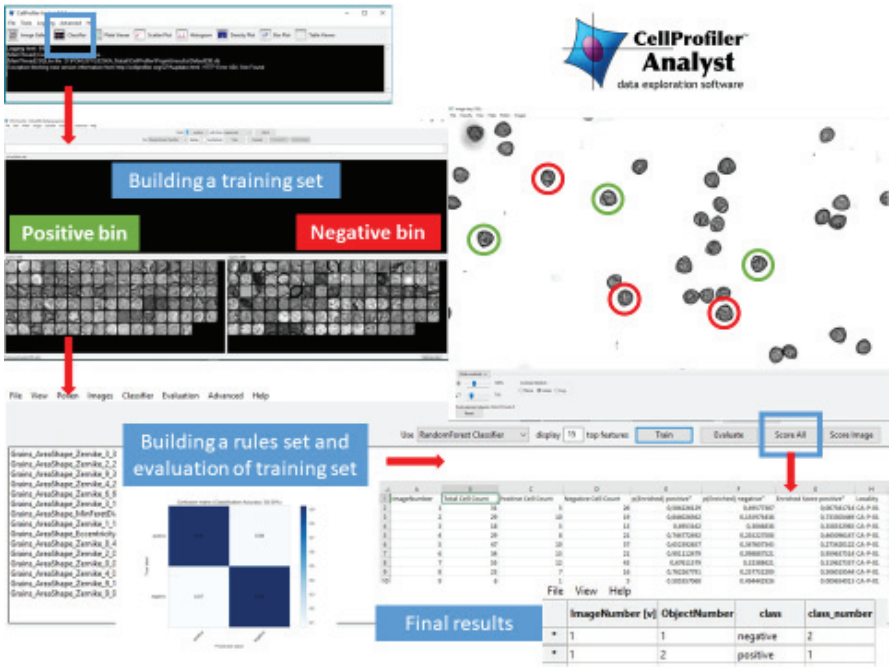


Fig. 10

Pipeline for classifying *C. avellana* L. pollen grains by software CellProfiler Analyst

DOI: https://doi.org/10.15414/2020.9788055222622

3.5 The software used in the processing of experimental activities

In our research we used the next software:

- MS Excel – creating tables, graphs, processing of experimental data;
- MS Word – text processing;
- SmartSEM – image acquisition;
- JSM Imagine system – image acquisition;
- CellProfiler and CellProfiler Analyst - image analysis software, machine learning software;
- STATISTIKA 10 – statistical analysis (descriptive statistics, ANOVA – Duncan's test and Kruscall-Wallis test (non-parametric statistics), Principal component analysis – PCA);

DOI: https://doi.org/10.15414/2020.9788055222622

4 Results and discussion of monograph

4.1 Morphometric analysis of *Corylus avellana* L. pollen grains

In our study a total 100 pollen grains from 14 genotypes were evaluated (Tab. 2). We measured the polar, equatorial axis, and diameter of aperture (µm). Extremes were deleted. Based on morphological parameters of pollen grains from different locations the statistically significant differences were exposed by ANOVA. The average values of polar axis, equatorial axis and aperture diameter were analyzed.

The polar axis, equatorial axis and aperture are one of the important characteristics of the pollen grain of *C. avellana* and respond for species affiliation.

4.1.1 Polar axis

In our results the average value of polar axis was 24.91 µm. Our value is higher than average value of measurements in the Erdtman's study (1952) and Hofman and Michalik (1998), they mentioned 14 – 15 µm and 18 µm, respectively. The possible reasons of inconsistency are different chemical solution for pollen sample preparation and technical equipment. For instance, we used dry pollen, and Blackmore et al. (2003) used glycerine jelly for pollen preparation. But our results had found out confirmation among results (18 – 25 µm) by Blackmore et al. (2003).

Basic descriptive characteristics of polar axis of pollen grains of *C. avellana* are listed in Tab. 6. Graphical presentation of descriptive statistics is displayed in Fig. 11. Minimum average value of polar axis was determined in cultivar Lombardská biela (CA-P-11). It was 21.43 µm. Maximum average value (26.96 µm) of polar axis was determined in plants sampled at Čertova pec locality (CA-P-01). The most samples from natural zones were in the range from 23.82 µm to 26.96 µm. The most samples from urban zones were in the range from 24.63 µm to 26.76 µm. Samples from industrial zone (cultivars) were in the range 21.43 µm to 22.04 µm.

DOI: https://doi.org/10.15414/2020.9788055222622

Tab. 6
Descriptive statistics of hazelnut pollen grains' parameter (polar axis, µm) at different locations

Sample	Location	n	$\bar{x}$	min	max	StDev	StEr	CV, 100%
CA-P-01	Čertova pec (N)	100	26.96	24.46	29.49	1.06	0.10	3.94
CA-P-02	Banka-1 (U)	100	26.63	23.54	29.01	0.95	0.09	3.56
CA-P-03	Banka-2 (U)	100	24.96	20.37	28.28	1.53	0.15	6.15
CA-P-04	Zemianske Podhradie (U)	99	26.76	23.94	29.49	1.21	0.12	4.53
CA-P-05	Moravské Lieskové (U)	97	25.61	21.03	29.57	1.90	0.19	7.45
CA-P-06	Ivanka pri Nitre (U)	97	24.63	20.80	29.73	1.97	0.20	8.03
CA-P-07	Jelenec-1 (N)	97	26.95	22.80	29.68	1.54	0.15	5.73
CA-P-08	Jelenec-2 (N)	98	25.80	20.85	28.99	1.71	0.17	6.64
CA-P-09	Žirany (U)	100	25.56	21.22	28.80	1.45	0.14	5.70
CA-P-10	Nitra (Hallská obrovská) (I)	98	22.04	20.13	24.73	1.00	0.10	4.54
CA-P-11	Nitra (Lombardská biela) (I)	78	21.43	20.02	24.64	1.22	0.13	5.73
CA-P-12	Nitra (Webbova) (I)	95	21.98	19.99	25.91	1.15	0.11	5.26
CA-P-13	Nitra (Zobor 1) (N)	99	25.72	20.16	29.53	1.95	0.19	7.58
CA-P-14	Nitra (Zobor 2) (N)	99	23.82	20.07	27.68	1.80	0.18	7.56

Legend: n – count of pollen grains for one sample; $\bar{x}$ – average value; min – minimum; max – maximum; StDev – standard deviation; StEr – standard error. N – nature zones; U – urban zones; I – industrial zones.

Samples from industrial zones with 21.43 – 22.04 µm of polar axis, CA-P-10, CA-P-11 and CA-P-12, were from territories with neutral to weakly alkaline pH (Annexes). Precipitation totals were 700 – 800 mm. According to climate region the sum of average daily temperatures above 10 °C was from > 3000 to 2500 with length of time with air temperature above 5 °C in days was from 242 to 231 (Annexes).

Samples from urban zones with 24.63 – 26.76 µm of polar axis, CA-P-02, CA-P-03, CA-P-05, CA-P-06, CA-P-08 were from territories with neutral to weakly alkaline pH, sample CA-P-09 was from soils with very strong alkaline pH, and sample CA-P-13 was from weakly alkaline pH soils (Annexes). Precipitation totals were 700 – 800 mm. The sum of average daily temperatures above 10 °C was from 3000 to 2500 with length of time with air temperature above 5 °C in days was from 237 to 231 (Annexes).

For samples with 23.82 – 26.96 µm polar axis from natural zones (CA-P-01, CA-P-07, CA-P-08, CA-P-13, CA-P-14) the difference was in precipitation totals, climate region and pH (H_2O).

Significant differences between pairs of samples were identified by Duncan's test. The statistically significant differences in polar axis were found in the most cases (Tab. 7). We looked on genotypes, which differ from all other genotypes, or on genotypes where was not confirmed

DOI: https://doi.org/10.15414/2020.9788055222622

significant difference in one case. According to this scheme we selected genotypes – CA-P-03, CA-P-06, CA-P-10, CA-P-11, CA-P-12, and CA-P-14. We can confirm our hypothesis, that polar axis is discriminating genotypes, but we did not find influence of environmental conditions on phenotypic expression.

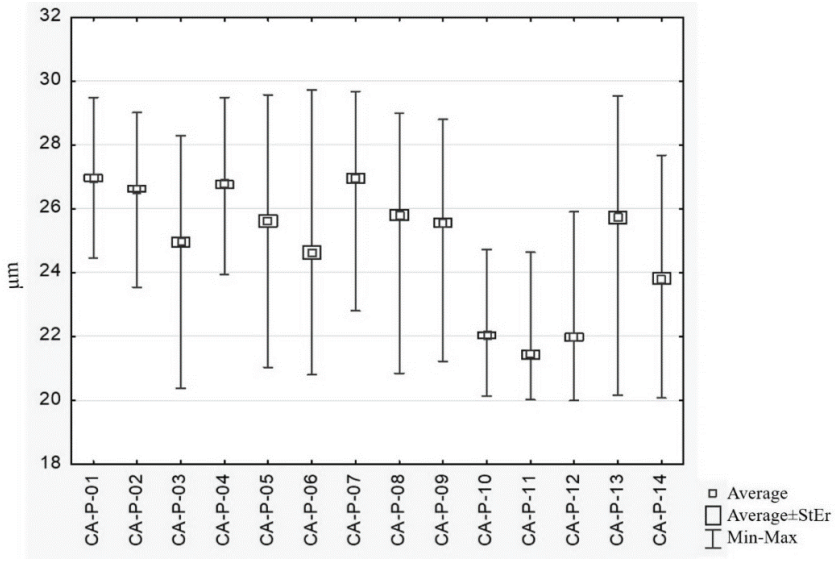


Fig. 11

Comparison of average value and variation of polar axis of *C. avellana* L. pollen grains in different locations

Tab. 7

Results of Duncan's post-hoc test of differences among averages for *C. avellana* L. sample (location) as independent variables and polar axis as a dependent variable

Sample	(1) M=26.9	(2) M=26.6	(3) M=24.9	(4) M=26.7	(5) M=25.6	(6) M=24.6	(7) M=26.9	(8) M=25.8	(9) M=25.5	(10) M=22.0	(11) M=21.4	(12) M=21.9	(13) M=25.7	(14) M=23.8
01 (1)		0.170	**0.000**	0.383	**0.000**	**0.000**	0.981	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**
02 (2)	0.170		**0.000**	0.559	**0.000**	**0.000**	0.160	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**
03 (3)	**0.000**	**0.000**		**0.000**	**0.003**	0.126	**0.000**	**0.000**	**0.006**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**
04 (4)	0.383	0.559	**0.000**		**0.000**	**0.000**	0.363	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**
05 (5)	**0.000**	**0.000**	**0.003**	**0.000**		**0.000**	**0.000**	0.406	0.808	**0.000**	**0.000**	**0.000**	0.615	**0.000**
06 (6)	**0.000**	**0.000**	0.126	**0.000**	**0.000**		**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**
07 (7)	0.981	0.160	**0.000**	0.363	**0.000**	**0.000**		**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**
08 (8)	**0.000**	**0.000**	**0.000**	**0.000**	0.406	**0.000**	**0.000**		0.309	**0.000**	**0.000**	**0.000**	0.699	**0.000**
09 (9)	**0.000**	**0.000**	**0.006**	**0.000**	0.808	**0.000**	**0.000**	0.309		**0.000**	**0.000**	**0.000**	0.486	**0.000**
10 (10)	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**		**0.006**	0.787	**0.000**	**0.000**
11 (11)	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.006**		**0.010**	**0.000**	**0.000**
12 (12)	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	0.787	**0.010**		**0.000**	**0.000**
13 (13)	**0.000**	**0.000**	**0.000**	**0.000**	0.615	**0.000**	**0.000**	0.699	0.486	**0.000**	**0.000**	**0.000**		**0.000**
14 (14)	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	

Legend: statistically significant differences marked out by red color; $p<0.05$ – statistically significant difference. N – nature zones (1, 7, 8, 13, 14); U – urban zones (2, 3, 4, 5, 6, 9); I – industrial zones (10, 11, 12).

DOI: https://doi.org/10.15414/2020.9788055222622

4.1.2 Equatorial axis

In our study we evaluated the average value of equatorial axis. It was 26.65 µm. Our result responds to Hofman and Michalik (1998) – 26 µm, and Blachmore et al. (2003) – 24 – 30 µm. In case with equatorial axis, our results were higher than Erdtman's values, too (Erdtman, 1952).

Basic descriptive characteristics of equatorial axis pollen grains are listed in Table 8. Graphical presentation of descriptive statistics for equatorial axis was displayed in Fig. 12. Minimum average value of equatorial axis was determined in Nitra in cultivar Lombardská biela (CA-P-11) with 22.24 µm, respectively. Maximum average value (29.63 µm) of equatorial axis was determined in plants sampled at Zemianske Podhradie locality (CA-P-04). The most samples originated from natural zones were in the range from 26.12 µm to 28.26 µm. The most samples obtained from urban zones were in the range from 25.41 µm to 29.63 µm. Samples collected from industrial zone (cultivars) were in the range from 22.24 µm to 23.34 µm.

Tab. 8

Descriptive statistics of hazelnut pollen grains' parameter (equatorial axis, µm) at different locations

Sample	Location	n	$\bar{x}$	min	max	StDev	StEr	CV, 100%
CA-P-01	Čertova pec (N)	100	28.13	23.58	31.97	1.71	0.17	6.08
CA-P-02	Banka-1 (U)	100	29.01	26.67	32.42	1.01	0.10	3.49
CA-P-03	Banka-2 (U)	100	27.94	23.46	31.95	1.64	0.16	5.88
CA-P-04	Zemianske Podhradie (U)	98	29.63	25.41	32.38	1.69	0.17	5.71
CA-P-05	Moravské Lieskové (U)	100	28.44	23.52	31.93	1.65	0.16	5.80
CA-P-06	Ivanka pri Nitre (U)	96	25.41	19.81	29.35	2.15	0.21	8.47
CA-P-07	Jelenec-1 (N)	98	28.26	21.17	32.36	2.46	0.24	8.71
CA-P-08	Jelenec-2 (N)	97	26.97	20.60	32.42	2.66	0.27	9.86
CA-P-09	Žirany (U)	100	27.24	20.97	31.14	1.97	0.19	7.23
CA-P-10	Nitra (Hallská obrovská) (I)	86	23.01	17.79	26.57	1.50	0.16	6.55
CA-P-11	Nitra (Lombardská biela) (I)	82	22.24	20.50	25.93	1.22	0.13	5.50
CA-P-12	Nitra (Webbova) (I)	95	23.34	20.52	26.94	1.43	0.14	6.13
CA-P-13	Nitra (Zobor 1) (N)	100	27.45	22.88	32.30	2.00	0.20	7.30
CA-P-14	Nitra (Zobor 2) (N)	99	26.12	20.77	29.81	2.07	0.20	7.92

Legend: n – count of pollen grains for one sample; $\bar{x}$ – average value; min – minimum; max – maximum; StDev – standard deviation; StEr – standard error. N – nature zones; U – urban zones; I – industrial zones.

DOI: https://doi.org/10.15414/2020.9788055222622

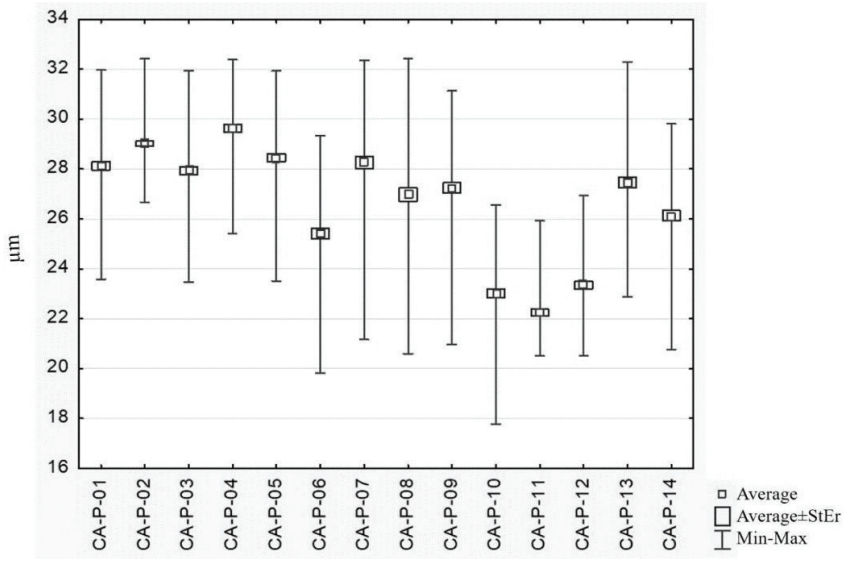


Fig. 12

Comparison of average value and variation of equatorial axis of *C. avellana* L. pollen grains in different locations

For samples with 26.12 – 28.26 µm equatorial axes from natural zones (CA-P-01, CA-P-07, CA-P-08) the difference was in precipitation totals and climate region. pH (H_2O) was the same for all samples.

For samples from urban zones (CA-P-03, CA-P-05, CA-P-06, CA-P-09, CA-P-13, CA-P-14) the difference was in pH (H_2O) soils (from neutral to weakly alkaline pH and very strong alkaline pH for sample CA-P-09) and climate region (the sum of average daily temperatures above 10 °C was from 3000 to 2500 with length of time with air temperature above 5 °C in days was from 237 to 231, but for sample CA-P-09 the sum of average daily temperatures above 10 °C was from >3000 to 2800 with length of time with air temperature above 5 °C in days was from 242 to 237). Precipitation totals was the same for all samples (Annexes).

Samples from industrial zones with 22.24 – 23.34 µm equatorial axis, CA-P-10, CA-P-11 and CA-P-12, were from territories with neutral to weakly alkaline pH (Annexes). Precipitation totals were 700 – 800 mm. According to climate region the sum of average daily temperatures above 10 °C was from > 3000 to 2500 with length of time with air temperature above 5 °C in days was from 242 to 237 (Annexes).

Significant differences between pairs of samples identified by Duncan's test. The statistically significant differences in equatorial axis were found in the most cases (Tab. 9). Again, we looked on genotypes, which differ from all other genotypes, or on genotypes where was not confirmed

DOI: https://doi.org/10.15414/2020.9788055222622

significant difference in one case. According to this scheme we selected genotypes – CA-P-02, CA-P-04, CA-P-06, CA-P-10, CA-P-11, CA-P-12, and CA-P-14. We can confirm our hypothesis, that equatorial axis is discriminating genotypes, but we did not find influence of environmental conditions on phenotypic expression.

Tab. 9

Results of Duncan's post-hoc test of differences among averages for *C. avellana* L. sample (location) as independent variables and equatorial axis as a dependent variable

Sample	(1) M=28.13	(2) M=29.01	(3) M=27.94	(4) M=29.63	(5) M=28.45	(6) M=25.41	(7) M=28.26	(8) M=26.97	(9) M=27.24	(10) M=23.01	(11) M=22.24	(12) M=23.34	(13) M=27.45	(14) M=26.12
01 (1)		**0.001**	0.474	**0.000**	0.266	**0.000**	0.610	**0.000**	**0.001**	**0.000**	**0.000**	**0.000**	**0.015**	**0.000**
02 (2)	**0.001**		**0.000**	**0.020**	**0.036**	**0.000**	**0.007**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**
03 (3)	0.474	**0.000**		**0.000**	0.082	**0.000**	0.250	**0.000**	**0.013**	**0.000**	**0.000**	**0.000**	0.071	**0.000**
04 (4)	**0.000**	**0.020**	**0.000**		**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**
05 (5)	0.266	**0.036**	0.082	**0.000**		**0.000**	0.499	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**
06 (6)	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**		**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.007**
07 (7)	0.610	**0.007**	0.250	**0.000**	0.499	**0.000**		**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.004**	**0.000**
08 (8)	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**		0.314	**0.000**	**0.000**	**0.000**	0.092	**0.001**
09 (9)	**0.001**	**0.000**	**0.013**	**0.000**	**0.000**	**0.000**	**0.000**	0.314		**0.000**	**0.000**	**0.000**	0.438	**0.000**
10 (10)	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**		**0.004**	0.216	**0.000**	**0.000**
11 (11)	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.004**		**0.000**	**0.000**	**0.000**
12 (12)	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	0.216	**0.000**		**0.000**	**0.000**
13 (13)	**0.015**	**0.000**	0.071	**0.000**	**0.000**	**0.000**	**0.004**	0.092	0.438	**0.000**	**0.000**	**0.000**		**0.000**
14 (14)	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.007**	**0.000**	**0.001**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	

Legend: statistically significant differences marked out by red color; $p<0.05$ – statistically significant difference. N – nature zones (1, 7, 8, 13, 14); U – urban zones (2, 3, 4, 5, 6, 9); I – industrial zones (10, 11, 12).

4.1.3 Aperture diameter

Among palynological researches the diameter of aperture wasn't study enough. The average value of aperture for hazelnut pollen grains was 3.25 µm. Our data respond to database Polleninfo (2003), which states the values 2 – 3 µm.

Basic descriptive characteristics of aperture diameter of pollen grains are listed in Table 10. Graphical presentation of descriptive statistics for equatorial axis was displayed in Fig. 13. Minimum average value (2.84 µm) of aperture diameter determined in plants sampled at Banka-2 locality (CA-P-03). Maximum average value (3.60 µm) aperture diameter was found in plants of cultivar Hallská obrovská sampled in Nitra locality (CA-P-04). The most of samples from natural zones were in the range from 2.90 µm to 3.54 µm. The most of samples from urban zones were in the range from

DOI: https://doi.org/10.15414/2020.9788055222622

2.84 µm to 3.20 µm. Samples from industrial zone (cultivars) were in the range from 3.36 µm to 3.60 µm.

Tab. 10

Descriptive statistics of hazelnut pollen grains' parameter (aperture, µm) at different locations

Sample	Location	n	$\bar{x}$	min	max	StDev	StEr	CV, 100%
CA-P-01	Čertova pec (N)	98	2.90	2.01	4.02	0.39	0.04	13.67
CA-P-02	Banka-1 (U)	100	3.20	2.24	4.06	0.39	0.03	12.27
CA-P-03	Banka-2 (U)	98	2.84	2.23	3.58	0.36	0.03	12.72
CA-P-04	Zemianske Podhradie (U)	99	3.04	2.23	3.94	0.42	0.04	14.02
CA-P-05	Moravské Lieskové (U)	98	3.06	2.23	4.25	0.45	0.04	14.98
CA-P-06	Ivanka pri Nitre (U)	98	3.06	2.17	4.18	0.44	0.04	14.68
CA-P-07	Jelenec-1 (N)	86	3.54	2.30	4.38	0.47	0.05	13.50
CA-P-08	Jelenec-2 (N)	91	3.51	2.68	4.43	0.37	0.03	10.76
CA-P-09	Žirany (U)	99	3.18	2.20	4.38	0.47	0.04	14.87
CA-P-10	Nitra (Hallská obrovská) (I)	96	3.36	2.40	4.37	0.39	0.04	11.77
CA-P-11	Nitra (Lombardská biela) (I)	90	3.60	2.64	4.75	0.44	0.04	12.37
CA-P-12	Nitra (Webbova) (I)	88	3.57	2.30	4.37	0.43	0.04	12.28
CA-P-13	Nitra (Zobor 1) (N)	96	3.33	2.29	4.43	0.46	0.04	13.85
CA-P-14	Nitra (Zobor 2) (N)	96	3.41	2.30	4.40	0.51	0.05	14.93

Legend: n – count of pollen grains for one sample; $\bar{x}$ – average value; min – minimum; max – maximum; StDev – standard deviation; StEr – standard error. N – nature zones; U – urban zones; I – industrial zones.

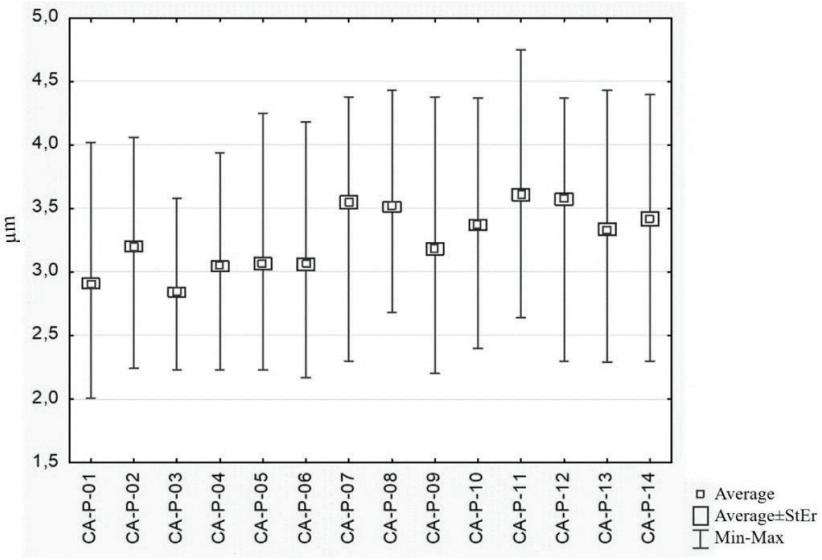


Fig. 13

Comparison of average value and variation of aperture of *C. avellana* L. pollen grains in different locations

DOI: https://doi.org/10.15414/2020.9788055222622

For samples from nature zones (CA-P-01, CA-P-07, CA-P-08, CA-P-13, CA-P-14) with 2.90 – 3.54 μm aperture diameter the difference was in pH (H_2O) soils, precipitation totals and climate region (Annexes).

For samples from urban zones (CA-P-02, CA-P-03, CA-P-04, CA-P-05, CA-P-06, CA-P-09) with 2.84 – 3.20 μm of aperture diameter the difference was in pH (H_2O) soils and climate region, as in the previous equatorial axis characteristic. Precipitation totals was the same for all samples (Annexes).

Samples from industrial zones with 3.36 – 3.60 μm of aperture diameter (CA-P-10, CA-P-11 and CA-P-12) were from territories with neutral to weakly alkaline pH. Precipitation totals were 700 – 800 mm. According to climate region the sum of average daily temperatures above 10 °C was from > 3000 to 2500 with length of time with air temperature above 5 °C in days was from 242 to 237 (Annexes).

Significant differences between pairs of samples were identified by Duncan's test. The statistically significant differences in aperture diameter were found in the most cases (Tab. 11). According to the scheme from previous traits we selected only genotypes – CA-P-01, CA-P-02, and CA-P-03. In this case there were more genotypes where we did not confirm significant differences. According to the results we could not confirm the influence of environmental conditions on phenotypic expression.

Tab. 11
Results of Duncan's post-hoc test of differences among averages for *C. avellana* L. sample (location) as independent variables and aperture as a dependent variable

Sample	(1) M=2.908	(2) M=3.203	(3) M=2.841	(4) M=3.046	(5) M=3.065	(6) M=3.060	(7) M=3.548	(8) M=3.514	(9) M=3.184	(10) M=3.369	(11) M=3.609	(12) M=3.574	(13) M=3.336	(14) M=3.417
01 (1)		**0.000**	0.285	**0.029**	**0.021**	**0.021**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**
02 (2)	**0.000**		**0.000**	**0.024**	**0.037**	**0.037**	**0.000**	**0.000**	0.764	**0.011**	**0.000**	**0.000**	**0.034**	**0.001**
03 (3)	0.285	**0.000**		**0.001**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**
04 (4)	**0.029**	**0.024**	**0.001**		0.783	0.819	**0.000**	**0.000**	**0.044**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**
05 (5)	**0.021**	**0.037**	**0.000**	0.783		0.947	**0.000**	**0.000**	0.058	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**
06 (6)	**0.021**	**0.037**	**0.000**	0.819	0.947		**0.000**	**0.000**	0.063	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**
07 (7)	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**		0.588	**0.000**	**0.008**	0.366	0.681	**0.001**	**0.049**
08 (8)	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	0.588		**0.000**	**0.029**	0.172	0.373	**0.008**	0.127
09 (9)	**0.000**	0.764	**0.000**	**0.044**	0.058	0.063	**0.000**	**0.000**		**0.006**	**0.000**	**0.000**	**0.021**	**0.000**
10 (10)	**0.000**	**0.011**	**0.000**	**0.000**	**0.000**	**0.000**	**0.008**	**0.029**	**0.006**		**0.000**	**0.002**	0.605	0.444
11 (11)	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	0.366	0.172	**0.000**	**0.000**		0.579	**0.000**	**0.005**
12 (12)	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	0.681	0.373	**0.000**	**0.002**	0.579		**0.000**	**0.021**
13 (13)	**0.000**	**0.034**	**0.000**	**0.000**	**0.000**	**0.000**	**0.001**	**0.008**	**0.021**	0.605	**0.000**	**0.000**		0.229
14 (14)	**0.000**	**0.001**	**0.000**	**0.000**	**0.000**	**0.000**	**0.049**	0.127	**0.000**	0.444	**0.005**	**0.021**	0.229	

Legend: statistically significant differences marked out by red color; $p<0.05$ – statistically significant difference. N – nature zones (1. 7. 8. 13. 14); U – urban zones (2. 3. 4. 5. 6. 9); I – industrial zones (10. 11. 12).

DOI: https://doi.org/10.15414/2020.9788055222622

4.1.4 Pollen grain shape

One of the important pollen characteristics is P/E ratio. The P/E determinates pollen shape. For our pollen samples, P/E was in the frame from 0.9 to 1.00. We determined the shape of hazelnut pollen grains in equatorial view ranges from (P/E = 0.90 – 1.00) oblate spheroidal to spheroidal or prolate spheroidal (Fig. 14). Our results exceeded Frenguelli et al. (1997) results – 0.88 but were consistent with the Blackmore et al. (2003) data – 0.92.

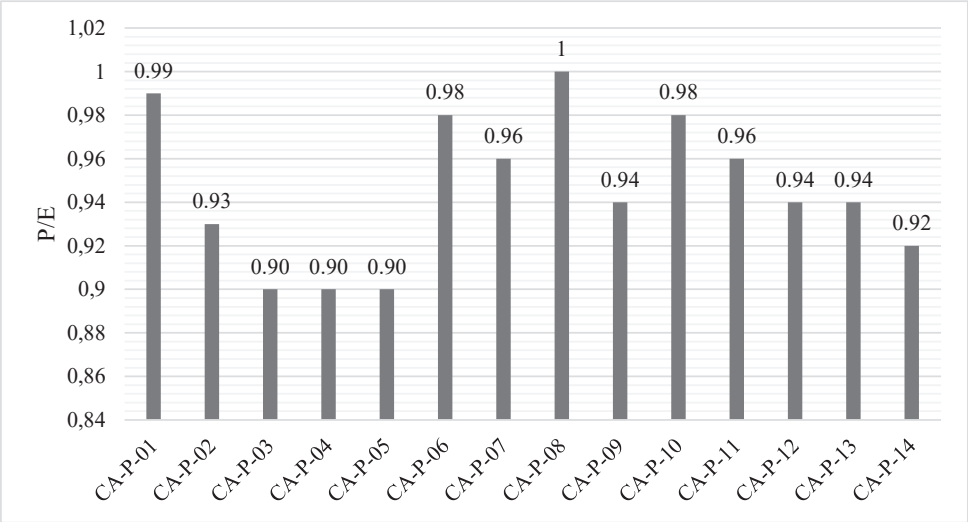


Fig. 14
***C. avellana* L. pollen shape to P/E ratio**

4.2 Pollen grains area measured by Image analysis software

Nowadays the development of information and communication technologies brought the new possibilities of automated microscopy and quantitative image analysis. The "big-data revolution" is now common also for biology. Microscopic techniques can bring thousands of microscopy images. Looking at and interpreting the results from these images by only human eye would be impossible and very subjective. In our study we used validated open-source software CellProfiler Analyst. The suite of image-based measurements was generated by CellProfiler and the machine-learning functionality by CellProfiler Analyst. These methods were not applied for morphological characterization of pollen grains, yet. There are some papers dealing with automated pollen grains counting (Chen et al., 2016; Novara et al., 2017; Tello et al., 2018), but using image analysis software for classification and further measuring the pollen grains was not used yet.

Due to mentioned information we displayed in this chapter the results of the image analysis, where the analyzed objects were the *C. avellana* L. pollen grains. Firstly, we have prepared the

DOI: https://doi.org/10.15414/2020.9788055222622

database where 27 286 RegionsOfInterest (ROI's) (pollen grains) were selected and they were measured by 126 characters for intensity, size, shape, and texture of segmented objects.

The second step was classification of selected ROI's into two classes. We decided to classify segmented objects into the first class (positive) composed by pollen grains in polar axis view, and into the second class (negative) where all other pollen grains and objects, which were not displayed in polar axis view, were classified. During this step we built manually the training set and traced its performance by setting the rules set and correct errors. The finally we created the training set which consists of 100 objects (*C. avellana* L. pollen grains in polar view) in positive bin, and 100 objects classified into the negative bin. The important result was the classification accuracy, which was checked by two methods RandomForest Clasifier and Fast Gentle Boosting. The Fig. 15 presents the Confusion matrix where we can see that software classified the positive objects as positive and negative objects as negative with classification accuracy 92.5 %. The chart Cross-validation accuracy shows that the performance of 15 rules set is more than 0.9. The number 1 means the ideal situation which is impossible to reach. In this chart is also the other accuracy test which is displayed for two versions of cross-validation, with 50% or 95% of the examples used for training and the remainder for testing. If the two accuracies are essentially the same, adding more cells to the training set is unlikely to improve performance.

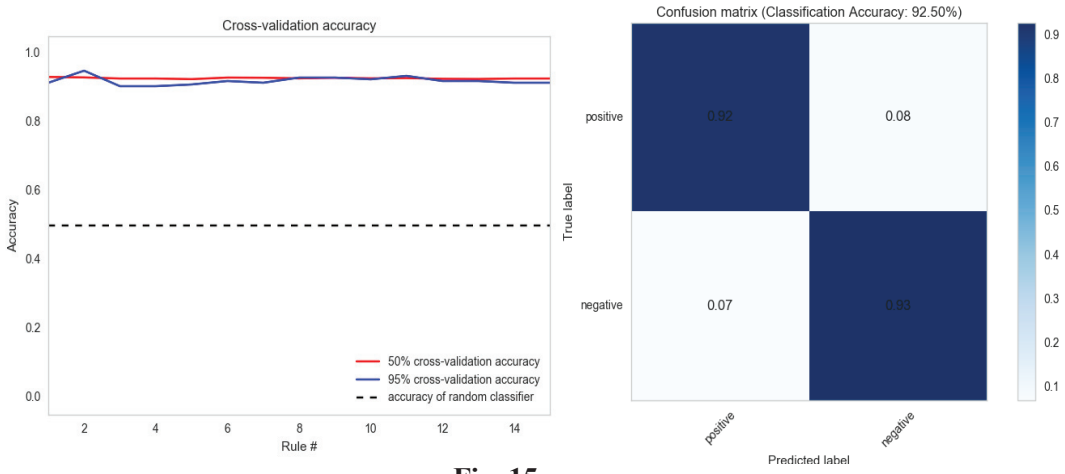


Fig. 15

Performance of training set (positive bin – 100 *C. avellana* L. pollen grains and negative bin – 100 *C. avellana* L. pollen grains) and rules set (15 rules)

After refining the training set evaluation, we could score all images and all included objects (27 286). All selected objects from all images (760) of experiment were automatically classified and each object was scored according the described phenotype (positive, negative bin). Then it was easy

DOI: https://doi.org/10.15414/2020.9788055222622

to select only objects scored as positive and combined these results with measurement data for each object of the experiment. The set of positive class contained 4078 pollen grains.

In the Tab. 12 are displayed the descriptive statistics parameters for pollen grains which were scored as positive (polar axis view). Graphical presentation of descriptive statistics for pollen grain area was displayed in Fig. 16. The extremes were deleted. We chose only area (μm^2) trait to see variability among selected samples. The highest average value reached the sample CA-P-04 (urban zone) and the lowest average number had sample CA-P-01 (natural zone). The variation coefficient was for all analyzed samples less than 10 %.

Significant differences between pairs of samples were identified by Duncan's test. The statistically significant differences in area were found in the most cases (Tab. 13). The significant differences were not found only between following pairs of samples – CA-P-09 and CA-P-05, CA-P-10 and CA-P-07. Again, we can confirm our hypothesis, that the trait area is discriminating genotypes, but we did not find influence of environmental conditions on phenotypic expression.

Tab. 12

Descriptive statistics of hazelnut pollen grains' parameter (area μm^2) at different locations

Samle	Location	N	$\bar{x}$	Min	Max	StDev	StEr	CV 100%
CA-P-01	Čertova Pec (N)	482	470.06	414.23	549.21	26.22	1.19	5.58
CA-P-04	Zemianske Podhradie (U)	214	530.34	441.27	613.39	39.37	2.69	7.42
CA-P-05	Moravské Lieskové (U)	241	507.64	434.23	581.45	33.86	2.18	6.67
CA-P-07	Jelenec-1 (N)	1271	495.27	404.33	585.64	40.17	1.13	8.11
CA-P-09	Žirany (U)	277	502.57	435.96	570.84	31.09	1.87	6.19
CA-P-10	Nitra (Hallská obrovská) (I)	823	493.23	401.38	588.80	40.90	1.43	8.29
CA-P-12	Nitra (Webbova) (I)	137	477.72	392.09	566.05	38.41	3.28	8.04
CA-P-14	Nitra (Zobor 2) (N)	633	515.01	430.25	603.49	39.41	1.57	7.65

Legend: n – count of pollen grains for one sample; $\bar{x}$ – average value; min – minimum; max – maximum; StDev – standard deviation; StEr – standard error. N – nature zones; U – urban zones; I – industrial zones.

DOI: https://doi.org/10.15414/2020.9788055222622

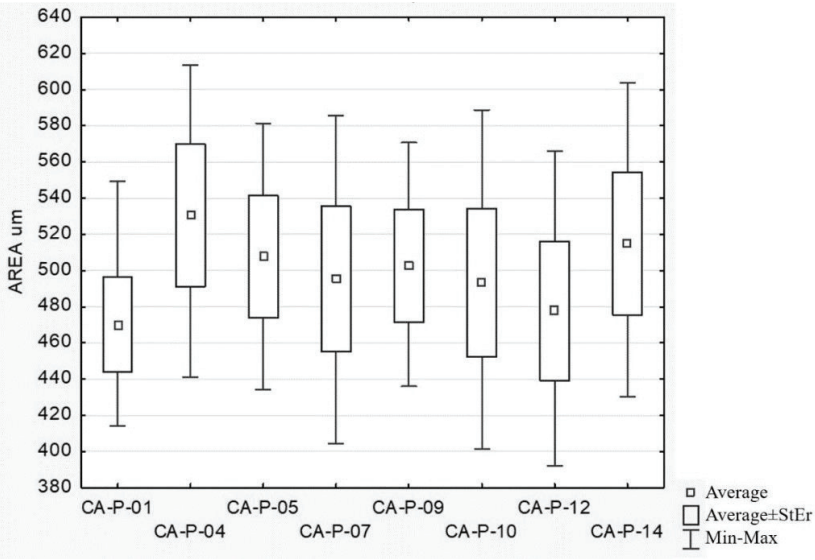


Fig. 16
Comparison of average value and variation of *C. avellana* L. pollen grains area in different locations

Tab. 13

Results of Duncan's post-hoc test of differences among averages for sample (location) as independent variable and area of *C. avellana* L. pollen grain as a dependent variable

Locations	(1) M=470.06	(4) M=530.34	(5) M=507.64	(7) M=495.27	(9) M=502.57	(10) M=493.23	(12) M=477.72	(14) M=515.01
CA-P-01 (1)		**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.010**	**0.000**
CA-P-04 (4)	**0.000**		**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**
CA-P-05 (5)	**0.000**	**0.000**		**0.000**	0.091	**0.000**	**0.000**	**0.014**
CA-P-07 (7)	**0.000**	**0.000**	**0.000**		**0.015**	0.499	**0.000**	**0.000**
CA-P-09 (9)	**0.000**	**0.000**	0.091	**0.015**		**0.002**	**0.000**	**0.000**
CA-P-10 (10)	**0.000**	**0.000**	**0.000**	0.499	**0.002**		**0.000**	**0.000**
CA-P-12 (12)	**0.010**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**		**0.000**
CA-P-14 (14)	**0.000**	**0.000**	**0.014**	**0.000**	**0.000**	**0.000**	**0.000**	

Legend: statistically significant differences marked out by red color; $p<0.05$ – statistically significant difference. N – nature zones (1, 7, 14); U – urban zones (4, 5, 9); I – industrial zones (10, 12).

4.3 Morphometric and weight analysis of *Corylus avellana* L. catkins

Human selection has had different consequences on the agro-morphological characteristics of *C. avellana*. Phenotypic observation is a traditional method to characterize and identify hazelnut cultivars (Boccacci et al., 2008). Male inflorescence traits such as, the amount, length, time of flowering, pollen shedding is using for molecular quantitative trait loci controlling of hazelnut species (Frary et al., 2019).

DOI: https://doi.org/10.15414/2020.9788055222622

In our research a total 50 catkins from 14 genotypes were evaluated. We measured the length (mm), diameter (mm) and weight (g) (Tab. 14). Extremes were deleted. Based on morphological parameters of catkins from different locations the statistically significant differences were exposed by ANOVA. The average values of length, diameter and weight were analyzed.

Basic descriptive characteristics of length, diameter and weight of catkins of *C. avellana* are listed in Tab. 14. Graphical presentation of descriptive statistics is displayed in Fig. 17.

In different *Corylus* species male catkin length variated from 3 cm to 12 cm (Turrill, 1962; Kasapligil, 1963; Davis, 1982). Sharma (2003) carried out that the male catkin length was from 7.02 cm to 9.50 cm. And the variation of this trait correlated with yield, trunk diameter, trunk cross section area, leaf area and degree of dichogamy. In our research male catkin length was in average 26.87 – 80.80 mm. That is responded to data previous authors.

The average value of length catkin was 40.68 mm. Minimum average value of length was determined in plants at Nitra settlement (Zobor-1) (CA-P-13). It was 20.83 mm. Maximum average value (80.8 mm) of length was determined in plants collected at Zemianske Podhradie locality (CA-P-04). Most samples from natural zones were in the range from 22.83 mm to 58.57 mm. Most samples from urban zones were in the range from 26.87 mm to 80.80 mm. Samples from industrial zone (cultivars) were in the range 31.89 mm to 41.36 mm.

The average value of catkin diameter was 5.04 mm. Minimum average value of diameter was determined in samples sampled at Žirany locality (CA-P-09). It was 4.38 mm. Maximum average value (6.20 mm) of diameter was determined in plants collected at Jelenec-2 locality (CA-P-08). Most samples from natural zones were in the range from 4.57 to 6.20 mm. Most samples from urban zones were in the range from 4.38 mm to 5.20 mm. Samples from industrial zone (cultivars) were in the range 4.80 mm to 5.59 mm.

The average value of weight of catkin was 0.43 g. Minimum average value of weight was determined in plants sampled at Nitra locality (Zobor-2) (CA-P-14). It was 0.20 g. Maximum average value (0.75 g) of weight was determined in plants from Jelenec-2 locality (CA-P-08). Most samples from natural zones were in the range from 0.2 g to 0.75 g. Most samples from urban zones were in the range from 0.28 g to 0.50 g. Samples from industrial zone (cultivars) were in the range 0.43 g to 0.68 g. Our results of pollen weigh were higher than Piotrowska (2008) data. Researcher evaluated hazelnut pollen weigh in 66 mg.

DOI: https://doi.org/10.15414/2020.9788055222622

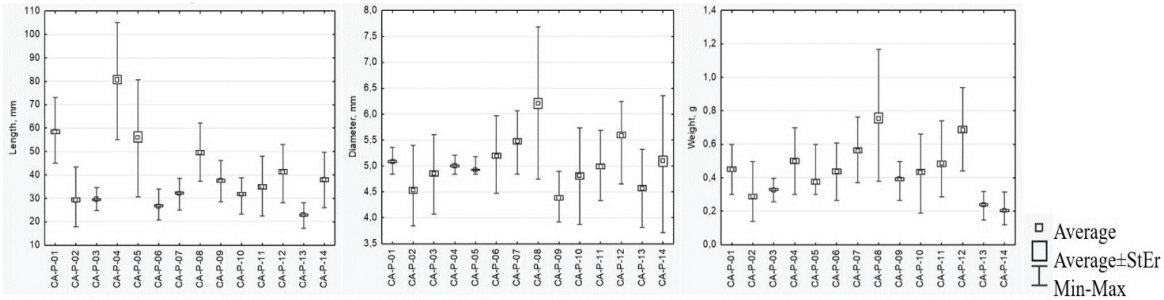
Tab. 14

Descriptive statistics of hazelnut catkins' parameters at different locations

Sample	Location	n	$\bar{x}$	min	max	StDev	StEr	CV, %
Length, mm								
CA-P-01	Čertova pec (N)	46	58.57	45.01	73.01	0.13	0.02	10.30
CA-P-02	Banka-1 (U)	45	29.43	17.91	43.32	0.37	0.05	18.49
CA-P-03	Banka-2 (U)	47	29.49	24.77	34.60	0.34	0.05	8.25
CA-P-04	Zemianske Podhradie (U)	47	80.80	55.00	105.00	0.11	0.01	15.00
CA-P-05	Moravské Lieskové (U)	47	56.15	30.60	80.70	0.07	0.01	26.83
CA-P-06	Ivanka pri Nitre (U)	47	26.87	20.74	33.89	0.35	0.05	11.54
CA-P-07	Jelenec-1 (N)	48	32.30	24.91	38.64	0.33	0.04	9.97
CA-P-08	Jelenec-2 (N)	45	49.57	37.38	62.26	0.64	0.09	12.00
CA-P-09	Žirany (U)	47	37.55	28.48	46.16	0.26	0.03	11.88
CA-P-10	Nitra (Hallská obrovská) (I)	47	31.89	23.22	38.81	0.40	0.05	13.25
CA-P-11	Nitra (Lombardská biela) (I)	48	34.90	22.52	48.05	0.34	0.05	18.90
CA-P-12	Nitra (Webbova) (I)	45	41.36	28.08	53.00	0.35	0.05	15.49
CA-P-13	Nitra (Zobor 1) (N)	50	22.83	17.30	28.14	0.37	0.05	12.11
CA-P-14	Nitra (Zobor 2) (N)	47	37.94	25.92	49.67	0.66	0.09	15.72
Diameter, mm								
Sample	**Location**	**n**	**$\bar{x}$**	**min**	**max**	**StDev**	**StEr**	**CV, %**
CA-P-01	Čertova pec (N)	48	5.08	4.84	5.36	0.13	0.02	2.63
CA-P-02	Banka-1 (U)	46	4.53	3.84	5.40	0.37	0.05	8.24
CA-P-03	Banka-2 (U)	47	4.85	4.07	5.60	0.34	0.05	7.06
CA-P-04	Zemianske Podhradie (U)	49	5.00	4.84	5.21	0.12	0.01	2.16
CA-P-05	Moravské Lieskové (U)	49	4.93	4.84	5.18	0.07	0.01	1.49
CA-P-06	Ivanka pri Nitre (U)	47	5.20	4.47	5.97	0.35	0.05	6.71
CA-P-07	Jelenec-1 (N)	48	5.47	4.84	6.07	0.33	0.05	6.05
CA-P-08	Jelenec-2 (N)	47	6.20	4.75	7.68	0.64	0.09	10.34
CA-P-09	Žirany (U)	48	4.38	3.92	4.90	0.26	0.03	5.99
CA-P-10	Nitra (Hallská obrovská) (I)	50	4.80	3.87	5.74	0.40	0.05	8.44
CA-P-11	Nitra (Lombardská biela) (I)	49	4.99	4.33	5.69	0.34	0.05	6.96
CA-P-12	Nitra (Webbova) (I)	46	5.59	4.65	6.24	0.35	0.05	6.28
CA-P-13	Nitra (Zobor 1) (N)	49	4.57	3.82	5.32	0.37	0.05	8.07
CA-P-14	Nitra (Zobor 2) (N)	47	5.09	3.71	6.36	0.66	0.09	12.99
Weight, g								
Sample	**Location**	**n**	**$\bar{x}$**	**min**	**max**	**StDev**	**StEr**	**CV, %**
CA-P-01	Čertova pec (N)	46	0.45	0.30	0.60	0.08	0.013	19.63
CA-P-02	Banka-1 (U)	48	0.28	0.12	0.49	0.09	0.013	32.45
CA-P-03	Banka-2 (U)	47	0.33	0.25	0.39	0.04	0.005	11.74
CA-P-04	Zemianske Podhradie (U)	48	0.50	0.30	0.70	0.11	0.016	22.21
CA-P-05	Moravské Lieskové (U)	44	0.37	0.30	0.60	0.09	0.014	25.47
CA-P-06	Ivanka pri Nitre (U)	48	0.43	0.26	0.61	0.10	0.014	23.13
CA-P-07	Jelenec-1 (N)	47	0.56	0.37	0.76	0.10	0.014	17.79
CA-P-08	Jelenec-2 (N)	47	0.75	0.37	1.16	0.20	0.029	26.50
CA-P-09	Žirany (U)	49	0.39	0.26	0.49	0.06	0.008	15.90
CA-P-10	Nitra (Hallská obrovská) (I)	50	0.43	0.19	0.66	0.11	0.015	25.88
CA-P-11	Nitra (Lombardská biela) (I)	45	0.48	0.28	0.74	0.10	0.015	21.57
CA-P-12	Nitra (Webbova) (I)	45	0.68	0.44	0.93	0.13	0.019	19.00
CA-P-13	Nitra (Zobor 1) (N)	49	0.23	0.15	0.32	0.05	0.006	20.12
CA-P-14	Nitra (Zobor 2) (N)	43	0.20	0.12	0.31	0.04	0.006	21.50

Legend: $\bar{x}$ – average value; min – minimum; max – maximum; StDev – standard deviation; StEr – standard error; CV, % – coefficient of variance. N – nature zones; U – urban zones; I – industrial zones.

DOI: https://doi.org/10.15414/2020.9788055222622



a **b** **c**

Fig. 17

Comparison of average value and variation of length (a), diameter (b) and weigh (c) of hazelnut catkins in different locations

For samples from natural zones the difference was in pH (H_2O) soils from neutral to weakly alkaline pH for CA-P-01, CA-P-07, CA-P-08 (with catkin's length from 32.30 mm to 49.57 mm), and to weakly acidic pH for CA-P-13 and CA-P-14 (with catkin's length from 22.83 mm to 37.94 mm) and in climate region with the sum of average daily temperatures above 10 °C was from >3000 to 2800 with length of time with air temperature above 5 °C in days was from 242 to 237 for CA-P-07 and CA-P-08. For sample CA-P-01 the sum of average daily temperatures above 10 °C was from 3000 to 2500 with length of time with air temperature above 5 °C in days was from 237 to 231. For samples CA-P-13 and CA-P-14 the sum of average daily temperatures above 10 °C was from 3000 to 2500 with length of time with air temperature above 5 °C in days was from 237 to 231. Also, difference was in precipitation totals (700 – 800 mm) for samples CA-P-07, CA-P-08, CA-P-13, CA-P-14, but for sample CA-P-01 was 600 – 700 mm. The detailed characterization of localities is placed at Annexes.

For samples from urban zones the difference was in pH (H_2O) soils from neutral to weakly alkaline pH soils for CA-P-02, CA-P-03, CA-P-04, CA-P-05 and CA-P-06 (with catkin's length from 26.87 mm to 80.80 mm), and to very strong pH for CA-P-09 (with catkin's length 37.55 mm) and climate region with the sum of average daily temperatures above 10 °C was from 3000 to 2500 with length of time with air temperature above 5 °C in days was from 237 to 231 for CA-P-02, CA-P-03, CA-P-04, CA-P-05. For sample CA-P-06 and CA-P-09 the sum of average daily temperatures above 10 °C was from > 3000 to 2800 with length of time with air temperature above 5 °C in days was from 242 to 237. Precipitation totals was the same for all samples. The detailed characterization of localities is placed at Annexes.

DOI: https://doi.org/10.15414/2020.9788055222622

Samples from industrial zone (CA-P-10, CA-P-11 and CA-P-12 with catkin's length from 31.89 mm to 41.36 mm) were in the same conditions – pH (H_2O) soils (weakly acidic), the sum of average daily temperatures above 10 °C was from 3000 to 2500 with length of time with air temperature above 5 °C in days was from 237 to 231. Precipitation totals were 700 – 800 mm (Annexes).

Significant differences between pairs of samples identified by Duncan's test. The statistically significant differences in catkin's length were found in the most cases (Tab. 15). We found 4 genotypes (CA-P-04, CA-P-08, CA-P-12, and CA-P-13) which significantly differed from all other genotypes. The other 3 genotypes (CA-P-01, CA-P-05, and CA-P-14) were not significantly different only from one genotype. We can confirm our hypothesis, that catkin's length is discriminating genotypes, but we did not find influence of environmental conditions on phenotypic expression.

Tab. 15

Results of Duncan's post-hoc test of differences among averages for *C. avellana* L. sample (location) as independent variables and catkin's length as a dependent variable

Sample	(1) M=58.57	(2) M=29.43	(3) M=29.49	(4) M=80.80	(5) M=56.16	(6) M=26.87	(7) M=32.30	(8) M=49.57	(9) M=37.55	(10) M=31.89	(11) M=34.91	(12) M=41.36	(13) M=22.83	(14) M=37.94
01 (1)		**0.000**	**0.000**	**0.000**	0.089	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**
02 (2)	**0.000**		0.968	**0.000**	**0.000**	0.072	0.065	**0.000**	**0.000**	0.103	**0.000**	**0.000**	**0.000**	**0.000**
03 (3)	**0.000**	0.968		**0.000**	**0.000**	0.082	0.061	**0.000**	**0.000**	0.091	**0.000**	**0.000**	**0.000**	**0.000**
04 (4)	**0.000**	**0.000**	**0.000**		**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**
05 (5)	0.089	**0.000**	**0.000**	**0.000**		**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**
06 (6)	**0.000**	0.072	0.082	**0.000**	**0.000**		**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.004**	**0.000**
07 (7)	**0.000**	0.065	0.061	**0.000**	**0.000**	**0.000**		**0.000**	**0.000**	0.772	0.067	**0.000**	**0.000**	**0.000**
08 (8)	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**		**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**
09 (9)	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**		**0.000**	0.063	**0.010**	**0.000**	0.786
10 (10)	**0.000**	0.103	0.091	**0.000**	**0.000**	**0.000**	0.772	**0.000**	**0.000**		**0.044**	**0.000**	**0.000**	**0.000**
11 (11)	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	0.067	**0.000**	0.063	**0.044**		**0.000**	**0.000**	**0.043**
12 (12)	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.010**	**0.000**	**0.000**		**0.000**	**0.016**
13 (13)	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.004**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**		**0.000**
14 (14)	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	0.786	**0.000**	**0.043**	**0.016**	**0.000**	

Legend: statistically significant differences marked out by red color; $p<0.05$ – statistically significant difference. N – nature zones (1, 7, 8, 13, 14); U – urban zones (2, 3, 4, 5, 6, 9); I – industrial zones (10, 11, 12).

Significant differences between pairs of samples identified by Duncan's test. The statistically significant differences in catkin's diameter were found in the most cases (Tab. 16). In the case of the trait catkin's diameter there was only one genotype (CA-P-08) which significantly differed from all other genotypes. The genotypes (CA-P-07, CA-P-09, CA-P-12, and CA-P-13) were not significantly

DOI: https://doi.org/10.15414/2020.9788055222622

different only from one genotype. We can confirm our hypothesis, that catkin's diameter is discriminating genotypes, but we did not find influence of environmental conditions on phenotypic expression.

Tab. 16

Results of Duncan's post-hoc test of differences among averages for *C. avellana* L. sample (location) as independent variables and catkin's diameter as a dependent variable

Sample	(1) M=5.085	(2) M=4.532	(3) M=4.853	(4) M=5.001	(5) M=4.930	(6) M=5.200	(7) M=5.476	(8) M=6.206	(9) M=4.384	(10) M=4.809	(11) M=4.991	(12) M=5.596	(13) M=4.575	(14) M=5.092
01 (1)		**0.000**	**0.005**	0.273	0.063	0.160	**0.000**	**0.000**	**0.000**	**0.000**	0.248	**0.000**	**0.000**	0.930
02 (2)	**0.000**		**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	0.054	**0.000**	**0.000**	**0.000**	0.579	**0.000**
03 (3)	**0.005**	**0.000**		0.078	0.319	**0.000**	**0.000**	**0.000**	**0.000**	0.560	0.090	**0.000**	**0.000**	**0.004**
04 (4)	0.273	**0.000**	0.078		0.382	**0.016**	**0.000**	**0.000**	**0.000**	**0.022**	0.892	**0.000**	**0.000**	0.267
05 (5)	0.063	**0.000**	0.319	0.382		**0.001**	**0.000**	**0.000**	**0.000**	0.137	0.425	**0.000**	**0.000**	0.058
06 (6)	0.160	**0.000**	**0.000**	**0.016**	**0.001**		**0.000**	**0.000**	**0.000**	**0.000**	**0.013**	**0.000**	**0.000**	0.160
07 (7)	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**		**0.000**	**0.000**	**0.000**	**0.000**	0.120	**0.000**	**0.000**
08 (8)	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**		**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**
09 (9)	**0.000**	0.054	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**		**0.000**	**0.000**	**0.000**	**0.017**	**0.000**
10 (10)	**0.000**	**0.000**	0.560	**0.022**	0.137	**0.000**	**0.000**	**0.000**	**0.000**		**0.028**	**0.000**	**0.002**	**0.000**
11 (11)	0.248	**0.000**	0.090	0.892	0.425	**0.013**	**0.000**	**0.000**	**0.000**	**0.028**		**0.000**	**0.000**	0.234
12 (12)	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	0.120	**0.000**	**0.000**	**0.000**	**0.000**		**0.000**	**0.000**
13 (13)	**0.000**	0.579	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.017**	**0.002**	**0.000**	**0.000**		**0.000**
14 (14)	0.930	**0.000**	**0.004**	0.267	0.058	0.160	**0.000**	**0.000**	**0.000**	**0.000**	0.234	**0.000**	**0.000**	

Legend: statistically significant differences marked out by red color; $p<0.05$ – statistically significant difference. N – nature zones (1, 7, 8, 13, 14); U – urban zones (2, 3, 4, 5, 6, 9); I – industrial zones (10, 11, 12).

Significant differences between pairs of samples identified by Duncan's test. The statistically significant differences in catkin's weight were found in the most cases (Tab. 17). We found 3 genotypes (CA-P-07, CA-P-08, and CA-P-12) which significantly differed from all other genotypes. The other 7 genotypes (CA-P-02, CA-P-03, CA-P-04, CA-P-05, CA-P-09, CA-P-13, and CA-P-14) were not significantly different only from one genotype. We can confirm our hypothesis, that catkin's weight is discriminating genotypes, but we did not find influence of environmental conditions on phenotypic expression.

DOI: https://doi.org/10.15414/2020.9788055222622

Tab. 17

Results of Duncan's post-hoc test of differences among averages for *C. avellana* L. sample (location) as independent variables and catkin's weight as a dependent variable

Sample	(1) M=0.452	(2) M=0.287	(3) M=0.032	(4) M=0.500	(5) M=0.377	(6) M=0.437	(7) M=0.563	(8) M=0.757	(9) M=0.391	(10) M=0.435	(11) M=0.483	(12) M=0.687	(13) M=0.238	(14) M=0.203
01 (1)		**0.000**	**0.000**	**0.032**	**0.001**	0.494	**0.000**	**0.000**	**0.007**	0.454	0.142	**0.000**	**0.000**	**0.000**
02 (2)	**0.000**		0.051	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.023**	**0.000**
03 (3)	**0.000**	0.051		**0.000**	**0.022**	**0.000**	**0.000**	**0.000**	**0.004**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**
04 (4)	**0.032**	**0.000**	**0.000**		**0.000**	**0.006**	**0.003**	**0.000**	**0.000**	**0.005**	0.437	**0.000**	**0.000**	**0.000**
05 (5)	**0.001**	**0.000**	**0.022**	**0.000**		**0.008**	**0.000**	**0.000**	0.505	**0.009**	**0.000**	**0.000**	**0.000**	**0.000**
06 (6)	0.494	**0.000**	**0.000**	**0.006**	**0.008**		**0.000**	**0.000**	**0.039**	0.906	**0.040**	**0.000**	**0.000**	**0.000**
07 (7)	**0.000**	**0.000**	**0.000**	**0.003**	**0.000**	**0.000**		**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**
08 (8)	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**		**0.000**	**0.000**	**0.000**	**0.001**	**0.000**	**0.000**
09 (9)	**0.007**	**0.000**	**0.004**	**0.000**	0.505	**0.039**	**0.000**	**0.000**		**0.040**	**0.000**	**0.000**	**0.000**	**0.000**
10 (10)	0.454	**0.000**	**0.000**	**0.005**	**0.009**	0.906	**0.000**	**0.000**	**0.040**		**0.036**	**0.000**	**0.000**	**0.000**
11 (11)	0.142	**0.000**	**0.000**	0.437	**0.000**	**0.040**	**0.000**	**0.000**	**0.000**	**0.036**		**0.000**	**0.000**	**0.000**
12 (12)	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.001**	**0.000**	**0.000**	**0.000**		**0.000**	**0.000**
13 (13)	**0.000**	**0.023**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**		0.094
14 (14)	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	0.094	

Legend: statistically significant differences marked out by red color; $p<0.05$ – statistically significant difference. N – nature zones (1, 7, 8, 13, 14); U – urban zones (2, 3, 4, 5, 6, 9); I – industrial zones (10, 11, 12).

4.4 Number of flowers per catkin and pollen production

In our research a total of 25 catkins from 14 genotypes were evaluated. We counted flowers number from one hazelnut catkin. Based on flowers counting from different locations the statistically significant differences were exposed by ANOVA. The average number of flowers were analyzed.

Basic descriptive characteristics of flowers number in catkin of *C. avellana* is listed in Tab. 18. Graphical presentation of descriptive statistics was displayed in Fig. 18. Average value of flowers number for one catkin was 252.58 flowers. Piotrowska (2008) defined the number of flowers (240) from one catkin. In comparison with our study it was lower. The largest average number of flowers (268.96) determined at Nitra locality – cultivar Webbova (CA-P-12). The smallest average number of flowers (239.68) was again at Nitra locality (Zobor-1, sample CA-P-13). Most samples from natural zones were in the range from 239.68 flowers to 260.64 flowers. Most samples from urban zones were in the range from 244 flowers to 254.64 flowers. Samples from industrial zone (cultivars) were in the range from 246.64 flowers to 268.96 flowers.

DOI: https://doi.org/10.15414/2020.9788055222622

Tab. 18

Descriptive statistics of flower number in hazelnut catkin from different locations

Sample	Location	n	$\bar{x}$	min	max	StDev	StEr	CV %
CA-P-01	Čertova pec (N)	25	251.52	180.00	308.00	32.95	6.59	13.10
CA-P-03	Banka-2 (U)	25	250.60	100.00	300.00	41.63	8.32	16.61
CA-P-04	Zemianske Podhradie (U)	25	251.24	120.00	320.00	43.55	8.71	17.33
CA-P-05	Moravské Lieskové (U)	25	244.00	180.00	310.00	36.07	7.21	14.78
CA-P-06	Ivanka pri Nitre (U)	25	257.08	210.00	312.00	27.34	5.46	10.63
CA-P-08	Jelenec-2 (N)	25	260.64	200.00	300.00	27.50	5.50	10.55
CA-P-09	Žirany (U)	25	254.64	116.00	310.00	42.78	8.55	16.80
CA-P-10	Nitra (Hallská obrovská) (I)	25	246.64	157.00	330.00	40.14	8.02	16.27
CA-P-11	Nitra (Lombardská biela) (I)	25	253.40	60.00	324.00	54.72	10.94	21.59
CA-P-12	Nitra (Webbova) (I)	25	268.96	220.00	342.00	29.25	5.85	10.87
CA-P-13	Nitra (Zobor 1) (N)	25	239.68	170.00	300.00	37.45	7.49	15.62

Legend: $\bar{x}$ – average value; SE – standard error; min – minimum; max – maximum; StDev – standard deviation; StEr – standard error of the mean. N – nature zones; U – urban zones; I – industrial zones.

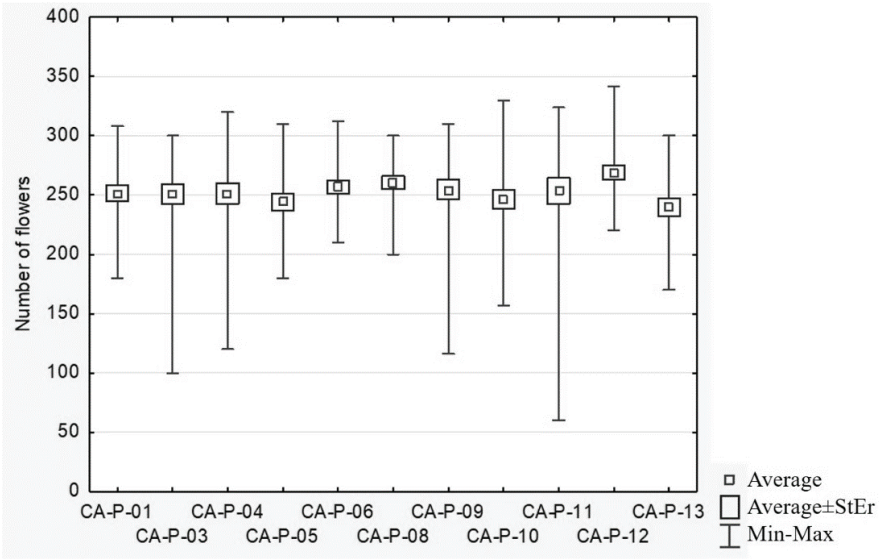


Fig. 18

The average meaning of flower numbers in one hazelnut catkin from different location

Significant differences between pairs of samples identified by Duncan's test. The statistically significant differences in flowers number were found in the pair CA-P-12 (cultivar "Webbova", industrial zone) and CA-P-13 (Nitra, Zobor-1, urban zone) (Tab. 19). We can conclude that the trait number of flowers per catkin is not discriminating genotypes, and the environmental conditions are not influencing phenotypic expression.

DOI: https://doi.org/10.15414/2020.9788055222622

Tab. 19

Results of Duncan's post-hoc test of differences among averages for *C. avellana* L. sample (location) as independent variables and flower numbers as a dependent variable

Sample	(1) M=251.5	(3) M=250.6	(4) M=251.2	(5) M=244.0	(6) M=257.0	(8) M=260.6	(9) M=254.6	(10) M=246.6	(11) M=253.4	(12) M=268.9	(13) M=239.6
CA-P-01 (1)		0.937	0.979	0.547	0.647	0.465	0.789	0.688	0.862	0.165	0.351
CA-P-03 (3)	0.937		0.953	0.570	0.612	0.438	0.747	0.715	0.818	0.155	0.366
CA-P-04 (4)	0.979	0.953		0.551	0.641	0.461	0.780	0.693	0.853	0.165	0.353
CA-P-05 (5)	0.547	0.570	0.551		0.317	0.205	0.411	0.807	0.461	0.053	0.690
CA-P-06 (6)	0.647	0.612	0.641	0.317		0.743	0.822	0.420	0.752	0.305	0.184
CA-P-08 (8)	0.465	0.438	0.461	0.205	0.743		0.606	0.283	0.551	0.443	0.109
CA-P-09 (9)	0.789	0.747	0.780	0.411	0.822	0.606		0.531	0.909	0.234	0.250
CA-P-10 (10)	0.688	0.715	0.693	0.807	0.420	0.283	0.531		0.589	0.084	0.550
CA-P-11 (11)	0.862	0.818	0.853	0.461	0.752	0.551	0.909	0.589		0.207	0.287
CA-P-12 (12)	0.165	0.155	0.165	0.053	0.305	0.443	0.234	0.084	0.207		**0.022**
CA-P-13 (13)	0.351	0.366	0.353	0.690	0.184	0.109	0.250	0.550	0.287	**0.022**	

Legend: statistically significant differences marked out by red color; $p<0.05$ – statistically significant difference. N – nature zones (1, 7, 8, 13, 14); U – urban zones (2, 3, 4, 5, 6, 9); I – industrial zones (10, 11, 12).

In laboratory conditions pollen production was determinate from 50 catkins (Fig. 19). The average value of pollen production of 50 catkins was 0.28 g. The maximum value of pollen production was 0.56 g from Jelenec-2 locality (CA-P-08). The minimum value of pollen production was 0.09 g from Nitra Zobor-1 locality (CA-P-13). Most samples from natural zones were in the range from 0.09 g to 0.56 g. Most samples from urban zones were in the range from 0.15 g to 0.44 g. Samples from industrial zone (cultivars) were in the range from 0.20 to 0.45 g.

Samples CA-P-07, CA-P-13 and CA-P-14 (from natural zones), CA-P-05, CA-P-06 and CA-P-09 (from urban zone), and CA-P-10 (from industrial zone) were below average value of pollen production (Fig. 19). Samples CA-P-01 and CA-P-08 (from natural zones), CA-P-02, CA-P-03, CA-P-04 (urban zones) and CA-P-11 and CA-P-12 (from industrial zone) were above average value of pollen production.

DOI: https://doi.org/10.15414/2020.9788055222622

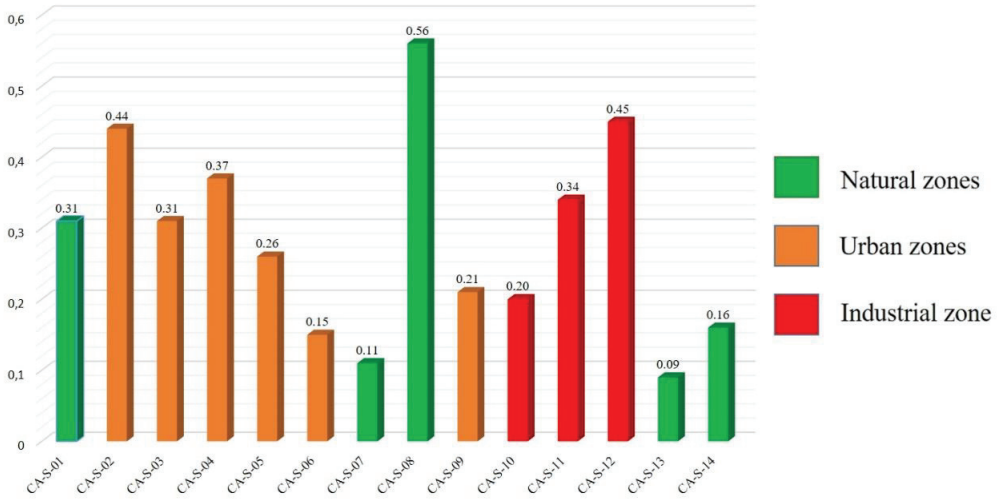


Fig. 19

Pollen production (g) from 50 *C. avellana* L. catkins

4.5 Principal component analysis of pollen and catkins

4.5.1 Principal component analysis for pollen and catkins traits

The data acquired from morphological analysis we applied also for Principal component analysis (PCA). We look on our data from two sides. Firstly, we used these traits – length, diameter, and weight of catkins, together with traits measured on pollen grains – length of polar, equatorial axis, and diameter of aperture. There were chosen the three components which explained the largest variation in the data (Fig. 20). The first factor elucidated the 45.97 %, the second 32.74 %, and the third 11.66 % of variation in the data. At the first chart (Fig. 20) we can see that the parameter aperture had the large positive loading on the factor 1, and the length had the large negative loading on the factor 1 and 2. The pairs of traits (diameter and weight, and equatorial and polar axis) are strongly correlated, so they are very coherent on principal components. Again, at the second and third charts the parameter length had the large negative loading on the factor 2 and 3.

DOI: https://doi.org/10.15414/2020.9788055222622

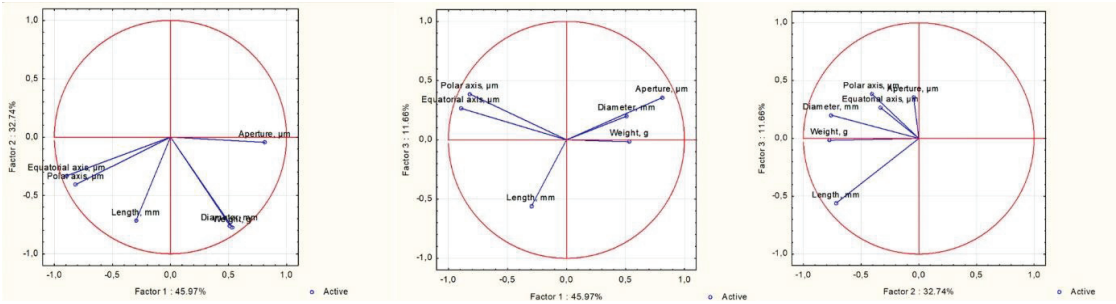


Fig. 20

Graphical visualization of Principal component analysis for *C. avellana* L. pollen (traits – polar axis, equatorial axis, aperture) and catkins (length, diameter)

The following charts (Fig. 21) are displaying similarities and dissimilarities among samples (genotypes). At all charts the genotypes CA-P-07, CA-P-08 (circle of red color) are always standing out of others, and we can suppose that these genotypes have different phenotype expression in analyzed traits. The genotypes CA-P-10, CA-P-11, CA-P-12 (cultivars) making also separated group (circle of red color), what is visible at the first two charts.

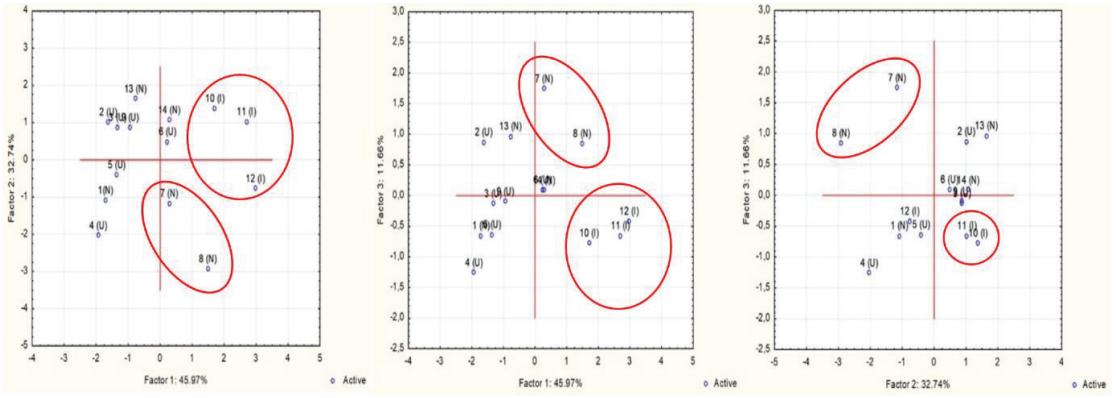


Fig. 21

***C. avellana* L. samples grouping by Principal component analysis**

4.5.2 Principal component analysis for pollen traits

In the second point of view we used only traits measured on pollen grains (length of polar and equatorial axis, and aperture diameter). In this case we chose again three factors, where the first factor explained 78.91 %, the second 19.60 %, and third only 1.49 % of variation in the data. The both charts (Fig. 22) display the trait aperture diameter which has the large positive loading on factor 1.

DOI: https://doi.org/10.15414/2020.9788055222622

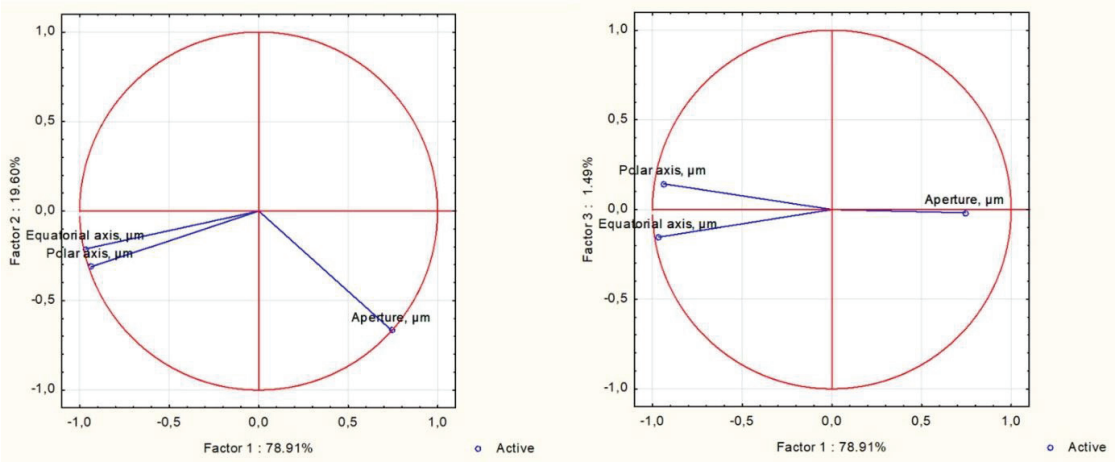


Fig. 22

Graphical visualization of Principal component analysis for *C. avellana* L. pollen (traits – polar axis, equatorial axis, aperture)

The charts presenting the similarities and dissimilarities among genotypes (Fig. 23) revealed that the genotypes CA-P-10, CA-P-11, CA-P-12 (cultivars – circle of red color), and the genotypes CA-P-07, CA-P-08 (locality Jelenec – circle of red color) made again the separated groups. In this point of view (traits on pollen grains) also the genotype CA-P-14 (locality Zobor) has interesting position towards the cultivars (standing closer to this group – circle of red color).

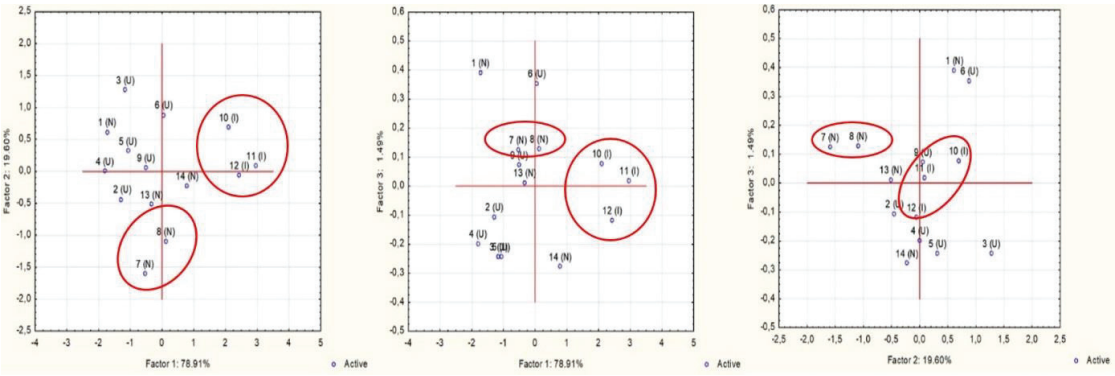


Fig. 23

***C. avellana* L. samples grouping by Principal component analysis**

DOI: https://doi.org/10.15414/2020.9788055222622

Conclusions

Solving the experimental activities about characterization and evaluation of male inflorescences of *Corylus avellana* L. selected genotypes by morphological traits we obtained the following conclusions:

Ecological location features

1. Selected samples of *C. avellana* were classified to groups – natural zones (CA-P-01, CA-P-07, CA-P-08, CA-P-13 and CA-P-14), urban zones (CA-P-02, CA-P-03, CA-P-04, CA-P-05, CA-P-06 and CA-P-09); industrial zone (CA-P-10, CA-P-11 and CA-P-12) in Slovakian conditions;
2. Detailed characteristics of sampling locations were created (Annexes).

Morphological characterization of pollen grains and catkins

Pollen morphology

Hazelnut pollen samples from different locations of Slovakia were compared by average values of polar axis (µm), equatorial axis (µm) and aperture diameter (µm).

1. The average value of polar axis of 14 selected genotypes was 24.91 µm;
2. The polar axis of samples from industrial zone was in the range from 21.43 µm to 22.04 µm, and in comparison to other observed zones, it was the smallest. The genotypes from natural zone (23.82 – 26.96 µm) and from urban zone (24.63 – 26.76 µm) reached pretty the same range of average values of pollen grain polar axis.
3. The average value of equatorial axis of 14 selected genotypes was 26.65 µm;
4. The equatorial axis of samples from industrial zone had reached again the smallest range from 22.24 µm to 23.34 µm, in comparison with samples from other zones. We found out that the genotypes from natural zone (26.12 – 28.26 µm) and from urban zone (25.41 – 29.63 µm) reached again pretty the same range of average values of equatorial axis.
5. The average value of aperture diameter of 14 selected genotypes was 3.25 µm;
6. The situation with the aperture diameter was repeated as in the previous traits measured on pollen grains. The smallest range was observed on genotypes from industrial zone (3.36 – 3.60 µm). The aperture diameter of samples from natural zones was in the range

DOI: https://doi.org/10.15414/2020.9788055222622

from 2.90 μm to 3.54 μm, and the aperture diameter of samples from urban zones was in the range from 2.84 μm to 3.20 μm.

7. From previously reported results we can conclude that the cultivars (CA-P-10, CA-P-11 and CA-P-12), genotypes from industrial zone, acquired the smallest average values in parameters measured on pollen grains.
8. The statistically significant differences in polar axis, equatorial axis and aperture diameter were found in the most cases by Duncan's test at a significant level of 0.05. The genotypes CA-P-02, CA-P-03, CA-P-06, CA-P-10, CA-P-11, CA-P-12, and CA-P-14 were significantly different from almost all or from all other genotypes repeatedly. The mentioned traits have power to discriminate genotypes.
9. The shape of pollen grains (P/E = 0.9) was determined. It was from oblate spheroidal to prolate spheroidal.
10. We used innovative approach of pollen grains morphology characterization by the tools of the image analysis software. From a wide range of open source options, we selected the software CellProfiler and CellProfiler Analyst. We used the automated measuring, machine-learning, and classification functionality of selected software.
11. The target phenotype was the pollen grains in polar point of view, and we got the set of 4078 objects, which were automatically classified, and measured. Using a traditional way of measuring the pollen grains, for example in polar and equatorial axis, it would never be possible to evaluate such many objects.
12. The variability among genotypes was analyzed on the new trait area (μm^2) of segmented object (pollen grain in polar point of view). The variation coefficient was for all analyzed samples less than 10 %, which can be a good indication that this trait (area μm^2) is suitable for determining genotypic differences. The statistically significant differences in the area were found in the most cases by Duncan's test at a significant level of 0.05.

Catkins morphometric features and weight

Hazelnut catkins samples from different locations of Slovakia were compared by average values of length (mm), diameter (mm) and weight (g):

1. The average value of length was 40.68 mm.
2. The samples from industrial zone reached the range of catkin length from 31.89 mm to 41.36 mm, what was the smallest value of the longest catkin. The samples from urban zone

DOI: https://doi.org/10.15414/2020.9788055222622

reached the length of catkin from 26.87 mm to 80.80 mm and it was the longest catkin value. The samples from natural zone obtained the length of catkin from 22.83 mm to 58.57 mm.

3. The average value of catkin's diameter was 5.04 mm.
4. The samples from all selected locations had the pretty the same average value of catkin's diameter, and that was from 4.38 mm to 5.20 mm.
5. The average value of catkin's weight was 0.43 g.
6. The smallest range of catkin's weight had the samples from urban zone (0.28 – 0.50 g). The samples from natural zone reached the weight of catkin from 0.2 g to 0.75 g and samples from industrial zone from 0.43 g to 0.68 g.
7. The cultivars (CA-P-10, CA-P-11 and CA-P-12) acquired the average values around or above the total average in parameters – diameter and weight of catkins.
8. The statistically significant differences in catkin's length, diameter and weight were found in the most cases by Duncan's test at a significant level of 0.05. The genotypes CA-P-04, CA-P-05, CA-P-07, CA-P-08, CA-P-09, CA-P-12, CA-P-13, and CA-P-14 were significantly different from almost all or from all other genotypes repeatedly. The mentioned traits have power to discriminate genotypes.
9. The average value of flowers number per one catkin was 252.58.
10. The highest number of flowers per catkin had samples from industrial zone (246.64 to 268.96). The samples from natural zone had from 239.68 to 260.64 flowers, and samples from urban zone had from 244 to 254.64 flowers per catkin.
11. The statistically significant differences in flower numbers were found in the pair CA-P-12 (cultivar "Webbova", industrial zone, the highest average number of flowers) and CA-P-13 (Nitra, Zobor-1, urban zone, the least average number of flowers).
12. The average value of pollen production of 50 catkins was 0.28 g.
13. The pollen production of samples from natural zone was in the range from 0.09 g to 0.56 g. The samples from urban zone had production from 0.15 g to 0.44 g, and samples from industrial zone had production from 0.20 g to 0.45 g.

Principal component analysis of pollen and catkins morphology

1. The morphological data (on catkins and pollen grains) were combined by Principal component analysis. The traits the length of catkins and aperture diameter had the largest loading on selected factors.

DOI: https://doi.org/10.15414/2020.9788055222622

2. From the results was revealed that the genotypes CA-P-10, CA-P-11, CA-P-12 (cultivars), and CA-P-07, CA-P-08, CA-P-14 made the separated groups, which may suggest the different phenotype expression in analyzed traits.
3. By looking at locations' characteristics (Annexes) all mentioned genotypes belong to region with precipitations totals in 2016 - 701 – 800 mm. The genotype CA-P-07, CA-P-08, CA-P-14 grew in locations characterized as natural (Jelenec, Zobor). The pH was weakly acidic (6.1 – 6.5), medium acidic (5.6 – 6.0) for locality Zobor (genotype CA-P-14), and neutral (6.6 – 7.3), weakly alkaline (7.4 – 7.8) for localities Nitra (CA-P-10, CA-P-11, CA-P-12) and Jelenec (CA-P-07, CA-P-08). We can suppose that the location characteristics had not influence on separation of genotypes by principal component analysis.
4. We assume that the genotypes CA-P-10, CA-P-11, CA-P-12 form separated group because they are cultivars and it means that they phenotypic expression was formed by breeding process. It is interesting that genotype CA-P-14 (Zobor) on charts was always placed close to this group.
5. We do not know way the genotypes CA-P-07, CA-P-08 (locality Jelenec) had formed separated group. We can conclude that in the monitored traits on pollen and catkins they form original phenotype compared to other analyzed genotypes. There is needed further experimental work on these genotypes.

DOI: https://doi.org/10.15414/2020.9788055222622

Proposes and recommendation for practice

In the following chapter there are summarized recommendations for practice.

1. Extension of theoretical knowledge about some biological features *C. avellana* (pollen and catkin morphology) can complement the species characteristics in the Slovak Republic.
2. The evaluation of hazelnut on pollen and catkins levels bring a new outlook on the assessment of natural values of the country and it might be use for country typology in Slovakia.
3. Applying a visual analyzing of pollen grain – size, shape, polar and equatorial axes, aperture it might be helpful for *C. avellana* pollen identification as a source of the appropriate botanical taxonomic characterization.
4. As the *Corylus* species represent a large and significant portion of park plant communities, they can be used in biomonitoring programs of the urban environment in which human activity contributes to the increase in pollution and, consequently, affects the growth of plants. Studies of the impact of human-induced stress on the morphology of pollen are necessary in order to estimate the reproductive potential of plants in natural, urban and industrial areas.
5. The increased pollution leads to the reduced size and germination rate of pollen. That is why some authors consider that pollen is a good indicator of the state of the environment. The share of phenotypic variability determined by environmental differences within the zones (types of growth) is statistically significant for the analyzed pollen properties. The effects of environmental factors in the urban areas on the morphological properties of pollen of deciduous woody species, i.e., the use of pollen properties as the bioindicator of the environment conditions.
6. We proposed the new way of pollen morphology characterization by automated system of image analysis software. The advantages of this system are following – more than 100 pollen grains can be simply measured, new quantitative traits can be used and bring the new exact data for pollen morphology, which can help for pollen origin identification.
7. The modern scientific approach in biological sciences is to combine the results of different experimental methods. In our work we combined the results gathered by Principal component analysis, expression levels of profilin, and genetic diversity analysis, which revealed the interesting conclusions. The genotypes CA-P-08 (locality Jelenec), and CA-P-14 (locality Zobor) had separated position towards the rest of selected genotypes. Also, cultivars (CA-P-10, CA-P-11, CA-P-12) from the morphological analysis point of view had separated position, which can be caused by breeding manipulation with morphological traits.

DOI: https://doi.org/10.15414/2020.9788055222622

References

ALDÉN, B. – RYMAN, S. – HJERTSON, M. 2012. *Svensk Kulturväxtdatabas*, [online] SKUD. Swedish Cultivated and Utility Plants Database, updated 2012. [cit. 2017-11-02]. Available at: <www.skud.info>.

APG III. 2009. An update of the Angiosperm Phylogeny Group classification for the orders and families of flowering plants: APG III. In *Botanical Journal of the Linnean Society*, vol. 161, pp. 105–121.

BEDINGER, P. 1992. The remarkable biology of pollen. In *The Plant Cell,* vol. 4, pp. 879- 887.

BLACKMORE, S. – BARNES, S.H. 1990. Pollen wall development in angiosperms. In S. Blackmore and R. B. Knox (eds.). *Microspores: evolution and ontogeny*. London : Academic Press, vol. 173–192.

BLACKMORE, S. – STEINMANN, J.A.J. – HOEN, P.P. – PUNT, W. 2003. *Betulaceae* and *Corylaceae*. In *The Northwest European Pollen Flora 65. Review of Palaeobotany and Palynology,* vol. 123, pp. 71-98.

BOCCACCI P. – BOTTA, R. – ROVIRA, M. 2008. Genetic diversity of hazelnut (*Corylus avellana* L.) germplasm in northeastern Spain. In *Horticultural Science*, vol. 43, pp. 667–672.

BORG, M. – TWELL, D. 2011. Pollen: Structure and Development. In *eLS (Encyclopedia of Life Sciences),* ed. Agró A. F., editor. (Chichester: John Wiley & Sons, Ltd.). DOI: 10.1002/9780470015902.a0002039.pub2.

BOTU, I. – TURCU, E. 2001. Evaluation of ecological conditions and prospects for growing hazelnuts in Romania. Proc. 5th Int. Cong. on Hazelnuts. In *Acta Horticulturae,* vol. 556, pp.117-123.

BRAY, M.A. – CARPENTER, A.E. 2018. Quality Control for High-Throughput Imaging Experiments Using Machine Learning in Cellprofiler. In *Methods in Molecular Biology* (Clifton, N.J.), vol. 1683, pp. 89-112, DOI: 10.1007/978-1-4939-7357-6_7.

HUNT, A.W.J. 2001. *Pruning Kentish Cobnuts*. Kentish Cobnuts Association, 32 p. ISBN-13: 978-0954160500

CARPENTER, A. E. – JONES, T. R. 2019. CellProfiler cell image analysis software. [online] [Cit. 2019-05-10]. Available at: <www.cellprofiler.org>

CARPENTER, A.E. – JONES, T.R. – LAMPRECHT, M.R. – CLARKE, C. – KANG, I.H. – FRIMAN O. – GUERTIN, D.A. – CHANG, J.H. – LINDQUIST, R.A. – MOFFAT, J. – GOLLAND, P. – SABATINI D.M. 2006. CellProfiler: image analysis software for identifying and quantifying cell phenotypes. In *Genome Biology,* vol. 7, iss. 10, article R100, 11 p. DOI: 10.1186/gb-2006-7-10-r100.

CHEN W. – XIA X. – HUANG Y. – CHEN X. – HAN J-D J. 2016. Bioimaging for quantitative phenotype analysis. In *Methods*, Volume 102, 2016, p. 20-25, ISSN 1046-2023. https://doi.org/10.1016/j.ymeth.2016.01.017.

CLÉMENT, C. – LAPORTE, P. – AUDRAN, J.C. 1998. The loculus content and tapetum during pollen development in *Lilium*. In *Sex Plant Reproduction*, vol. 11, pp. 11–94.

DOI: https://doi.org/10.15414/2020.9788055222622

ČREPINŠEK, Z. – ŠTAMPAR, F. – KAJFEŽ-BOGATAJ, L. – SOLAR, A. 2012. The response of *Corylus avellana* L. phenology to rising temperature in north-eastern Slovenia. In *International Journal of Biometeorology,* vol. 56, pp. 681–694.

ČURLÍK, J. – ŠEFČÍK, P. 1999. Mapa pôdnej reakcie (pH). In GRANEC, M. – ŠURINA, B. 1999. *Atlas pôd SR.* Bratislava : Výskumný ústav pôdnej úrodnosti, 60 s. [online]. [cit. 2019-05-30] Dostupné na: http://www.podnemapy.sk/portal/prave_menu/atlas_pod_sr/Atlas_pod_SR.pdf

D'CRUZ, S.C. – VAITHINATHAN, S. – JUBENDRADASS, R. – MATHUR, P.P. 2010. Effect of plants and plant products on the testis. In *Asian journal of Andrology*, vol. 12, is. 4, pp. 468-479.

DAO, D. – FRASER, A.N. – UNG, J. – LJOSA, V. – SINGH, S. – CARPENTER, A.E. 2016. CellProfiler Analyst: interactive data exploration, analysis and classification of large biological image sets. In *Bioinformatics*, Vol. 32, is. 20, pp. 3210–3212.

DAVIS, P.H. 1982. *Flora of Turkey and the east region islands*. Flora of Turkey, Edinburgh University Press, vol. 7, 948 p. https://www.jstor.org/stable/10.3366/j.ctvxcrhxk

DUKE, J.A. 2000. *Handbook of Nuts: Herbal reference library*. CRC Press; 1 edition, 360 p. ISBN-13: 978-0849336379

ĎURIŠOVÁ, Ľ. 2018. *Vývin, vitalita a morfológia peľu zástupcov rodu Prunus L*. Nitra : Slovenská poľnohospodárska univerzita, 2018. 116 p. ISBN 978-80-552-1945-5

DYAKOWSKA J. 1959. *Podrezcznik palinologii. Metody i problemy*. Warszawa : Wydawnictwa Geologiczne, Wyd. 1, p. 325.

ENCKE, F. – BUCHHEIM, G. – SEYBOLD, S. 1993. *Zander: Handwörterbuch der Pflanzennamen* = Zander : dictionary of plant names. Verlag E. Ulmer Stuttgart, 770 p. ISBN 3800150638.

ERDTMAN G. 1952. *Pollen morphology and plant taxonomy*. Angiosperms. Stockholm : Almqvist and Wiksell, 539 p. ISBN 90-04-08122-4

ERHARDT, W. – GÖTZ, E. – BÖDEKER, N. – SEYBOLD, S. 2000. *Zander. Handwörterbuch der Pflanzennamen* = Zander : dictionary of plant names. Verlag E. Ulmer Stuttgart, 16. edn., 990 p.

FRARY, A. – ÖZTÜRK, S. – BALIK, H.I. – BALIK, S.K. – KIZILCI, G. – DOĞANLAR, S. – FRARY, A. 2019. Association mapping of agro-morphological traits in European hazelnut (*Corylus avellana* L.). In *Euphytica*, vol. 215, pp. 215-221.

FRENGUELLI, G. – FERRANTI, F. –FORMACIARI, M. – ROMANO, B. 1994. Dormaci and chilling requirement for catkins in hazel (*Corylus avellana* L., *Corylaceae*). In *Gionale Botanico Italiano*, vol. 128, is. 1. DOI: 10.1080/11263509409437055.

FRENGUELLI, G. – TEDESCHINI, E. 1997. Volume changes in the pollen grain of *Corylus avellana* L. (*Corylaceae*) during development. In *Grana*, vol. 36, is. 5, pp. 289-292.

GERMAIN, E. – SARRAQUIGNE, J.P. 2004. *Le Noisetier*. Centre Technique Interprofessionnel des Fruits et Legumes, Paris : CTIFL, 295 p. ISBN 2879111595 9782879111599

DOI: https://doi.org/10.15414/2020.9788055222622

GERMAIN, E. 1994. The reproduction of hazelnut (*Corylus avellana* L.): a review. In *Acta Horticulturaea,* vol. 351, pp. 195–209.

GOLDBERG, R.B. – BEALS, T.P. – SANDERS, P.M. 1993. Anther development: basic principles and practical applications. In *Plant Cell*, vol. 5, pp. 1217–1229.

GRANEC, M. – ŠURINA, B. 1999. *Atlas pôd SR*. Bratislava : Výskumný ústav pôdoznalectva a ochrany pôdy, 60 s.

GOOGLE MAPS. 2019. [online]. [cit. 2019-05-30] Dostupné na: <https://www.google.com/maps/>

GRAU, P. – FRANCE, A. – GERDING, M. – LAVIN, A. – TORRES, A. 2001. Preliminary evaluation of hazelnut performance in Chile, Proc. 5th Int. Cong. on Hazelnut. In *Acta Horticulturaea,* vol. 556, pp. 49- 57.

HÁJKOVÁ, L. – NEKOVÁŘ, J. – RICHTEROVÁ, D. 2009. Temporal and spatial variability in allergy-triggering phenological phases of hazel and alder in Czechia. In *Folia oecologica*, vol. 36, no. 1, pp. 8-19.

HANELT, P. (eds.). 2001. *Mansfeld's Encyclopedia of Agricultural and Horticultural Crops* (except Ornamentals) (6 volumes). [online database]. [cit. 2019-04-21] and Institute of Plant Genetics and Crop Plant Research (IPK). Berlin : Springer Verlag. Available at: < http://mansfeld.ipk-gatersleben.de/apex/f?p=185:3:::::: >.

HESSE, M. – HALBITTER, H. – ZETTER, R. – WEBER, M. – BUCHNER, R. – FROSCH-RADIVO, A. – URLICH S. 2009. *Pollen terminology*. Wien, New York : Springer, 263 p.

HOFMAN J. – MICHALIK T. 1998. *Alergia pyłkowa*. Poznań : TOM, Ed. 1, 133 p. ISBN: 83-907607-0-3

IANOVICI, N. – BIRSAN, M. – TUDORICA, D. – BALIŢA, A. 2013. Fagales pollen in the atmosphere of Timişoara, Romania (2000-2007). In *Annals of West University of Timişoara*, ser. Biology, vol. XVI, is. 2, pp. 115-134.

JONES, T.R. – CARPENTER, A.E. – LAMPRECHT, M.R. – MOFFAT, J. – SILVER, S. – GRENIER, J. – ROOT, D. – GOLLAND, P. – SABATINI, D.M. 2009. Scoring diverse cellular morphologies in image-based screens with iterative feedback and machine learning. In *Proceedings of the National Academy of Sciences,* vol. 106, is. 6, pp. 1826-1831. doi: 10.1073/pnas.0808843106. PMID: 19188593 PMCID: PMC2634799.

JONES, T.R. – KANG, I.H. – WHEELER, D.B. – LINDQUIST, R.A. – PAPALLO, A. – SABATINI, D.M. – GOLLAND, P. – CARPENTER, A.E. 2008. CellProfiler Analyst: data exploration and analysis software for complex image-based screens. In *BMC Bioinformatics* vol. 9, is. 1, p. 482. /doi: 10.1186/1471-2105-9-482. PMID: 19014601 PMCID: PMC2614436.

KASAPLIGIL, B. 1963. Corylus colurna and its varieties. In *California Horticultural Society Journal*, vol. 24, pp. 95-104.

KHOKHLOV, S. 2001. Growing hazelnuts on the south coast of Crimea. Proc. 5th Int. Cong. on Hazelnut. In *Acta Horticulturaea*, vol. 556, pp. 141-142.

KREMP, G.O.U. 1967. *Palinologicheskaya entsiklopediya* (Palynological Encyclopedy), Moscow : Mir, 263 p.

DOI: https://doi.org/10.15414/2020.9788055222622

KRUCZEK, A. – PUC, M. 2012. Phenology of flowering and pollen release of selected herbaceous plants in Szczecin and Gudowo (Western Pomerania) and the risk of pollen allergy. In *Acta agrobotanica*, vol. 65, is. 4, pp. 49-60.

LAMPRECHT, M.R. – SABATINI, D.M. – CARPENTER, A.E. 2007. CellProfiler: free, versatile software for automated biological image analysis. In *Biotechniques*, vol. 42, is. 1, pp. 71-75. PMID: 17269487.

LINKEŠ, V. – PESTÚN, V. – DŽATKO, M. 1996. *Príručka pre používanie máp bonitovaných pôdno-ekologických jednotiek.* Príručka pre bonitáciu poľnohospodárskych pôd. Tretie upravené vydanie. Bratislava : Výskumný ústav pôdnej úrodnosti, 104 s.

McNEIL, D.L. 1999. *Walnuts, Chestnuts & Hazelnuts.* A compilation of general information, NZ situation and NZ research results [online resource]. [cit. 2020-04-23], Lincoln University, New Zealand, pp. 80. Available at: <http://ecite.utas.edu.au/41719>.

McQUIN, C. – GOODMAN, A. – CHERNYSHEV, V. – KAMENTSKY, L. – CIMINI, B.A. – KARHOHS, K.W. – DOAN, M. – DING, L. – RAFELSKI, S.M. – THIRSTRUP, D. – WIEGRAEBE, W. – SINGH, S. – BECKER, T. – CAICEDO, J.C. – CARPENTER, A.E. 2018. CellProfiler 3.0: Next-generation image processing for biology. In *PLoS Biology,* vol. 16, is. 7, p. e2005970.

MEHLENBACHER, S.A. 1991. Hazelnuts (*Corylus*). Genetic resources of temperate fruit and nut crops. In *Acta Horticulture,* vol. 290, pp. 791–836.

MEHLENBACHER, S.A. 2005. The hazelnut situation in Oregon, Proc. 6th Int. Cong. on Hazelnut. In *Acta Horticulturaea*, vol. 686, pp. 665 – 667.

MERCEL, F. 1988. Rozšírenie a variabilita zástupcov rodov *Cornus L., Swida. Opiz* a *Corylus* L. na Slovensku. In *Acta Dendrologica,* Bratislava: SAV, 1988, 164 s.

MERCEL, F. 2006. *Corylus* L. Lieska. In Goliašová, K. & Michalková, E. (eds): *Flóra Slovenska V/3*. Bratislava : Veda, pp. 180–186.

MEYER–MELYKYAN, N.R. 1987. Morphology of spores and pollen. In *Methodological aspects of palynology*. Moscow : Nedra, pp. 17-40.

MEYER-MELYKYAN, N.R. – SEVEROVA, E.E. – GAPOCHKA, G.P. – POLEVOVA, S.V. – TOKAREV, P.I. – BOVINA, I.YU. 1999. Principles and methods of aeropalynological investigations. Moscow. (in Russian)

MILLER, P. 1908. *The Gardeners dictionary*. London, Mo. Bot. Garden. [online]. B. m. : b. v. [cit. 2017-09-05]. 1367 p. Available at: < https://archive.org/details/mobot31753002894894>.

MIMET, A. – PELLISSIER, V. – QUÉNOL, H. – AGUEJDAD, R. – DUBREUIL, V. – ROZÉ, F. 2009. Urbanisation induces early flowering: Evidence from Platanus acerifolia and Prunus cerasus. In *International Journal of Biometeorology*, vol.53, pp. 287–298.

MINGEAU, M. – AMEGLIO, T. – PONS, B. – ROUSSEAU, P. 1994. Effects of water stress on development, growth and yield of hazelnut trees. In *Acta Horticul Ture*, vol. 351, pp. 305–314.

DOI: https://doi.org/10.15414/2020.9788055222622

MOORE, P.D. – WEBB, J.A. - COLLINSON, M.E. 1991. *Pollen Analysis*. Oxford : Blackwell, 2nd Edition, 216 p. ISBN-13: 978-0632021765

MYSZKOWSKA, D. – JENNER, B. – PUC, M. – STACH, A. – NOWAK, M. – MALKIEWICZ, M. – CHŁOPEK, K. – URUSKA, A. – RAPEIEJKO, P. – MAJKOWSKA-WOJCIECHOWSKA, B. – WERYSZKO-CHMIELEWSKA, E. – PIOTROWSKA, K. – KASPRZYK, I. 2010. Spatial variations in the dynamics of the *Alnus* and *Corylus* pollen season in Poland. In *Aerobiologia*, vol. 26, pp. 209-221.

NOVARA, C. – ASCARI, L. – LA MORGIA, V. – REALE, L. – GENRE, A. – SINISCALCO, C. 2017. Viability and germinability in long term storage of *Corylus avellana* pollen. In *Scientia Horticulturae*, Vol. 214, pp. 295-303, ISSN 0304-4238, Available at: <https://doi.org/10.1016/j.scienta.2016.11.042>.

INTERNATIONAL NUT AND DRIED FRUIT. NUTS AND DRIED FRUITS. 2017 – 2018. Statistical yearbook 2017/2018 [online resource]. [cit. 2020-04-23], Available at: <https://doi.org/www.nutfruit.org/who-we-are/news-and.../inc-2017-2018-statistical-yearbook (accessed on 18th May 2018) >.

PACINI, E. 1990. Tapetum and microspore function. In Blackmore, S. and Knox, R.B. (Ed.) *Microspores: evolution and ontogeny.* London : Academic press limited, p. 213-238. ISBN 0-12-103458-5.

PALACIOS, A.G. 2015. Corylus avellana: a new biotechnological source of anticancer agents. Dissertation thesis. Barselona : Universitat Pompeu Farba. 246 p.

PIOTROWSKA, K. 2008. Ecological features of flowers and the amount of pollen released in *Corylus avellana* (L.) and *Alnus glutinosa* (L.) Gaertn. In *Acta Agrobotanica*, vol. 61, is. 1, pp. 33-39.

POLLENATLAS 2013. *Corylus avellana* L. [online resource]. [cit. 2020-04-23], Available at: <http://www.polleninfo.org/SK/sk/allergy-infos/aerobiologics/pollen-atlas.html?letter=C>.

PUC, M. – KASPRZYK, I. 2013. The patterns of *Corylus* and *Alnus* pollen seasons and pollination periods in two Polish cities located in different climatic regions. In *Aerobiologia* (Bologna), vol. 29, pp. 495-511.

REHM, S. 1994. *Multilingual Dictionary of Agronomic Plants*. Springer Netherlands, 286 p. ISBN 978-0-7923-2970-1.

REMIŠOVÁ, V. – VINCEOVÁ, A. 2007. Kvitnutie liesky obyčajnej (*Corylus avellana* L.) na Slovensku za obdobie 1987 – 2006. In *Vyskot, I. (ed): „Klima lesa"*, Křtiny, 11. – 12.4.2007. ISBN 978-80-86690-40-7.

ROMANOVSKAJA, D. – BAKŠIENĖ, E. – RAŽUKAS, A. – TRIPOLSKAJA, L. 2012. Influence of climate change on the European hazel (*Corylus avellana* L.) and Norway maple (*Acer platanoides* L.) phenology in Lithuania during the period 1961-2010. In *Baltic forestry*, vol. 18, no. 2, pp. 228-236.

SARRAQUIGNE, J.P. 2005. Hazelnut production in France. In *Acta horticulturae*. vol. 686, pp. 669–672. DOI: 10.17660/ActaHortic.2005.686.89

DOI: https://doi.org/10.15414/2020.9788055222622

SCOTT, R.J. 1994. Pollen exine: the sporopollenin enigma and the physics of pattern. In *Molecular and Cellular Aspects of Plant Reproduction*. Cambridge University Press, pp. 49–81.

SCOTT, R.J. – SPIELMAN, M. – DICKINSON, H.G. 2004. Stamen structure and function. In *Plant Cell*, vol. 16, pp. 46–60.

SHARMA, V.K. 2003. *Studies on variability and selection in hazelnut in Himachal Pradesh* – Dissertation Thesis in Horticulture. [online resource]. [cit. 2020-04-23], College of Horticulture, Dr Yashwant Singh Parmar University of Horticulture and Forestry, Nauni, Solan-173 230, India. Available at: <http://krishikosh.egranth.ac.in/handle/1/5810017024>.

SILVESTRI, C. 2015. *Hazelnut (Corylus avellana L.) genetic resources and nursey industry improvement by biothechnological approaches).* The PhD Programme in Plant Biotechnology – XXVII Cycle [online resource]. [cit. 2020-04-23], Italy, 86 p. Available at: <https://pdfs.semanticscholar.org/d8a8/3324975c3eae2019928d6b1ac0b11a421541.pdf>.

SOLAR, A. – STAMPAR, F. 1997. First experiences with some foreign hazelnut cultivars (Corylus avellana L.) in Slovenia. In *Acta Horticulturae*, vol. 445, pp. 83-90. DOI: 10.17660/ActaHortic.1997.445.12

STACE, C. 1997. *New flora of the British Isles*. Cambridge university press. Ed. 2, 1232 p. ISBN 0521589335

STĘPALSKA, D. – MYSZKOWSKA, D. – PIOTROWICZ, K. – KASPRZYK, I. 2016. The phenological phases of flowering and pollen seasons of spring flowering tree taxa against a background of meteorological conditions on Kraków, Poland. In *Acta Agrobotanica*, vol. 65, is. 2, pp. 1678.

SUSZKA, B. 1980. Rozmnazanie generatywne. In *S. Białobok (Ed.) Olsze Alnus Mill,* Warszawa–Poznań: PWN. pp. 99–144.

TELLO, J. – MONTEMAYOR, M.I. – FORNECK, A. – IBÁÑEZ, J. 2018. A new image-based tool for the high throughput phenotyping of pollen viability: evaluation of inter- and intra-cultivar diversity in grapevine. In *Plant Methods*, Vol. 14, pp. 3, ISSN 1746-4811. Available at: <https://doi.org/10.1186/s13007-017-0267-2>.

THOMPSON, M.M. – LAGERSTEDT, H.B. – MEHLENBACHER, S.A. 1996. *Hazelnuts.* In *J. Janick and J.N. Moore (eds.). Fruit breeding*. New York : Wiley, Nuts. vol. 3, pp. 125–184.

TIYAYON, C. 2008. *A microscopic and phenological study of pollen development and bloom in selected cultivars of hazelnut (Corylus avellana)* – PhD thesis [online resource]. [cit. 2017-09-05], Oregon State University. Available at: <http://hdl.handle.net/1957/9976>.

TOKAREV, P.I. 2002. *Morphology and ultrastructure of pollen grains*. Moscow : Partnership of scientific publications KMK, 51 p.

TOMBESI, A. 2005. World hazelnut situation and perspectives: Italy. In *Acta horticulturae*. vol. 845, pp. 39–52. DOI: 10.17660/ActaHortic.2005.686.86

TURCU, E. – BOTU, I. 1997. Behaviour of some hazelnut varieties in the subcarpathian area of Oltenia - Romania. In *Acta Horticulturae*, vol. 445, pp. 135-144 DOI: 10.17660/ActaHortic.1997.445.18

DOI: https://doi.org/10.15414/2020.9788055222622

TURRILL, W.B. 1962. *Corylus jacquemontii* (Betulaceae). In *Curtis's Botanical Magazine,* vol. 173, is. 4, p. 391.

ÚHRNY ATMOSFÉRICKÝCH ZRÁŽOK NA SLOVENSKU 12/2013 V % NORMÁLU 1961-1990. Slovenský hydrometeorologický ústav, Odbor Klimatologická služba, 2013.

U.S. NATIONAL PLANT GERMPLASM SYSTEM. 2003. *Corylus avellana*. [online]. [cit. 2017-09-05]. Available at: <://npgsweb.ars-grin.gov/gringlobal/taxonomydetail.aspx?11643>

UOTILA, P. 2009. *Corylaceae*. In *Euro+Med Editorial Committee Euro+Med Plantbase: the information resource for Euro-Mediterranean plant diversity* [online resource]. [cit. 2017-09-05]. Available at: < http://www.emplantbase.org/home.html>.

ZINKL, G.M. – ZWIEBEL, B.I. – GRIER, D.G. – PREUSS, D. 1999. Pollen–stigma adhesion in *Arabidopsis*: a species-specific interaction mediated by lipophilic molecules in the pollen exine. In *Development,* vol. 126, pp. 5431–5440.

ZVERKO, J. – VIDO, J. – ŠKVARENINOVÁ, J. – ŠKVARENINA, J. 2014. Early onset of spring phenological phases in the period 2007-2012 compared to the period 1931-1960 as a potential bioindicator of environmental changes in The National Nature Reserve Boky (Slovakia). In ROŽNOVSKY, J., LITSCHMAN, T., (eds). 2014. *Mendel a bioklimatologie*. Brno, 3-5.09.2014. ISBN 978-80-210-6983-1.

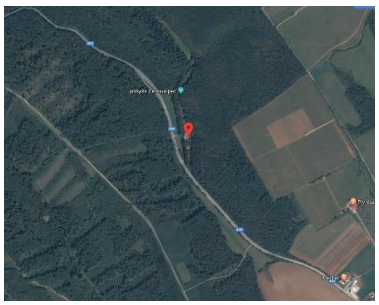
DOI: https://doi.org/10.15414/2020.9788055222622

Annexes

Physical and geographical data about locations were prepared by using the following sources: Linkeš et al., 1996; Granec and Šurina, 1999; Čurlík and Šefčík, 1999; Úhrny atmosférických zrážok na Slovensku 12/2013 v % normálu 1961-1990. All images of selected localities were copied from Google maps (2019).

1. **Čertova Pec** (48°33'29.1276" N; 17°54'57.3912" E).

   District Topoľčany, Radošina Cadaster.

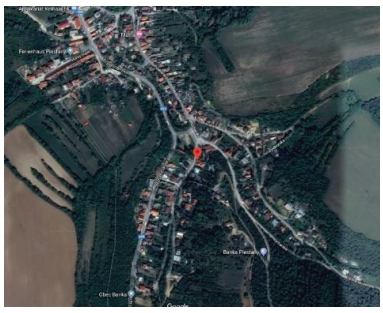



   Forest, roadhouse, road is approximately 50 m.

   Soil: fluvic gleysols; pH (H_2O): neutral (6.6 – 7.3), weakly alkaline (7.4 – 7.8). Climate region: from warm, dry to very dry, from lowland to hilly, sum of average daily temperatures above 10 °C is from 3000 to 2500; length of time with air temperature above 5 °C in days is from 237 to 231; precipitations totals in 2016: 600 – 700 mm.

2. **Banka-1** (48°34'45.624" N; 17°51'13.9788" E).

   District Piešťany




   Village, near road (approximately 3 m), built-up area.

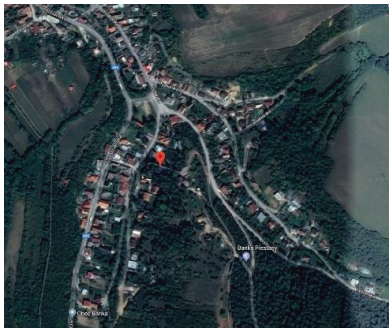
   Soil: eutric fluvisols, chernozems, mollic fluvisols, haplic luvisols, albic luvisols, arenosols, cambisols, rendzic leptosols, planosols and stagnosols, histosols, leptosols; pH (H_2O): neutral

DOI: https://doi.org/10.15414/2020.9788055222622

(6.6 – 7.3), weakly alkaline (7.4 – 7.8). Climate region: warm, dry or very dry, from lowland to hilly (sum of average daily temperatures above 10 °C is from 3000 to 2500; length of time with air temperature above 5 °C in days is from 237 to 231; precipitations totals in 2016: 701 – 800 mm.

3. **Banka-2** (48°34'43.6332" N; 17°51'14.0184" E).

District Piešťany

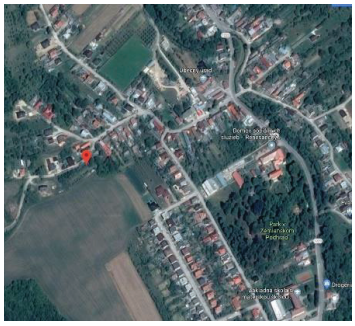


Village, on the hill, built-up area.

Soil: eutric fluvisols, chernozems, mollic fluvisols, haplic luvisols, albic luvisols, arenosols, cambisols, rendzic leptosols, planosols and stagnosols, histosols, leptosols; pH (H_2O): neutral (6.6 – 7.3), weakly alkaline (7.4 – 7.8). Climate region: warm, dry or very dry, from lowland to hilly (sum of average daily temperatures above 10 °C is from 3000 to 2500; length of time with air temperature above 5 °C in days is from 237 to 231; precipitations totals in 2016: 701 – 800 mm.

4. **Zemianske Podhradie** (48°50'24.2304" N; 17°49'42.5172" E)

District Nové Mesto nad Váhom. Region Trenčín. Biele Karpaty Protected landscape area.



Village, built-up area, field is approximately 20 m.

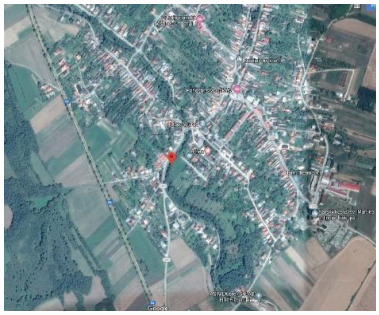
Soil: eutric cambisols and eutric fluvisols; ph (H_2O): neutral (6.6 – 7.3), weakly alkaline (7.4 – 7.8). Climate region: warm, dry or very dry, from lowland to hilly; sum of average daily

DOI: https://doi.org/10.15414/2020.9788055222622

temperatures above 10 °C is from 3000 to 2500; length of time with air temperature above 5 °C in days is from 237 to 231; precipitations totals in 2016: 801 – 900 mm.

5. **Moravské Lieskové** (48°49'1.2108" N; 17°47'28.5684" E)

District Nové Mesto nad Váhom. Region Trenčín. Biele Karpaty Protected landscape area.

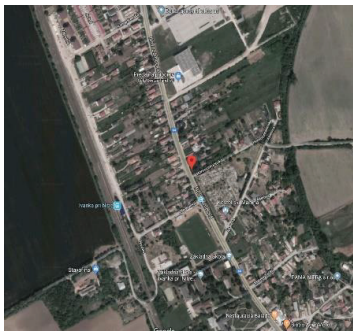



Village, on the highest hill. The main road is approximately 300 m.

Soil: eutric cambisols and eutric fluvisols; pH (H_2O): neutral (6.6 – 7.3), weakly alkaline (7.4 – 7.8). Climate region: warm, dry or very dry, from lowland to hilly; sum of average daily temperatures above 10 °C is from 3000 to 2500; length of time with air temperature above 5 °C in days is from 237 to 231; precipitations totals in 2016: 701 – 800 mm.

6. **Ivanka pri Nitre** (48°14'42.8928" N; 18°6'44.7732" E)

Southeastern part of the Nitra city. Loess hills on the floodplain of the old Nitra river.



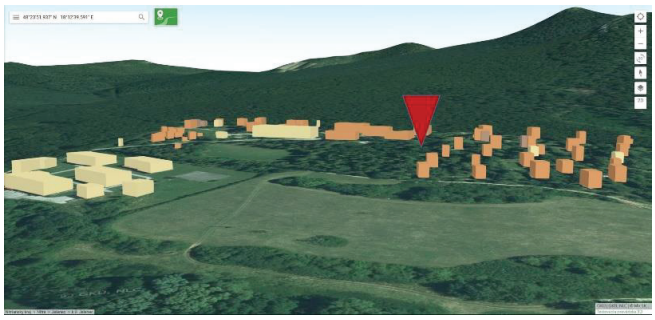

Village. Near road (approximately 3 m).

Soil: mollic fluvisols, arenosols, haplic luvisols, fluvisols, haplic chernozems; pH (H_2O): weakly alkaline (7.4 – 7.8), neutral (6.6 – 7.3). Climate region: from very warm, very dry, lowland to warm, very dry lowland, sum of average daily temperatures above 10 °C is from >3000 to 2800; length of time with air temperature above 5 °C in days is from 242 to 237; precipitations totals in 2016: 701 – 800 mm.

DOI: https://doi.org/10.15414/2020.9788055222622

7. **Jelenec-1** (48°23'51.937" N; 18°12'39.591" E)

Region Nitra, District Nitra.

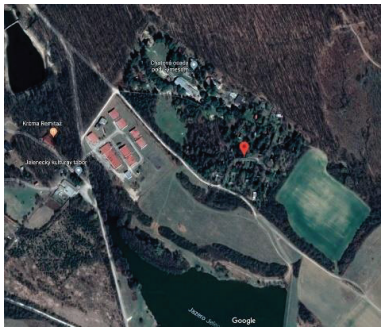
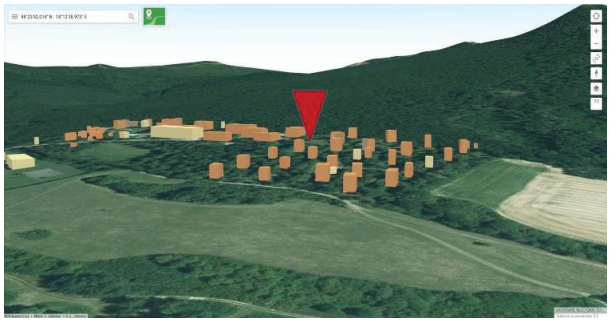



Recreation cottage area. On the hill, near forest and field (approximately 500 m).

Soils: eutric fluvisols, stagni-haplic luvisols, luvic stagnosols and planosols; pH (H_2O): neutral (6.6 – 7.3), weakly alkaline (7.4 – 7.8). Climate region: from very warm, very dry, lowland to warm, very dry lowland, sum of average daily temperatures above 10 °C is from >3000 to 2800; length of time with air temperature above 5 °C in days is from 242 to 237; precipitations totals in 2016: 701 – 800 mm.

8. **Jelenec-2** (48°23'52.014" N; 18°12'38.973" E)

Region Nitra, District Nitra.

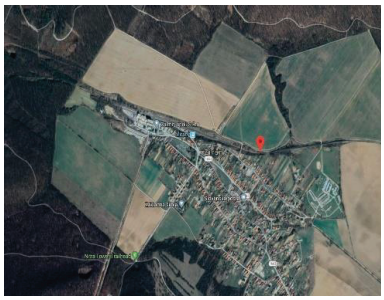
Recreation cottage area. On the hill, near forest and field (approximately 500 m).

Soils: eutric fluvisols, stagni-haplic luvisols, luvic stagnosols and planosols; pH (H_2O): neutral (6.6 – 7.3), weakly alkaline (7.4 – 7.8). Climate region: from very warm, very dry, lowland to warm, very dry lowland, sum of average daily temperatures above 10 °C is from >3000 to 2800; length of time with air temperature above 5 °C in days is from 242 to 237; precipitations totals in 2016: 701 – 800 mm.

DOI: https://doi.org/10.15414/2020.9788055222622

9. **Žirany** (48°22' 52.887" N; 18°10'36.864" E)

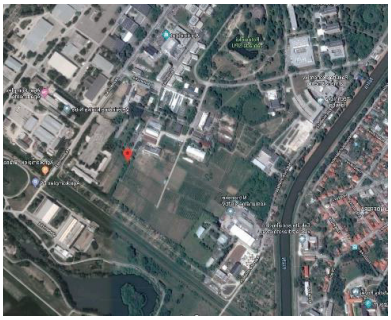
Region Nitra, District Nitra, Ponitrie Landscape Area.




Near road, built-up area. Approximately 400 m from whitewash factory.

Soil: fluvisols, mollic fluvisols, chernozems, arenosols, haplic luvisols, cambisols; pH (H_2O): from very strongly acidic (4.5 – 5.0) to strongly acidic (5.1 – 5.5). Climate region: from very warm, very dry, lowland to warm, very dry lowland, sum of average daily temperatures above 10 °C is from >3000 to 2800; length of time with air temperature above 5 °C in days is from 242 to 237; precipitations totals in 2016: 701 – 800 mm.

10. **Nitra** (48°18'8.646" N, 18°6'5.473" E; 48°18'8.363" N, 18°6'5.647" E; 48°18'8.222" N, 18°6'5.724" E)




Botanical garden of Slovak University of agriculture. Experimental bases of Botanical Garden.

Soil: mollic fluvisols, arenosols, haplic luvisols, fluvisols, haplic chernozems; pH (H_2O): neutral (6.6 – 7.3), weakly alkaline (7.4 – 7.8). Climate region: from very warm, very dry, lowland to warm, very dry lowland, sum of average daily temperatures above 10 °C is from >3000 to 2800; length of time with air temperature above 5 °C in days is from 242 to 237; precipitations totals in 2016: 701 – 800 mm.

DOI: https://doi.org/10.15414/2020.9788055222622

11. Nitra (Zobor-1) (48°20'45.137" N; 18°5'31.929" E)

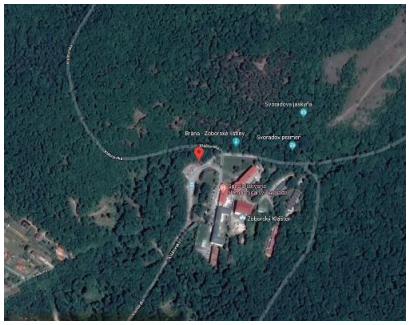
Region Nitra, District Nitra, National Nature Reserve Zoborská lesostep.

Treatment Institute. Forest

Soil: mollic fluvisols, arenosols, haplic luvisols, fluvisols, haplic chernozems; pH (H_2O): weakly acidic (6.1 – 6.5), medium acidic (5.6 – 6.0). Climate region: from very warm, very dry, lowland to warm, very dry lowland, sum of average daily temperatures above 10 °C is from 3000 to 2500; length of time with air temperature above 5 °C in days is from 237 to 231; precipitations totals in 2016: 701 – 800 mm.

12. Nitra (Zobor-2) (48° 20' 46.241" N; 18° 5' 25.576" E)

Region Nitra, District Nitra, National Nature Reserve Zoborská lesostep.



Parking of Treatment Institute. Forest.

Soil: mollic fluvisols, arenosols, haplic luvisols, fluvisols, haplic chernozems; pH (H_2O): weakly acidic (6.1 – 6.5), medium acidic (5.6 – 6.0). Climate region: from very warm, very dry, lowland to warm, very dry lowland, sum of average daily temperatures above 10 °C is from 3000 to 2500; length of time with air temperature above 5 °C in days is from 237 to 231; precipitations totals in 2016: 701 – 800 mm.

DOI: https://doi.org/10.15414/2020.9788055222622

Contacts

Mgr. Nataliia Nikolaieva, PhD.
Department of Genetics and Plant Breeding
Faculty of Agrobiology and Food Resources
Slovak University of Agriculture in Nitra
Tr. A. Hlinku 2, 949 01 Nitra
n.nikolaeva703@gmail.com

doc. Ing. Janka Nôžková, PhD.
Department of Genetics and Plant Breeding
Faculty of Agrobiology and Food Resources
Slovak University of Agriculture in Nitra
Tr. A. Hlinku 2, 949 01 Nitra
janka.nozkova@uniag.sk

Ing. Ján Gažo, PhD.
Department of Genetics and Plant Breeding
Faculty of Agrobiology and Food Resources
Slovak University of Agriculture in Nitra
Tr. A. Hlinku 2, 949 01 Nitra
jan.gazo@uniag.sk

DOI: https://doi.org/10.15414/2020.9788055222622

Authors: Mgr. Nataliia Nikolaieva, PhD.
doc. Ing. Janka Nôžková, PhD.
Ing. Ján Gažo, PhD.

Title: *Corylus avellana* L. pollen and catkins characterization on genotypes from selected localities of west Slovakia

Publisher: Slovak University of Agriculture (SUA) in Nitra

Edition: First

Form of publication: Online

Author of pictures on the book cover: Nataliia Nikolaieva**,** Radovan Ostrovský**,** Alexej Oravec, www.pixabay.com

Book cover design: Martin Lopušný

AQ – PQ: 4.61 – 4.76

Printed: Publishing center of SUA in Nitra

Not edited in the Publishing center of SUA in Nitra.

ISBN 978-80-552-2262-2

DOI: https://doi.org/10.15414/2020.9788055222622