Intraspecific bimodal variability in eye lenses of two Devonian trilobites

Catherine Crônier, Petr Budil, Oldřich Fatka and Lukáš Laibl

Abstract.—Arthropods are known to display a variable number of eye lenses and this number mostly increases during their development. In trilobites, most species possessing schizochroal eyes exhibit a notable intraspecific variation in the number of dorso ventral files of eye lenses that can be age related (i.e., growth) or not (i.e., living environment). Several previous studies have shown that some trilobite groups (e.g. phacopids) tend to have fewer lenses/files in representatives from the deeper habitats than those from shallower habitats. In this study, we analyzed the pattern of variation in the number of dorso ventral files of eye lenses in two Devonian phacopid trilobites from the Prague Basin of the Czech Republic. We quantified their intraspecific variability. To better understand the patterning, we compared more than 120 individuals. Data first reveal evidence of a bimodal distribution of lens/file number without intermediate forms among each of two studied populations of *Prokops prokopi* (Chlupáč, 1971) and throughout the ontogeny of *Pedinopariops insequens* (Chlupáč, 1977). Our results indicate that caution must be taken for taxonomical affiliation and biodiversity analyses of taxa in which the intraspecific variability is unclear. Additionally, we investigated possible relations of these bimodalities to the stratigraphical position of studied populations and to the paleoenvironment. In *Prokops prokopi*, a slightly different age of both populations, together with supposed differences in the local environments can be responsible for observed variability. In *Pedinopariops insequens*, stress conditions possibly related to the approaching onset of the Basal Choteč Event can be responsible for surprising intrapopulation variability. We speculate that the stress conditions could cause a bimodal selection and possibly also the change of ontogenetic trajectory within this species. *Pedinopariops insequens* was the only phacopid in the Prague Basin that crosses the Lower/Middle Devonian boundary and survived also the onset of Basal Choteč Event.

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Petr Budil. Czech Republic & Faculty of Environmental Science, Czech University of Life Sciences, Kamýcká 129, CZ-165 21 Praha 6 – Suchdol, Czech Republic and Czech Geological Survey, Klárov 3, 118 21 Prague 1, Czech Republic. E-mail: phacopidina@seznam.cz, petr.budil@geology.cz.
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Introduction

Numerous arthropods are known to display ommatidia, which are clustered together to form compound eyes (Oakley 2003; Waloszek et al. 2005; Harzsch and Hafner 2006; Nilsson and Kelber 2007; Aberhan et al. 2012), including the extinct trilobites (Clarkson 1979; Levi-Setti et al. 1998; Clarkson et al. 2006). Among these, phacopids are known to display a variable number of eye lenses or dorso ventral files (Thomas 1998; Crônier and Clarkson 2001), thereby providing variation for evolution and natural selection to act upon such a character (Darwin and Wallace 1958; Hallgrímsson and Hall 2005; Hunt 2007). Separating the variation

resulting from a phenotypic heredity of the variation resulting from a plastic environmental response is, however difficult, notably in extinct organisms (Urdy et al. 2010). Furthermore, knowledge of intraspecific variability is an important requirement for taxonomic studies as well as for studying of spatio temporal patterns of diversity through time and their relation with environmental changes (Crônier et al. 2004; Brayard et al. 2006; De Baets et al. 2012; Månsson and Clarkson 2012).

In trilobites, only a few studies documented an intraspecific variability in the number of files or lenses (for example Novák in Perner 1918; Clarkson 1966a, 1966b, 1971; Clarkson

and Tripp 1982; Thomas 1998; Budil 1999; Crônier and Clarkson 2001). Phacopid trilobite species exhibit a relatively moderate morphological intraspecific variation in the number of dorso ventral files that can be age related (Crônier and Clarkson 2001) or not (Crônier et al. 2004). In particular, some previous studies have shown that eyes in phacopine populations from deeper environments tend to be smaller and to have fewer lenses/files than those from shallower environment (Crônier et al. 2004; Feist et al. 2009). However, the intraspecific variability in these phacopid trilobites has hardly been studied in the respect to reveal a continuous spread of morphotypes with a common intermediate shape and a bimodal or polymodal distribution.

A large sample of well preserved phacopid trilobites from the Barrandian area gives us the opportunity to study the pattern of the variation in the number of dorso ventral files of eye lenses for two Devonian species and to quantify their intraspecific variability. In order to better understand the developmental patterning, to investigate the frequency distribution change through ontogeny and to explore the evolutionary significance of intraspecific variability in the lens number, we compared more than 120 individuals in two Barrandian species.

Additionally, we investigated possible relations of phenotypic variation with environmental characteristics of the studied area in an attempt to test whether the bimodality can persist and possibly be a factor in enabling organisms to adapt to environmental changes. For example, in organisms affected by sea level fluctuations, an eye lens polymorphism matching altered environments in space and time is of selective importance. Environmental fluctuations are known to influence the evolution of natural populations and to trigger long term evolutionary diversification. According to Schluter (2000), the diversification is driven by an adaptation to different ecological conditions. Thus, a divergent/disruptive selection causes adaptive divergence between conspecific groups that occupy different environments or use different resources, and suggested by a bimodal distribution of morphological traits. Such bimodal distribution can bear upon or control the origin of new species. Numerous

organisms are able to maintain a high genetic variability, and a rapid alteration of phenotypic traits is necessary for their survival as a response to environmental fluctuations (Ramel 1998; Frankham et al. 2002). Adaptive divergence and ecological speciation are believed to take part in the evolution of biodiversity (Skúlason and Smith 1995; Schluter 2000; Kristjánsson et al. 2002; Rundle and Nosil 2005).

Material and Methods

Data collection.—For our study, we analyzed individuals of two phacopid species, *Prokops prokopi* (Chlupáč, 1971) and *Pedinopariops insequens* (Chlupáč, 1977).

Prokops prokopi is locally abundant in the current lowermost Emsian (= classical upper Pragian; Chlupáč et al. 1998) in the near vicinity of Prague, in the Barrandian area (Figs. 1 and 2). This species is represented by two populations in our study: (1) the first population, of 22 individuals, comes from the Malá Chuchle locality (old quarry, 50°1'30.399"N, 14°22'46.758"E, in a suburb SW of Prague; Figs. 1, 2 herein; this locality has been discussed especially by Růžička 1940; Chlupáč 1957, 1983; Havlíček and Vaněk 1998; Vaněk 1999; Frýda et al. 2013), and (2) the second population originates from the Hlubočepy/St-Prokop's Quarry (called also Schwarzenberg's Quarry, 50°2'37.876"N, 14°22'34.475"E, in the Prokopské Údolí Valley, SW suburb of Prague; Figs. 1, 2 herein; this locality has been discussed especially by Woldřich 1919; Růžička 1941; Chlupáč 1957, 1983, 1988, 1999; Kříž 1999; Havlíček and Vaněk 1998; Vaněk 1999; Frýda et al. 2013). For this study, 12 individuals were used, all coming from several paleontological collections (Růžička, Vaněk, Chlupáč amongst others) housed in the National Museum Prague and in the Czech Geological Survey, Prague. All studied material originates from "white beds," i.e. the decalcified limestones (see Suchý 2002), from the Dvorce and Prokop Limestone facies, Prokop Limestone subfacies of Praha Formation (Fig. 2).

Prokops prokopi (Fig. 3A,B) is clearly distinguished from the other phacopids of the Prague Basin, including the ancestral and partially

contemporaneous *Prokops hoeninghausi* (Barrande, 1846) by the characteristic configuration of the visual surface that is reduced and asymmetrically kidney shaped, with 19–34 lenses as well as by vaulted glabella and steep genae.

Pedinopariops insequens is locally abundant in the interval of the uppermost Emsian to lowermost Eifelian in the near vicinity of Prague. This species is represented by 142 specimens in our study, including 42 individuals with well preserved eyes, coming from one population from the Praha Holyně Section in a small quarry NE of the village of Holyně and W of the Prastav quarry (50°1'57.627"N, 14°21'5.471"E) in a SW suburb of Prague (Figs. 1, 2 herein; this locality has been discussed especially by Bouček 1931; Růžička 1941; Chlupáč 1959, 1985, 1988, 1999; Kříž 1999; Frýda et al. 2013). All studied material comes from "white beds," i.e. the decalcified limestones (see above), and was collected by J. Vaněk, partially also by F. Hanuš (both collections are housed in the Czech Geological Survey, Prague and in the National Museum, Prague). This locality yielded a rich fauna comprising about 350 invertebrate species (Bouček 1931; Růžička 1941) and comes from topmost part of the Třebotov Limestone (uppermost Emsian/lowermost Eifelian). The exact dating of these strata has been a subject of discussion (Chlupáč 1959; Chlupáč 1988, 1999; Kříž 1999; Frýda et al. 2013). The boundary between the Lower and Middle Devonian was placed by Chlupáč et al. (1979) and Chlupáč (1982, 1993) within the topmost Třebotov Limestone, 2.8 m below the base of the overlying Choteč Limestone, in the nearby Prastav
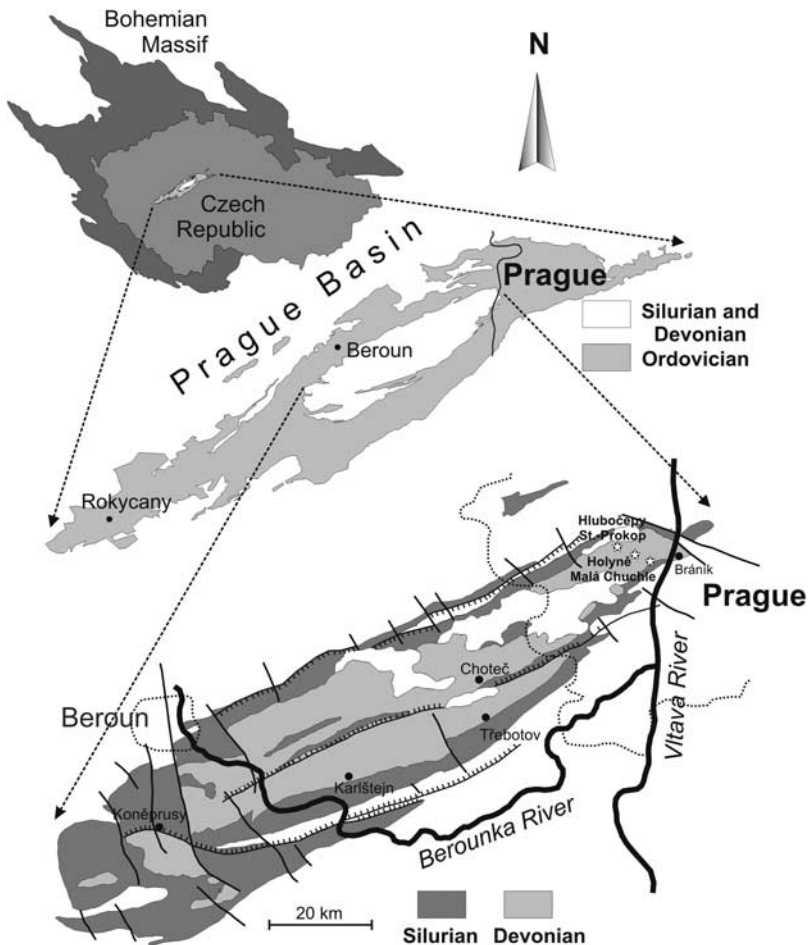

FIGURE 1. A sketch map of the distribution of the Devonian rocks in the Barrandian area. The position of both studied localities is indicated by stars.
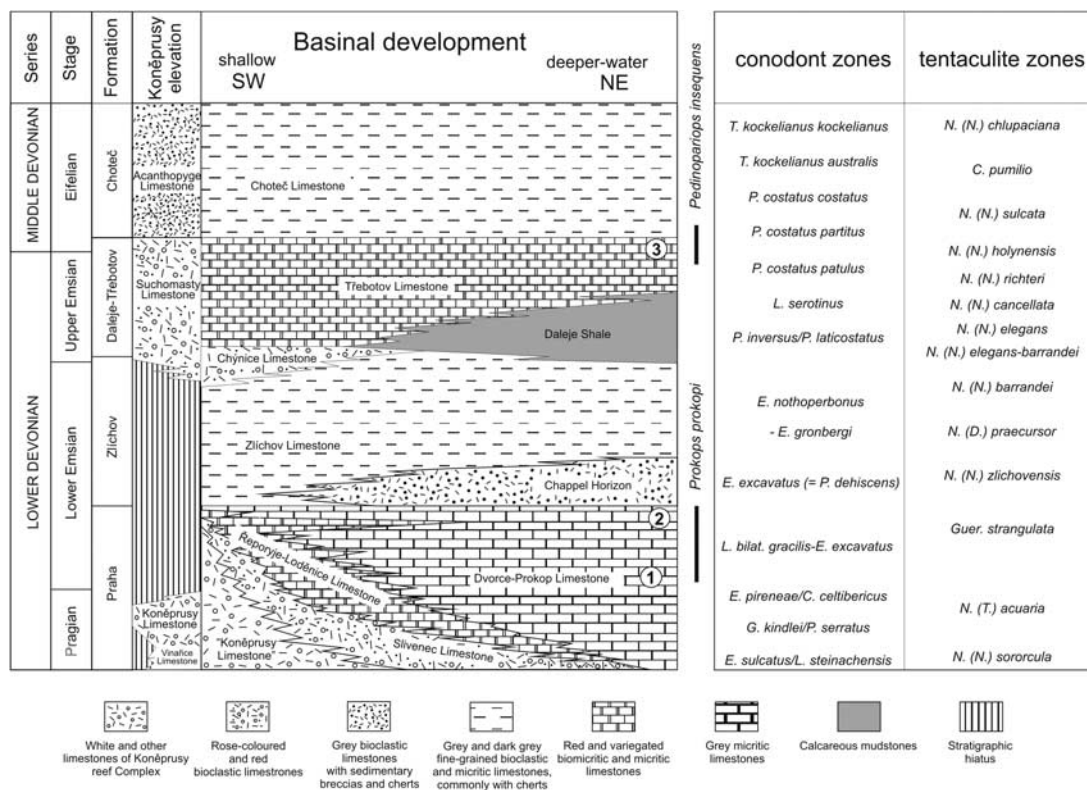
FIGURE 2. Pragian to Eifelian stratigraphy of the Prague Basin (after Chlupáč et al. 1998, modified by Budil et al. 2009). Conodont biozones of Pragian after Slavík (2004a, 2004b), Slavík et al. (2007), and Carls et al. (2008). Upper Emsian to Eifelian biozones after Gradstein et al. (2012), Ferová et al. (2012), Klapper and Vodrážková (2013) and Vodrážková et al. (2013). Key to the localities: (1) Malá Chuchle; (2) St. Prokop's Quarry; (3) Praha-Holyně Section.

Quarry (Chlupáč 1985). Berkyová (2009) moved this boundary here to ca. 5 m below the base of the Choteč Limestone, e.g. immediately above the supposed stratigraphical position of the "white beds" outcrop. Frýda et al. (2013) suggest that the weathered "white beds" at Holyně may belong already to the *Polygnathus costatus partitus* conodont Biozone of early Eifelian age.

By its characteristic configuration of the dorsal exoskeleton, *Pedinopariops insequens* (Fig. 3C,D) is regarded as closely related to *P. superstes* (Barrande, 1852), a stratigraphically older species from the Daleje Shale and lower part of the Třebotov Limestone (upper Emsian).

Methods and Measured Parameters.—The studied phacopids have been investigated using quantitative methods. Lenses and dorso ventral files for each preserved eye have been numbered using binocular microscopes NIKON MCZ 1500 and Olympus. Cephalic width and cephalic length (in mm) have been measured. Additionally, in order to describe the morphology of these organisms, a set of 21 landmarks was selected on the cephalon to summarize the general cephalic shape, along with the eye, for 33 individuals of *Prokops prokopi* and 88 individuals of *Pedinopariops insequens*. Landmarks are points among which the number and the position should be strictly identical for all individuals (Bookstein 1991). The x- and y-coordinates of 21 landmarks were obtained by an optical image analyzer (TPSdig) and were automatically extracted on cephala (Fig. 3E). Only the two dimensional projections of the dorsal views have been considered here. The Barrandian trilobite material (see above) is suitable for such analyses because this material is quite abundant, preserved in three dimensions, and came from a restricted stratigraphic interval, thus minimizing time averaging.
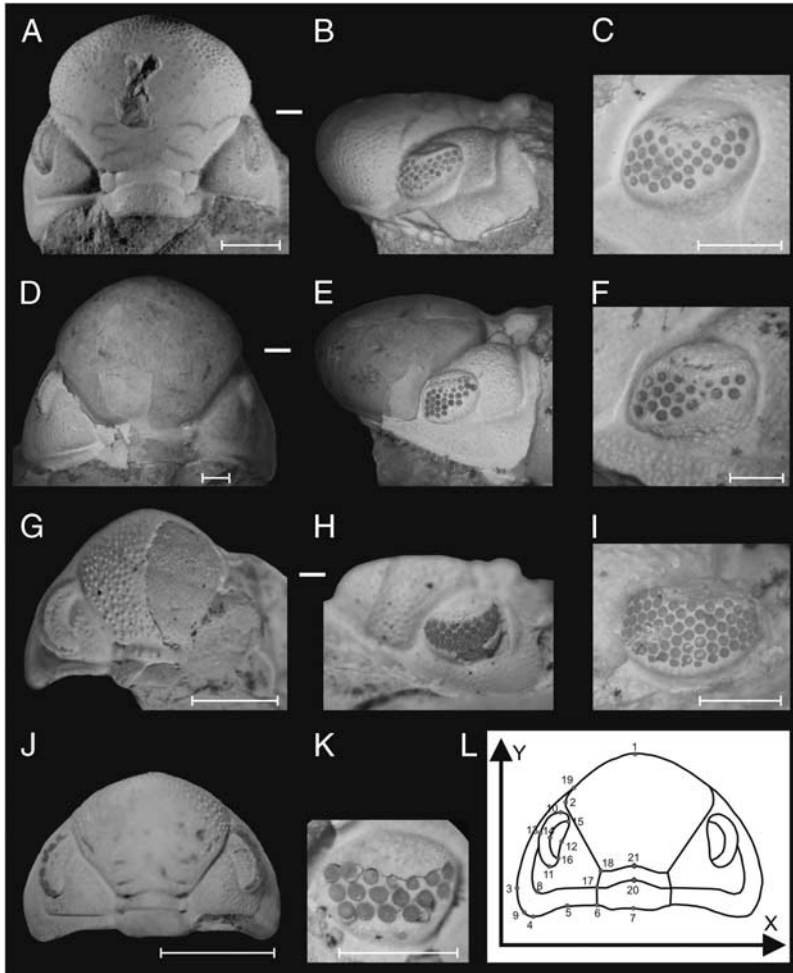
FIGURE 3. A–C, *Prokops prokopi* (Chlupáč, 1971) from the Malá Chuchle locality, in the vicinity of Praha: (A, B) cephalon in dorsal and lateral views respectively with 13 dorso ventral files, C, detail of an eye with 13 dorso ventral files. D–F, *Prokops prokopi* (Chlupáč, 1971) from the Hlubočepy/St-Prokop's Quarry, in the vicinity of Praha: D–E, cephalon in dorsal and lateral views respectively with 8/9 dorso ventral files, F, detail of an eye with nine dorso ventral files. G–K, *Pedinopariops insequens* (Chlupáč, 1977) from the Praha-Holyně Section, at a SW suburb of Praha: G–H, cephalon in dorsal and lateral views respectively with 18 dorso ventral files, I, detail of an eye with 18 dorso ventral files, J, cephalon in dorsal view with 12 dorso ventral files, K, detail of an eye with 12 dorso ventral files. L, Location of 21 landmarks on the cephalon. Scale: 2 mm.

Frequency histograms.—Frequency distributions have been investigated by both 'traditional' and smoothed histograms (via kernel density estimation, KDE) in order to detect any multimodality in the distribution. While the 'traditional' parametric histograms represent categorical variables and depend on the fixed width of the bins (divided into equal subintervals in which the whole data interval is divided) and the end points of the bins, the nonparametric Kernel density estimators represent noncategorical variables, which can reveal asymmetry and mutimodality (Salgado-Ugarte et al. 1994, 2002). Plots histograms and KDE were achieved using the data analysis software PAST v2.08 (Hammer et al. 2001; Hammer and Harper 2006). KDE uses an optimal Gaussian bin width according to the rule given by Silverman (1986).

Geometric morphometry.—To complete the biometrical study, a geometric morphometric method based on a set of landmarks has been used for an accurate description of specific features such as cephala. These landmarks are tracked down in a system of axes in Cartesian

coordinates and have been compared by fit methods (see Adams et al. 2004 for details on procedures).

Each cephalon is defined by a configuration of landmarks containing both size and shape information. Size being independently analyzed; all configurations of landmarks were scaled to unit centroid size. The centroid size is the square root of the mean squared distance between the centroid and each landmark of the configuration (Bookstein 1991). The scaled configurations are then superimposed, i.e., translated and rotated through the Generalized Least Square (GLS) Procrustes procedure (Rohlf and Slice 1990). The superimposed configurations constitute a data set corresponding to Procrustes residuals, i.e., x- and y-coordinates of each landmark of configurations superimposed on a reference configuration. This method requires: (1) the translation of all configurations in order to superimpose their centroid and (2) the rotation of all configurations in order to minimize the scatter of landmarks, i.e., the sum of squared distances between landmarks of each configuration to the corresponding landmarks of a consensus configuration, here the average of all configurations.

The GLS superimposition is computed by the *tpsSuper* software v. 1.14 (Rohlf 2003a). Five replicating landmark measurements carried out on the same individual demonstrate that within individual measurement error is negligible. Univariate Analyses of Variance (ANO-VAs) on each coordinate of landmarks indicate no significant morphological differentiation among replicas ($P = 0.05$ NS).

To examine the major trend of shape variation and to estimate the intraspecific variability, the residual matrix is then subjected to a Thin Plate Spline (TPS) procedure, i.e., a Relative Warp Analysis (RWA, see Rohlf 1993), which fits an interpolated function to the consensus configuration. This is a principal components analysis of the partial warp scores that describe localized departures from the average configuration (Bookstein 1991; Rohlf 1993). The RWA allows the emergence of principal independent axes according to a decreasing order of importance in the explanation of the shape variability. Significant RWs define a shape space into which it is possible to locate each

individual according to its coordinates on these RWs. Analysis of variance is then applied on the matrix of individual scores on the significant RW in order to assess the shape difference between the populations/groups. Moreover, thin plate spline deformation grids have been plotted on each extremity of the first Significant RWs. The RWA is computed by the *tpsRelw* program (Rohlf 2003b).

Additionally, in order to compare size and shape variations of these individuals, a morphological space defined by both the size, estimated by the centroid size of cephala, and the shape, estimated by the first axis of the relative warp, was constructed to visualize the relationships between size and shape and to determine the correlation between the scores on the first axis and centroid size. The size with the shape describes the complete morphology of the cephala.

Range of Intraspecific Variability.—In order to determine trends in the amount of deviation from the mean lens number and the global shape variability, the coefficient of variation (CV) that is a measure of the range of intraspecific variability of these morphological characters, has been calculated and compared among populations by using an F-max test. For the global shape variability, the degrees of freedom have been modified to take into account the number of coordinates and the loss of four degrees of freedom due to the Procrustes Superimposition procedure (Klingenberg and McIntyre 1998).

Variability within and between populations of *Prokops prokopi*

Frequency Distributions.—The frequency distribution of the number of dorso ventral files of lenses for *Prokops prokopi* from the Malá Chuchle locality and the Hlubočepy/St-Prokop's quarry is presented in figure 4A. Estimations from the 'traditional' and the smoothed (Gaussian KDE) histograms result in a multimodal distribution; at least two modes can be distinguished (Fig. 4A). Additionally, bivariate plots of lenses versus files number, of cephalic width (tr.) versus cephalic length (sag.) and, of files versus cephalic width (tr.) are respectively presented in figure 4B, 4C, and 4D.
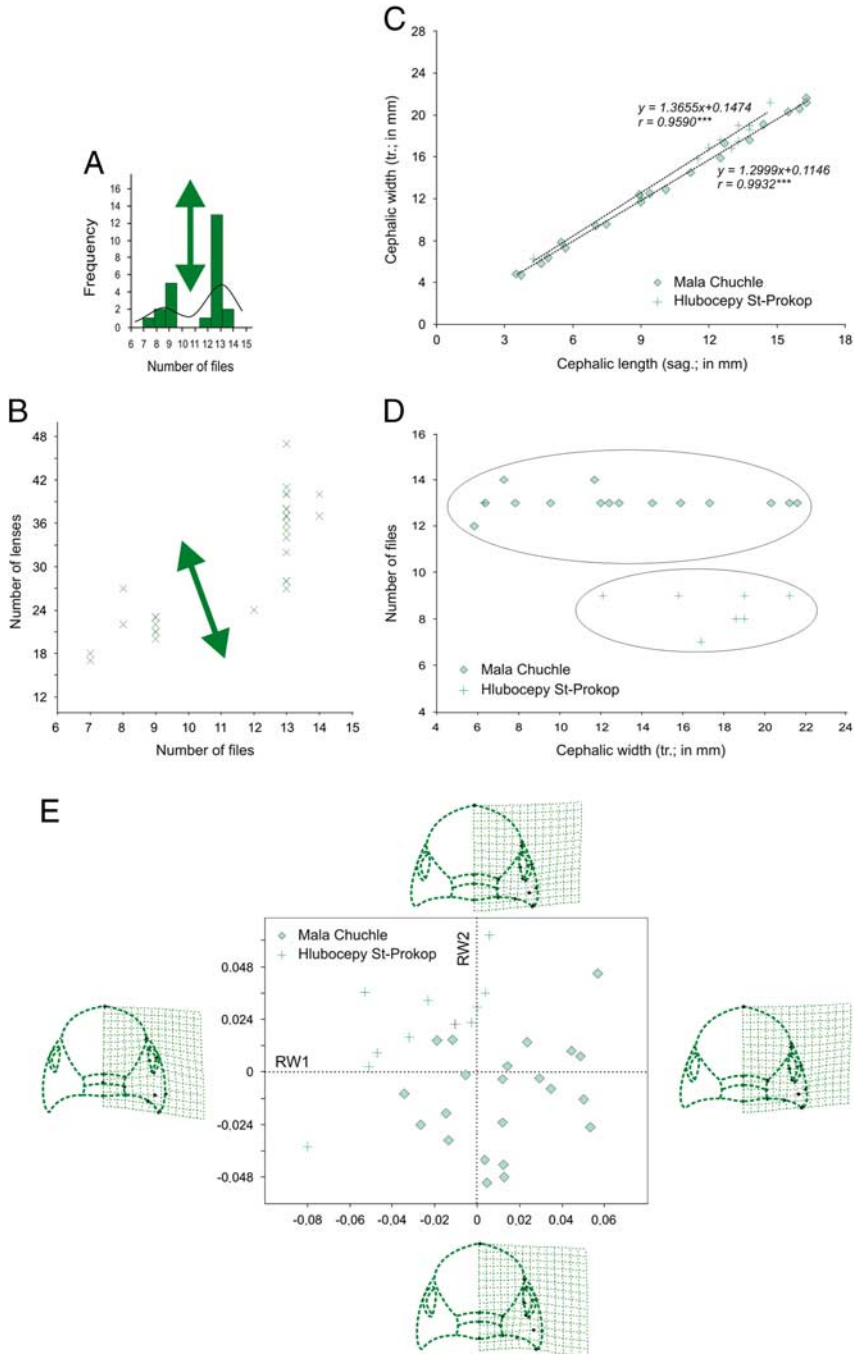
FIGURE 4. A, The frequency distribution of the file number of lenses for all individuals of *Prokops prokopi* (Chlupáč, 1971). The 'traditional' histogram and the Kernel density estimate distribution of file number using the optimal Gaussian bin width. The frequency distribution shows two modes, i.e. a bimodality in the file number of lenses; B, scatter plot of lens number versus file number for all individuals; C, scatter plot of cephalic width (tr.) in mm versus cephalic length (tr.) in mm for all individuals showing a discontinuous and a typical isometric distribution for each population; D, scatter plot of file number versus cephalic width (tr.) in mm for all individuals; E, location of 33 cranidia/cephala of *Prokops prokopi* (Chlupáč, 1971) in the morphospace defined according to the first two relative warps RW1 and RW2. Axes RW1 and RW2 represent 25.5% and 18.5%, respectively, of the total variance. Thin plate spline deformation grids depicting patterns of shape change between the both populations of *Prokops prokopi*. Double arrows in (A) and (B) underline the gap.

TABLE 1. Univariate Analyses of Variance performed (A, B) on cephalic width (in mm) and cephalic length (in mm) showing no significant morphological differentiation between the two populations of *Prokops prokopi* (Chlupáč, 1971); C, D, on the file and lens number showing a significant differentiation among the two populations of *Prokops prokopi* (Chlupáč, 1971); E–G, on the first three significant axes showing a significant morphological differentiation among the two populations of *Prokops prokopi* (Chlupáč, 1971) according to RW1 and RW2.

Source of variation	ddl	Sum of squares	Mean square	Fisher F	$P > F$
(A) Width	1	87.354	87.354	3.289	*P = 0.079 NS*
(B) Length	1	30.166	30.166	1.969	*P = 0.170 NS*
(C) Lens number	1	1081.600	1081.600	48.000	*P < 0.0001*
(D) File number	1	93.025	93.025	82.081	*P < 0.0001*
(E) RW1	1	0.011	0.011	14.0976	*P < 0.001*
(F) RW2	1	0.007	0.007	12.943	*P < 0.001*
(G) RW3	1	0.001	0.001	1.112	*P = 0.300 NS*

In comparing two independent groups, an F-test provides a measure of the probability that they have the same variance or regression model (Lomax 2007). However, in our groups, the cephalic length (sag.) does not meet the assumptions of normality of distribution and / or homogeneity of variances. Therefore, a nonparametric Mann-Whitney U-test was used (Mann and Whitney 1947), and shows that the mean of this variable does not differ between the two studied groups ($Z = 1.279$, $p = 0.2$NS). And, no significant differences between the ontogenetic trajectories of *Prokops prokopi* from the Malá Chuchle locality and the Hlubočepy/St-Prokop's quarry, were detected by an F-test on the slopes ($F = 1.561$, $p = 0.22$NS).

The size range of cephala in the Malá Chuchle locality as in the Hlubočepy/St-Prokop's quarry is almost the same from 3.5 to 17 mm for the length and from 4.5 to 22 mm for the width (Fig. 4C). The ANOVAs performed on the cephalic width and length confirms the existence of a size unity among groups previously defined according to their provenance. All tests are insignificant (Table 1A,B). Furthermore, in the Malá Chuchle locality, the lowest values of lens file number are 12, with the maximum values at 14. In the Hlubočepy/St-Prokop's quarry, the lowest values are seven, with the maximum values at nine, with one exception at 13 (Fig. 4D). Apart from the exception, in Malá Chuchle as in Hlubočepy/St-Prokop´s quarry, the lens file number range is three. ANOVAs performed on the file and lens number confirms the existence of a differentiation among groups previously defined according to their provenance. All tests are significant (Table 1C–D). Thus, the reduction in the lens files number in the St-Prokop population is independent of body size. Additionally, the coefficient of variation in the number of eye lenses is more important in this St-Prokop population (CV = 18.43%) than in the Malá Chuchle population (CV = 3.50%). The comparison of the coefficient of variation by a F-max test indicates a significant heterogeneity of variance among groups ($F_{vmax/vmin} = 5.27$; $ddl_1 = 13$; $ddl_2 = 7$; $p = 0.018$) due to the presence of one individual with 13 lenses in the St-Prokop's quarry.

Morphological space.—Morphological relationships among individuals can be displayed by their representation in the morphological space (Fig. 4E) defined by the first two principal axes, which provide a valuable approximation of the morphospace (56% of the total variance). The first relative warp explained 25.5% of the observed variation among individuals, the second 18.5%. Analyses of variance applied on the matrix of individual scores on the first three RW indicate a significant morphological differentiation among populations according to RW1 and RW2 (Table 1E–G). The set of individuals is grouped together according to their locality in the morphospace along the first two axes (Fig. 4E). The morphological interpretation of the shape changes between individuals from these two localities is indicated by the TPS warp grids showing the direction of maximum variance (Fig. 4E). The first two relative warp axes show that the main shape change in these both populations contrasts more or less developed eyes: the individuals from Malá Chuchle with more developed eyes and the individuals from Hlubočepy/St-Prokop with less developed eyes.

The morphological relationships among individuals observed in the morphological space support the same general pattern as the bivariate plot of file number versus cephalic width (tr.), towards more files in individuals from Malá Chuchle area.

Furthermore, linear regressions performed on the size (width) and shape (RW1 and RW2) suggest that there is no relationship between size and shape throughout the studied span of growth among populations previously defined according to their provenance.

Variability within population of *Pedinopariops insequens*

Frequency Distributions.—The frequency distribution in the number of dorso ventral files of lenses in the individuals of *Pedinopariops insequens* is presented in figure 5A. Estimations from the 'traditional' and the smoothed (Gaussian KDE) histograms result in a multimodal distribution; at least two modes can be distinguished (Fig. 5A). Additionally, bivariate plots of lenses versus files number, of cephalic width (tr.) versus cephalic length (sag.) and, of files versus cephalic width (tr.) are respectively presented in figure 5B, 5C, and 5D.

As detected in other phacopids (Crônier and Clarkson 2001; Crônier and Fortey 2006), the scatter plot of cephalic width (tr.) versus cephalic length (sag.) shows a linear growth for all considered individuals. Within the population of *Pedinopariops insequens*, the relative proportions of cephala remain constant $(y = ax + b, r = 0.9765, p < 0.001)$ whatever the degree of development of individuals (Fig. 5C).

However, two major groups can be distinguished according to their lens file number: a first group 1 of generally smaller individuals where the lowest values are nine, with the maximum values at 13; a second group 2 where the lowest values are 16, with the maximum values at 21. In each group, the lens file number range is from five to six (Fig. 5D). These higher values correspond well with the diagnosis and the description of adult *P. insequens* by Chlupáč (1977). In addition, a third group is represented by a very small individual with only four files of lenses (Fig. 5A–D).

Within this population, a possible break in the linear dimensions between the individuals with a low lens file number versus a high lens file number seems to exist with a modification of the origin, i.e. a back and a persistence of the same slope (Fig. 5C). In order to test for structural change in these both groups, a Chow test (Chow 1960) which is a variation of the F-test (Fisher 1970) and requires the sum of squared errors from three regressions, has been used to test whether a single regression line or two separate regression lines fit the data best and to detect for equality of the coefficients in these two separate linear regressions. The test indicates a structural break $(F = 43.41, p < 0.001)$. Moreover, no significant difference between the two slopes while a significant difference between the constants exists (slope: $F = 0.2559$, $p = 0.61$NS; constant: $F = 49.86$, $p < 0.0001$).

Analyses of variance performed on the cephalic width and length (Table 2A,B) and on the file and lens number (Table 2C–D) confirm the existence of size heterogeneity and a differentiation among groups previously defined according to their file number (group 1 with nine to 13 lenses; group 2 with 16–20 lenses; group 3 with four lenses). All tests are significant (Table 2A–D).

Thus, the reduction of the lens files number is dependent on body size. Additionally, the coefficient of variation in the number of eye lenses is somewhat more important in the population with 13 lenses or less (CV = 8.80%) than in the population with 16 lenses or more (CV = 2.44%). The comparison of the coefficient of variation by a *F*-max test indicates a significant heterogeneity of variance among groups $(F_{vmax/vmin} = 3.62; ddl_1 = 55; ddl_2 = 19; p = 0.016)$.

Morphological space.—Morphological relationships among individuals can be displayed by their representation in the morphological space (Fig. 5E) defined by the first two principal axes, which provide a valuable approximation of the morphospace (61% of the total variance). The first relative warp explained 37% of the observed variation among individuals, the second 14.5%. Analyses of variance applied on the matrix of individual scores on the first three significant axes indicate a significant morphological differentiation among groups according to RW1
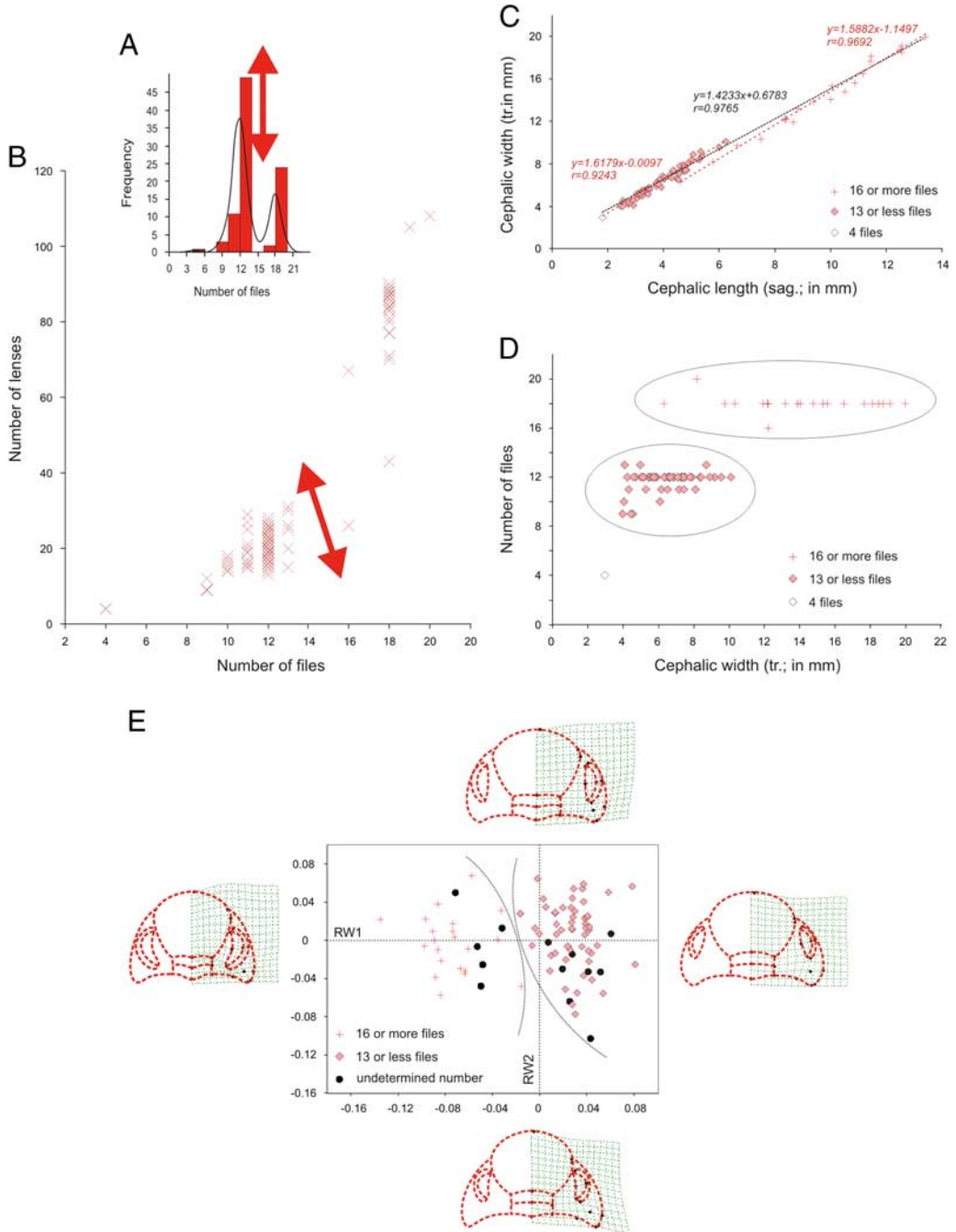
FIGURE 5. A, The frequency distribution of the file number of lenses for all individuals of *Pedinopariops insequens* (Chlupáč, 1977). The 'traditional' histogram and the Kernel density estimate distribution of file number using the optimal Gaussian bin width. The frequency distribution shows two modes, i.e. a bimodality in the file number of lenses; B, scatter plot of lens number versus file number for all individuals; C, scatter plot of cephalic width (tr.) in mm versus cephalic length (tr.) in mm for all individuals showing a discontinuous and a typical isometric distribution for each group previously defined according to their file number (group 1 with nine to 13 lenses; group 2 with 16–20 lenses; group 3 with four lenses); D, scatter plot of file number versus cephalic width (tr.) in mm for all individuals; E, location of 88 cranidia/cephala of *Pedinopariops insequens* (Chlupáč, 1977) in the morphospace defined according to the first two relative warps RW1 and RW2. Axes RW1 and RW2 represent 37% and 14.5%, respectively, of the total variance. Thin plate spline deformation grids depicting patterns of shape change between the both populations of *Pedinopariops insequens* (Chlupáč, 1977). Double arrows in (A) and (B) underline the gap.

TABLE 2. Univariate Analyses of Variance performed (A, B) on cephalic width (in mm) and cephalic length (in mm), on the file and lens number (C, D), on centroid size (E), and on the first three significant axes showing a significant morphological differentiation among groups of *Pedinopariops insequens* (Chlupáč, 1977) according to size, file and lens number and RW1 (F–H).

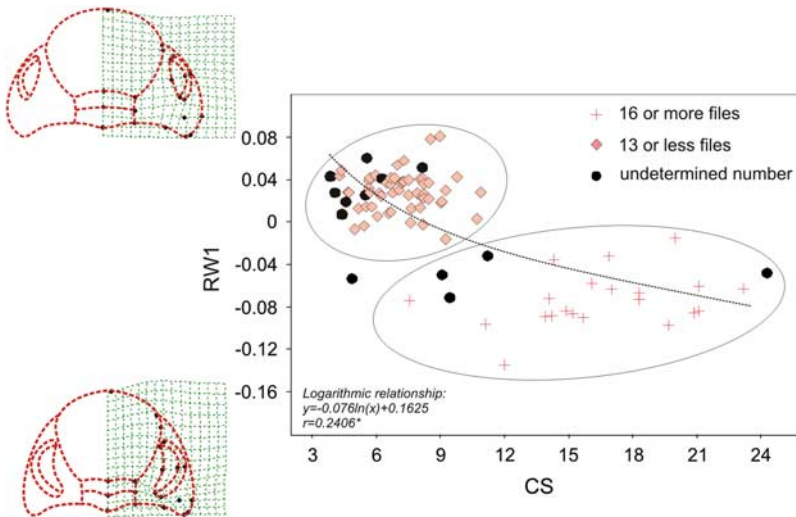| Source of variation	ddl	Sum of squares	Mean square	Fisher F	$P > F$
(A) Width (tr.)	2	954.580	477.290	90.101	$P < 0.0001$
(B) Length (sag.)	2	508.587	254.293	125.481	$P < 0.0001$
(C) Lens number	2	65231.939	32615.970	420.833	$P < 0.0001$
(D) File number	2	802.673	401.336	611.322	$P < 0.0001$
(E) CS	2	1696.345	848.173	168.236	$P < 0.0001$
(F) RW1	2	0.148	0.074	92.043	$P < 0.0001$
(G) RW2	2	0.006	0.003	2.683	$P = 0.074\ NS$
(H) RW3	2	0.005	0.002	4.085	$P = 0.020\ NS$

FIGURE 6. Scatter plot of size (centroid size CS) versus shape (RW1) for 88 individuals of *Pedinopariops insequens* (Chlupáč, 1977). A logarithmic regression suggests a slight relationship ($y = a ln(x) + b$; $p < 0.05$) between size and shape and shows a progressive shape change with size increase.

(Table 2F–H). The set of individuals are grouped together according to their file number in the morphospace along the first axis (Fig. 5E).

The morphological interpretation of the shape changes between individuals from the two major groups is indicated by the TPS warp grids showing the direction of maximum variance (Fig. 5E). The first relative warp axes shows that the main shape change in these both groups contrasts more or less developed eyes: the individuals from group 2 with more developed eyes and the individuals from group 1 with less developed eyes.

The morphological relationships among individuals observed in the morphological space support the same general pattern as the bivariate plot of file number versus cephalic width (tr.), towards more files in individuals from group 2.

Furthermore, a MANOVA performed on the size (CS) and the shape (RW1) suggests that there is a differentiation among groups previously defined according to their file number whereas a logarithmic regression suggests only a slight relationship ($y = a ln(x) + b$; $p < 0.05$) between size and shape (Fig. 6).

Discussion and conclusions

According to the number of files/lenses and the shape change, a bimodal distribution seems to differentiate individual populations of both studied species, *Prokops prokopi* and *Pedinopariops insequens*.

Changes observed in *Prokops prokopi* may correspond to changes in which extreme values for the number of files or lenses are favored over intermediate values. The variance of the feature increases and the population is then divided into two distinct groups. This process is known as disruptive selection, also called diversifying selection. This evolutionary process has long been viewed as important in increasing variation within natural populations (Rueffler et al. 2006) and initiating sympatric speciation (Maynard Smith 1962; Dieckmann and Doebeli 1999). This disruptive selection arises from intense intraspecific competition (Martin and Pfenning 2012).

Prokops prokopi is confined approximately to the upper half to the uppermost Dvorce-Prokop Limestone (see Chlupáč 1977; Vaněk 1999), which is mostly represented by the subfacies of Prokop Limestone (thin bedded limestone but with supposed mud mound structures). *Prokops hoeninghausi* occurs especially in the lower part of the formation (Dvorce Limestone facies with thick, massive beds) but it also co occurs with *P. prokopi* on both studied localities (see also Vaněk 1999). Both populations of *Prokops prokopi* come from two geographically close areas in the vicinity of Prague. There are two possible explanations for the observed bimodalities:

1. The population from Malá Chuchle is slightly older than the population from the St. Prokop's Quarry (see localities 1 and 2 in Fig. 2). In Malá Chuchle, approximately middle levels to upper third of Praha Formation are exposed (see Chlupáč 1957), most probably still within the upper part of *Nowakia acuaria* Dacryoconarid Biozone. The St. Prokop's quarry population, on the other hand, comes from the upper part of the Praha Formation (see Chlupáč 1957; Kříž 1999), corresponding to the *Guerichina strangulata* Dacryoconarid Biozone (correlation based on the occurrence of *Guerichina infindibulum* (Richter 1854), P. Lukeš personal communication 2015). We thus cannot exclude the possibility of some microevolutionary shift between these two populations.
2. In the St. Prokop's quarry, a supposed local mud mound structure characterized by a lens like body and by the thick bedded limestones with common fenestrate (Stromatactis) structure has been ascertained (see Chlupáč 1983: pp. 50; Chlupáč 1993; Galle and Parsley 2005); this structure was regarded as a reef by earlier authors. Disarticulated trilobite exoskeletons often form accumulations of lens like bodies with micritic matrix. It is, however, probable that similar mud mound structure existed also at Malá Chuchle (see Chlupáč 1983). The mud mound structures present a slightly different environment in comparison with the mostly muddy sea floor occurring in the NE part of the Prague Basin during middle–upper Pragian. The mud mound can offer a richer source of food and, especially caverns and cavities suitable for inhabiting. One can only speculate that some restricted populations of *P. prokopi* became adapted to such a protected environment as with other trilobite taxa in African Gondwana, Baltica and European Gondwana (Kaufmann 1997, 1998; Cavalazzi 2006; Popp and Pärnaste 2011; Bignon and Cronier 2014). The water depth at both localities was probably comparable (the muddy sea floor in the deeper shelf lay below the fair weather wave base, possibly also below the storm wave base but still within the photic zone, for discussion, see also Chlupáč et al. 1998). *Prokops prokopi* occurs at both localities together with diverse trilobites with well developed eyes (*Reedops cephalotes* Hawle et Corda, 1847; *Reedops bronni* Barrande, 1846; *Odontochile tenuigranulata* Budil et al., 2009; *Zlichovaspis* (Z.) *rugosa* Hawle et Corda, 1847etc.); trilobite associations established at both localities are very similar. It is interesting, that following Chlupáč (1983), the trilobite association at both localities contain more common scutelluids than established in typical *Reedops Odontochile* Assemblage.

The bimodal population of *Prokops prokopi* thus shows several signatures of ecologically maintained adaptive divergence and reproductive isolation. The two modes differ in eye size and visual capacities, presumably owing to specialization (adaptation?) on different photic conditions.

Here we demonstrate that bimodality might also be maintained by a selective loss of trilobites with intermediate eye sizes, i.e. disruptive selection between the modes (Fig. 4). Such a disruptive selection could be expected to be important during the radiation of phacopid trilobites.

What are the possible causes for the documented disruptive selection here? One possibility is that competition for shared habitats reduces the success of individuals specializing on intermediate habitats, causing them to die or emigrate. Another possibility is that intermediate phacopids fall into a gap in the underlying habitat distribution (i.e. independent of competition). Both of these mechanisms can cause a disruptive selection in theoretical models (Schluter 2000; Rueffler et al. 2006; Doebeli et al. 2007; Abrams et al. 2008) and could be implicated in the divergence of these phacopid trilobites. These considerations underline the importance of determining the conditions under which adaptive radiation can or cannot result by extrapolating processes observed within species.

In *Pedinopariops insequens*, the bimodal distribution reflects individuals from only one locality but rather two size classes. In a population, extreme values of the number of lenses and files can to be favored leading to an increase in the variance of these traits and to divide the population into two distinct groups. Could a disruptive selection cause an adaptive divergence between conspecific groups occupying different environments or using different resources?

For *Pedinopariops insequens*, there are two clear modes in the number of lenses and files of lenses (Figs. 5A, 7C) and there may be three modes in the cephalic width and length across the size range (Figs. 7A,B).

The first size mode corresponds to individuals with 13 or less than 13 files of lenses. The two following modes of size correspond to individuals with 16 or more than 16 files of lenses. It seems that there is a discontinuity in the establishment of visual lenses between the first and the second mode size. We prefer to maintain the validity of this species but have to admit its notable and surprising intraspecific variability. There are several explanations of this phenomenon:
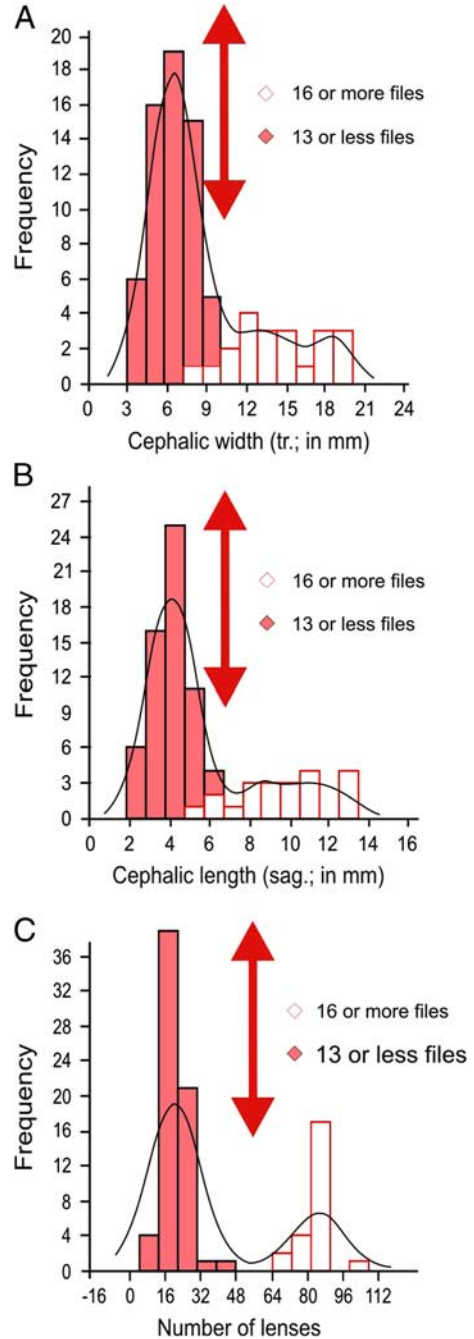

FIGURE 7. The frequency distribution of (A) the cephalic width (in mm), of (B) the cephalic length (in mm) and of (C) the number of lenses for all individuals of *Pedinopariops insequens* (Chlupáč, 1977). The 'traditional' histogram and the Kernel density estimate distribution using the optimal Gaussian bin width. The frequency distribution shows two clear major modes, i.e. a bimodality in the number of lenses and probably two modes in the cephalic length and width. Double arrows underline the gap.

1. We can confirm the relative homogeneity of the studied material. It comes from one locality and distinct horizon at Praha Holyně (for details, see above). We thus can eliminate any collecting bias or misidentification of the samples, perhaps with the exception of hypothetical mixing of different layers within the uppermost Třebotov Limestone and the lowermost Choteč Limestone. Such unlikely mixing, in our opinion, cannot cause the observed discontinuity in establishment of the visual surface during ontogenetic development but can explain well cephalic width/length differences.

2. It is noteworthy that the population comes from the uppermost part of the Třebotov Limestone, situated very close (possibly above) to the Lower/Middle Devonian boundary. Very slightly later, the Basal Choteč Event took place, representing quite important overturn in the trilobite communities worldwide (see Chlupáč and Kukal 1988; Chlupáč 1983, 1994; Chlupáč et al. 2000). The polymodality within a single population, as discussed, can be explained by a compensation of ecological stress caused by the onset of unfavorable conditions. It is noteworthy that *Pedinopariops insequens* is the only phacopid that crosses the Lower/Middle Devonian boundary and survived the Basal Choteč Event in the Prague Basin. It was apparently able to cope with extinctions and faunal changes accompanying this event. Perhaps, the stress conditions could cause changes of the ontogenetic trajectory and a certain type of the disruptive selection, possibly even with a short time acceleration of ontogenetic changes.

3. We have to admit the possibility that two or more species are hidden inside the traditional species of *Pedinopariops insequens*, especially the "large" and "small eyed" forms. One such form, however, lack adult specimens. Such an alternative explanation is, in our opinion, less probable.

Acknowledgments

Literature Cited

Aberhan, M., S. Nürnberg, and W. Kiessling. 2012. Vision and the diversification of Phanerozoic marine invertebrates. Paleobiology 38:187–204.

Abrams, P. A., C. Rueffler, and G. Kim. 2008. Determinants of the strength of disruptive and/or divergent selection arising from resource competition. Evolution 62:1571–1586.

Adams, D. C., F. J. Rohlf, and D. E. Slice. 2004. Geometric morphometrics: ten years of progress following the 'revolution'. Italian Journal of Zoology 71:5–16.

Barrande, J. 1846. Notice préliminaire sur le Système silurien et les trilobites de Bohême. Hirschfeld, Leipzig.

——. 1852. Système silurien du centre de la Bohême. Ière partie: Recherches Paléontologiques. I. Crustacés: Trilobites. Prague, Paris.

Berkyová, S. 2009. Lower-Middle Devonian (upper Emsian-Eifelian, *serotinus-kockelianus* zones) conodont faunas from the Prague Basin, the Czech Republic. Bulletin of Geosciences 84:667–686.

Bignon, A., and C. Crônier. 2014. Trilobite faunal dynamics on the Devonian continental shelves of the Ardenne Massif and Boulonnais (France, Belgium). Acta Palaeontologica Polonica. (doi: org/10.4202/app.00019.2013.

Bookstein, F. L. 1991. Morphometric tools for landmark data: geometry and biology. Cambridge University Press, Cambridge.

Bouček, B. 1931. O některých nových bohatých nalezištích zkamenělin ve starším paleozoiku středních Čech. Věda přírodní 12:136–144.

Brayard, A., H. Bucher, G. Escarguel, F. Fluteau, S. Bourquin, and T. Galfetti. 2006. The Early Triassic ammonoid recovery: paleoclimatic significance of diversity gradients. Palaeogeography, Palaeoclimatology, Palaeoecology 239:374–395.

Budil, P. 1999. Some comments on the genus *Ormathops* Delo from the Bohemian Ordovician. Acta Universitatis Carolinae, Geologica 43:373–376.

Budil, P., F. Hörbinger, and R. Mencl. 2009. Lower Devonian dalmanitid trilobites of the Prague Basin (Czech Republic). Earth and Environmental Science Transactions of the Royal Society of Edinburgh, Earth Sciences 99:61–100.

Carls, P., L. Slavík, and J. I. Valenzuela-Ríos. 2008. Comments on the GSSP for the basal Emsian stage boundary: the need for its redefinition. Bulletin of Geosciences 83:383–390.

Cavalazzi, B. 2006. Kess Kess carbonate mounds, Hamar Laghdad, Tafilalt, Anti-Atlas, SE Morocco – A Field Guide, Morocco. UNESCO Field Action, 01–05 December 2006, p. 20.

Chlupáč, I. 1957. Faciální vývoj a biostratigrafie středočeského spodního devonu (Facial development and biostratigraphy of the Lower Devonian of Central Bohemia). Sborník Ústředního ústavu geologického, oddíl geologický 23:369–448.

——. 1959. Faciální vývoj a biostratigrafie břidlic dalejských a vápenců hlubočepských (eifel) ve středočeském devonu (Facial development and biostratigraphy of the Daleje Shale and the Hlubočepy Limestone (Eifelian) in the Devonian of Central Bohemia). Sborník Ústředního ústavu geologického, oddělení geologické 25:446–511.

——. 1971. New phacopid trilobites from the Devonian of Czechoslovakia. Časopis pro mineralogii a geologii 16:255–261.

——. 1977. The phacopid trilobites of the Silurian and Devonian of Czechoslovakia. Rozpravy Ústředního ústavu geologického 43:1–172.

——. 1982. Preliminary submission for Lower-Middle Devonian boundary stratotype in the Barrandian area. Courier Forschungsinstitut Senckenberg 55:85–96.

——. 1983. Trilobite assemblages in the Devonian of the Barrandian area and their relations to palaeoenvironments. Geologica et Palaeontologica 17:45–73.

——. 1985. Comments of the Lower-Middle Devonian boundary. Courier Forschungsinstitut Senckenberg 75:389–400.

——. 1988. Geologické zajímavosti pražského okolí. Academia, Praha (in Czech).

——. 1993. Geology of the Barrandian. A field trip guide. Senckenberg-Buch 69. Verlag Waldemar Kramer, Frankfurt am Main.

——. 1994. Devonian trilobites – Evolution and events. Geobios 27:487–505.

——. 1999. Vycházky za geologickou minulostí pražského okolí, (second edition). Academia, Praha in Czech.

Chlupáč, I., R. Feist, and P. Morzadec. 2000. Trilobites and standard Devonian stage boundaries. Courier Forschungsinstitut Senckenberg 220:87–98.

Chlupáč, I., P. Lukeš, and J. Zikmundová. 1979. The Lower-Middle Devonian boundary beds in the Barrandian area, Czechoslovakia. Geologica et Palaeontologica 13:125–156.

Chlupáč, I., V. Havlíček, J. Kříž, Z. Kukal, and P. Štorch. 1998. Palaeozoic of the Barrandian (Cambrian to Devonian). Czech Geological Survey, Prague.

Chlupáč, I., and Z. Kukal. 1988. Possible global events and the stratigraphy of the Palaeozoic of the Barrandian (Cambrian-Middle Devonian, Czechoslovakia). Sborník geologických věd, Geologie 43:83–146.

Chow, G. C. 1960. Tests of equality between sets of coefficients in two linear regressions. Econometrica 28:591–605.

Clarkson, E. N. K. 1966a. Schizochroal eyes and vision in some Silurian acastid trilobites. Palaeontology 9:1–29.

——. 1966b. Schizochroal eyes and vision in some phacopid trilobites. Palaeontology 9:464–487.

——. 1971. On the early schizochroal eyes of Ormathops (Trilobita: Zeliszkellinae). Mémoires du Bureau des Recherches Géologiques et Minières 73:51–63.

——. 1979. The visual system of trilobites. Palaeontology 22:1–22.

Clarkson, E. N. K., and R. P. Tripp. 1982. The Ordovician trilobites Calyptaulax brongniartii (Portlock). Transactions of the Royal Society of Edinburgh, Earth Sciences 72:287–294.

Clarkson, E. N. K, R. Levi-Setti, and G. Horvath. 2006. The eyes of trilobites: the oldest preserved visual system. Arthropod Structure Development 35:247–259.

Crônier, C., and E. N. K. Clarkson. 2001. Variation of eye-lens distribution in a new Late Devonian phacopid trilobite. Transactions of the Royal Society of Edinburgh 92:103–113.

Crônier, C., and R. A. Fortey. 2006. Morphology and ontogeny of an Early Devonian Phacopid trilobite with reduced sight from southern Thailand. Journal of Paleontology 80:529–536.

Crônier, C., R. Feist, and J.-C. Auffray. 2004. Variation in the eye of Acuticryphops (Phacopina, Trilobita) and its evolutionary significance: a biometric and morphometric approach. Paleobiology 30:470–480.

Darwin, C., and A. R. Wallace. 1958. Evolution by natural selection: a centenary commemorative volume. Cambridge University Press, Cambridge.

Dieckmann, U., and M. Doebeli. 1999. On the origin of species by sympatric speciation. Nature 400:354–357.

De Baets, K., C. Klug, and C. Monnet. 2012. Intraspecific variability through ontogeny in early ammonoids. Paleobiology 39:75–94.

Doebeli, M., H. J. Blok, O. Leimar, and U. Dieckmann. 2007. Multimodal pattern formation in phenotype distributions of sexual populations. Proceedings of the Royal Society B 274:347–357.

Feist, R., K. J. McNamara, C. Crônier, and R. Lerosey-Aubril. 2009. Patterns of extinction and recovery of phacopid trilobites during the Frasnian-Famennian (Late Devonian) mass extinction event, Canning Basin, Western Australia. Geological Magazine 146: 12–33.

Fisher, F. M. 1970. Tests of equality between sets of coefficients in two linear regressions: An expository note. Econometrica 38: 361–366.

Frankham, R., J. D. Ballou, and D. A. Briscoe. 2002. Introduction to Conservation Genetics. Cambridge: Cambridge University Press.

Frýda, J., L. Ferrová, and B. Frýdová. 2013. Review of palaeozygopleurid gastropods (Palaeozygopleuridae, Gastropoda) from Devonian strata of the Perunica microplate (Bohemia), with a re-evaluation of their stratigraphic distribution, notes on their ontogeny, and descriptions of new taxa. Zootaxa 3669:469–489.

Galle, A., and R. L. Parsley. 2005. Epibiont relationships on hyolithids demonstrated by Ordovician trepostomes (Bryozoa) and Devonian tabulates (Anthozoa). Bulletin of Geosciences 80:125–138.

Gradstein, F. M., J. G. Ogg, M. Schmitz, and G. Ogg. 2012. The Geologic Time Scale 2012. Elsevier.

Hallgrímsson, B., and B. K. Hall. 2005. Variation: a central concept in biology. Elsevier, Amsterdam.

Hammer, Ø., D. A. T. Harper, and P. D. Ryan. 2001. PAST: Paleontological Statistics Software Package for Education and Data Analysis. Palaeontologia Electronica 4:1–9.

Hammer, Ø., and D. A. T. Harper. 2006. Paleontological Data Analysis. Blackwell.

Harzsch, S., and G. Hafner. 2006. Evolution of eye development in arthropods: phylogenetic aspects. Arthropod Structure & Development 35:319–340.

Havlíček, V., and J. Vaněk. 1998. Pragian brachiopods, trilobites, and principal biofacies in the Prague Basin (Lower Devonian, Bohemia). Sborník geologických věd, Řada P, Paleontologie 34:27–109.

Hawle, I., and A. J. C. Corda. 1847. Prodrom einer Monographie der böhmischen Trilobiten. J.G. Calve, Prague. Reprint, 1848. Abhandlungen der königlichen böhmischen Gesellschaft der Wissenschaften 5:117–292.

Hunt, G. 2007. Variation and early evolution. Science 317:459–460.

Kaufmann, B. 1997. Middle Devonian reef and mud mounds on a carbonate ramp: Mader Basin (eastern Anti-Atlas, Morocco). Geological Society, London, Special Publications 149:417–435.

——. 1998. Facies, stratigraphy and diagenesis of Middle Devonian reef- and mud-mounds in the Mader (eastern Anti-Atlas, Morocco). Acta Geologica Polonica 48:43–106.

Klapper, G., and S. Vodrážková. 2013. Ontogenetic and intraspecific variation in the late Emsian-Eifelian (Devonian) conodonts Polygnathus serotinus and P. bultyncki in the Prague Basin (Czech Republic) and Nevada (western U.S.). Acta Geologica Polonica 63:153–174.

Klingenberg, C. P., and G. S. McIntyre. 1998. Geometric morphometrics of developmental instability: analyzing patterns of fluctuating asymmetry with Procrustes methods. Evolution 52:1363–1375.

Kristjánsson, B. K., S. Skúlason, and D. L. G. Noakes. 2002. Morphological segregation of Icelandic threespine stickleback

(*Gasterosteus aculeatus* L). Biological Journal of the Linnean Society 76:247–257.

Kříž, J. 1999. Geologické památky Prahy. Český geologický ústav, Praha.

Levi-Setti, R., E. N. K. Clarkson, and G. Horwáth. 1998). Paleontologia dell'occhio. Pp. 365–379 in Frontiere della Vita: Enciclopedia Italiana.

Lomax, R. G. 2007. Statistical Concepts: A Second Course for Education and the Behavioral Sciences, (3rd edition). Lawrence Erlbaum Associates, Mahwah, New Jersey.

Mann, H. B., and D. R. Whitney. 1947. On a Test of Whether one of Two Random Variables is Stochastically Larger than the Other. Annals of Mathematical Statistics 18:50–60.

Månsson, K., and E. N. K. Clarkson. 2012. Ontogeny of the Upper Cambrian (Furongian) olenid trilobite *Protopeltura aciculata* (Angelin, 1854) from Skåne and Våstergötland, Sweden. Palaeontology 55:887–901.

Martin, R. A., and D.W. Pfenning. 2012. Widespread disruptive selection in the wild is associated with intense resource competition. BMC. Evolutionary Biology 12:136–149.

Maynard Smith, J. 1962. Disruptive selection, polymorphism and sympatric speciation. Nature 195:60–62.

Nilsson, D. E., and A. Kelber. 2007. A functional analysis of compound eye evolution. Arthropod Structure and Development 36:373–385.

Oakley, T. H. 2003. On Homology of Arthropod Compound Eyes. Integrative and Comparative Biology 43:522–530.

Perner, J. 1918. Trilobiti pasma D-d1, z okoli pražského. Palaeontographica Bohemiae 9:1–51.

Popp, A., and H. Pärnaste. 2011. Biometry and life style of the Ordovician proetide trilobite *Cyamella stensioei* Owens, 1979. GFF 133:111–123.

Ramel, C. 1998. Biodiversity and intraspecifiic genetic variation. Pure and Applied Chemistry 70:2079–2084.

Richter, R. 1854. Thüringische Tentaculiten. Zeitschrift der Deutschen Geologischen Gesellschaft 6:275–290.

Rohlf, F. J. 1993. Relative warps analysis and an example of its application to mosquito wings. Pp. 131–159 in L. F. Marcus, E. Bello, and A. Garcia-Valdecasas, eds. Contributions to morphometrics. Madrid, Museu Nacional de Ciencias Naturales.

——. 2003a. TpsSuper. Version 1.06. Department of Ecology and Evolution, State University of New York, Stony Brook, NY.

——. 2003b. TpsRelw. Version 1.21. Department of Ecology and Evolution, State University of New York, Stony Brook, NY.

Rohlf, F. J., and D. E. Slice. 1990. Extensions of the Procrustes methods for the optimal superimposition of landmarks. Systematic Zoology 39:49–50.

Rueffler, C., T. , J. M. Van Dooren, O. Leimar, and P. A. Abrams. 2006. Disruptive selection and then what? Trends in Ecology and Evolution 21:238–245.

Rundle, H. D., and P. Nosil. 2005. Ecological speciation. Ecology Letters 8:336–352.

Růžička, R. 1940. Faunistické seznamy z Barrandienu ze souvrství gα v okolí pražském. Věstník Královské české společnosti nauk, Třída mathematicko přírodovědná, 1–25.

——. 1941. Faunistické seznamy z Barrandienu ze souvrství gα (část II) a ze dvou lokalit gγ v okolí pražském. Věstník Královské české společnosti nauk, Třída mathematicko přírodovědná, 1–16.

Salgado-Ugarte, I. H., M. Shimizu, and T. Taniuchi. 1994. Exploring the shape of univariate data using Kernel density estimators. Stata Technical Bulletin 16:8–19.

Salgado-Ugarte, I. H., M. Shimizu, T. Taniuchi, and K. Matsushita. 2002. Nonparametric Assessment of Multimodality for Size Frequency Distributions. Asian Fisheries Science 15:295–303.

Schluter, D. 2000. The Ecology of Adaptive Radiation. New York: Oxford University Press.

Silverman, B. W. 1986. Density estimation for statistics and data analysis. Chapman & Hall.

Skúlason, S., and T. B. Smith. 1995. Resource polymorphisms in vertebrates. Trends in Ecology & Evolution 10:366–370.

Slavík, L. 2004a. A new conodont zonation of the Pragian Stage (Lower Devonian) in the Stratotype area (Barrandian, Central Bohemia). Newsletters on Stratigraphy 40:39–71.

——. 2004b. The Pragian-Emsian conodont successions of the Barrandian area: search of an alternative to the GSSP polygnathid-based. Geobios 37:454–470.

Slavík, L., J. I. Valenzuaela-Ríos, J. Hladil, and P. Carls. 2007. Early Pragian conodont-based correlations between the Barrandian area and the Spanish Central Pyrenees. Geological Journal 42:499–512.

Suchý, V. 2002. The "white beds" – a fossil caliche of the Barrandian area: its origin and paleoenvironmental significance. Journal of the Czech Geological Society 47:45–54.

Thomas, A. T. 1998. Variation in the eyes of the Silurian trilobites *Eophacops* and *Acaste* and its significance. Palaeontology 41: 897–911.

Urdy, S., N. Goudemand, H. Bucher, and R. Chirat. 2010. Growth dependent phenotypic variation of molluscan shell shape: implications for allometric data interpretation. Journal of Experimental Zoology Part B 314:303–326.

Vaněk, J. 1999. Pražský stupeň (spodní devon) v Pražské pánvi a relativní stáří jeho faciií (Česká republika). Part 2. New taxa of trilobites from Pragian, Bohemia. Palaeontologia Bohemiae 5:39–67.

Vodrážková, S., J. Frýda, T. J. Suttner, L. Koptíková, and P. Tonarová. 2013. Environmental changes close to the Lower–Middle Devonian boundary; the Basal Chotec Event in the Prague Basin (Czech Republic). Facies 59:425–449.

Woldřich, J. 1919. Das Prokopital südlich von Prag. Jahrbuch der k.k. geologischen Reichsanstalt 68:63–112.

Waloszek, D., J.-Y. Chen, A. Maas, and X.-G. Wang. 2005. Early Cambrian arthropods - new insights into arthropod head and structural evolution. Arthropod Structure and Development 34:189–205.