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**ORIGINAL RESEARCH PAPER in TAXONOMY AND PHYTOGEOGRAPHY**

# Identification of New Taxa of *Portulaca oleracea* L. Aggregate From Poland Based on Seed Coat Micromorphological Characteristics

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## Abstract

This paper presents the results of micromorphological studies of *Portulaca oleracea* aggregate seeds deposited at the Herbarium of the Department of Systematic and Environmental Botany (POZ), Adam Mickiewicz University in Poznań, Poland, based on scanning electron microscopy. The seed surface ultrastructure was studied and the following seven morphotypes (species) were noted: *P. daninii*, *P. granulatostellulata*, *P. macrantha*, *P. nitida*, *P. oleracea* s. str., *P. papillatostellulata*, and *P. trituberculata*. New alien taxa for the flora of Poland, *P. daninii*, *P. macrantha*, and *P. papillatostellulata*, were recorded. The original photos of the seed surface ultrastructure of the studied morphotypes are included.

## Keywords

alien species; Poland; *Portulaca*; Portulacaceae; POZ; seed surface ultrastructure; Wielkopolska region

## 1. Introduction

*Portulaca* L. (Portulacaceae Juss.) comprises over 100 species that are distributed worldwide (Mabberley, 2017; *Plants of the World Online*, 2022). Presently, this genus forms its own family (The Angiosperm Phylogeny Group, 2016; Nyffeler & Eggli, 2010), but in older classification systems, it has been combined with many other genera into a widely treated family of about 450 species (Carolin, 1993; Takhtajan, 2009). This genus occurs mainly in tropical and subtropical regions, particularly in Africa, South America, and the western regions of North America (Carolin, 1993; Takhtajan, 2009). However, the genus *Portulaca* is poorly represented in Eurasia.

In Poland, depending on the taxonomical point of view, this genus is represented by three or four species (Broda & Mowszowicz, 2000; Mirek et al., 2020; Rutkowski, 2011; Stengl-Rejthar, 1992; Szafer et al., 1988): *P. oleracea* L. (= *P. oleracea* L. subsp. *oleracea*), *P. sativa* Haw. [= *P. oleracea* L. subsp. *sativa* (Haw.) Čelak], *P. grandiflora* Hooker, and *P. umbraticola* Kunth. In recent studies, *P. oleracea* and *P. sativa* have been recognized as subspecies (subsp. *oleracea* and subsp. *sativa*, respectively) of *P. oleracea* (among others, Mirek et al., 2020; Rutkowski, 2011). *Portulaca oleracea* L. subsp. *oleracea* is an accidentally introduced and naturalized anthropophyte (kenophyte) (Mirek et al., 2020; Tokarska-Guzik et al., 2012). It occurs mainly in central-western (Wielkopolska) and south-western (Śląsk) parts of Poland (Zajac &

Zajac, 2001), and is found in ruderal (among others, flower beds, roadsides, courtyards, garbage dumps, lawns, and railway grounds) and segetal (gardens, fields – mostly among root crops; sometimes as a troublesome weed) habitats (Mirek et al., 2020; Mowszowicz, 1986; Rutkowski, 2011; Stengl-Rejthar, 1992; Szafer et al., 1988; Tokarska-Guzik et al., 2012). *Portulaca oleracea* subsp. *sativa* has been reported from Poland only as a species cultivated as a vegetable (Broda & Mowszowicz, 2000; Mirek et al., 2020; Rutkowski, 2011; Stengl-Rejthar, 1992; Szafer et al., 1988).

*Portulaca grandiflora* Hook. was placed on a list of Polish ergasioephemeroophytes (Rostański & Sowa, 1986–1987). This species was reported from Poland only once in 1932, from a rubble heap in Legnica (western Poland), as a possible escapee from cultivation (Schalow, 1933). It is cultivated in Poland as an ornamental plant (Rutkowski, 2011). *Portulaca umbraticola* has been reported from Poland only as a crop plant (“*Portulaca umbraticola*,” 2022).

The common purslane (*Portulaca oleracea* L.) is an interesting object of research in many different areas. In agricultural and ecological sciences, it has attracted attention as a common weed and invasive species in many European countries (among others, Protopopova & Shevera, 2019; Tokarska-Guzik, 2005; Tokarska-Guzik et al., 2012). In pharmaceutical sciences, it is a source of vitamins and antioxidants (among others, Liu et al., 2000; Oliveira et al., 2009; Simopoulos, 2004; Simopoulos et al., 1995; Simopoulos & Salem, 1986; Uddin et al., 2012, 2014). The genus *Portulaca* is also an interesting object of taxonomical studies, mostly due to its polyploidy (particularly within the *Portulaca oleracea* aggr.).

Recently, this aggregate has become an object of thorough investigation, started by Danin et al. (1978). It was found that it is a heterogeneous polyploid complex consisting of at least 9–19 microspecies or, according to other terminology, morphotypes (mainly autogamic) (among others, Danin & Anderson, 1986; Danin et al., 1978, 2008, 2012, 2014, 2016; Danin & Reyes-Betancort, 2006; Danin in Greuter & Raus, 2012, p. 291; Danin in Greuter & von Raab-Straube, 2011, pp. 131–134; Soltis & Soltis, 1999; Walter et al., 2015). The results of these studies and the narrow interpretation of taxa have stimulated further research on *Portulaca oleracea* aggr. in different countries and, consequently, led to the discovery of new taxa (among others, Bulakh et al., 2019; Domina & Raimondo, 2009; Feráková et al., 2012; Danin in Greuter & Raus, 2012, p. 291; Danin in Greuter & von Raab-Straube, 2011, pp. 131–134; Matthews et al., 1993; Rad et al., 2017; Rupesh et al., 2015).

The morphological features of seeds are important traits that enable differentiation between newly separated taxa. Based on the seed size and morphology of epidermal cells of testa, descriptions of new taxa and keys to their identification were prepared (Danin et al., 2008; Ocampo, 2013). Furthermore, Danin (in Greuter & von Raab-Straube, 2011, pp. 131–134), based on the analyzed material of *P. oleracea* s. l. from Poland (KOR), found several new Polish morphotypes (species): *P. granulostellulata* (Poelln.) Ricceri & Arrigoni, *P. nitida* (Danin & HG Baker) Ricceri & Arrigoni, *P. trituberculata* Danin, Domina & Raimondo and *P. oleracea* L. s. str. All of these taxa are of Mediterranean origin.

The aim of this study was to analyze the seed structure of herbarium specimens of *P. oleracea* agg. deposited in the Herbarium of the Department of Systematic and Environmental Botany of Adam Mickiewicz University in Poznań (POZ; <https://amunatcoll.pl/>), to identify new Polish taxa in the studied collection.

## 2. Material and Methods

Herbarium specimens of *P. oleracea* aggr. from the POZ Herbarium of the Department of Systematic and Environmental Botany (formerly, Department of Plant Taxonomy) of Adam Mickiewicz University in Poznań were used in the present study (Table 1; <https://amunatcoll.pl/>).

In total, 14 seed samples (with 20 seeds each) were studied. The methodology used in this study follows that proposed by Danin et al. (1978, 2008, 2012), Danin and Raus (2012), Danin and Reyes-Betancort (2006), and Ocampo (2013), with modifications. The seed morphology, for example, the shape and peculiarity of the

**Table 1** Herbarium specimens of *Portulaca oleracea* aggr. deposited in the POZ Herbarium.

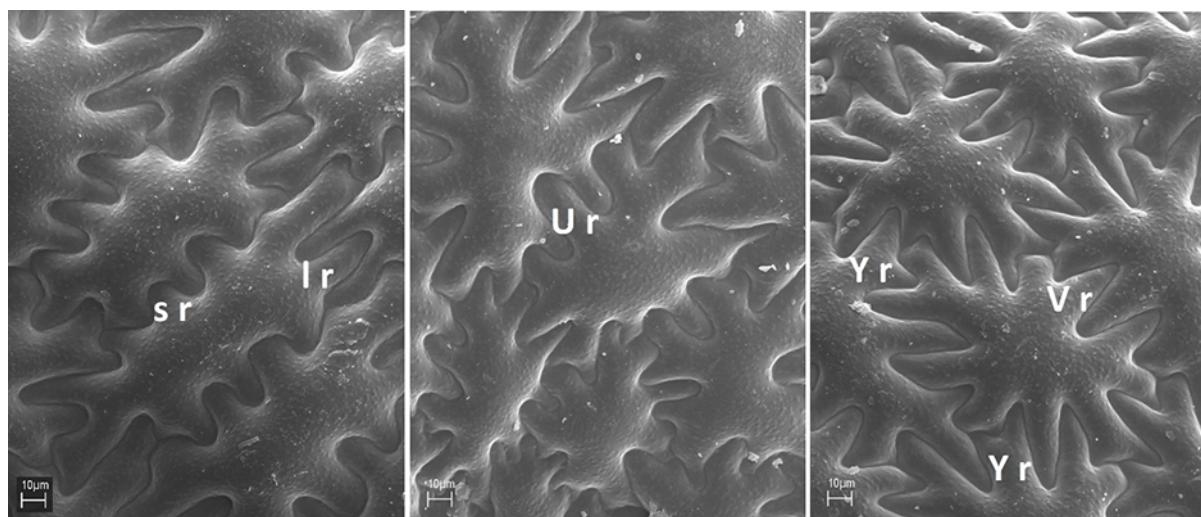
Species name on a herbarium label	Locality	Habitat	Sheet No.	Date of collection	Author of collection
<i>Portulaca oleracea</i> L.	Konin, Wielkopolska region	By the bus station wall	POZ-V-00075781	1973-07	Adamiak H.
<i>Portulaca oleracea</i> L.	Będów near Krosno Odrzańskie, Lubuskie region	Odra River alluvia	POZ-V-00075786	1976-08-27	Latowski K., Żukowski W.
<i>Portulaca oleracea</i> L.	Stare Dymaczewo, Wielkopolska National Park, Wielkopolska region	Roadside	POZ-V-00075777	1977-08-23	Mikołajczak E.
<i>Portulaca oleracea</i> L.	Railway station in Damasławek, Wielkopolska region	Trans-shipment siding track	POZ-V-00075780	1975-09-30	Latowski K.
<i>Portulaca oleracea</i> L.	Railway station in Książ Wlkp., Wielkopolska region	Between stone cobbles	POZ-V-00075775	1988-09-28	Czarna A.
<i>Portulaca oleracea</i> L.	Międzychód, Wielkopolska region	Gravel by buffer stop of siding track, near railway station	POZ-V-00075783	1970-09-30	Hantz J.
<i>Portulaca oleracea</i> L.	Środa Wlkp., Wielkopolska region	Siding track by ramp, opposite of railway station	POZ-V-00075784	1970-08-02	Hantz J.
<i>Portulaca oleracea</i> L.	Bielany, Toruń, Kujawsko-Pomorskie region	On sandy and gravelly ground by the playing-field	POZ-V-00075785	1979-09-20	Ceynowa-Gieldon M.
<i>Portulaca oleracea</i> L.	Czapury near Poznań, Wielkopolska region	Near compost heap along Warta River	POZ-C-0000826	2011-09-16	Stachnowicz W.
<i>Portulaca oleracea</i> L.	Wschowa, Wielkopolska region	Railway station at unused ramp, near station building	POZ-C-0000978	2000-09-01	Latowski K.
<i>Portulaca oleracea</i> L.	Krzyż, Wielkopolska region	Railway station, along the wall of station building on platform	POZ-C-0003211	1997-08-27	Latowski K.
<i>Portulaca oleracea</i> L.	Wschowa, railway station, Wielkopolska region	At unused reloading ramp	POZ-C-0003321	2000-09-01	Latowski K.
<i>Portulaca oleracea</i> L.	Closed railway line Czempiń-Śrem. Szymanowo, Wielkopolska region	On railway track	POZ-C-0003323	2003-07-20	Nowak K.
<i>Portulaca oleracea</i> L.	Leszno, railway station, Wielkopolska region	Along the wall on platform	POZ-C-0003385	1997-09-09	Latowski K.

surface, was studied using a scanning electron microscope (SEM) at magnifications of  $\times 100$ ,  $\times 200$ ,  $\times 400$ ,  $\times 800$ , and  $\times 1,000$ . For the scanning electron microscope (SEM, JSM-6060LA), the seeds were fixed on the brass table, then, the samples were sputter-coated with gold according to the standard method used at the Center of Electron Microscopy of the M. G. Kholodny Institute of Botany, NAS of Ukraine. For size determination, 20 measurements were taken along the polar and equatorial axes for each specimen using AxioVision Rel.4.8 program.

The following seed traits were analyzed: shape, size and diameter, shape of the testa cell rays (Vr, Ur, Yr) (Figure 1), and shape of the periclinal walls (PCW). The terminology of Danin et al. (1978, 2012) was used to describe the seed surface ultrastructure of the morphotypes and the key for its determination (Figure 1). For the identification of taxa, a seed diameter limit (0.85 mm) was used, according to which the groups or separate morphotypes were differentiated. The nomenclature of the studied taxa follows *Plants of the World Online* (2022).

### 3. Results

Seed morphology analysis in *Portulaca oleracea* aggr. shows that the analyzed material was very heterogeneous. It shows the differentiation between the studied



**Figure 1** Morphological features of testa epidermal cells. Abbreviations: sr – short rays; lr – long rays, Ur – U-rays, Vr – V-rays, Yr – bifurcated rays.

samples (herbarium sheets) and within the samples when multiple specimens were mounted on a single sheet. The seeds from the studied samples differed in size. The diameters ranged from 0.672 to 1.1 mm. The epidermal cells of the testa were star-shaped, with periclinal walls (either smooth or papillated). Analysis of seed microphotographs obtained under a scanning microscope enabled the description of the detailed traits of the individual samples (Table 2).

Based on the key for identification of *Portulaca* taxa according to seed traits presented in the article by Danin et al. (2008), the following taxa were distinguished: *P. daninii*, *P. granulatostellulata*, *P. macrantha*, *P. nitida*, *P. oleracea*, *P. papillatostellulata*, and *P. trituberculata*.

**Table 2** Identified taxa of *Portulaca oleracea* aggr. deposited in the POZ Herbarium.

Sheet No.	Taxon name	Seed length (μm)	Rays V	Rays U	Rays Y	Long rays	Short rays	CP pap.	Ray pap. 1	Ray pap. 2	ID cells	EN cells
POZ-V-00075775	<i>P. nitida</i>	840	0	+	+	+	+	0	0	0	+	+
POZ-V-00075777	<i>P. trituberculata</i>	950	0	+	+	+	+	+	+	0	+	+
POZ-V-00075780	<i>P. papillatostellulata</i>	900	0	+	+	0	+	+	+	+	+	0
POZ-V-00075780	<i>P. granulatostellulata</i>	800	0	+	+	0	+	+	+	0	+	0
POZ-V-00075781	<i>P. trituberculata</i>	950	0	+	+	0	+	+	+	0	0	+
POZ-V-00075783	<i>P. oleracea</i> s. str.	1,030	+	0	+	+	+	0	0	0	+	+
POZ-V-00075784	<i>P. oleracea</i> s. str.	930	+	0	+	+	0	0	0	0	+	+
POZ-V-00075785	<i>P. oleracea</i> s. str.	970	+	+	+	+	+	0	0	0	0	+
POZ-V-00075786	<i>P. daninii</i>	850	0	+	+	0	+	+	+	0	+	0
POZ-V-00075786	<i>P. macrantha</i>	1,000	0	+	+	+	+	+	+	+	+	+
POZ-V-00075786	<i>P. trituberculata</i>	950	0	+	+	0	+	+	+	+	0	+
POZ-C-0000826	<i>P. oleracea</i> s. str.	920	+	+	+	+	+	0	0	0	0	+
POZ-C-0000978	<i>P. nitida</i>	840	0	+	+	+	0	0	0	0	+	0
POZ-C-0003211	<i>P. nitida</i>	850	+	+	+	+	+	0	0	0	+	+
POZ-C-0003321	<i>P. nitida</i>	850	+	+	+	+	0	0	0	0	+	+
POZ-C-0003323	<i>P. oleracea</i> s. str.	840	+	+	+	+	0	0	0	0	+	0
POZ-C-0003385	<i>P. trituberculata</i>	920	0	+	+	+	+	+	+	0	+	+

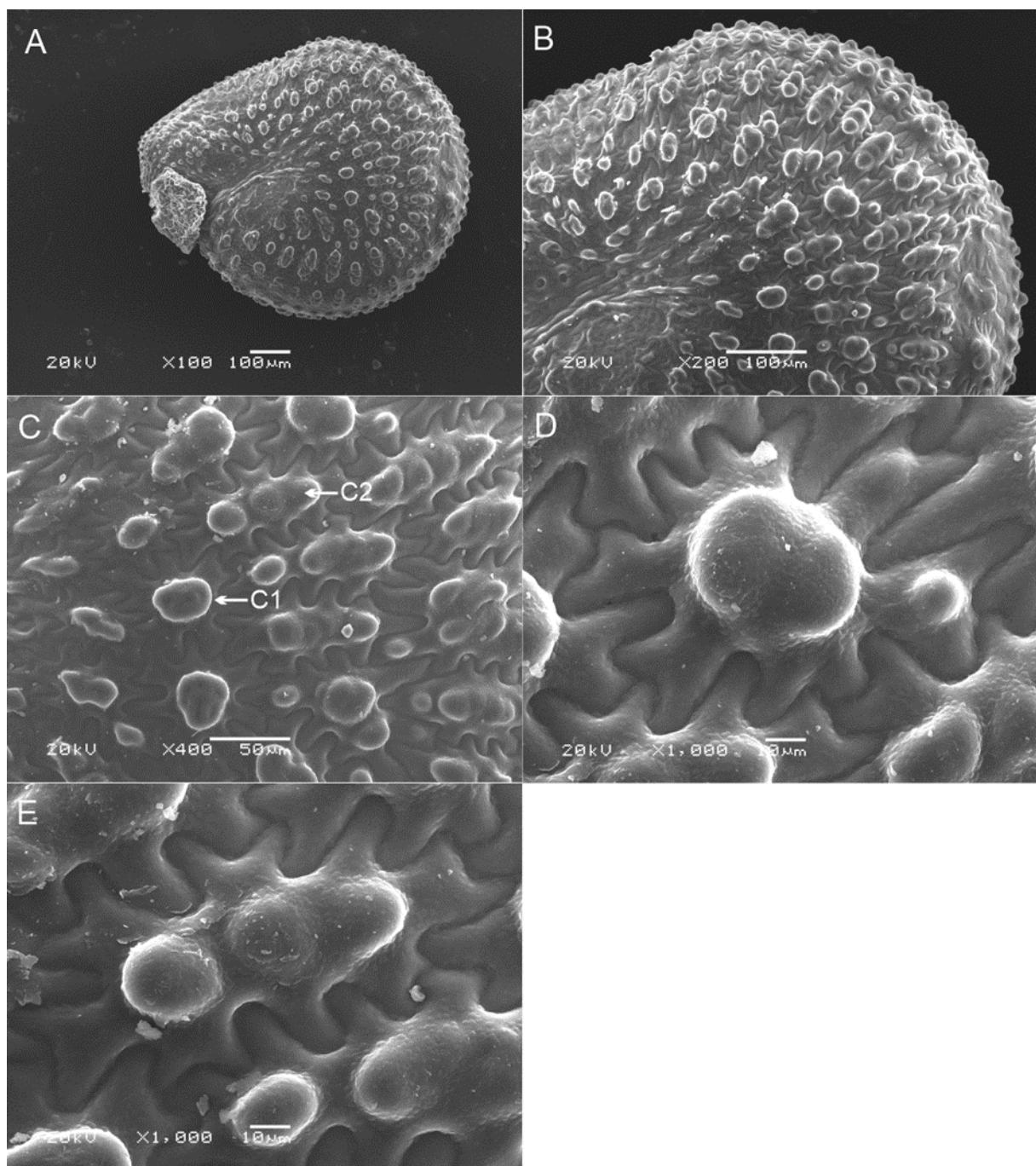
CP pap. – papillae in the central parts of rays; Ray pap. 1 – ray papillae rare (single); Ray pap. 2 – ray papillae frequent; ID cells – isodiametric cells; EN cells – elongated cells.

### 3.1. *Portulaca daninii* Galasso, Banfi & Soldano

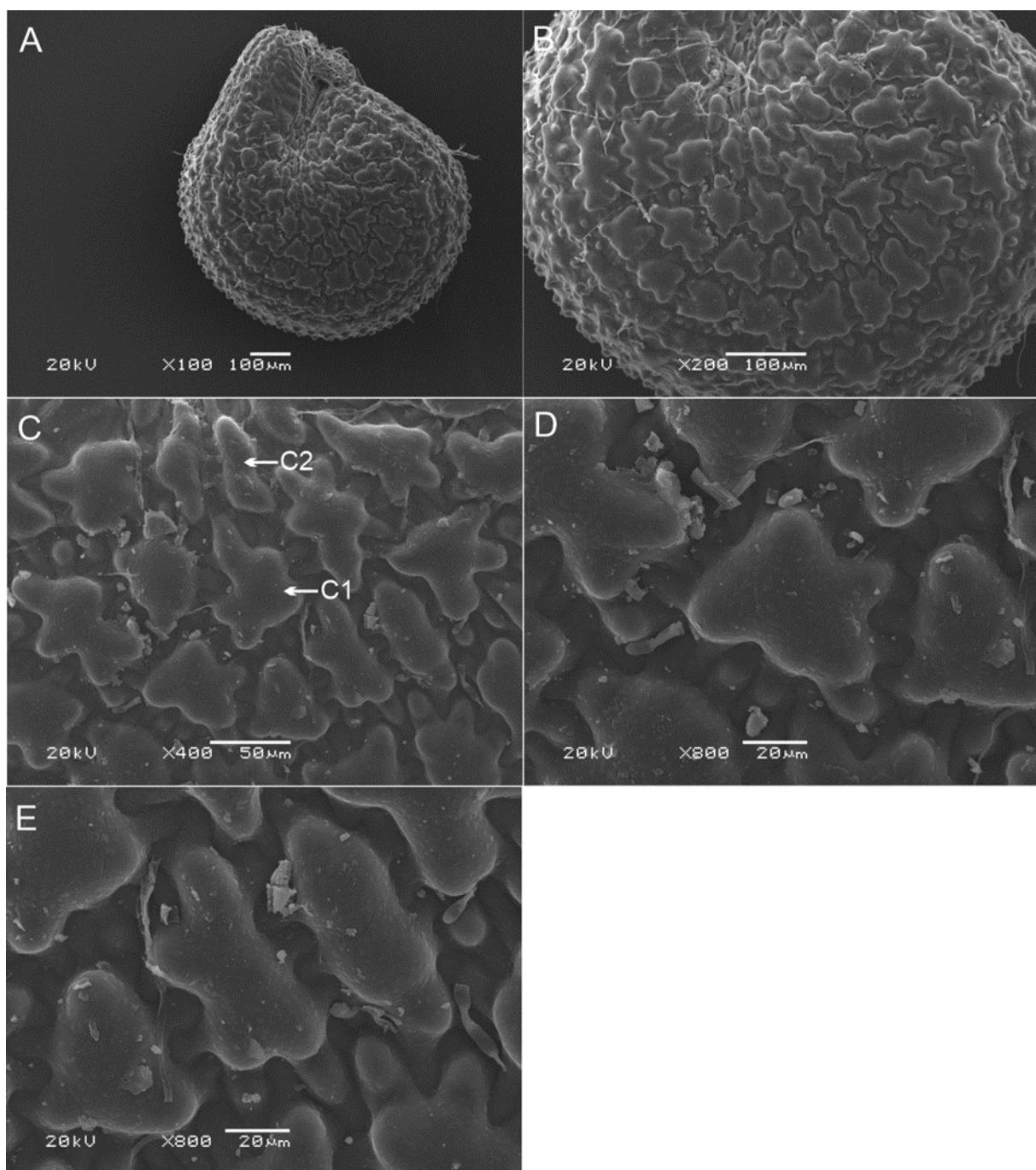
The seed is subreniform, black, with a glossy surface; the seeds are <0.85 mm in diameter and range from 0.672 mm to 0.823 mm (with an average of 0.819 mm). The individual cells were mostly elongated and isodiametric, mainly convex or domed, with tubercles in the center of the cells and papillae at the ends of some rays (Figure 2). The specimen examined originated from the Odra River alluvia in the vicinity of Krosno Odrzańskie (POZ 00075786; Table 1).

### 3.2. *Portulaca granulatostellulata* (Poelln.) Ricceri & Arrigoni

The seed is subreniform, black, with a glossy surface; the seeds are <0.85 mm in diameter and range from 0.703 mm to 0.849 mm and 0.733 mm on average.



**Figure 2** *Portulaca daninii* (SEM). General view of the seed (A). The seed surface (B,C). Isodiametric cell (C1), elongated cell (C2); general view of the cells (D,E).

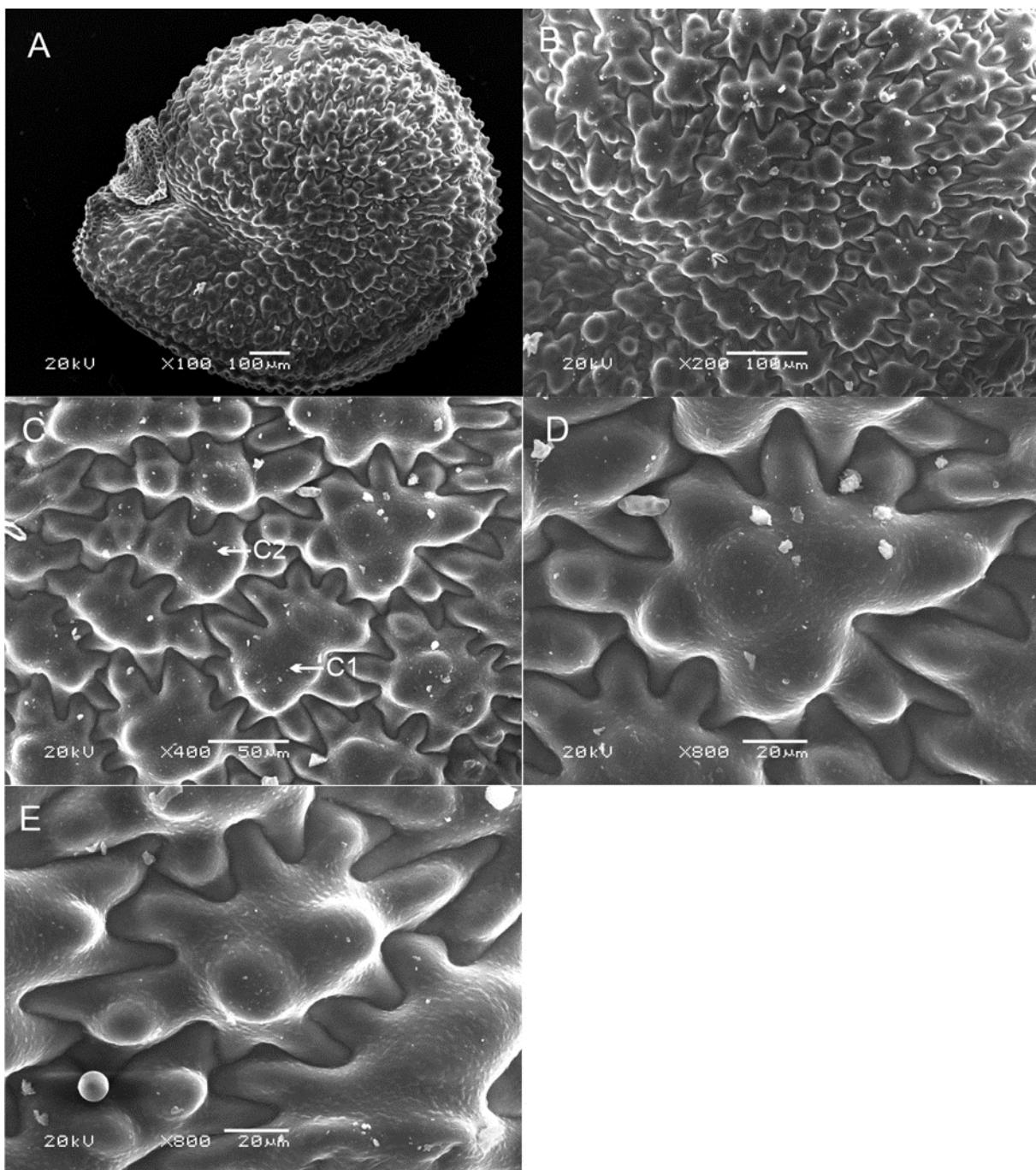


**Figure 3** *Portulaca granulatostellulata* (SEM). General view of the seed (A). The seed surface (B,C). Isodiametric cell (C1), elongated cell (C2); general view of the cells (D,E).

The individual cells of the surface are star-shaped and elongated, convex, nearly flat to low-convex; the length of the rays exceeds the width 1.5–2 times. The cells were papillated, with papillae located at the ends of the cell rays or on a few cells at the base of the rays (Figure 3). The examined specimen originated from the Damasławek railway station, Wielkopolska Province (POZ 00075780; Table 1).

### 3.3. *Portulaca macrantha* Ricceri & Arrigonii

The seed is subreniform, black, with a glossy surface; the seeds have a diameter of >0.85 mm and range from 0.850 mm to 1.03 mm; 0.916 mm on average. Individual cells on the surface were isodiametric and elongated. The testa cells are star-shaped, convex, or domed; the rays are short, with terminal papillae on the rays and



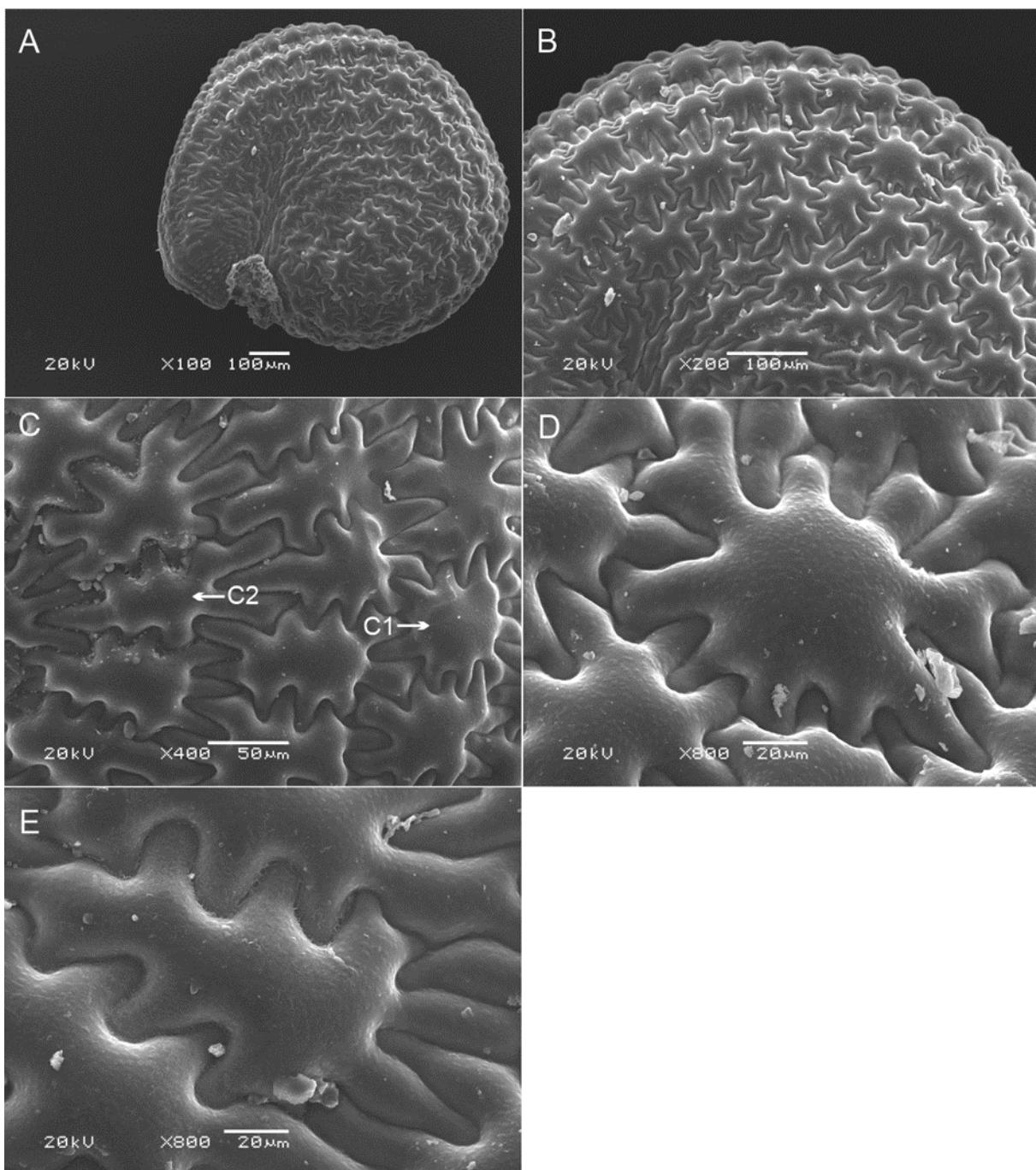
**Figure 4** *Portulaca macrantha* (SEM). General view of the seed (A). The seed surface (B,C). Isodiametric cell (C1), elongated cell (C2); general view of the cells (D,E).

two–seven scattered small tubercles or papillae in the center (Figure 4).

The specimen examined originated from the Odra River alluvia in the vicinity of Krosno Odrzańskie (POZ 00075786; Table 1).

### 3.4. *Portulaca nitida* (Danin & H. G. Baker) Ricceri & Arrigoni

The seeds are subreniform or rounded, black, with a glossy surface; the seeds are <0.85 mm in diameter and range from 0.691 mm to 0.848 mm, 0.808 mm on average. The individual cells were isodiametric, star-shaped, or elongated, with long rays, nearly flat to slightly convex, and without papillae and tubercles (Figure 5). The specimens examined originated from Książ Wlkp. (POZ 00075775), Wschowa

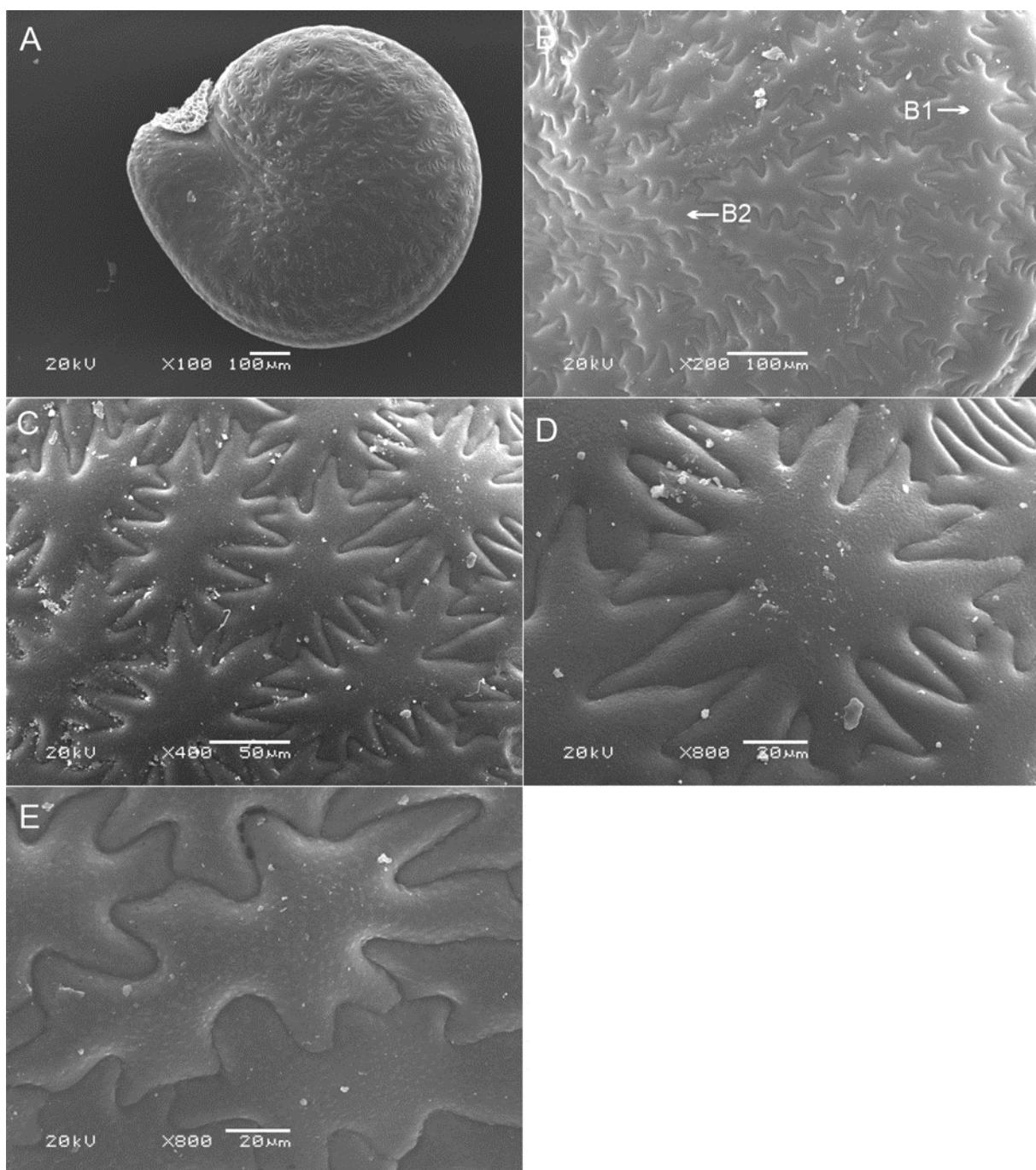


**Figure 5** *Portulaca nitida* (SEM). General view of the seed (A). The seed surface (B,C). Isodiametric cell (C1), elongated cell (C2); general view of the cells (D,E).

(POZ-C-0000978, POZ-C-0003321) and Krzyż (POZ-C-0003211) railway stations (Table 1).

### 3.5. *Portulaca oleracea* L. s. str.

The seed is subreniform or rounded, black, with a glossy surface; the seeds are approximately 0.85–1.1 mm in diameter and range from 0.812 mm to 1.076 mm; 0.943 mm on average. The individual cells were star-shaped, isodiametric, or elongated, mostly flat, with long rays, smooth, and without papillae and tubercles (Figure 6). The specimens examined originated from: Międzychód (POZ 00075783) and Środa Wlkp. railway stations (POZ 00075784), railway tracks in Szymanowo

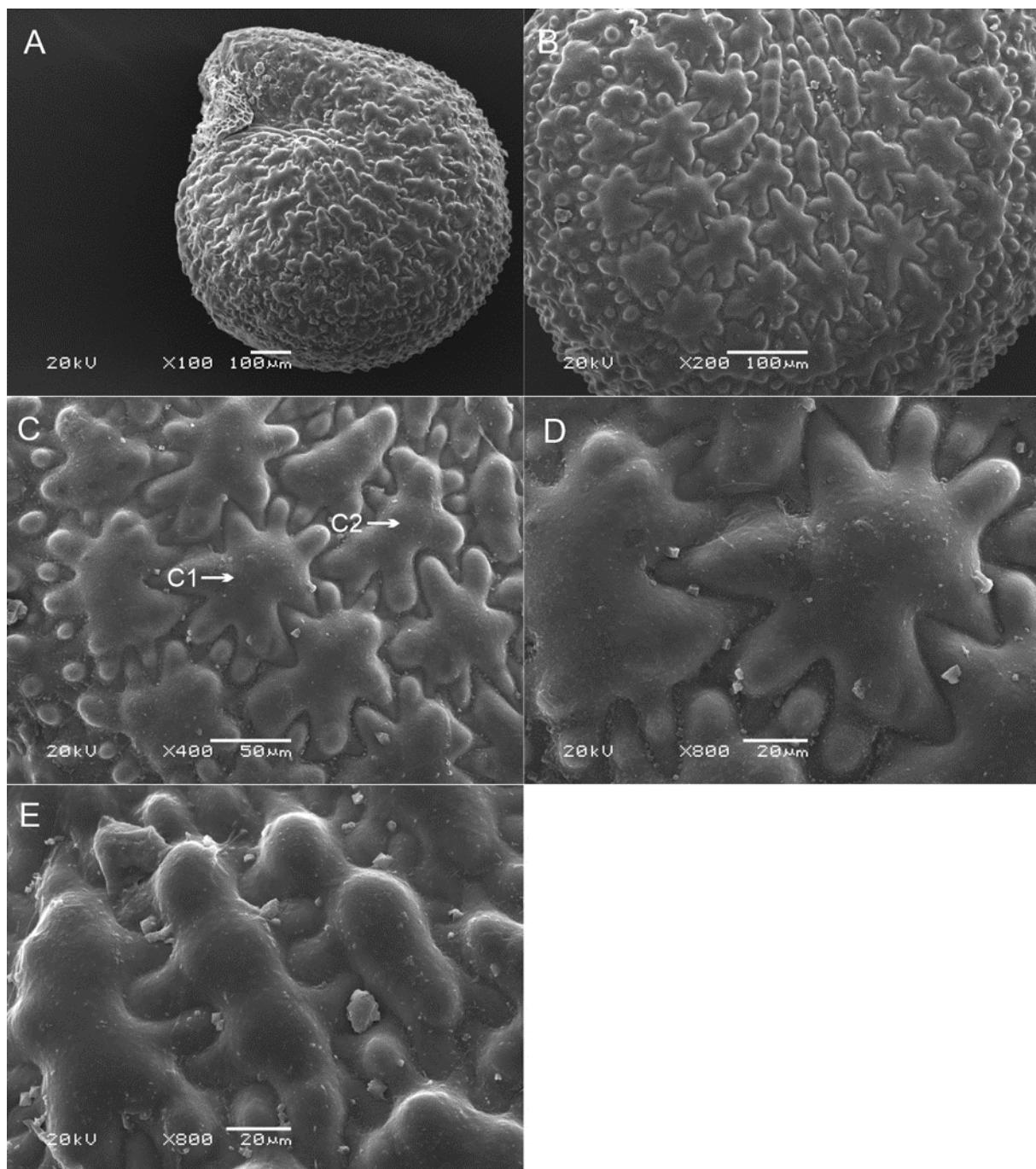


**Figure 6** *Portulaca oleracea* s. str. (SEM). General view of the seed (A). The seed surface (B,C). Isodiametric cell (B1), elongated cell (B2); general view of the cells (D,E).

(POZ-C-0003323), a sandy playing field in Toruń (POZ 00075785), and in the vicinity of a compost heap along the Warta River (POZ-C-0000826) (Table 1).

### 3.6. *Portulaca papillatostellulata* (Danin & H. G. Baker) Danin

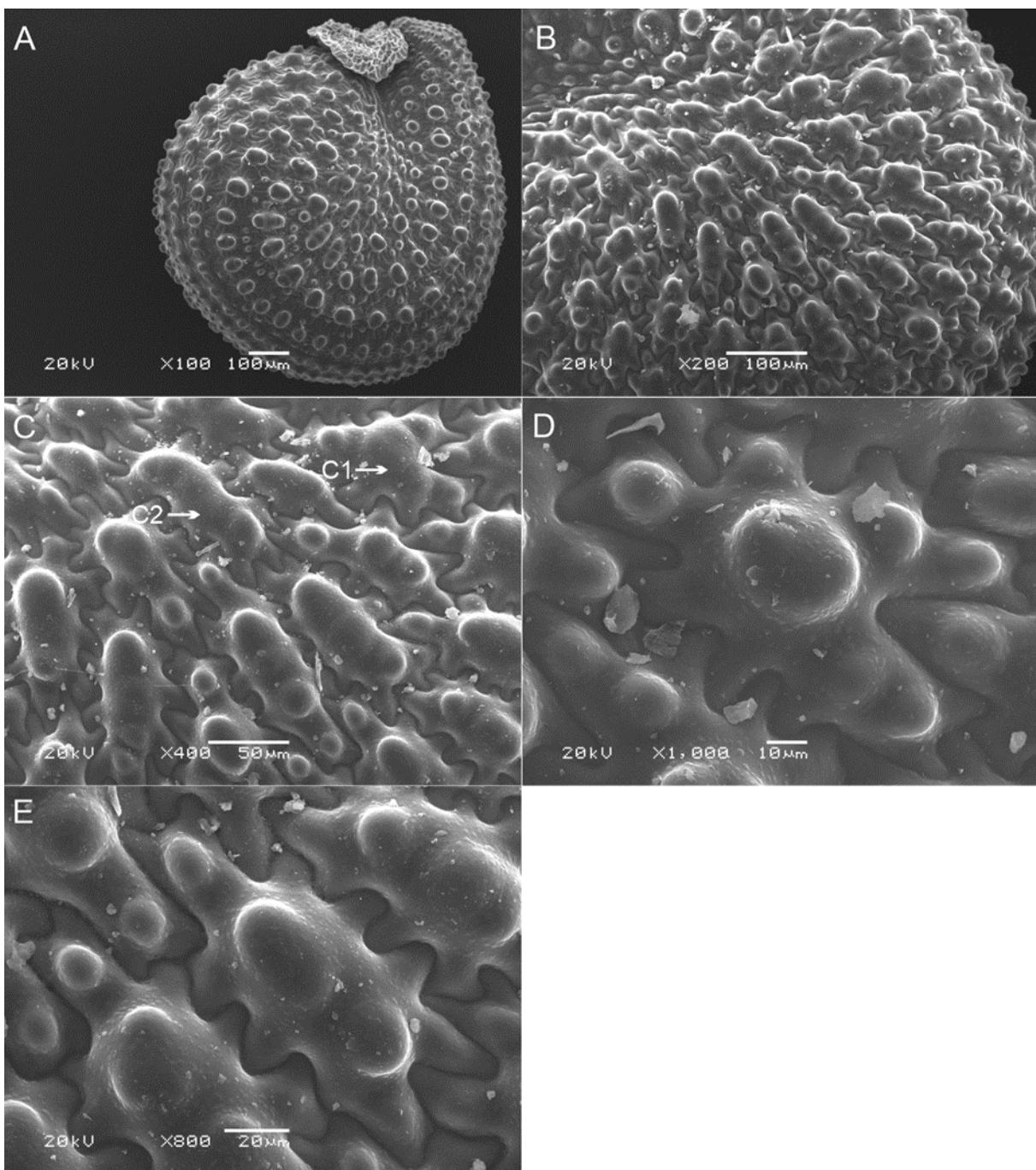
The seed is subreniform, black, with a glossy surface; the seeds are approx. 0.85–1.1 mm in diameter and range from 0.857 mm to 0.922 mm; 0.883 mm on average. The individual cells were star-shaped, isodiametric, and elongated; the central parts of the cells are convex or nearly flat to low-convex, smooth, without papillae, with long rays, many with terminal papillae, but not all were papillated; some papillae were more inflated than the cell bodies (Figure 7). The examined specimen originated from the Damasławek railway station (POZ 00075780; Table 1).



**Figure 7** *Portulaca papillastellulata* (SEM). General view of the seed (A). The seed surface (B,C). Isodiametric cell (C1), elongated cell (C2); general view of the cells (D,E).

### 3.7. *Portulaca trituberculata* Danin, Domina & Raimondo

The seed is subreniform, black, with a glossy surface; the seeds are approx. 0.85–1.1 mm in diameter and range from 0.857 mm to 1.03 mm; 0.931 mm on average. The individual cells were star-shaped and elongated; the central cell parts were convex or nearly flat to low convex, smooth, without papillae, with long rays, many with terminal papillae but not all were papillated, and papillae were more inflated than the cells (Figure 8). The specimens examined originated from the Konin (POZ 00075781) and Leszno (POZ-C-0003385) railway stations, roadsides in the Wielkopolski National Park (POZ 00075777), and the Odra River alluvia in the vicinity of Krosno Odrzańskie (POZ 00075786) (Table 1).



**Figure 8** *Portulaca trituberculata* (SEM). General view of the seed (A). The seed surface (B,C). Isodiametric cell (C1), elongated cell (C2); general view of the cells (D,E).

#### 4. Discussion

Among the 39 herbarium specimens, denoted as *P. oleracea* and deposited in the POZ Herbarium (Vascular Plant Collection, POZ-V; <https://amunatcoll.pl/>), only eight sheets contained seeds. Additionally, there were six samples of *P. oleracea* seeds in the carpological collection (POZ-C; <https://amunatcoll.pl/>). Fourteen seed samples were analyzed in total. All originated from central-western Poland (Wielkopolska Province). In this area, there is the highest concentration of *P. oleracea* localities in Poland (cf. Zajac & Zajac, 2001; <https://amunatcoll.pl/>).

Previously, four taxa have been distinguished within *Portulaca oleracea* aggr. in Poland: *P. granulatostellulata*, *P. nitida*, *P. trituberculata*, and *P. oleracea* s. str. (Danin in Greuter & von Raab-Straube, 2011, pp. 131–134). Micromorphological analysis of

the seed surface ultrastructure of *P. oleracea* aggr. material deposited in the POZ Herbarium, which was conducted as a part of this work, enabled the documentation of seven morphological types. Apart from those previously reported by Danin (in Greuter & von Raab-Straube, 2011, pp. 131–134), other morphotypes (species) are new records for Poland. They include *P. daninii*, *P. macrantha*, and *P. papillatostellulata*. Presently, there are 10 taxa of the genus *Portulaca* in the flora of Poland, seven of which have been distinguished within *P. oleracea* aggr. (Danin in Greuter & von Raab-Straube, 2011, pp. 131–134; Mirek et al., 2020; Rutkowski, 2011).

The most frequently identified taxa were *P. oleracea* L. s. str. (five samples), and *P. trituberculata* and *P. nitida* (four samples each) (Table 2), respectively. They mostly originated from railway areas (stations and tracks) and ruderal places (playing field and roadside), and less frequently from alluvial areas. These observations confirm that the taxa of *P. oleracea* aggr. mostly migrate to plant communities that develop in anthropogenic habitats (Tokarska-Guzik et al., 2012).

The nearest localities of the taxa documented in this study were observed in neighboring countries, such as Germany (*P. nitida*), Slovakia (*P. granulatostellulata*, *P. papillatostellulata*, *P. trituberculata*), Ukraine (*P. granulatostellulata*, *P. papillatostellulata*) and Belarus (*P. granulatostellulata*, *P. nitida*, *P. oleracea* s. str., *P. trituberculata*) (Bulakh et al., 2019; Feráková et al., 2012; Danin in Greuter & von Raab-Straube, 2011, pp. 131–134; Dzhus et al. in von Raab-Straube & Raus, 2015, pp. 457–459). Two out of the three new taxa distinguished for Poland in this study – *P. macrantha* and *P. tuberculate*, are also new records for Central and Eastern Europe.

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