

ORIGINAL ARTICLE

Clinical Mechanisms in Allergic Disease

Germination of pollen grains in the oesophagus of individuals with eosinophilic oesophagitis

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Summary

Background: Eosinophilic oesophagitis (EoE) is characterized by oesophageal dysfunction and, histologically, by eosinophilic inflammation. There is no a clear aetiological treatment. EoE exacerbations are often seasonal. We hypothesized that the inflammatory response of the oesophageal mucosa in patients with high levels of antibodies to pollen allergens and worsened seasonal EoE might be due to swallowing airborne pollen and the intrusion into the oesophageal mucosa of pollen allergens and pollen tubes, which encounter a pH and humidity resembling the stigma at pollination.

Objective: The aim of our study was to demonstrate the possible pathogenic role of environmental allergens in EoE through molecular and anatomopathological studies

Methods: One hundred and twenty-nine patients with EoE were tested for environmental and food allergens. Component resolved diagnosis (CRD), histological and botanical analysis was performed. Microscopic examination of oesophageal biopsies of 129 adults patients with EoE, 82 of them with seasonal exacerbation, and 100 controls, with gastroesophageal reflux without eosinophilic infiltrate, were made to verify the presence of callose (polysaccharide abundant in pollen tubes but absent in animal tissues) in the oesophagus.

Results: Component resolved diagnosis detected pollen allergens in 87.6% of patients with EoE. The predominant allergens were group 1 grass (55%), Art v 3 (11.3%) and lipid transfer proteins (LTPs) (19.4%) of common Mediterranean foods such as peach, hazelnuts, walnuts and wheat. Callose from pollen tubes was found in 65.6% of biopsies.

Conclusion: Alteration of the mucosal barrier in EoE might cause the penetration of pollen grains into the oesophageal tissues. In EoE patients, anatomopathological studies searching for intrusion to plant foods and pollen, and specific-guided diet and immunotherapy after plant structures detection in biopsies, might be effective.

Clinical relevance: It is possible to see the intrusion into animal tissues (oesophagus mucosa) of plant structures (pollen grains or pollen tubes) using an adequate histologic botanical analysis. Molecular and anatomopathological studies can help to demonstrate a possible pathogenic role of environmental allergens in EoE.

KEYWORDS

biopsies, callose, eosinophilic oesophagitis, germination, pollen tubes, seasonal oesophagitis

Eosinophilic oesophagitis (EoE) is an atopic disease of unclear aetiology.¹ As in allergic asthma, mucosal barrier dysfunction has been reported.² Many EoE patients present rhinoconjunctivitis, atopic dermatitis and associated asthma, in addition to dysphagia and food impaction. EoE exacerbations are often seasonal.³

We hypothesized that the inflammatory response of the oesophageal mucosa in patients with high levels of antibodies to pollen allergens and worsened seasonal EoE might be due to swallowing airborne pollen and the intrusion into the oesophageal mucosa of pollen allergens and pollen tubes emitted after pollen germination, which encounter a pH and humidity resembling the stigma at pollination. The aim of our study was to demonstrate the possible pathogenic role of environmental allergens in EoE through molecular and anatomopathological studies.

Microscopic examination of oesophageal biopsies of 129 adults patients with EoE (age 35 ± 16.24 years), 82 of them with seasonal exacerbation (see demographic and clinical data in supplemental materials) and 100 controls (34 ± 12.16 years) with gastroesophageal reflux without eosinophilic infiltrate was performed to verify the presence of callose (abundant in pollen tubes but absent in animal tissues) in the oesophagus.⁴ Callose was detected using histological sections stained with sirofluor fluorochrome. Oesophageal mucosal sections were analysed by scanning electron microscope to detect pollen or spores. Allergens involved in EoE were identified by prick, specific IgE and component resolved diagnosis by microarrays.

All 129 selected patients suffered from active oesophagitis at the biopsy time. In all these patients, conventional EoE treatment (PPI, diet or steroids) did not produce improvement. Callosa was detected in 82 (67%) EoE patients with positive CRD to group 1 grass pollen. All these patients suffered clinical exacerbation in pollen season. Exacerbation was observed at the same time only in 4 control patients. Clinical evaluations and oesophageal biopsies were made every 6 months. Gastrointestinal (dysphagia, heartburn, stomach upset, vomiting, constipation, diarrhoea, failure to thrive) and allergic (rhinitis, asthma, dermatitis, anaphylaxis) histories were reviewed.

Pollen allergens were detected in 87.6% of patients with EoE. The predominant allergens were group 1 grass (55%), Art v 3 (11.3%) and lipid transfer proteins (LTPs) (19.4%) of common Mediterranean foods such as peach, hazelnuts, walnuts and wheat.

Callose was found in 65.6% of biopsies of patient positive for pollen allergens but not in controls. We observed pollen tubes surrounded by eosinophils in the proximal and middle oesophageal mucosa (Figure 1), resulting in micro-abscesses. Eosinophils seemed to act as if they were responding to parasitic infections. Callosa detection was more important in spring than in winter months. We add in supplemental material more images from biopsies obtained during pollen season.

Alteration of the mucosal barrier might cause the penetration of pollen grains into the oesophageal tissues. The subsequent germination of pollen and the release of highly allergenic digestion-resistant molecules, such as group 1 grass (β -expansins) and LTPs, might be

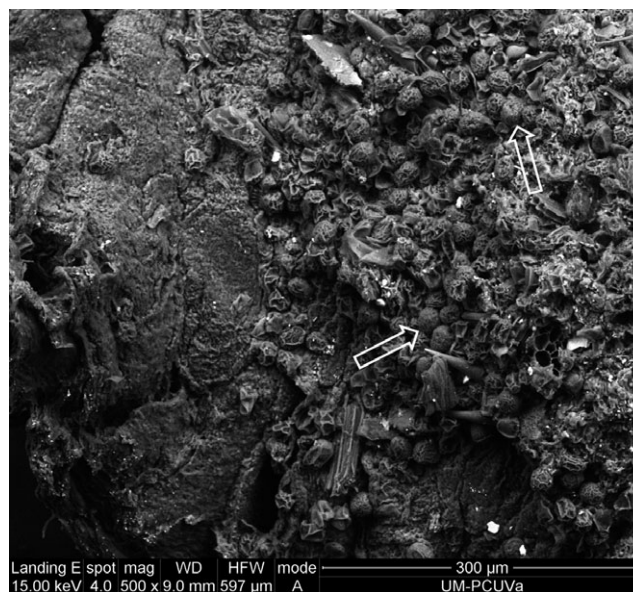


FIGURE 1 Epifluorescence shows pollen, spores and other plant elements on the surface of oesophageal biopsies. Plant impactions and oesophageal mucosae showing damaged spinous epithelial cells. Arrow shows microimpaction mainly composed of pollen grains of the Poaceae family infiltrating intercellular spaces using scanning electron microscope

responsible for the increase in symptoms in these patients during periods of greater pollination.

In EoE patients, anatomopathological studies searching for intrusion to plant foods and pollen, and specific-guided diet and immunotherapy after plant structures detection in biopsies, might be effective.⁵

CONFLICTS OF INTERESTS

The authors report no conflict of interest.

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SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section at the end of the article.

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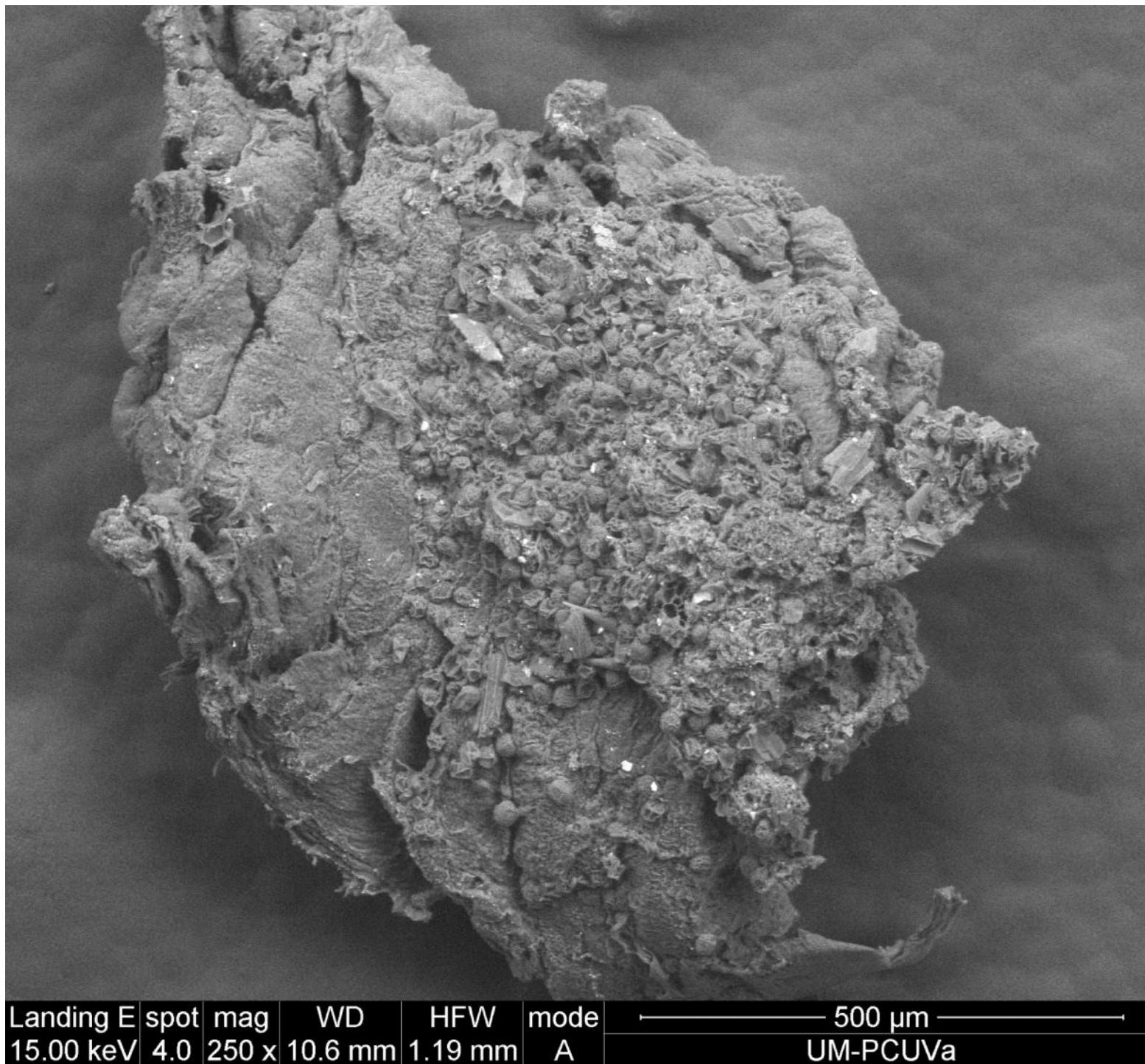
Supplemental materials

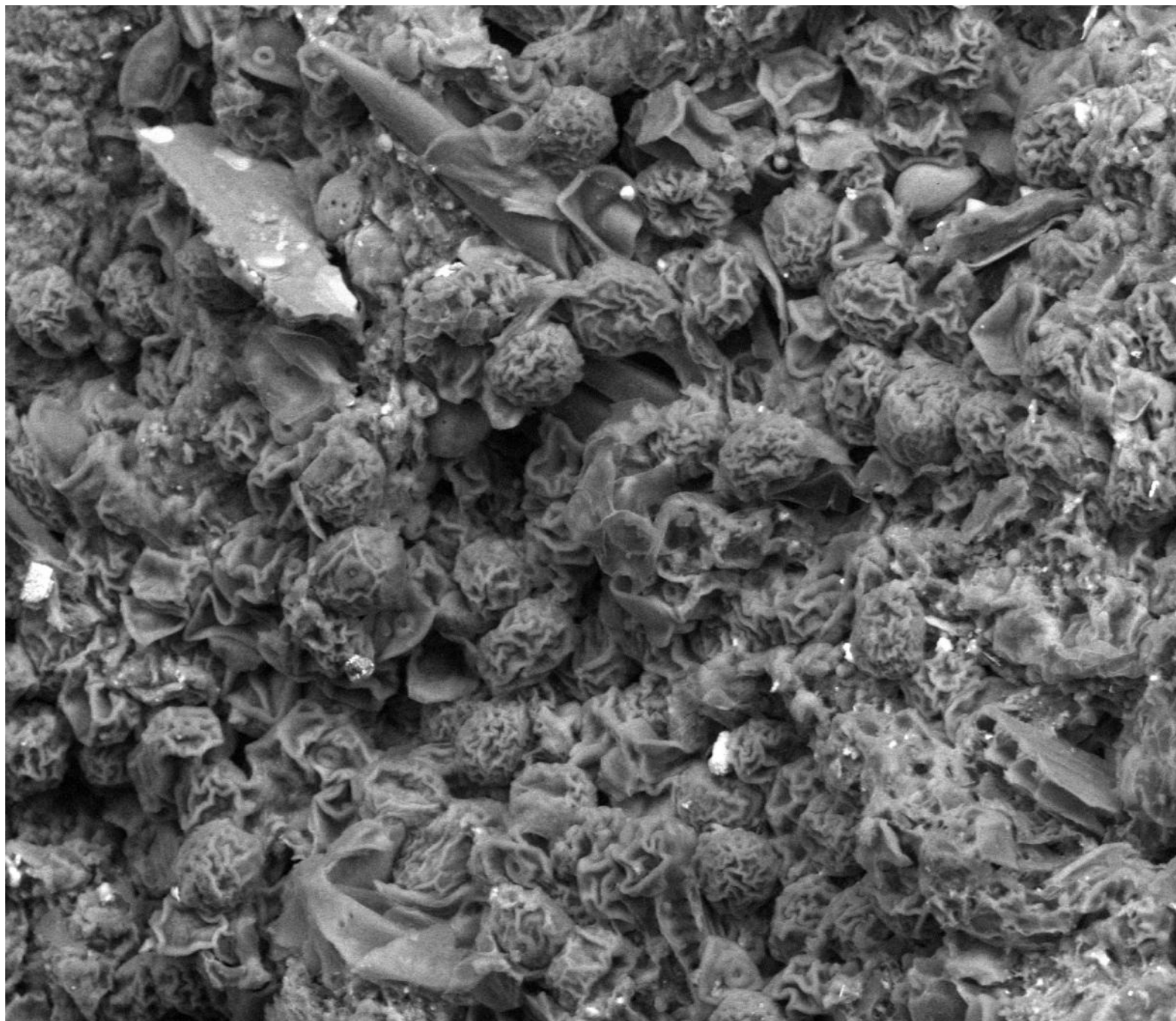
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Demographic and clinical characteristics of patients

	Number (%)
Patients with active EoE, with >15 eos/hpf	129
Males	83 (64%)
Mean age ($\pm$ SD) years	34.7 ($\pm$ 16)
EoE & Asthma	11 (8.5%)
EoE & other atopic symptoms (Rhinitis, Dermatitis, urticaria)	7 (5.4%)
EoE & Anaphylaxis	8 (6.2%)
EoE with Food sensitization	14 (10.9%)
Mean time (years) since EoE was diagnosed	10 ($\pm$ 16)
Mean EoE number at time biopsy	130/field (range 876-20)
Oral corticosteroid therapy	36 (27.9%)
6 food diet trial	20 (15.5%)
Proton pump inhibitor trial at least nine months	93 (72%)

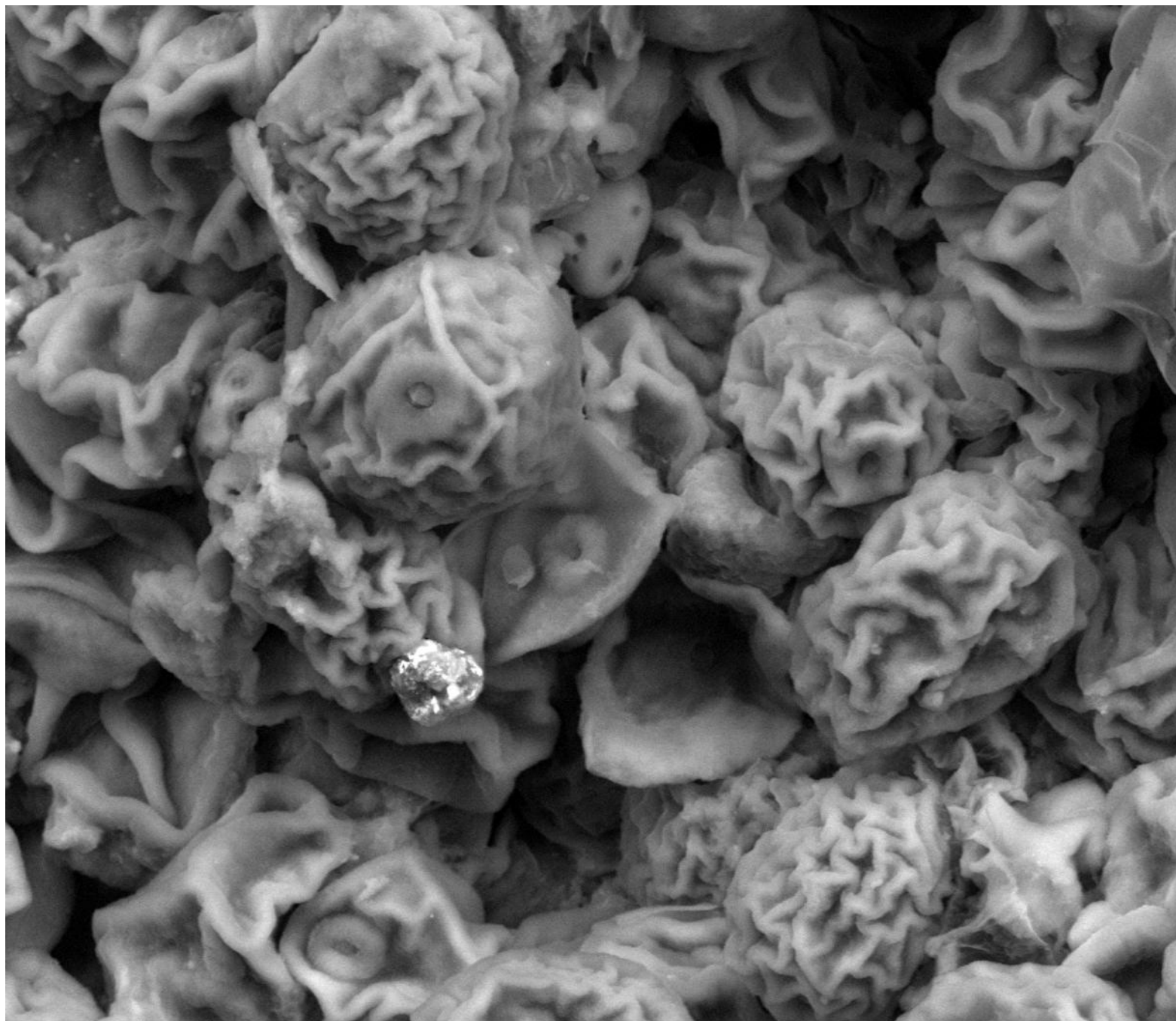




Landing E	spot	mag	WD	HFW	mode
15.00 keV	4.0	1 000 x	9.0 mm	298 μm	A

100 μm

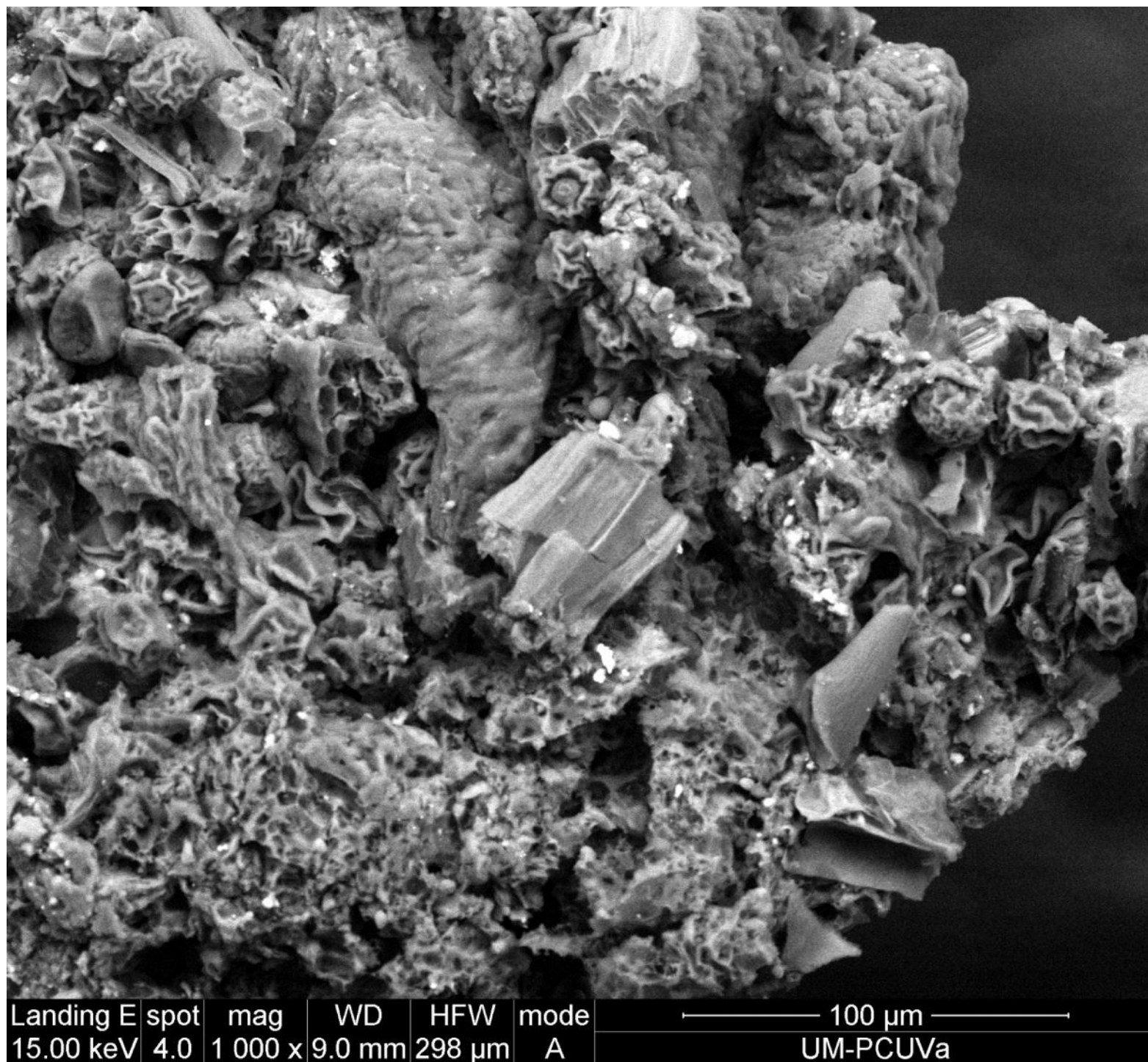
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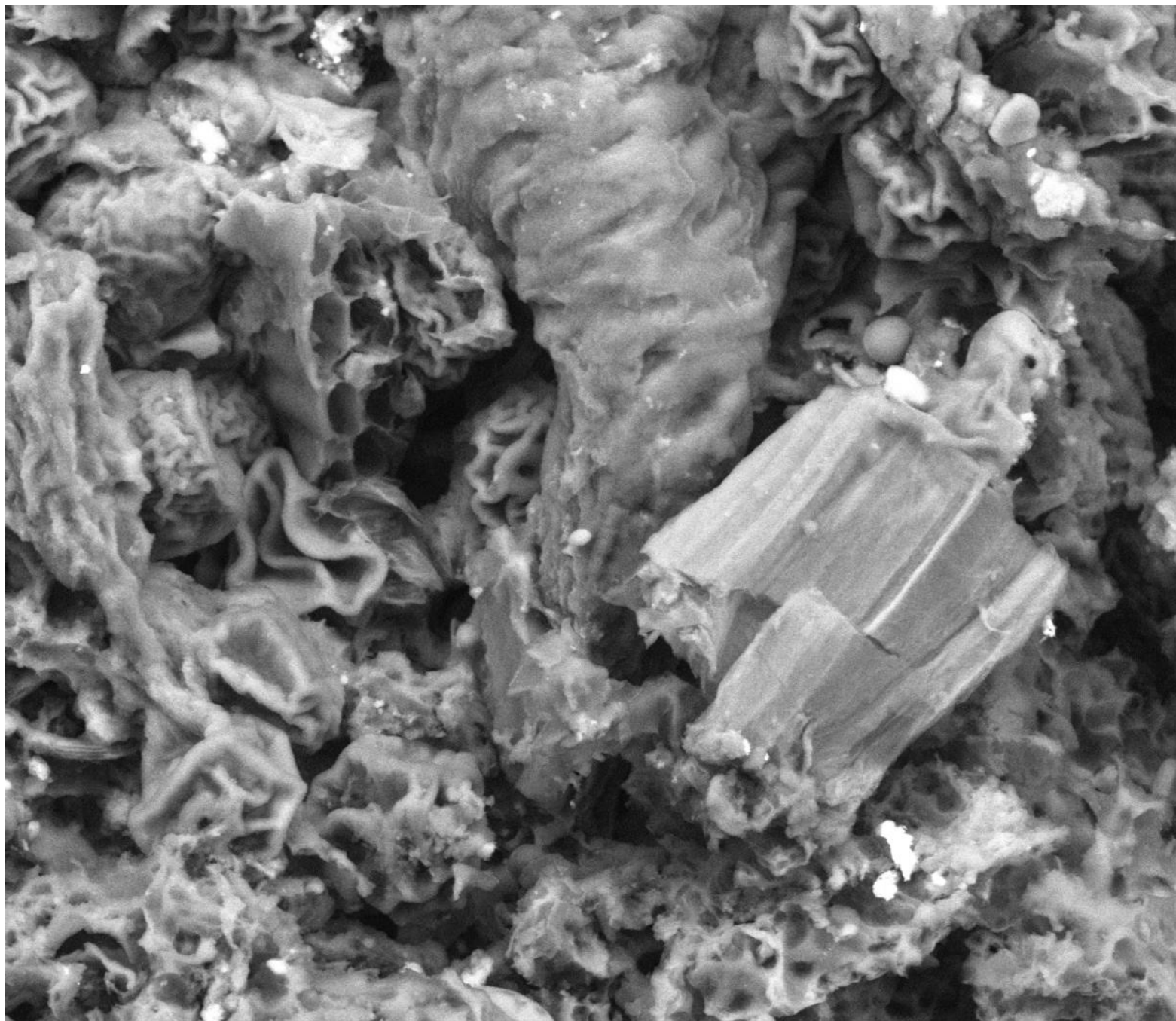


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40 μ m

UM-PCUVa

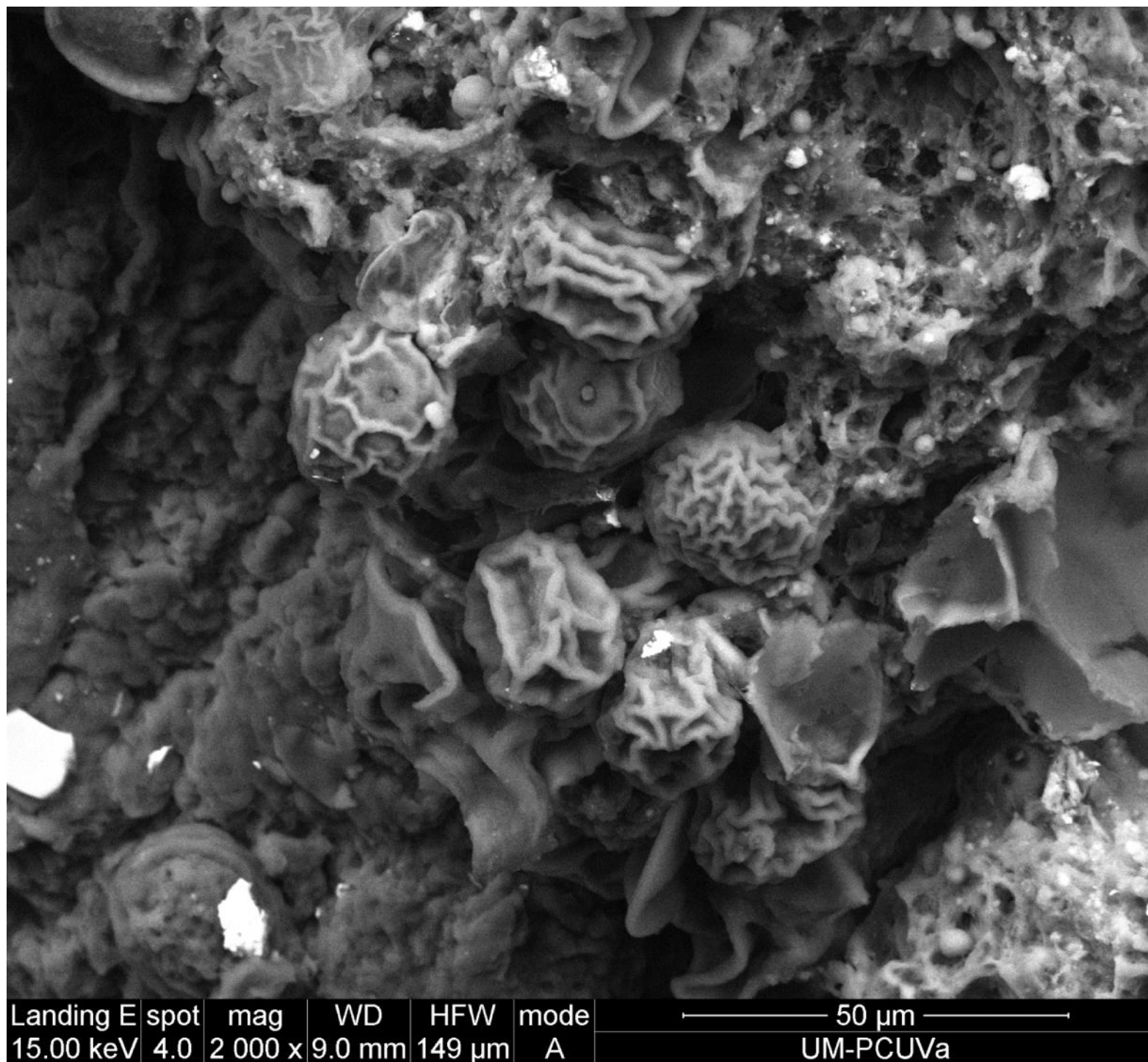




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15.00 keV	4.0	2 000 x	8.9 mm	149 μ m	A

50 μ m

UM-PCUVa

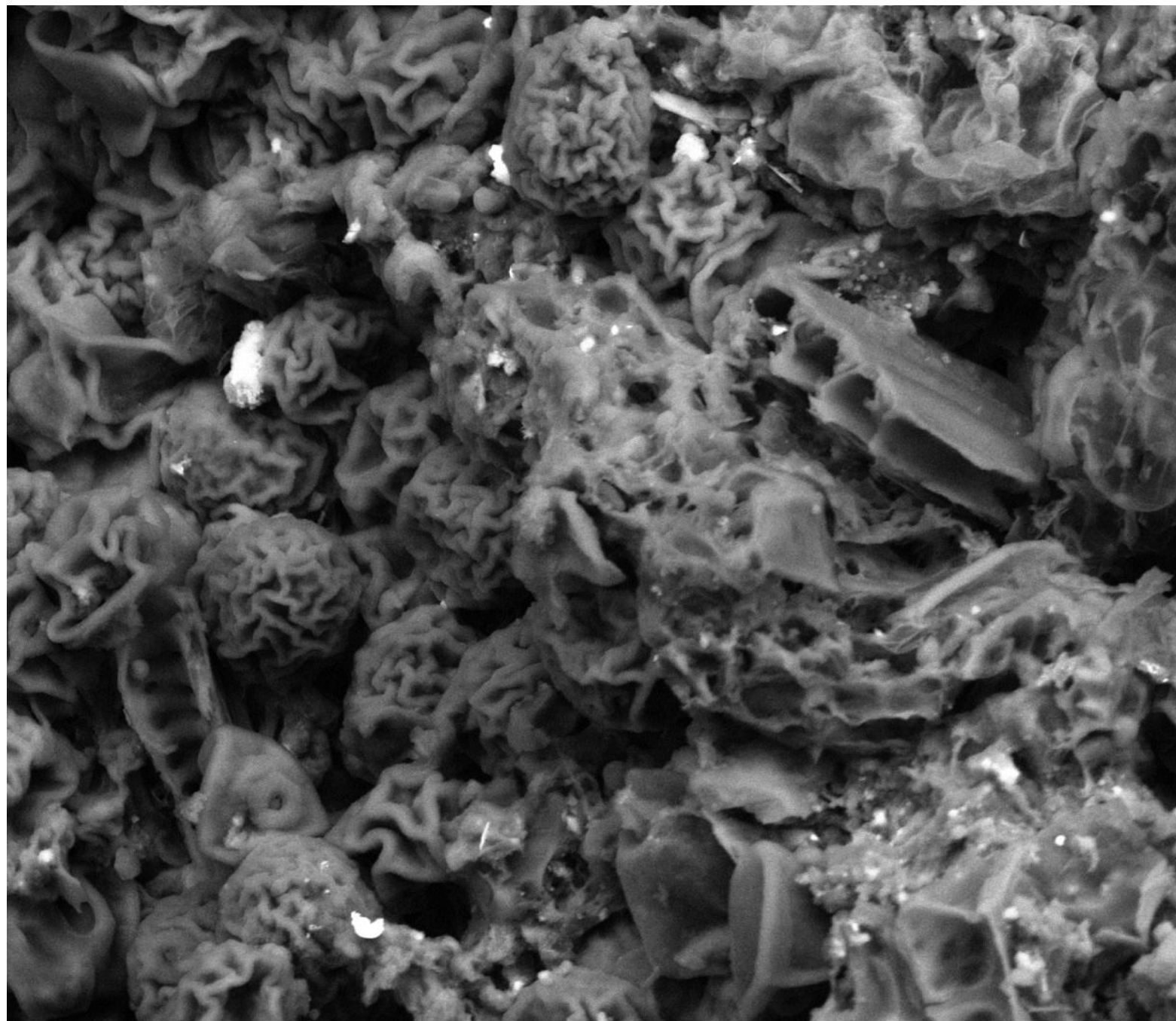


Landing E	spot	mag	WD	HFW	mode
15.00 keV	4.0	2 000 x	9.0 mm	149 µm	A

50 µm
UM-PCUVa



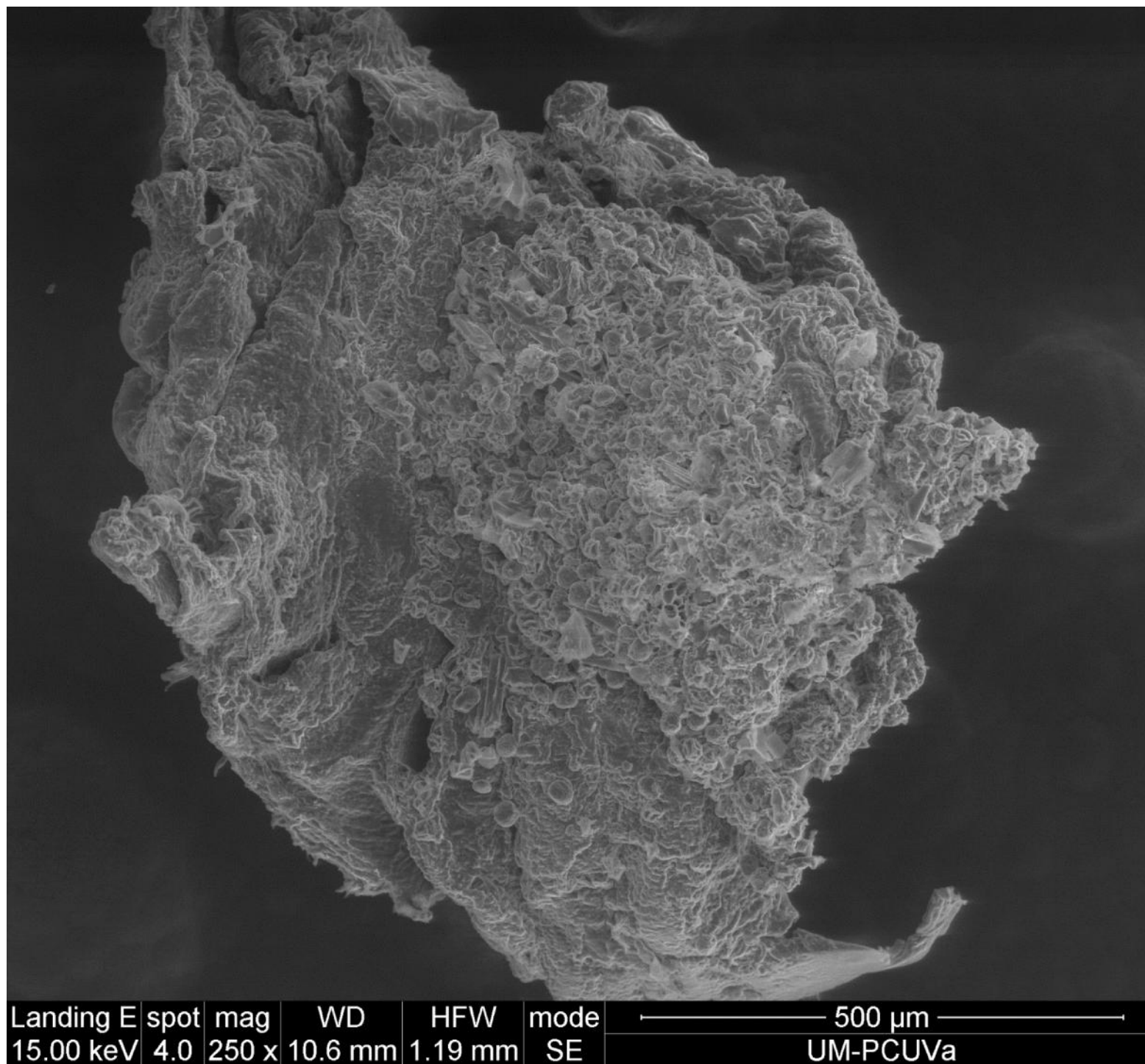
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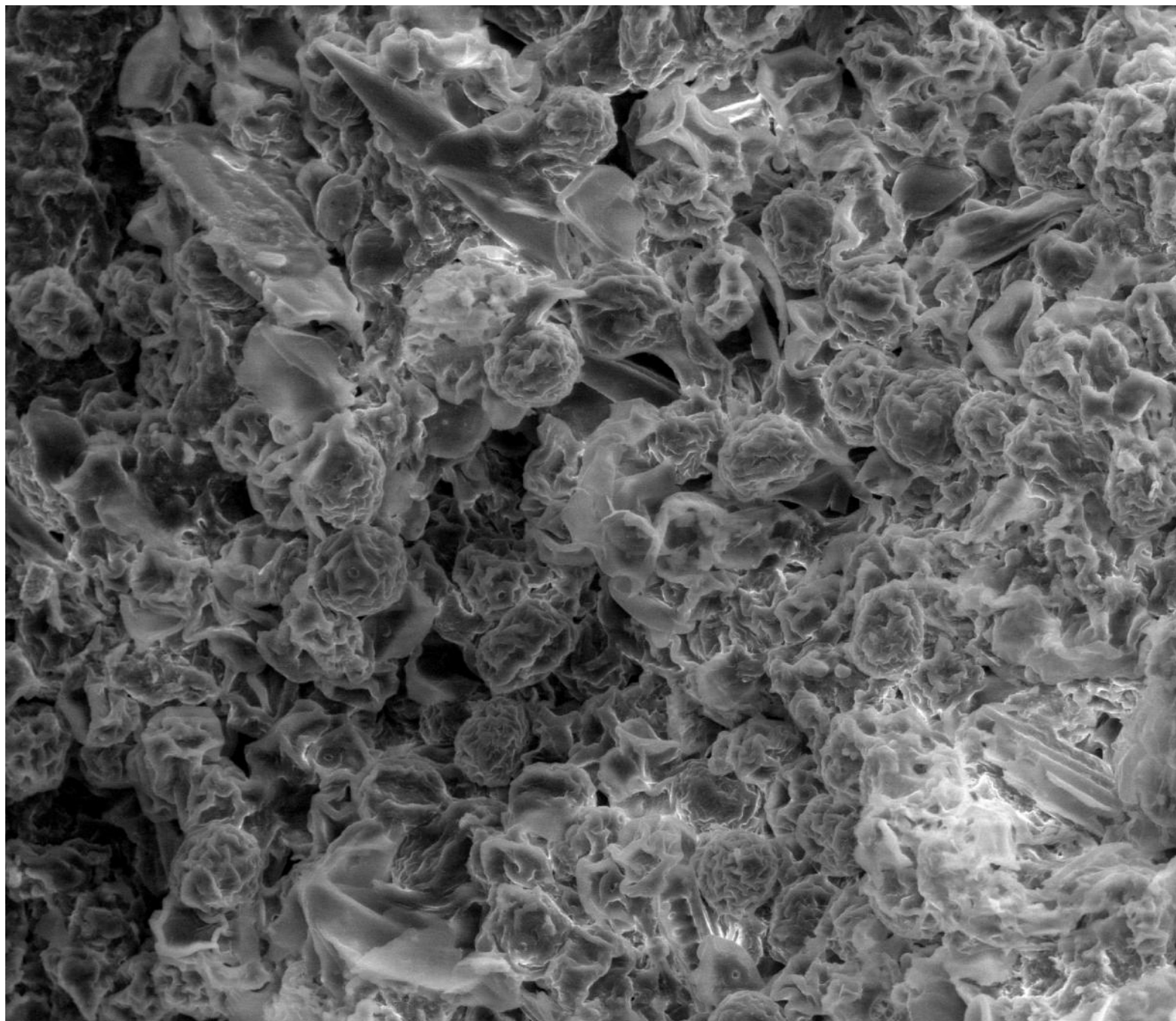


Landing E	spot	mag	WD	HFW	mode
15.00 keV	4.0	2 000 x	9.0 mm	149 μ m	A

50 μ m

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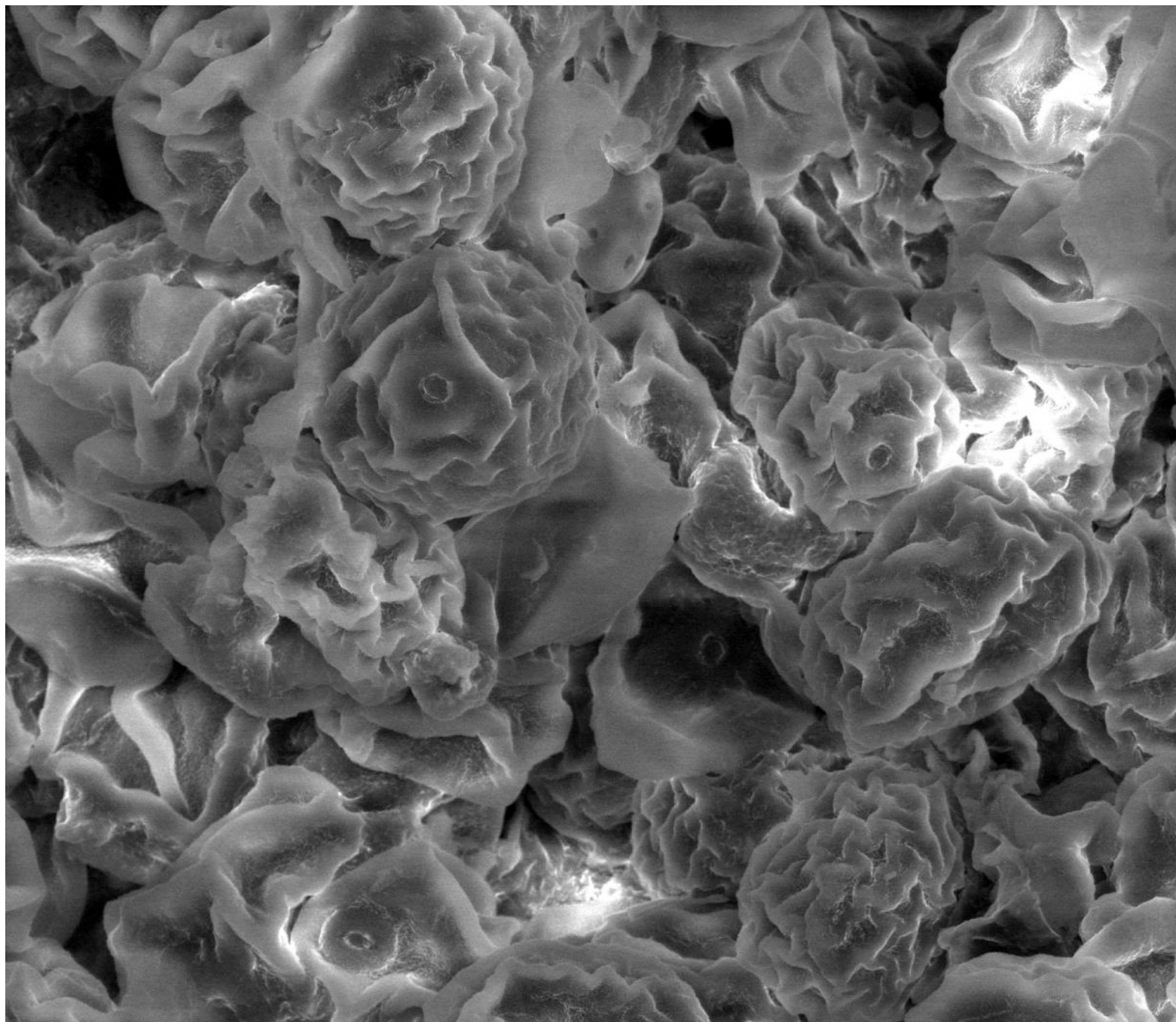




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15.00 keV	4.0	1 000 x	9.0 mm	298 μ m	SE

100 μ m

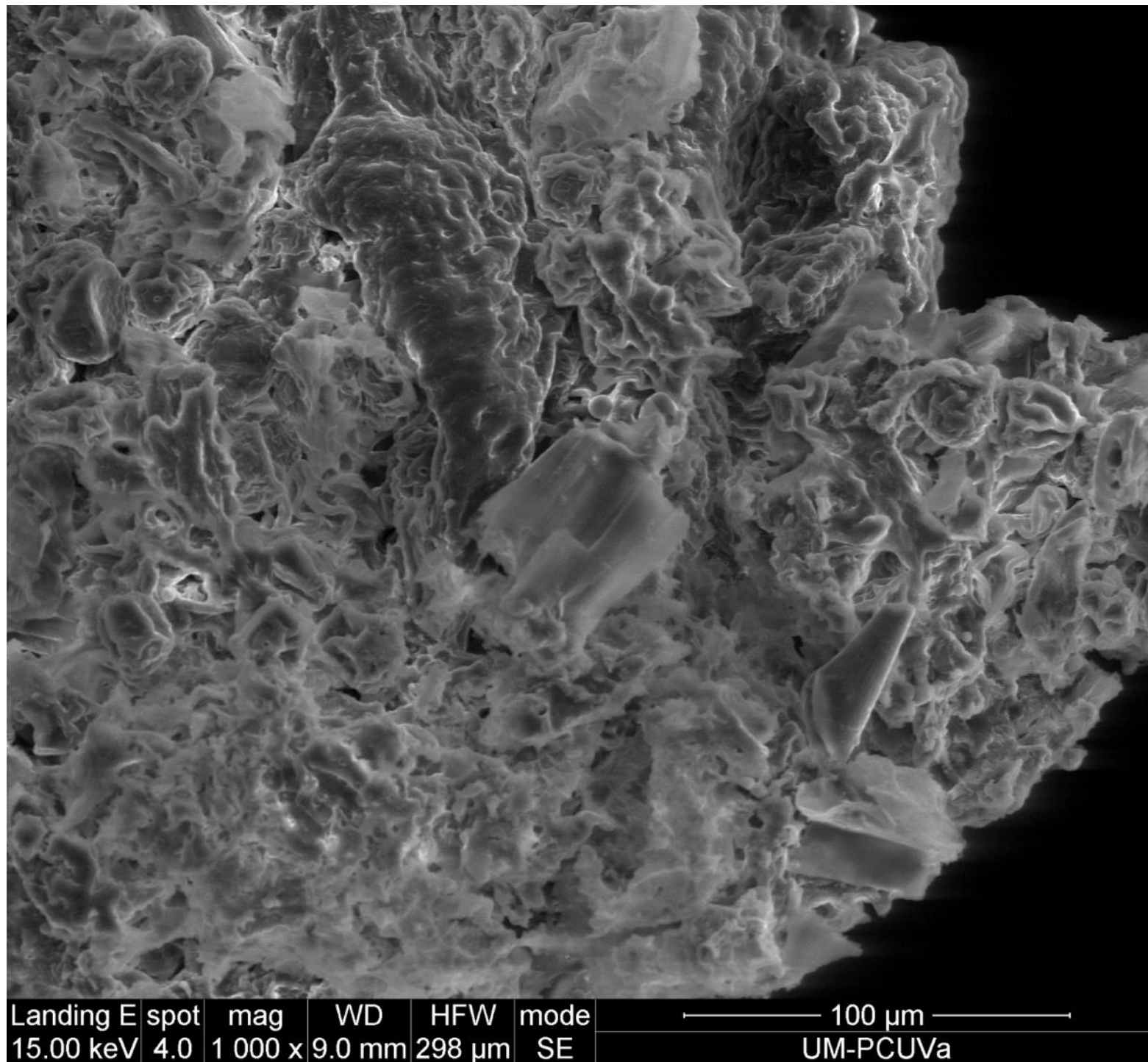
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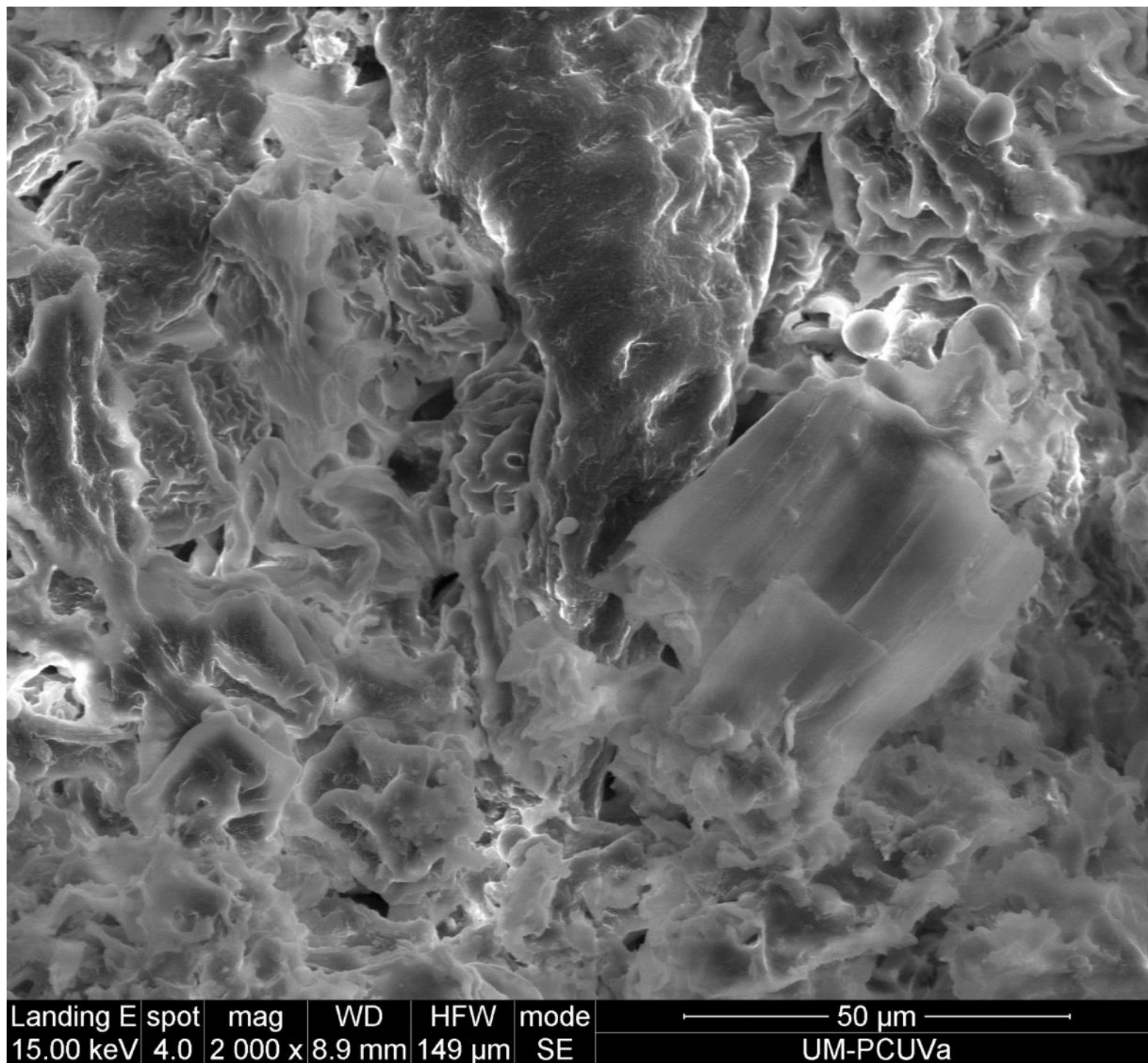


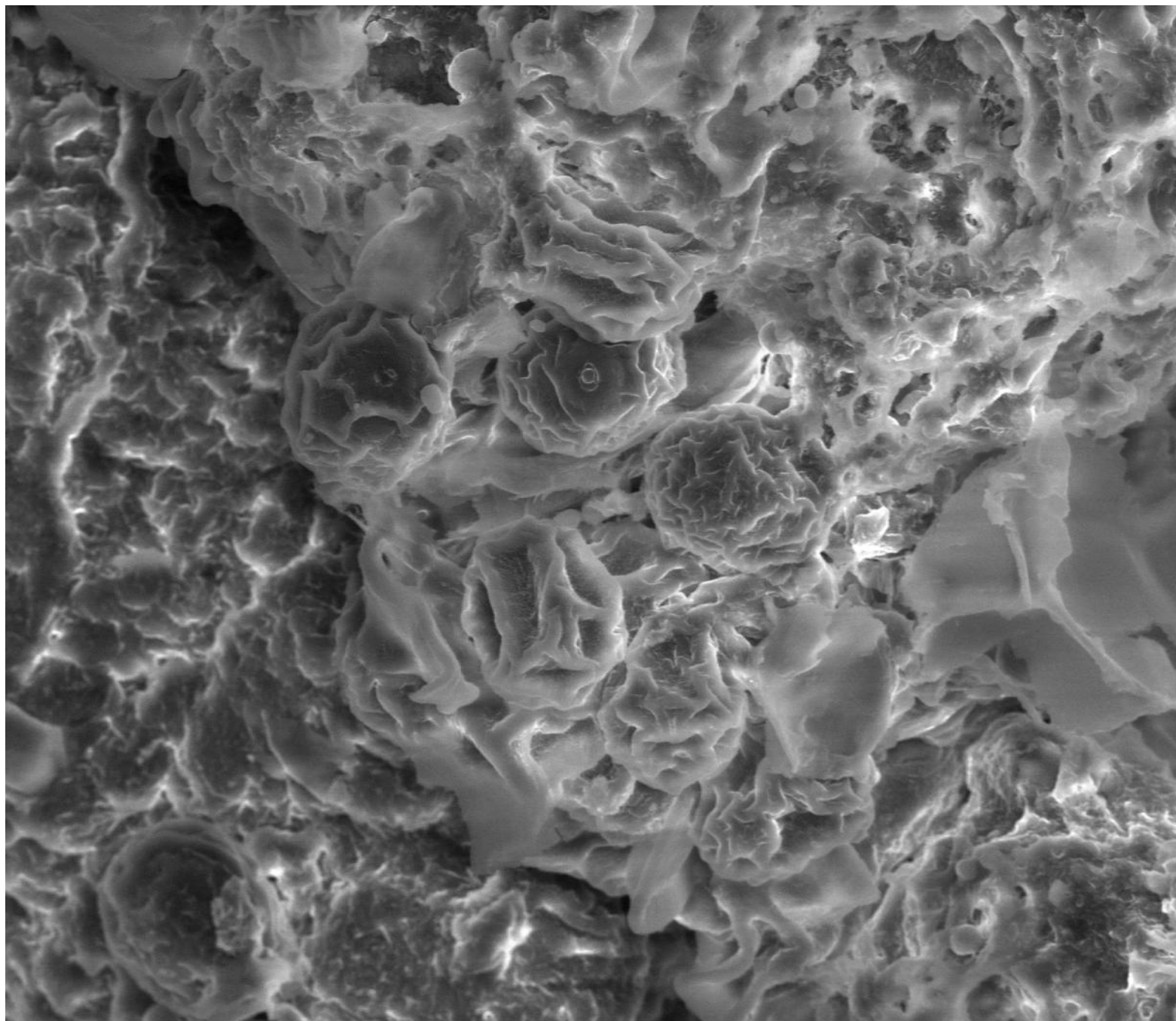
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15.00 keV	4.0	3 000 x	9.0 mm	99.5 μ m	SE

40 μ m

UM-PCUVa



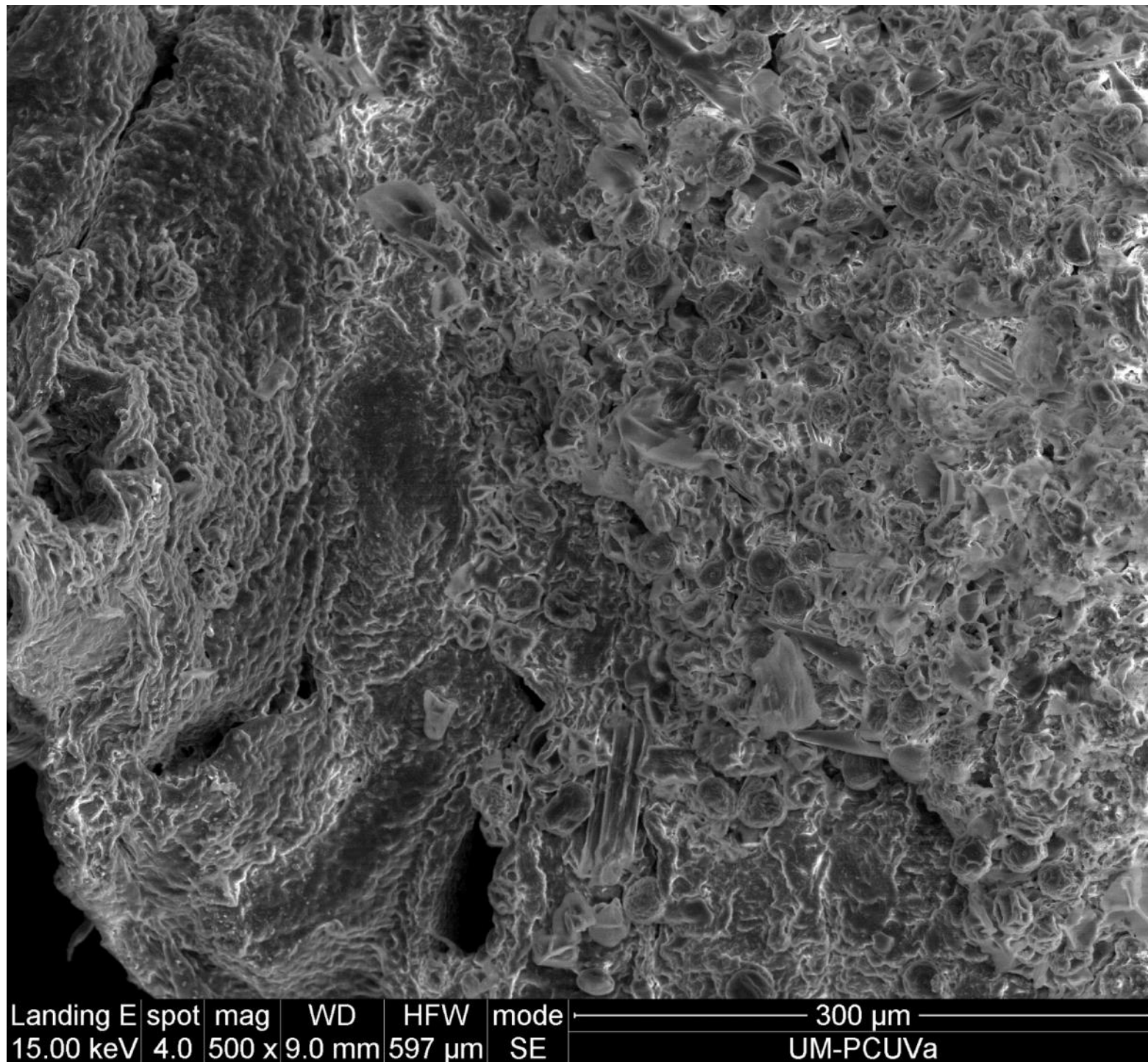


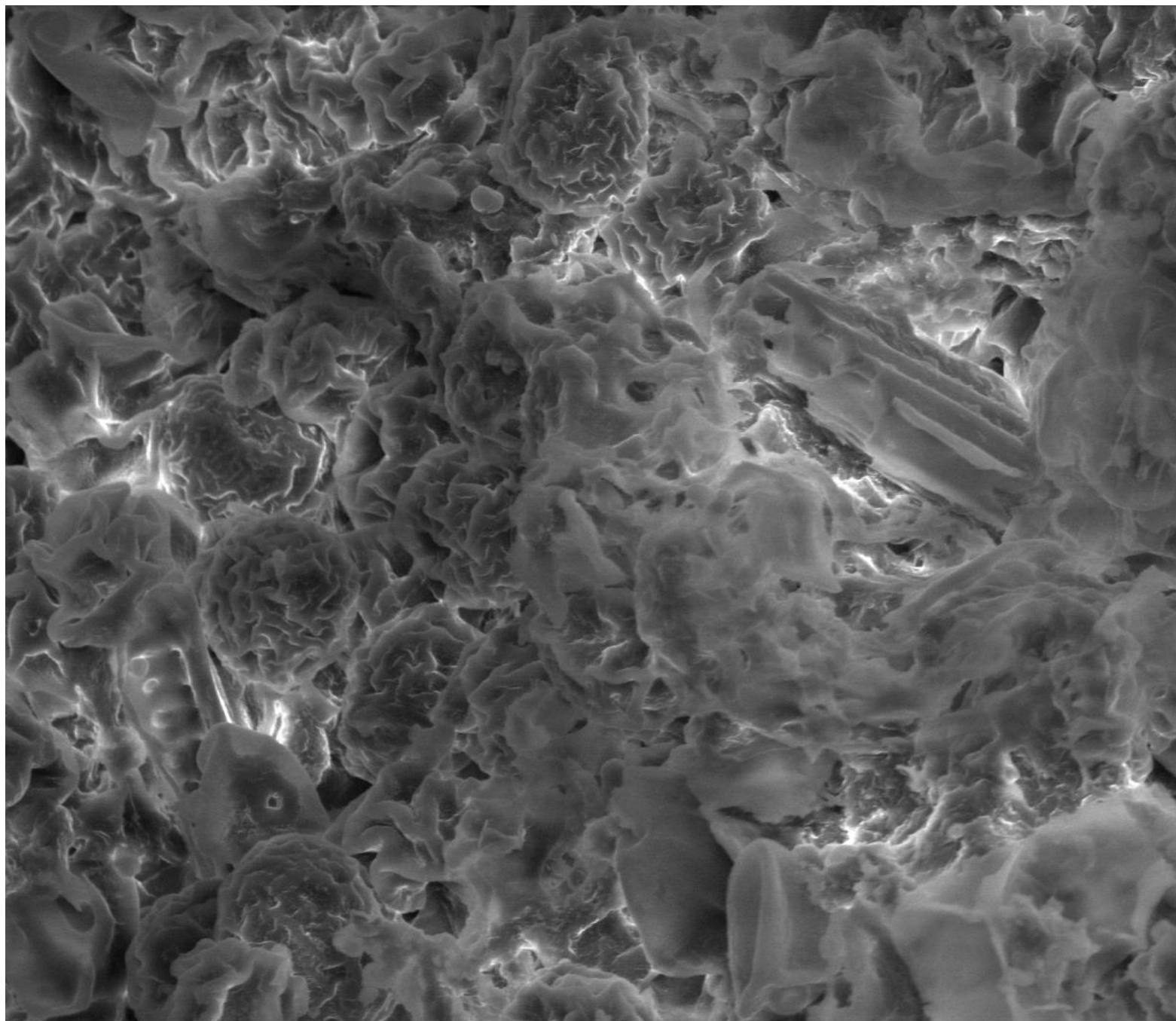


Landing E	spot	mag	WD	HFW	mode
15.00 keV	4.0	2 000 x	9.0 mm	149 μ m	SE

50 μ m

UM-PCUVa





Landing E	spot	mag	WD	HFW	mode
15.00 keV	4.0	2 000 x	9.0 mm	149 μm	SE

50 μm
UM-PCUVa