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Scanning electron microscope images of *Dunaliella tertiolecta* isolates of small cellular particles

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Abstract: Scanning electron microscope images of small cellular particles isolated from conditioned media of microalgae *Dunaliella tertiolecta* are presented. Each image is supplemented by description of the preparation of the sample and the data on the imaging technique and equipment.

The data curators of the repository are Veronika Kralj-Iglič and Anna Romolo. More data on experiments with microalgae small cellular particles can be found in (Adamo et al., 2021), (Picciotto et al., 2021) and (Božič et al., 2022).

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Keywords: Extracellular vesicles, Extracellular particles, Nanoalgosomes, Exosomes

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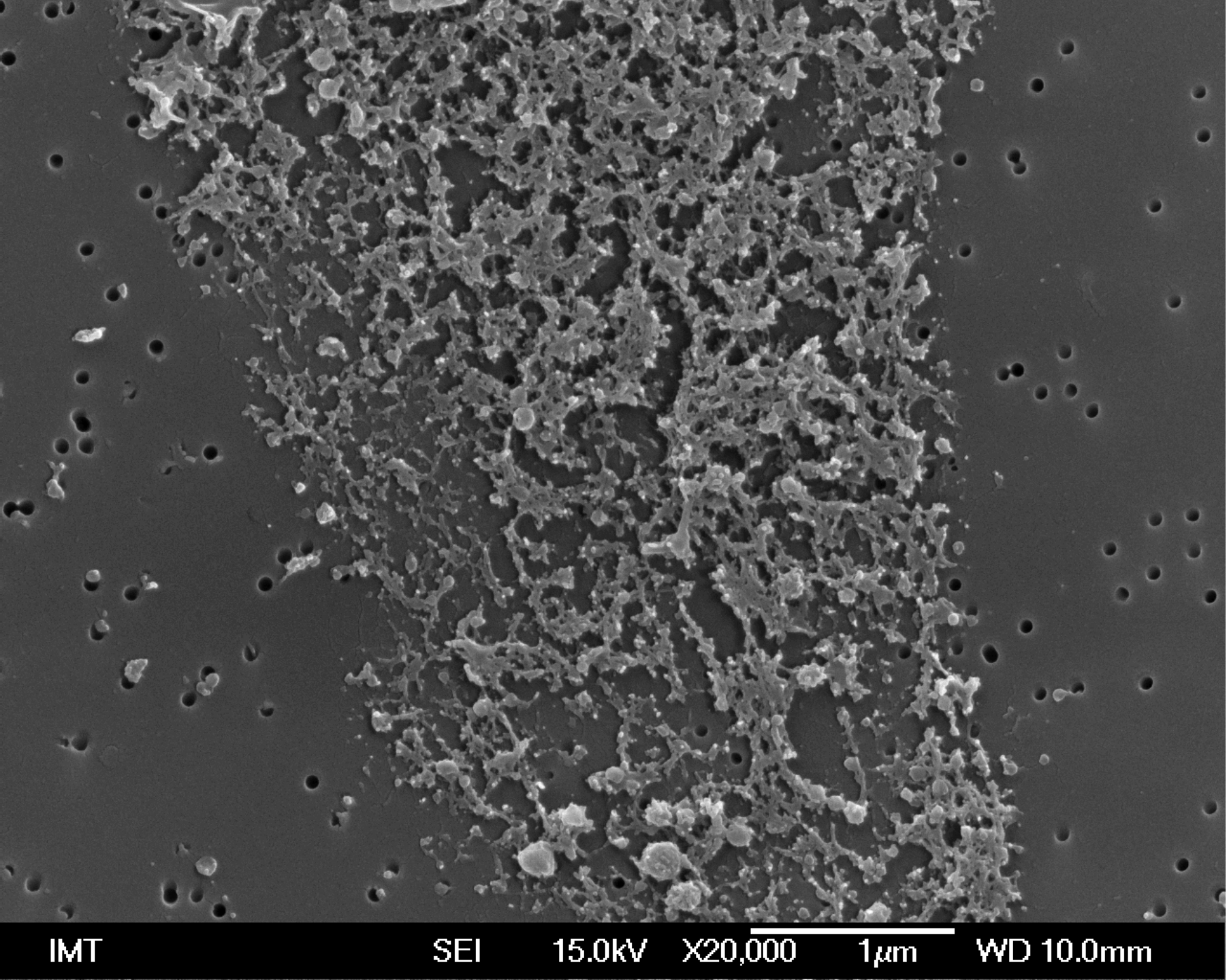

Figure *Dunaliella tertiolecta* isolate 100.000g SEM 1.

Isolation of nanoparticles (NPs)
NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. Then, the cell-depleted medium was centrifuged twice at 10 000 g and 4°C for 30 min (Beckman L8-70M ultracentrifuge, rotor SW55Ti (Beckman Coulter, USA)), using thin-wall polypropylene centrifuge tubes (ref. 326819, Beckman Coulter, USA) to remove larger cell debris. Finally, NPs were pelleted by ultracentrifugation at 118 000 g and 4°C, for 70 min in the same type of ultracentrifuge and ultracentrifuge tubes.
Scanning Electron Microscopy (SEM)
Samples were loaded onto 0.05-micron MCE filters (MF-Millipore™, ref. VMWP01300) and incubated in 2% OsO_4 for two hours. Then the osmium was removed, and the filter was taken out from the holder and further treated in a 24-well plate by changing the bath solution. After washing three times in distilled water, the samples were dehydrated in a graded series of ethanol (30%, 50%, 70%, 80%, 90%, absolute), treated with hexamethyldisilazane (30%, 50% mixtures with absolute ethanol, followed by pure hexamethyldisilazane), and air-dried. Samples were sputtered with Au/Pd (PECS Gatan 682) and examined with a JSM-6500F Field Emission Scanning Electron Microscope (JEOL Ltd., Tokyo, Japan).

From https://zenodo.org/record/6908895. Image 11

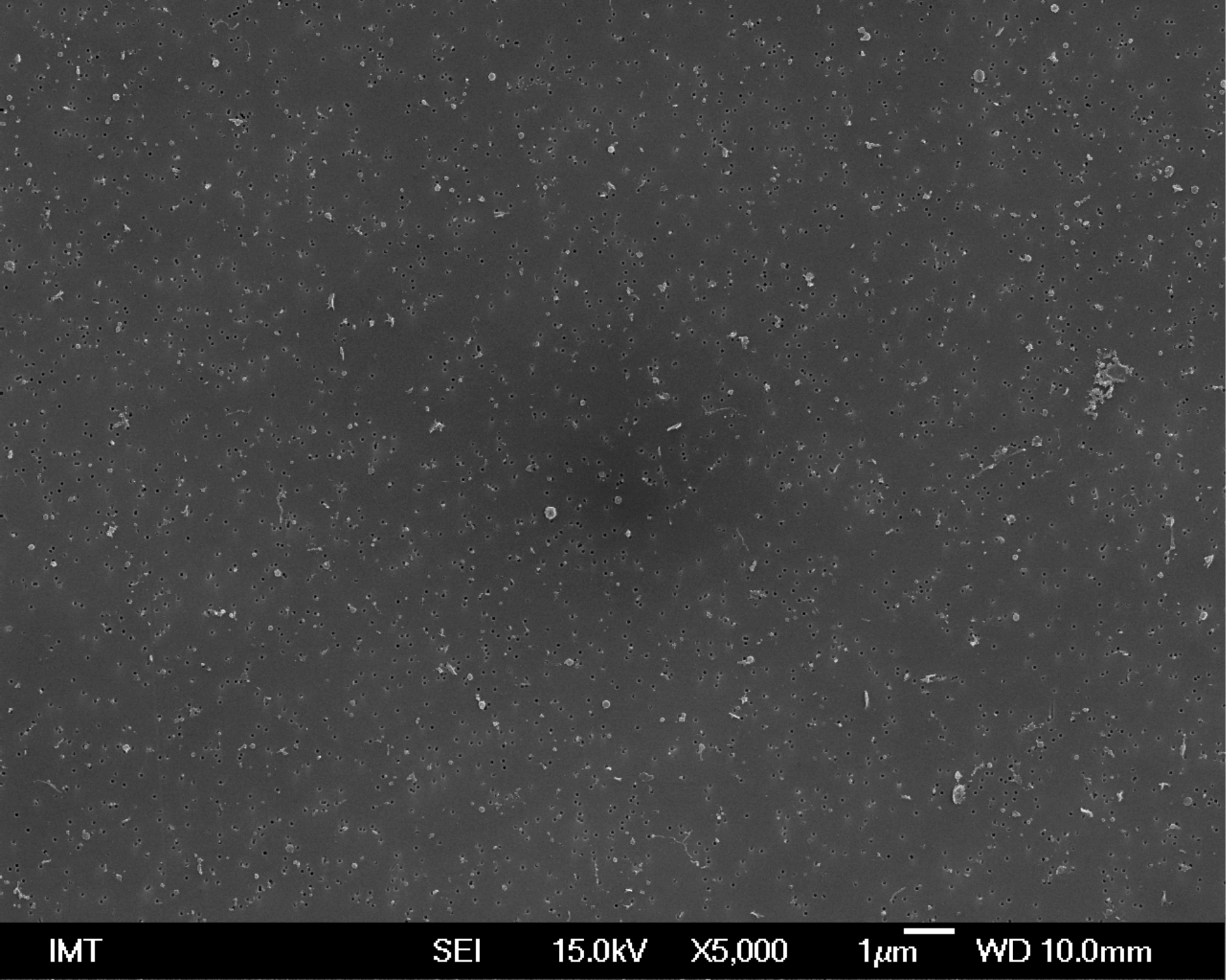

Figure *Dunaliela tertiolecta* isolate 100.000g SEM 2.

Isolation of nanoparticles (NPs)
NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. Then, the cell-depleted medium was centrifuged twice at 10 000 g and 4°C for 30 min (Beckman L8-70M ultracentrifuge, rotor SW55Ti (Beckman Coulter, USA)), using thin-wall polypropylene centrifuge tubes (ref. 326819, Beckman Coulter, USA) to remove larger cell debris. Finally, NPs were pelleted by ultracentrifugation at 118 000 g and 4°C, for 70 min in the same type of ultracentrifuge and ultracentrifuge tubes.
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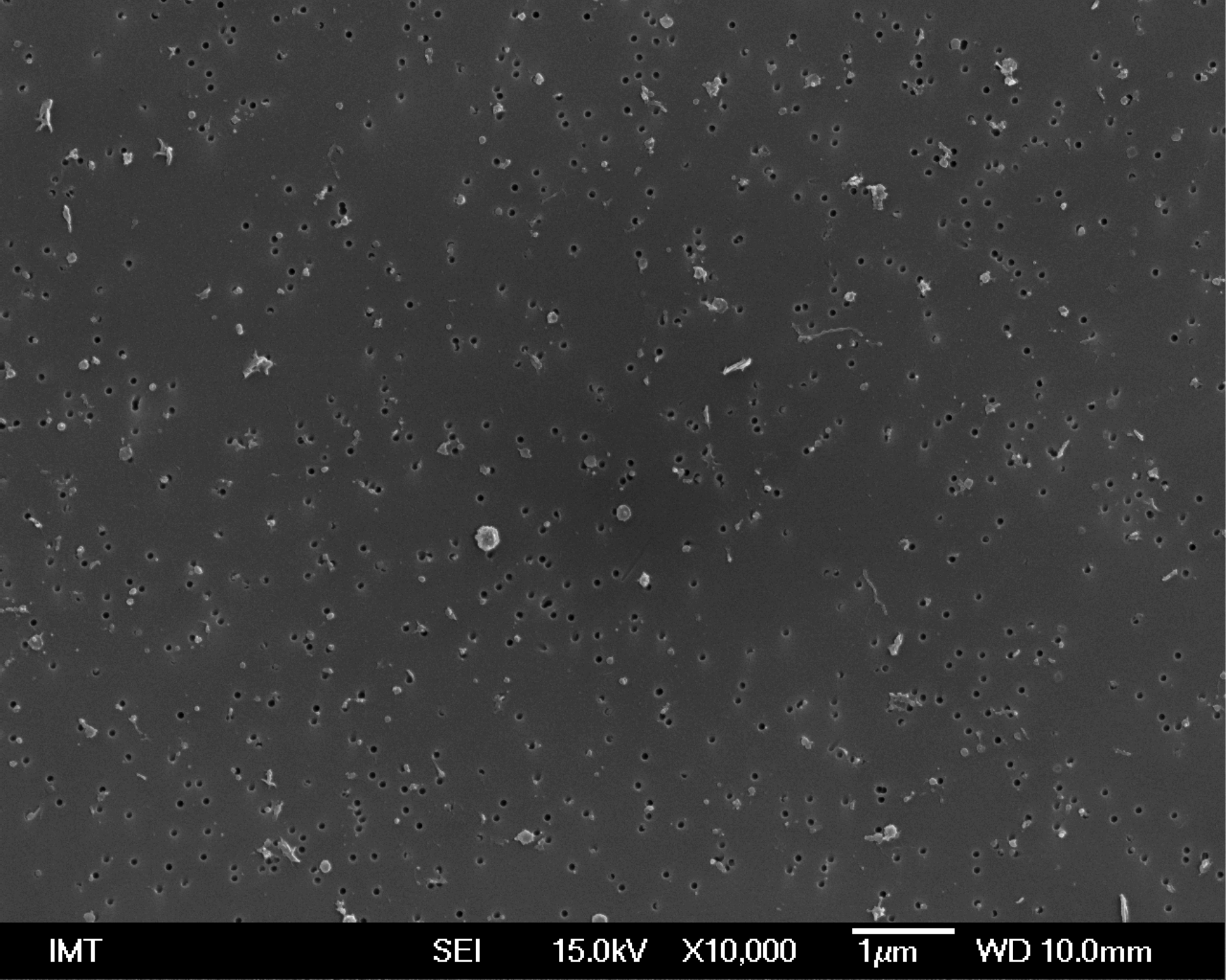

Figure *Dunaliella tertiolecta* isolate 100.000g SEM 3.

Isolation of nanoparticles (NPs)
NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. Then, the cell-depleted medium was centrifuged twice at 10 000 g and 4°C for 30 min (Beckman L8-70M ultracentrifuge, rotor SW55Ti (Beckman Coulter, USA)), using thin-wall polypropylene centrifuge tubes (ref. 326819, Beckman Coulter, USA) to remove larger cell debris. Finally, NPs were pelleted by ultracentrifugation at 118 000 g and 4°C, for 70 min in the same type of ultracentrifuge and ultracentrifuge tubes.
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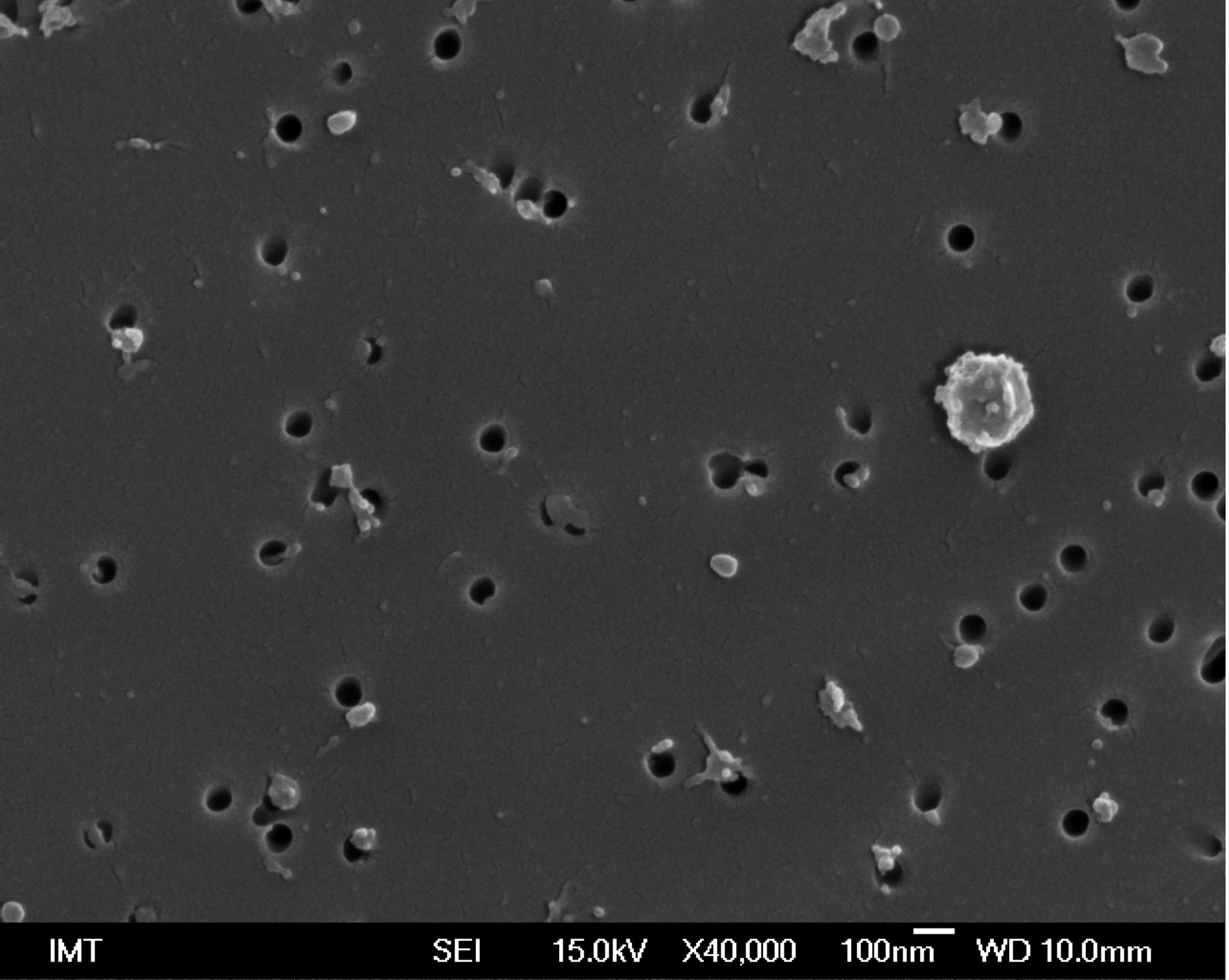

Figure *Dunaliella tertiolecta* isolate 100.000g SEM 4.

Isolation of nanoparticles (NPs)
NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. Then, the cell-depleted medium was centrifuged twice at 10 000 g and 4°C for 30 min (Beckman L8-70M ultracentrifuge, rotor SW55Ti (Beckman Coulter, USA)), using thin-wall polypropylene centrifuge tubes (ref. 326819, Beckman Coulter, USA) to remove larger cell debris. Finally, NPs were pelleted by ultracentrifugation at 118 000 g and 4°C, for 70 min in the same type of ultracentrifuge and ultracentrifuge tubes.
Scanning Electron Microscopy (SEM)
Samples were loaded onto 0.05-micron MCE filters (MF-Millipore™, ref. VMWP01300) and incubated in 2% OsO_4 for two hours. Then the osmium was removed, and the filter was taken out from the holder and further treated in a 24-well plate by changing the bath solution. After washing three times in distilled water, the samples were dehydrated in a graded series of ethanol (30%, 50%, 70%, 80%, 90%, absolute), treated with hexamethyldisilazane (30%, 50% mixtures with absolute ethanol, followed by pure hexamethyldisilazane), and air-dried. Samples were sputtered with Au/Pd (PECS Gatan 682) and examined with a JSM-6500F Field Emission Scanning Electron Microscope (JEOL Ltd., Tokyo, Japan).

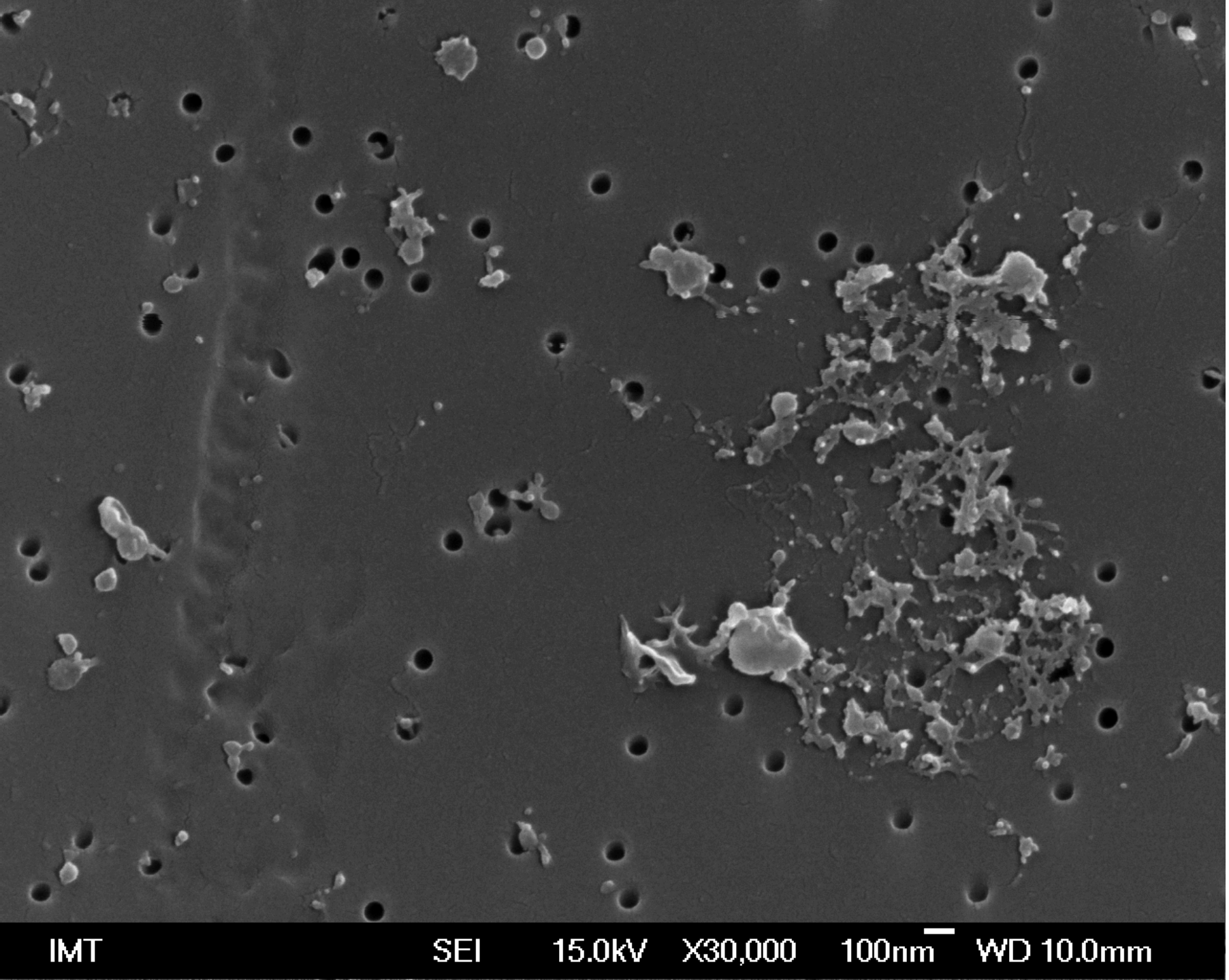

Figure *Dunaliela tertiolecta* isolate 100.000g SEM 5.

Isolation of nanoparticles (NPs)
NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. Then, the cell-depleted medium was centrifuged twice at 10 000 g and 4°C for 30 min (Beckman L8-70M ultracentrifuge, rotor SW55Ti (Beckman Coulter, USA)), using thin-wall polypropylene centrifuge tubes (ref. 326819, Beckman Coulter, USA) to remove larger cell debris. Finally, NPs were pelleted by ultracentrifugation at 118 000 g and 4°C, for 70 min in the same type of ultracentrifuge and ultracentrifuge tubes.
Scanning Electron Microscopy (SEM)
Samples were loaded onto 0.05-micron MCE filters (MF-Millipore™, ref. VMWP01300) and incubated in 2% OsO_4 for two hours. Then the osmium was removed, and the filter was taken out from the holder and further treated in a 24-well plate by changing the bath solution. After washing three times in distilled water, the samples were dehydrated in a graded series of ethanol (30%, 50%, 70%, 80%, 90%, absolute), treated with hexamethyldisilazane (30%, 50% mixtures with absolute ethanol, followed by pure hexamethyldisilazane), and air-dried. Samples were sputtered with Au/Pd (PECS Gatan 682) and examined with a JSM-6500F Field Emission Scanning Electron Microscope (JEOL Ltd., Tokyo, Japan).

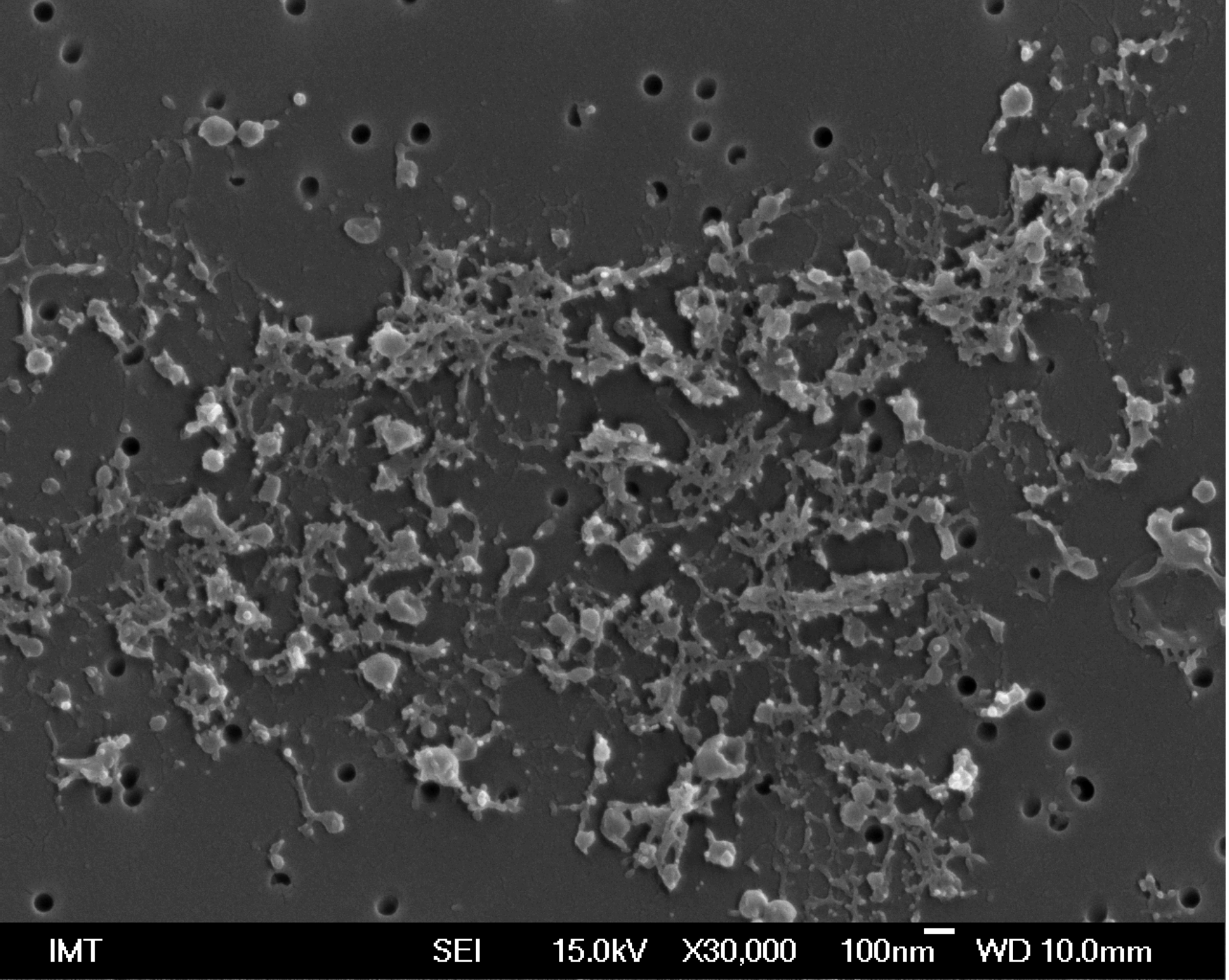

Figure *Dunaliela tertiolecta* isolate 100.000g SEM 6.

Isolation of nanoparticles (NPs)
NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. Then, the cell-depleted medium was centrifuged twice at 10 000 g and 4°C for 30 min (Beckman L8-70M ultracentrifuge, rotor SW55Ti (Beckman Coulter, USA)), using thin-wall polypropylene centrifuge tubes (ref. 326819, Beckman Coulter, USA) to remove larger cell debris. Finally, NPs were pelleted by ultracentrifugation at 118 000 g and 4°C, for 70 min in the same type of ultracentrifuge and ultracentrifuge tubes.
Scanning Electron Microscopy (SEM)
Samples were loaded onto 0.05-micron MCE filters (MF-Millipore™, ref. VMWP01300) and incubated in 2% OsO_4 for two hours. Then the osmium was removed, and the filter was taken out from the holder and further treated in a 24-well plate by changing the bath solution. After washing three times in distilled water, the samples were dehydrated in a graded series of ethanol (30%, 50%, 70%, 80%, 90%, absolute), treated with hexamethyldisilazane (30%, 50% mixtures with absolute ethanol, followed by pure hexamethyldisilazane), and air-dried. Samples were sputtered with Au/Pd (PECS Gatan 682) and examined with a JSM-6500F Field Emission Scanning Electron Microscope (JEOL Ltd., Tokyo, Japan).

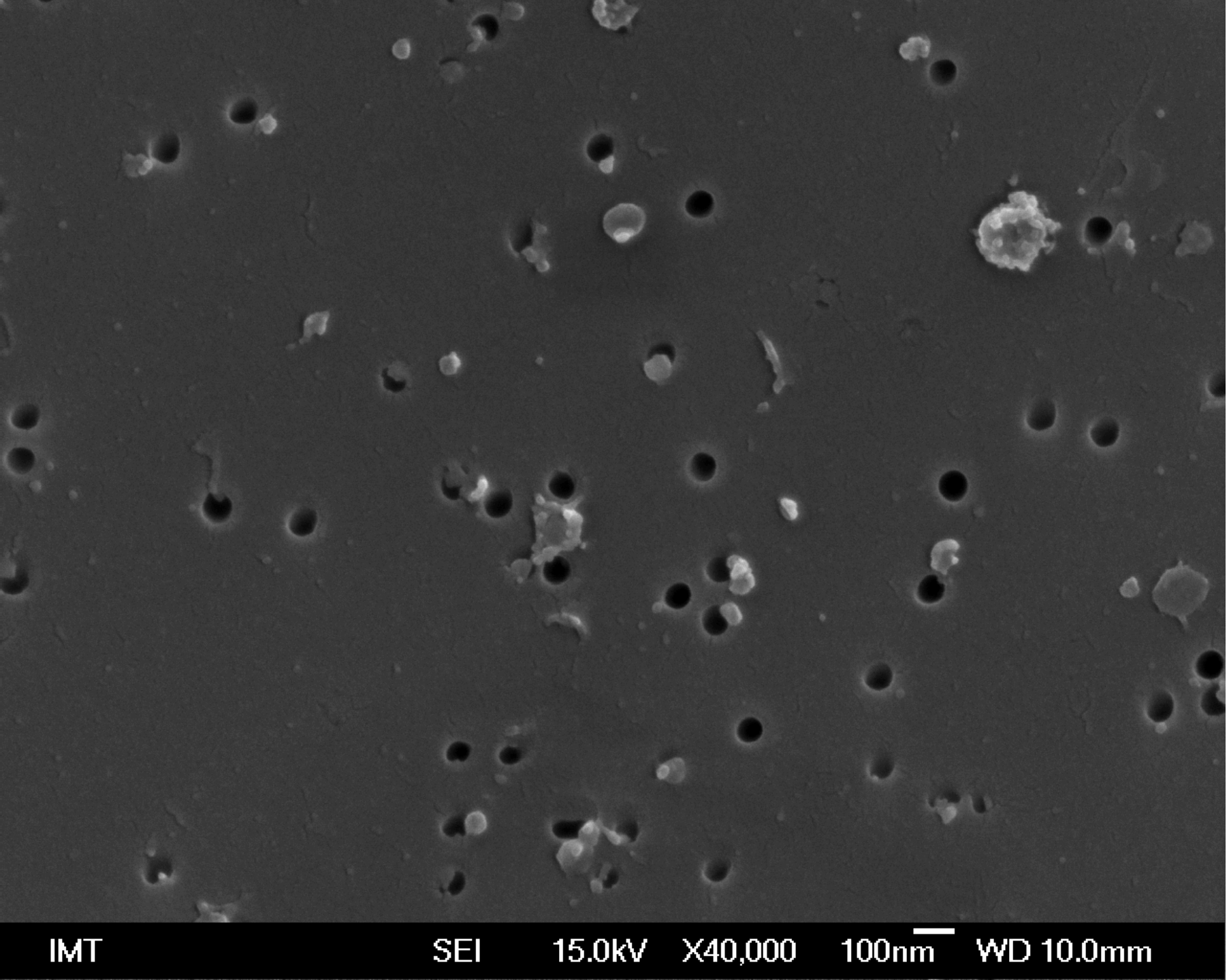

Figure *Dunaliella tertiolecta* isolate 100.000g SEM 7.

Isolation of nanoparticles (NPs)
NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. Then, the cell-depleted medium was centrifuged twice at 10 000 g and 4°C for 30 min (Beckman L8-70M ultracentrifuge, rotor SW55Ti (Beckman Coulter, USA)), using thin-wall polypropylene centrifuge tubes (ref. 326819, Beckman Coulter, USA) to remove larger cell debris. Finally, NPs were pelleted by ultracentrifugation at 118 000 g and 4°C, for 70 min in the same type of ultracentrifuge and ultracentrifuge tubes.
Scanning Electron Microscopy (SEM)
Samples were loaded onto 0.05-micron MCE filters (MF-Millipore™, ref. VMWP01300) and incubated in 2% OsO_4 for two hours. Then the osmium was removed, and the filter was taken out from the holder and further treated in a 24-well plate by changing the bath solution. After washing three times in distilled water, the samples were dehydrated in a graded series of ethanol (30%, 50%, 70%, 80%, 90%, absolute), treated with hexamethyldisilazane (30%, 50% mixtures with absolute ethanol, followed by pure hexamethyldisilazane), and air-dried. Samples were sputtered with Au/Pd (PECS Gatan 682) and examined with a JSM-6500F Field Emission Scanning Electron Microscope (JEOL Ltd., Tokyo, Japan).

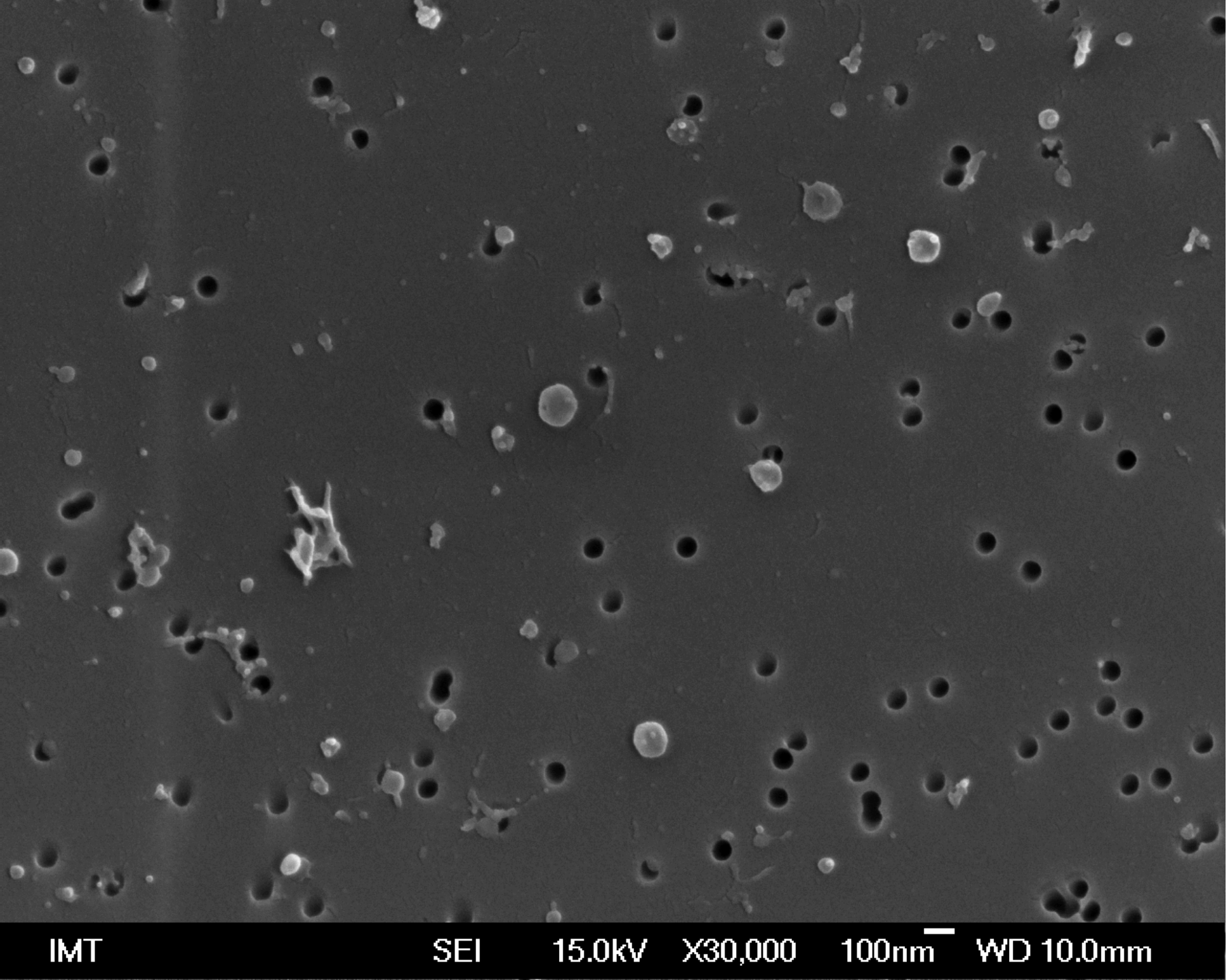

Figure *Dunaliela tertiolecta* isolate 100.000g SEM 8.

Isolation of nanoparticles (NPs)
NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. Then, the cell-depleted medium was centrifuged twice at 10 000 g and 4°C for 30 min (Beckman L8-70M ultracentrifuge, rotor SW55Ti (Beckman Coulter, USA)), using thin-wall polypropylene centrifuge tubes (ref. 326819, Beckman Coulter, USA) to remove larger cell debris. Finally, NPs were pelleted by ultracentrifugation at 118 000 g and 4°C, for 70 min in the same type of ultracentrifuge and ultracentrifuge tubes.
Scanning Electron Microscopy (SEM)
Samples were loaded onto 0.05-micron MCE filters (MF-Millipore™, ref. VMWP01300) and incubated in 2% OsO_4 for two hours. Then the osmium was removed, and the filter was taken out from the holder and further treated in a 24-well plate by changing the bath solution. After washing three times in distilled water, the samples were dehydrated in a graded series of ethanol (30%, 50%, 70%, 80%, 90%, absolute), treated with hexamethyldisilazane (30%, 50% mixtures with absolute ethanol, followed by pure hexamethyldisilazane), and air-dried. Samples were sputtered with Au/Pd (PECS Gatan 682) and examined with a JSM-6500F Field Emission Scanning Electron Microscope (JEOL Ltd., Tokyo, Japan).

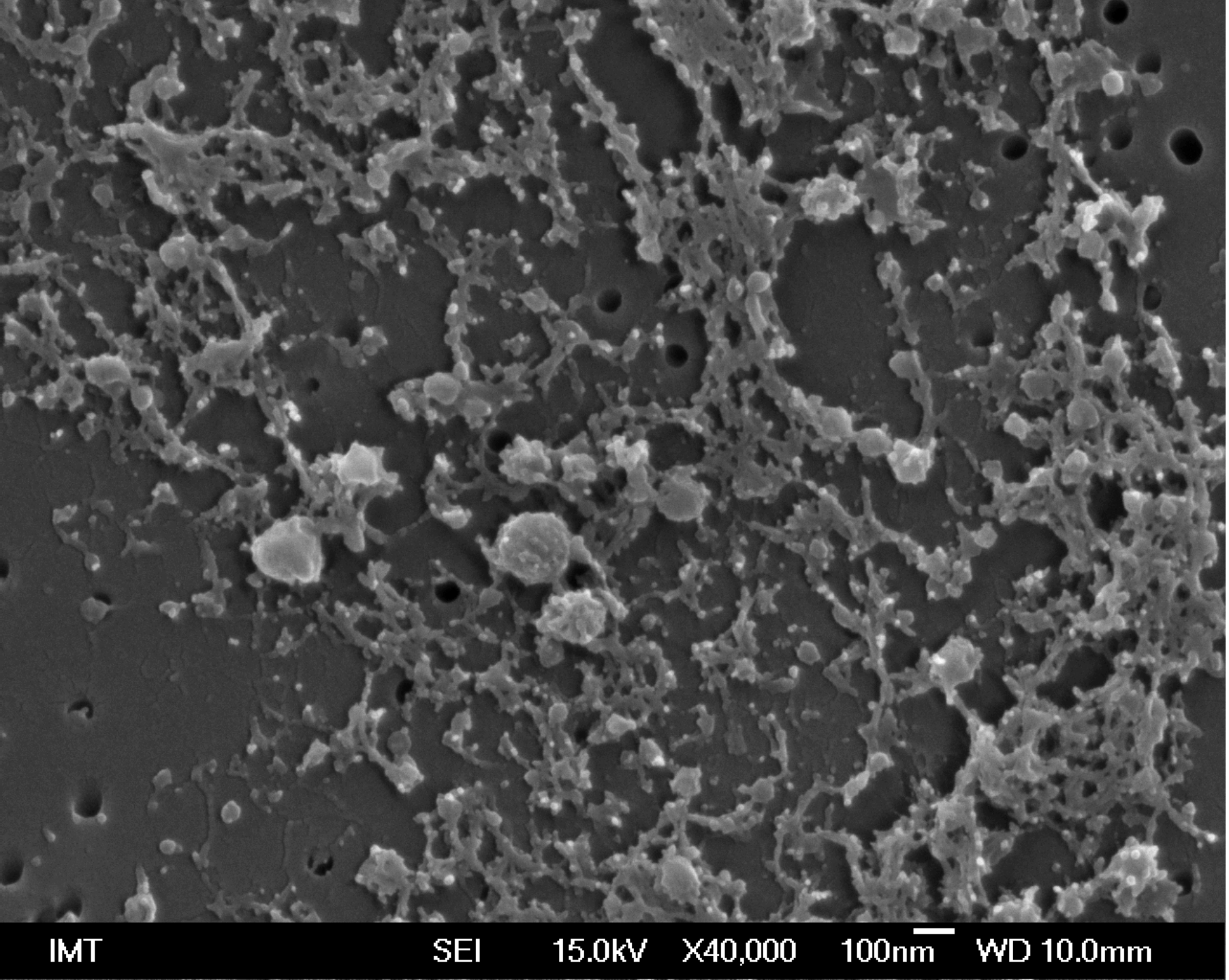

Figure *Dunaliela tertiolecta* isolate 100.000g SEM 9.

Isolation of nanoparticles (NPs)
NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. Then, the cell-depleted medium was centrifuged twice at 10 000 g and 4°C for 30 min (Beckman L8-70M ultracentrifuge, rotor SW55Ti (Beckman Coulter, USA)), using thin-wall polypropylene centrifuge tubes (ref. 326819, Beckman Coulter, USA) to remove larger cell debris. Finally, NPs were pelleted by ultracentrifugation at 118 000 g and 4°C, for 70 min in the same type of ultracentrifuge and ultracentrifuge tubes.
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Samples were loaded onto 0.05-micron MCE filters (MF-Millipore™, ref. VMWP01300) and incubated in 2% OsO_4 for two hours. Then the osmium was removed, and the filter was taken out from the holder and further treated in a 24-well plate by changing the bath solution. After washing three times in distilled water, the samples were dehydrated in a graded series of ethanol (30%, 50%, 70%, 80%, 90%, absolute), treated with hexamethyldisilazane (30%, 50% mixtures with absolute ethanol, followed by pure hexamethyldisilazane), and air-dried. Samples were sputtered with Au/Pd (PECS Gatan 682) and examined with a JSM-6500F Field Emission Scanning Electron Microscope (JEOL Ltd., Tokyo, Japan).

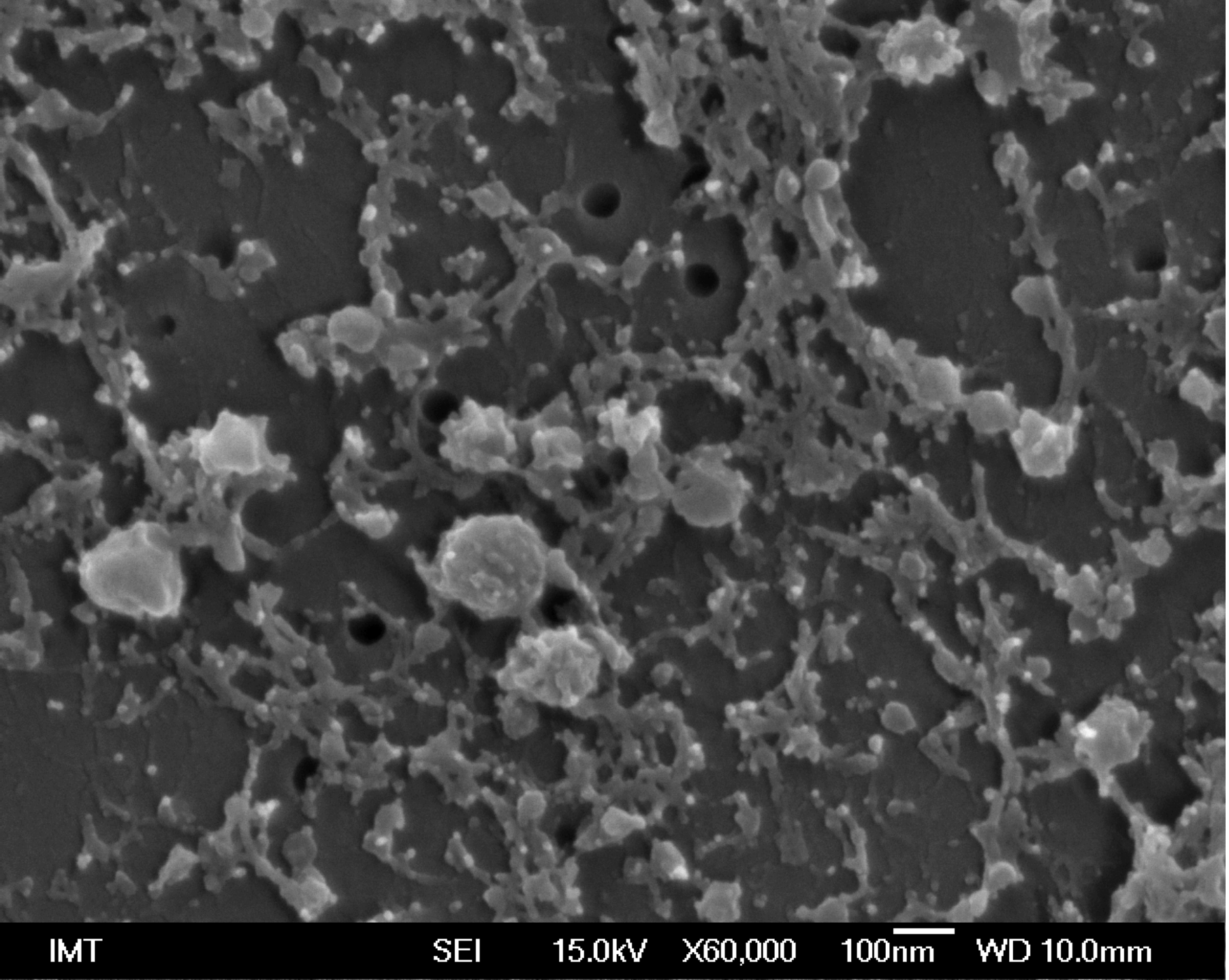

Figure *Dunaliella tertiolecta* isolate 100.000g SEM 10.

Isolation of nanoparticles (NPs)

NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. Then, the cell-depleted medium was centrifuged twice at 10 000 g and 4°C for 30 min (Beckman L8-70M ultracentrifuge, rotor SW55Ti (Beckman Coulter, USA)), using thin-wall polypropylene centrifuge tubes (ref. 326819, Beckman Coulter, USA) to remove larger cell debris. Finally, NPs were pelleted by ultracentrifugation at 118 000 g and 4°C, for 70 min in the same type of ultracentrifuge and ultracentrifuge tubes.

Scanning Electron Microscopy (SEM)

Samples were loaded onto 0.05-micron MCE filters (MF-Millipore™, ref. VMWP01300) and incubated in 2% OsO_4 for two hours. Then the osmium was removed, and the filter was taken out from the holder and further treated in a 24-well plate by changing the bath solution. After washing three times in distilled water, the samples were dehydrated in a graded series of ethanol (30%, 50%, 70%, 80%, 90%, absolute), treated with hexamethyldisilazane (30%, 50% mixtures with absolute ethanol, followed by pure hexamethyldisilazane), and air-dried. Samples were sputtered with Au/Pd (PECS Gatan 682) and examined with a JSM-6500F Field Emission Scanning Electron Microscope (JEOL Ltd., Tokyo, Japan).

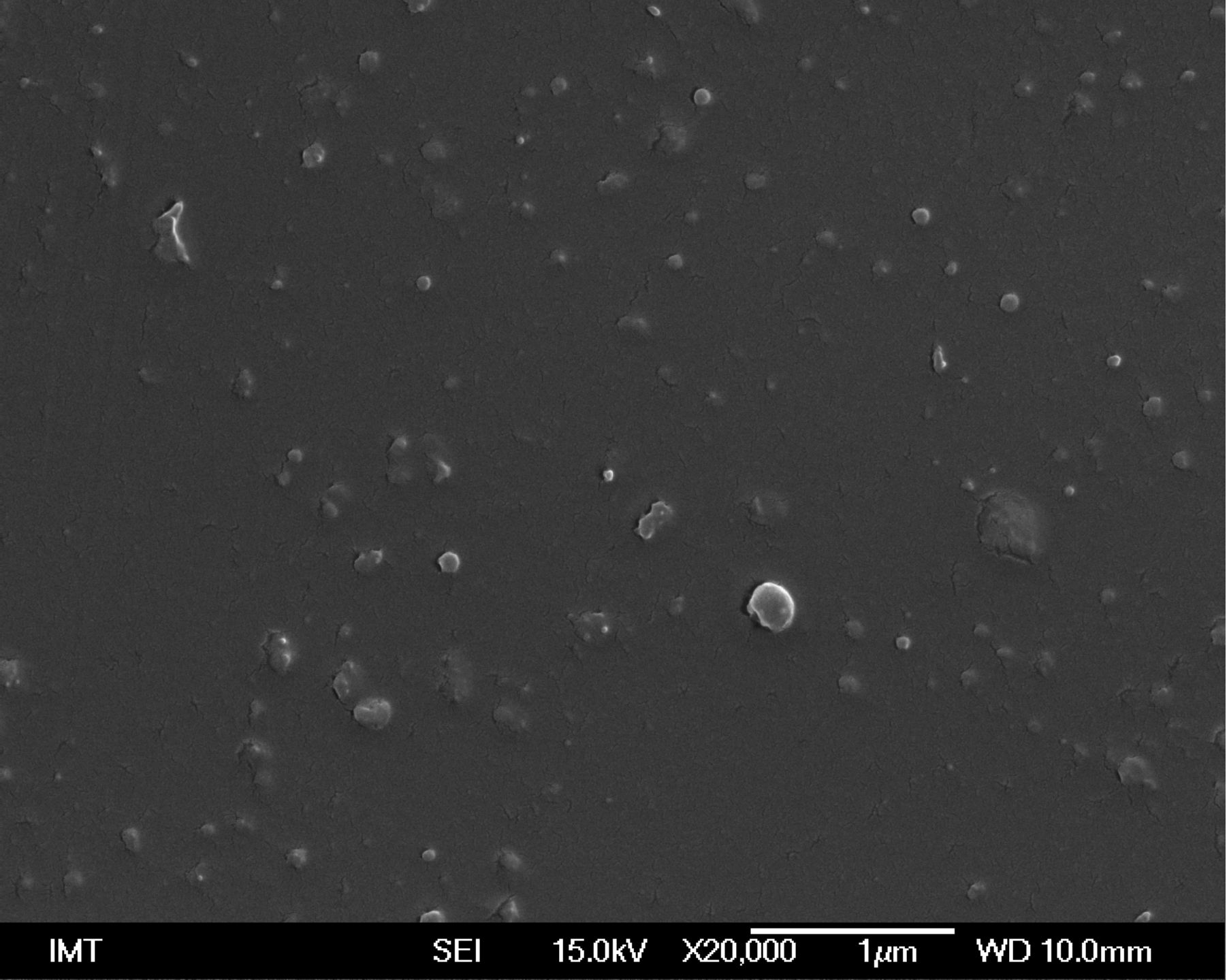

Figure *Dunaliella tertiolecta* isolate 100.000g SEM 11.

Isolation of nanoparticles (NPs)
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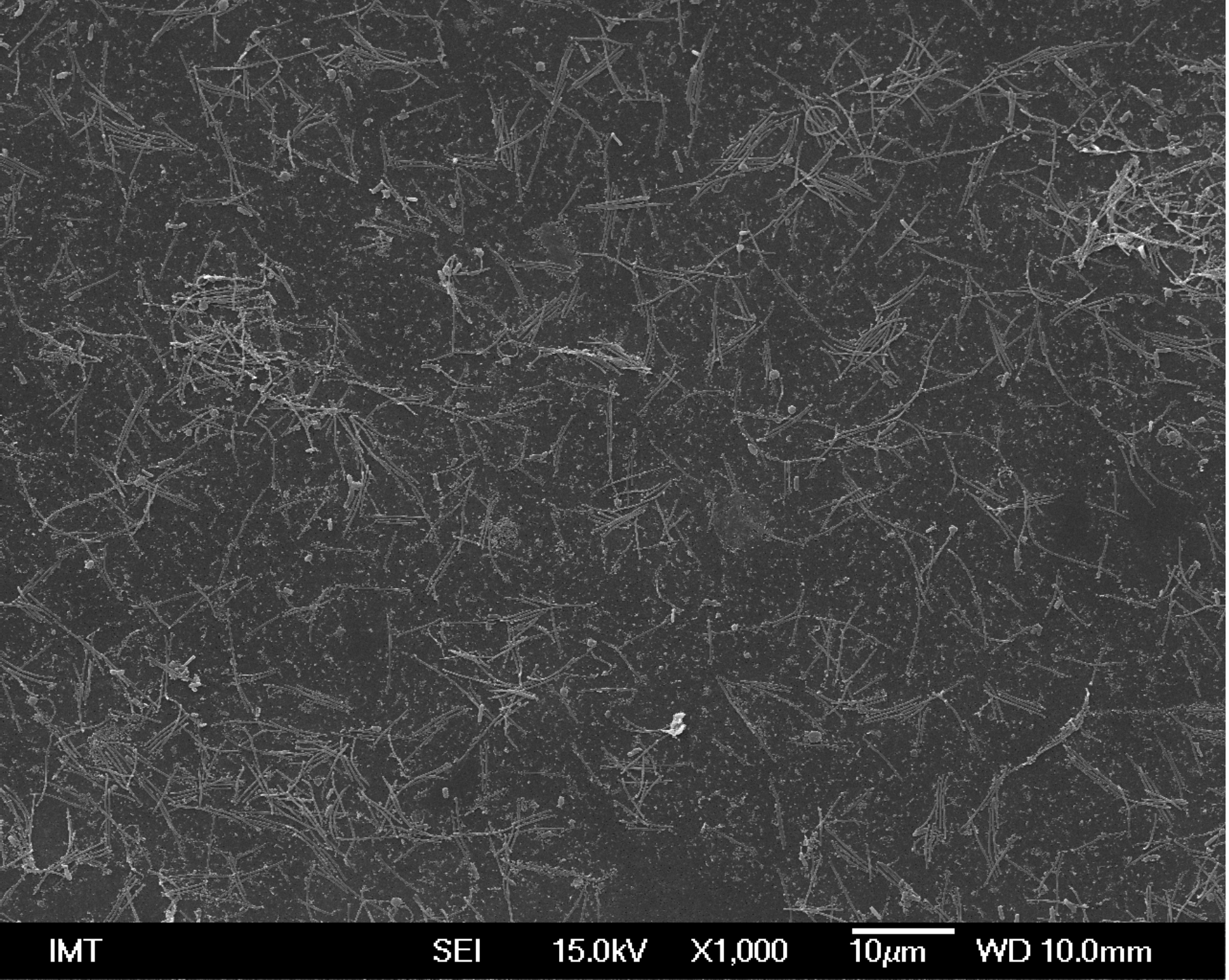

Figure *Dunaliela tertiolecta* isolate 10.000g SEM 1.

Isolation of nanoparticles (NPs)

NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. Then, the cell-depleted medium was centrifuged twice at 10 000 g and 4°C for 30 min (Beckman L8-70M ultracentrifuge, rotor SW55Ti (Beckman Coulter, USA)), using thin-wall polypropylene centrifuge tubes (ref. 326819, Beckman Coulter, USA). The isolate presents the pellet of the second centrifugation.

Scanning Electron Microscopy (SEM)

Samples were loaded onto 0.05-micron MCE filters (MF-Millipore™, ref. VMWP01300) and incubated in 2% OsO_4 for two hours. Then the osmium was removed, and the filter was taken out from the holder and further treated in a 24-well plate by changing the bath solution. After washing three times in distilled water, the samples were dehydrated in a graded series of ethanol (30%, 50%, 70%, 80%, 90%, absolute), treated with hexamethyldisilazane (30%, 50% mixtures with absolute ethanol, followed by pure hexamethyldisilazane), and air-dried. Samples were sputtered with Au/Pd (PECS Gatan 682) and examined with a JSM-6500F Field Emission Scanning Electron Microscope (JEOL Ltd., Tokyo, Japan).

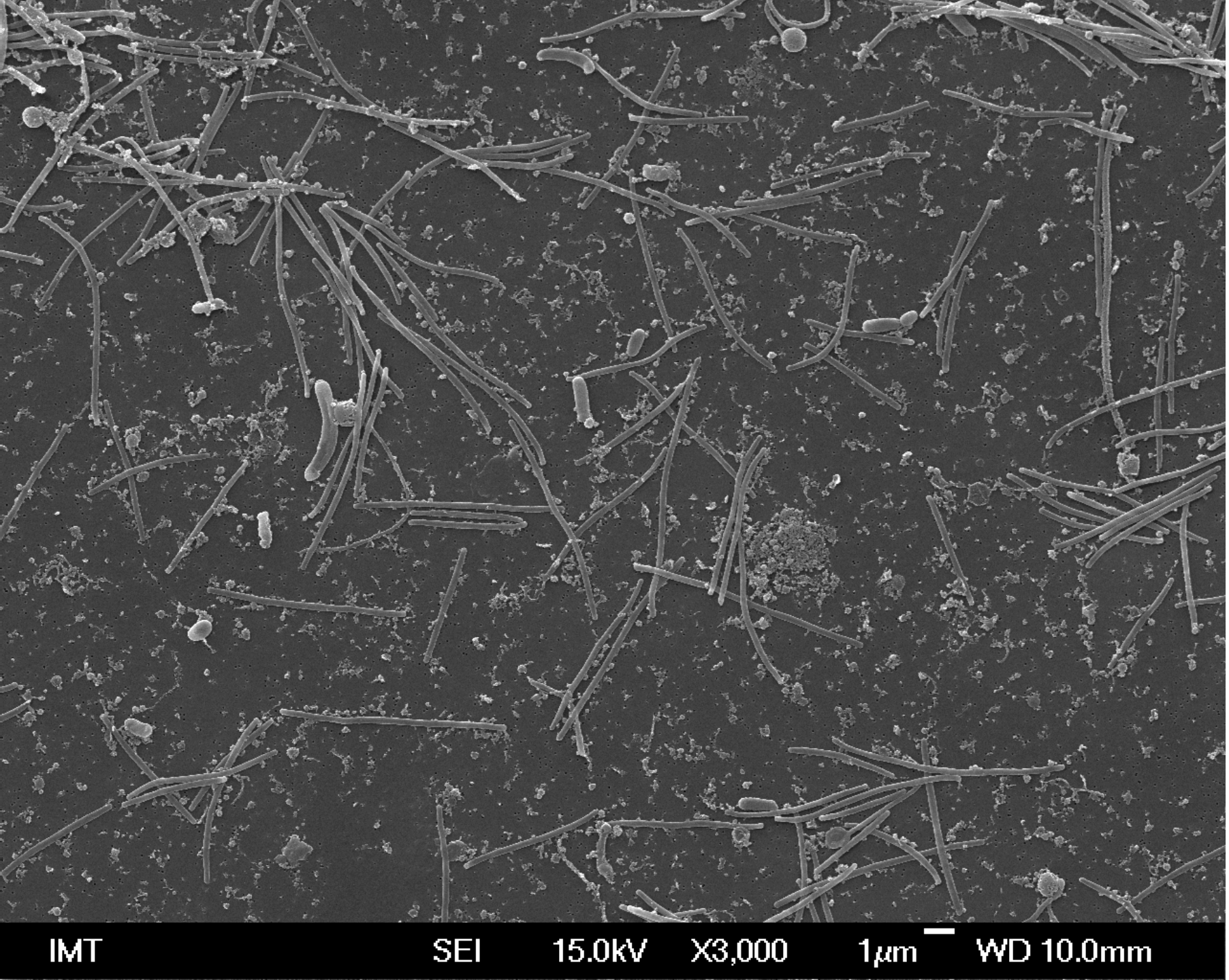

Figure *Dunaliela tertiolecta* isolate 10.000g SEM 2.

Isolation of nanoparticles (NPs)

NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. Then, the cell-depleted medium was centrifuged twice at 10 000 g and 4°C for 30 min (Beckman L8-70M ultracentrifuge, rotor SW55Ti (Beckman Coulter, USA)), using thin-wall polypropylene centrifuge tubes (ref. 326819, Beckman Coulter, USA). The isolate presents the pellet of the second centrifugation.

Scanning Electron Microscopy (SEM)

Samples were loaded onto 0.05-micron MCE filters (MF-Millipore™, ref. VMWP01300) and incubated in 2% OsO_4 for two hours. Then the osmium was removed, and the filter was taken out from the holder and further treated in a 24-well plate by changing the bath solution. After washing three times in distilled water, the samples were dehydrated in a graded series of ethanol (30%, 50%, 70%, 80%, 90%, absolute), treated with hexamethyldisilazane (30%, 50% mixtures with absolute ethanol, followed by pure hexamethyldisilazane), and air-dried. Samples were sputtered with Au/Pd (PECS Gatan 682) and examined with a JSM-6500F Field Emission Scanning Electron Microscope (JEOL Ltd., Tokyo, Japan).

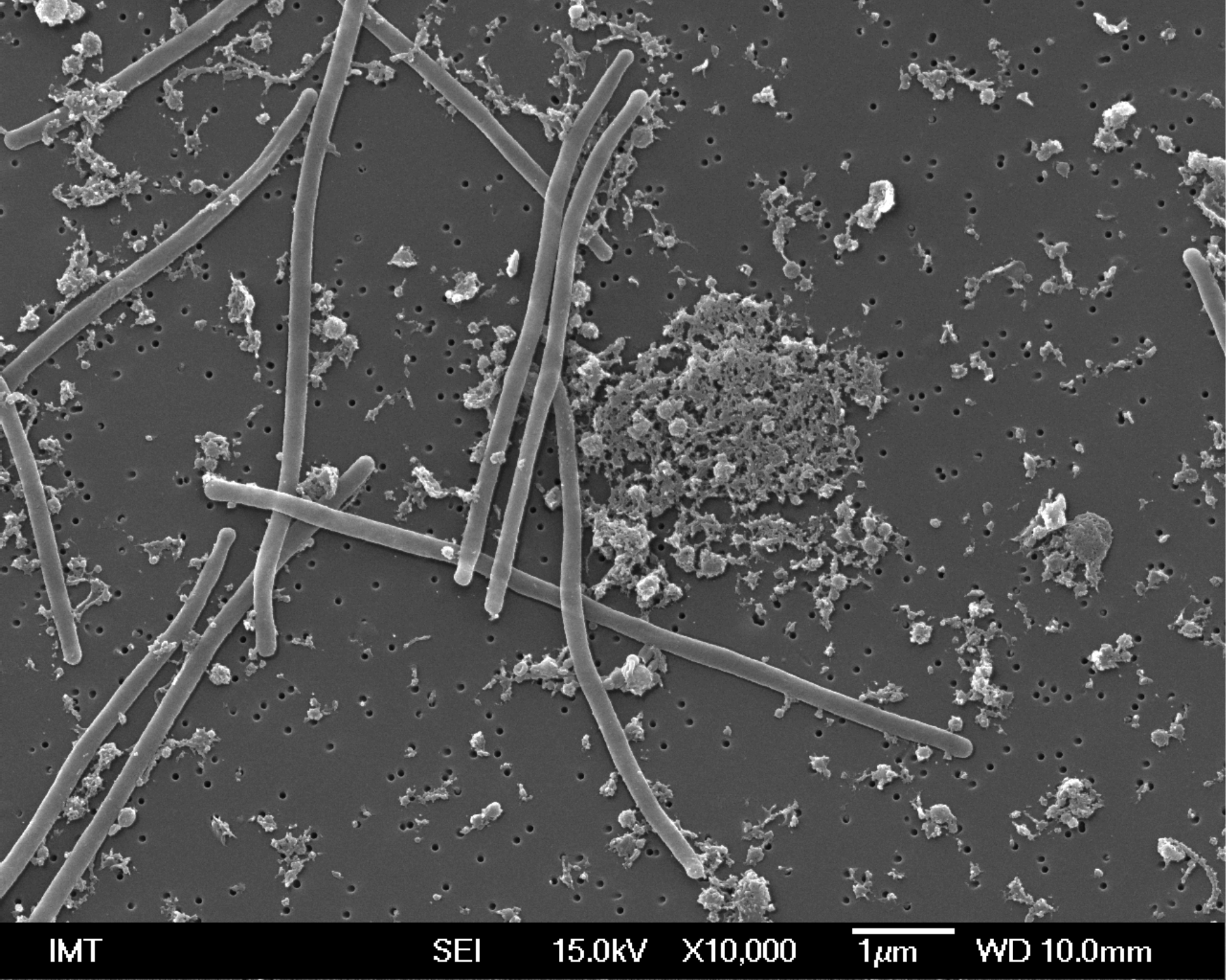

Figure *Dunaliela tertiolecta* isolate 10.000g SEM 3.

Isolation of nanoparticles (NPs)

NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. Then, the cell-depleted medium was centrifuged twice at 10 000 g and 4°C for 30 min (Beckman L8-70M ultracentrifuge, rotor SW55Ti (Beckman Coulter, USA)), using thin-wall polypropylene centrifuge tubes (ref. 326819, Beckman Coulter, USA). The isolate presents the pellet of the second centrifugation.

Scanning Electron Microscopy (SEM)

Samples were loaded onto 0.05-micron MCE filters (MF-Millipore™, ref. VMWP01300) and incubated in 2% OsO_4 for two hours. Then the osmium was removed, and the filter was taken out from the holder and further treated in a 24-well plate by changing the bath solution. After washing three times in distilled water, the samples were dehydrated in a graded series of ethanol (30%, 50%, 70%, 80%, 90%, absolute), treated with hexamethyldisilazane (30%, 50% mixtures with absolute ethanol, followed by pure hexamethyldisilazane), and air-dried. Samples were sputtered with Au/Pd (PECS Gatan 682) and examined with a JSM-6500F Field Emission Scanning Electron Microscope (JEOL Ltd., Tokyo, Japan).

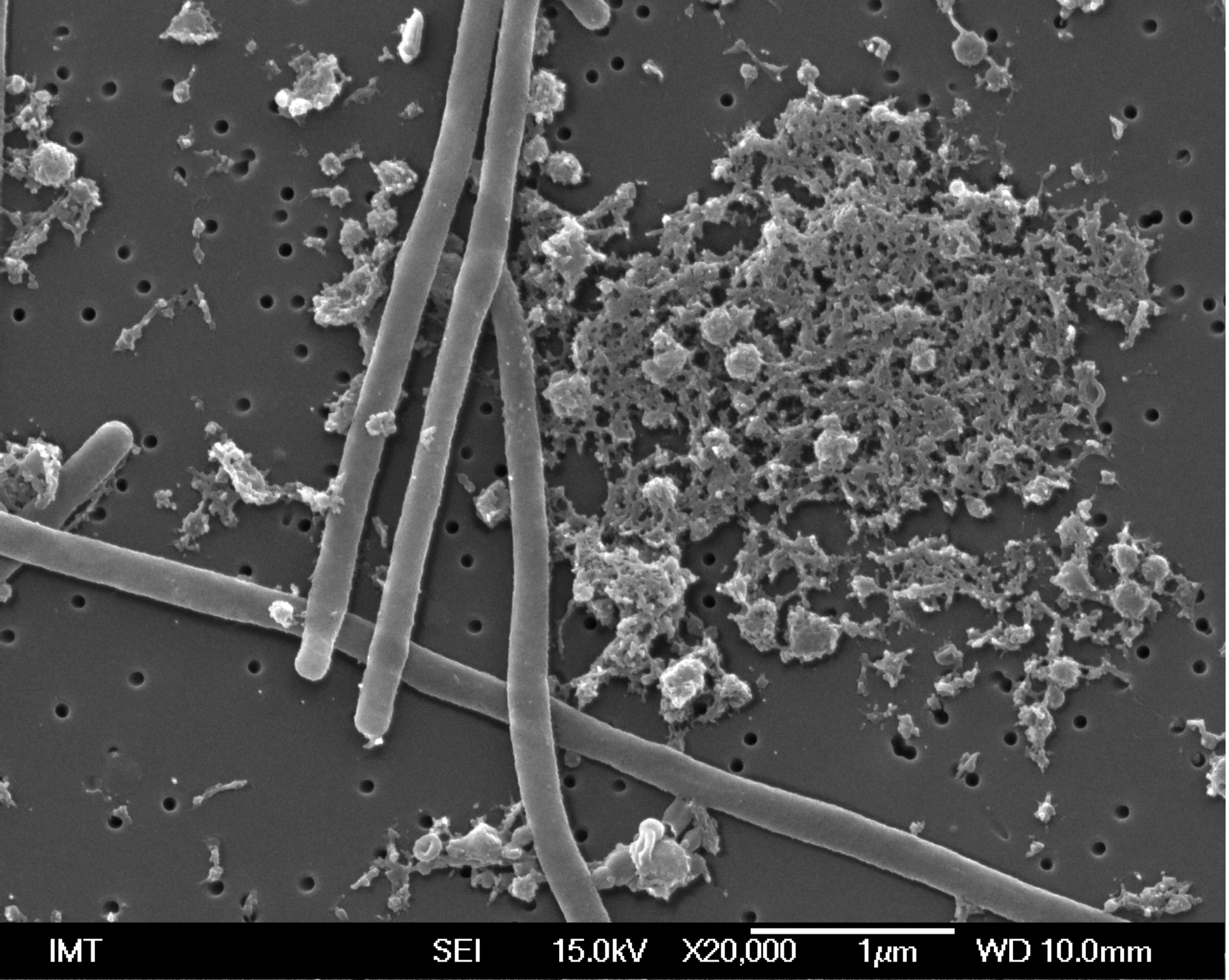

Figure *Dunaliela tertiolecta* isolate 10.000g SEM 4.

Isolation of nanoparticles (NPs)

NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. Then, the cell-depleted medium was centrifuged twice at 10 000 g and 4°C for 30 min (Beckman L8-70M ultracentrifuge, rotor SW55Ti (Beckman Coulter, USA)), using thin-wall polypropylene centrifuge tubes (ref. 326819, Beckman Coulter, USA). The isolate presents the pellet of the second centrifugation.

Scanning Electron Microscopy (SEM)

Samples were loaded onto 0.05-micron MCE filters (MF-Millipore™, ref. VMWP01300) and incubated in 2% OsO_4 for two hours. Then the osmium was removed, and the filter was taken out from the holder and further treated in a 24-well plate by changing the bath solution. After washing three times in distilled water, the samples were dehydrated in a graded series of ethanol (30%, 50%, 70%, 80%, 90%, absolute), treated with hexamethyldisilazane (30%, 50% mixtures with absolute ethanol, followed by pure hexamethyldisilazane), and air-dried. Samples were sputtered with Au/Pd (PECS Gatan 682) and examined with a JSM-6500F Field Emission Scanning Electron Microscope (JEOL Ltd., Tokyo, Japan).

Figure *Dunaliela tertiolecta* isolate 10.000g SEM 5.

Isolation of nanoparticles (NPs)
NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. Then, the cell-depleted medium was centrifuged twice at 10 000 g and 4°C for 30 min (Beckman L8-70M ultracentrifuge, rotor SW55Ti (Beckman Coulter, USA)), using thin-wall polypropylene centrifuge tubes (ref. 326819, Beckman Coulter, USA). The isolate presents the pellet of the second centrifugation.

Scanning Electron Microscopy (SEM)
Samples were loaded onto 0.05-micron MCE filters (MF-Millipore™, ref. VMWP01300) and incubated in 2% OsO_4 for two hours. Then the osmium was removed, and the filter was taken out from the holder and further treated in a 24-well plate by changing the bath solution. After washing three times in distilled water, the samples were dehydrated in a graded series of ethanol (30%, 50%, 70%, 80%, 90%, absolute), treated with hexamethyldisilazane (30%, 50% mixtures with absolute ethanol, followed by pure hexamethyldisilazane), and air-dried. Samples were sputtered with Au/Pd (PECS Gatan 682) and examined with a JSM-6500F Field Emission Scanning Electron Microscope (JEOL Ltd., Tokyo, Japan).

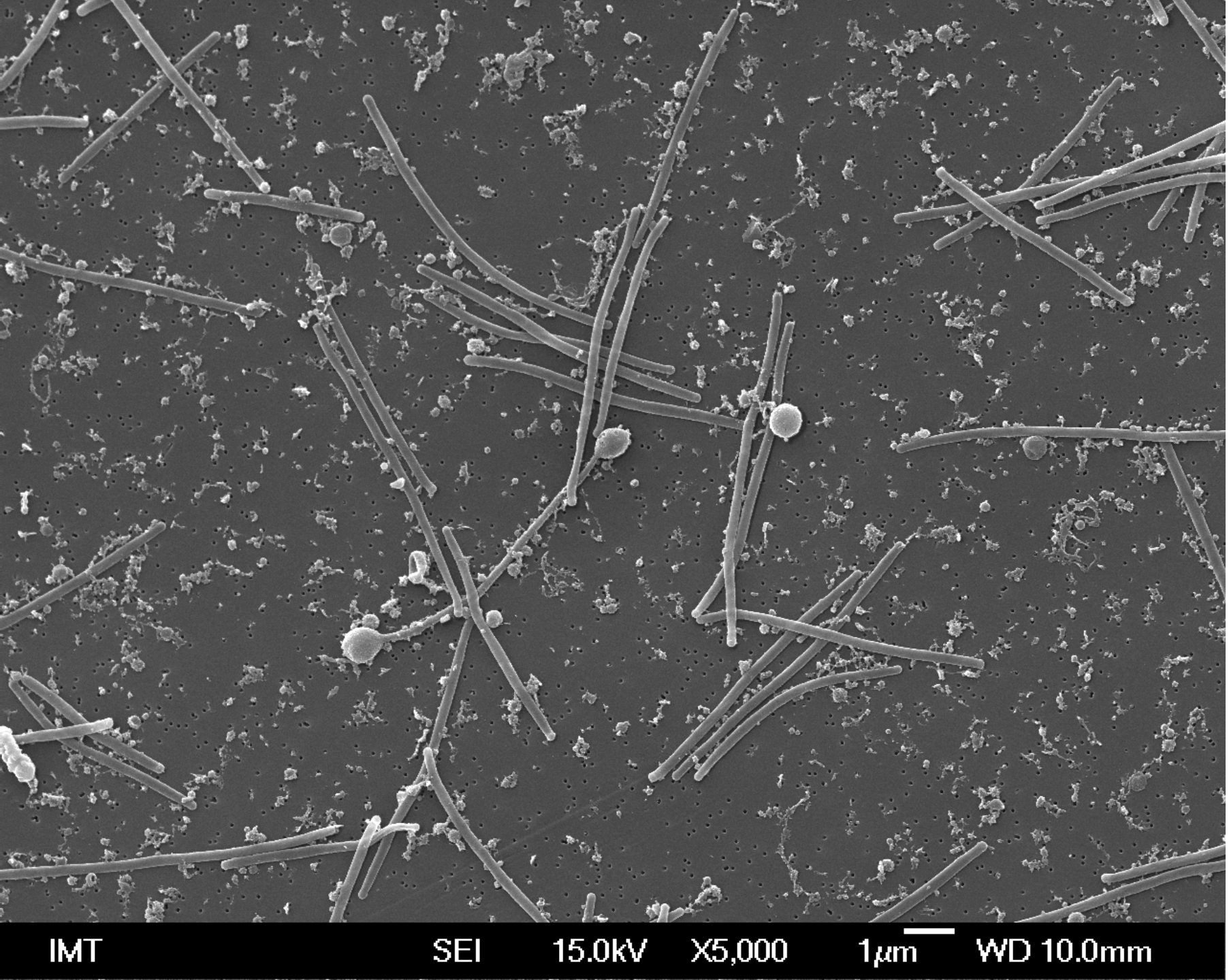

Figure *Dunaliela tertiolecta* isolate 10.000g SEM 6.

Isolation of nanoparticles (NPs)
NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. Then, the cell-depleted medium was centrifuged twice at 10 000 g and 4°C for 30 min (Beckman L8-70M ultracentrifuge, rotor SW55Ti (Beckman Coulter, USA)), using thin-wall polypropylene centrifuge tubes (ref. 326819, Beckman Coulter, USA). The isolate presents the pellet of the second centrifugation.
Scanning Electron Microscopy (SEM)
Samples were loaded onto 0.05-micron MCE filters (MF-Millipore™, ref. VMWP01300) and incubated in 2% OsO_4 for two hours. Then the osmium was removed, and the filter was taken out from the holder and further treated in a 24-well plate by changing the bath solution. After washing three times in distilled water, the samples were dehydrated in a graded series of ethanol (30%, 50%, 70%, 80%, 90%, absolute), treated with hexamethyldisilazane (30%, 50% mixtures with absolute ethanol, followed by pure hexamethyldisilazane), and air-dried. Samples were sputtered with Au/Pd (PECS Gatan 682) and examined with a JSM-6500F Field Emission Scanning Electron Microscope (JEOL Ltd., Tokyo, Japan).

Figure *Dunaliella tertiolecta* isolate 10.000g SEM 7.

Isolation of nanoparticles (NPs)
NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. Then, the cell-depleted medium was centrifuged twice at 10 000 g and 4°C for 30 min (Beckman L8-70M ultracentrifuge, rotor SW55Ti (Beckman Coulter, USA)), using thin-wall polypropylene centrifuge tubes (ref. 326819, Beckman Coulter, USA). The isolate presents the pellet of the second centrifugation.
Scanning Electron Microscopy (SEM)
Samples were loaded onto 0.05-micron MCE filters (MF-Millipore™, ref. VMWP01300) and incubated in 2% OsO_4 for two hours. Then the osmium was removed, and the filter was taken out from the holder and further treated in a 24-well plate by changing the bath solution. After washing three times in distilled water, the samples were dehydrated in a graded series of ethanol (30%, 50%, 70%, 80%, 90%, absolute), treated with hexamethyldisilazane (30%, 50% mixtures with absolute ethanol, followed by pure hexamethyldisilazane), and air-dried. Samples were sputtered with Au/Pd (PECS Gatan 682) and examined with a JSM-6500F Field Emission Scanning Electron Microscope (JEOL Ltd., Tokyo, Japan).

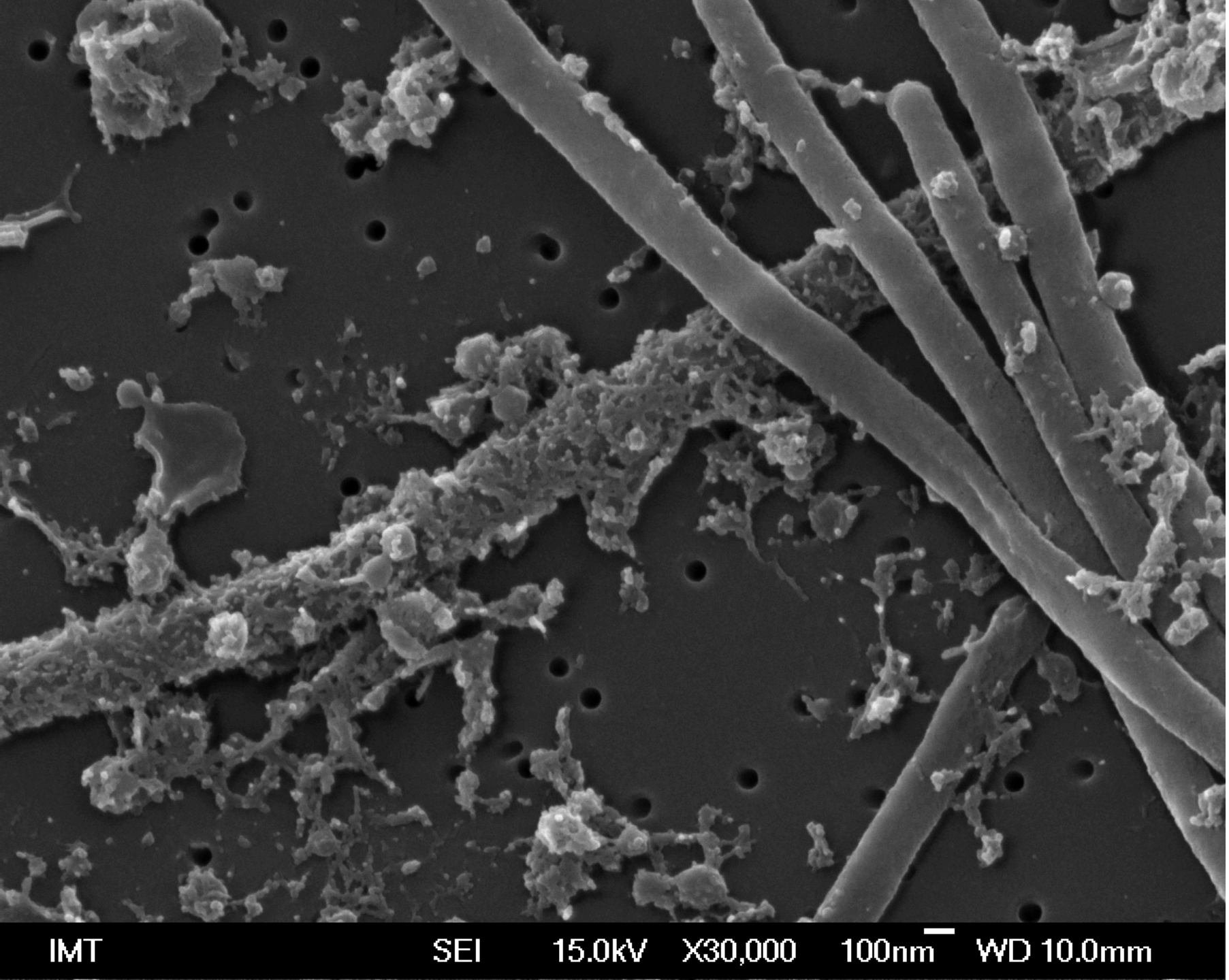

Figure *Dunaliella tertiolecta* isolate 10.000g SEM 8.

Isolation of nanoparticles (NPs)

NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. Then, the cell-depleted medium was centrifuged twice at 10 000 g and 4°C for 30 min (Beckman L8-70M ultracentrifuge, rotor SW55Ti (Beckman Coulter, USA)), using thin-wall polypropylene centrifuge tubes (ref. 326819, Beckman Coulter, USA). The isolate presents the pellet of the second centrifugation.

Scanning Electron Microscopy (SEM)

Samples were loaded onto 0.05-micron MCE filters (MF-Millipore™, ref. VMWP01300) and incubated in 2% OsO_4 for two hours. Then the osmium was removed, and the filter was taken out from the holder and further treated in a 24-well plate by changing the bath solution. After washing three times in distilled water, the samples were dehydrated in a graded series of ethanol (30%, 50%, 70%, 80%, 90%, absolute), treated with hexamethyldisilazane (30%, 50% mixtures with absolute ethanol, followed by pure hexamethyldisilazane), and air-dried. Samples were sputtered with Au/Pd (PECS Gatan 682) and examined with a JSM-6500F Field Emission Scanning Electron Microscope (JEOL Ltd., Tokyo, Japan).

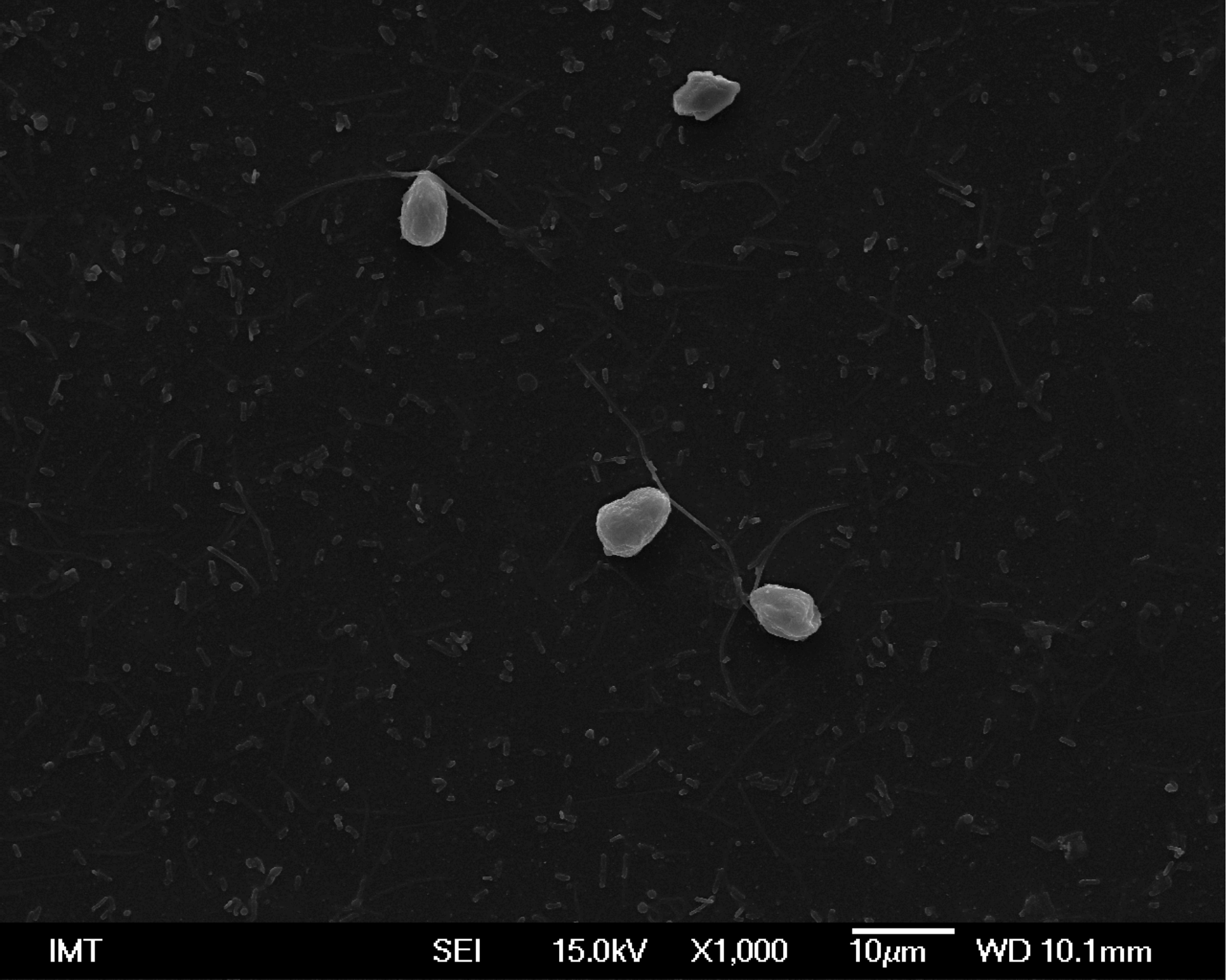

Figure *Dunaliela tertiolecta* isolate 2.000g SEM 9.

Isolation of nanoparticles (NPs)
NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. The isolate presents the pellet of the second centrifugation.

Scanning Electron Microscopy (SEM)
Samples were loaded onto 0.05-micron MCE filters (MF-Millipore™, ref. VMWP01300) and incubated in 2% OsO_4 for two hours. Then the osmium was removed, and the filter was taken out from the holder and further treated in a 24-well plate by changing the bath solution. After washing three times in distilled water, the samples were dehydrated in a graded series of ethanol (30%, 50%, 70%, 80%, 90%, absolute), treated with hexamethyldisilazane (30%, 50% mixtures with absolute ethanol, followed by pure hexamethyldisilazane), and air-dried. Samples were sputtered with Au/Pd (PECS Gatan 682) and examined with a JSM-6500F Field Emission Scanning Electron Microscope (JEOL Ltd., Tokyo, Japan).

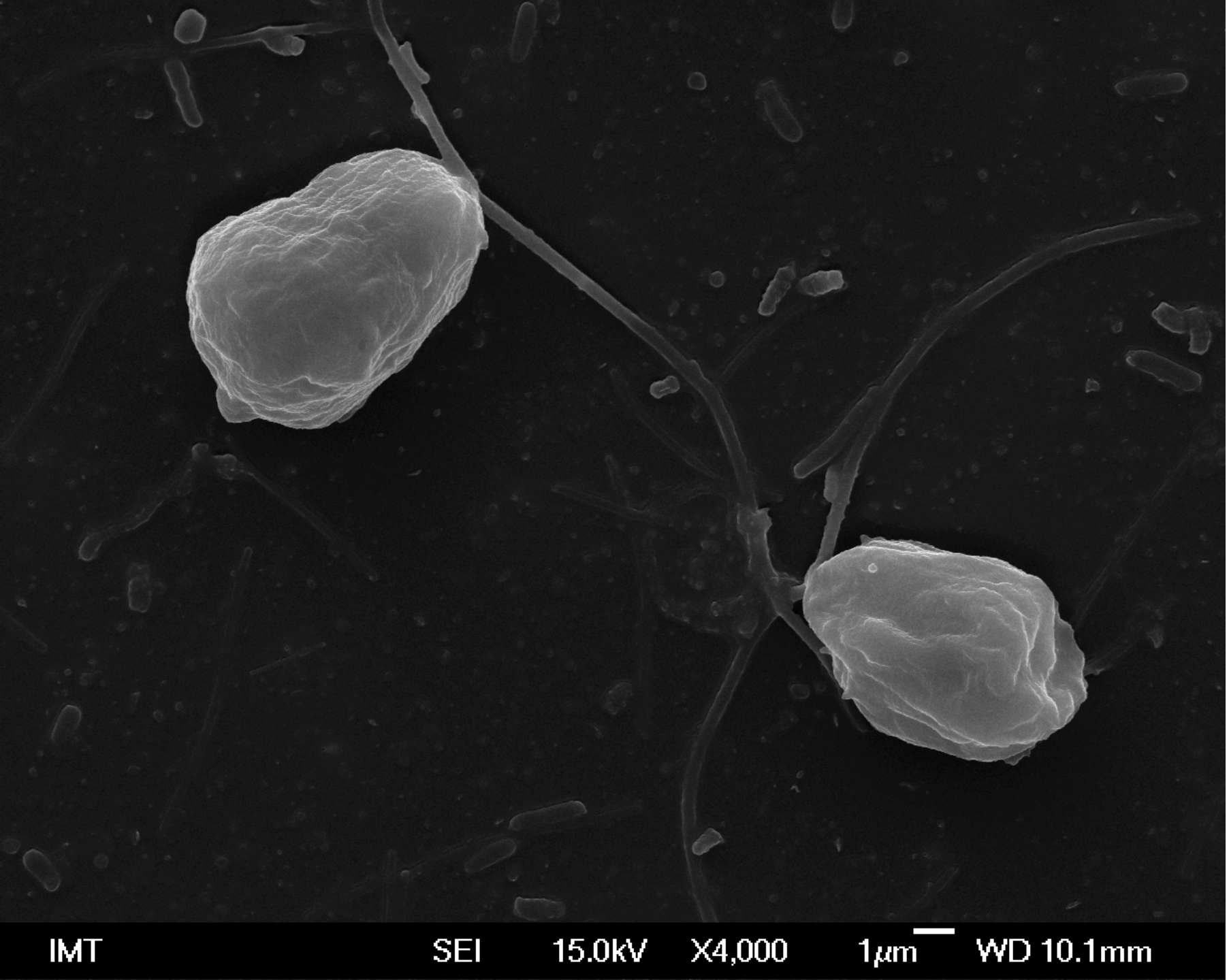

Figure *Dunaliela tertiolecta* isolate 2.000g SEM 10.

Isolation of nanoparticles (NPs)
NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. The isolate presents the pellet of the second centrifugation.

Scanning Electron Microscopy (SEM)
Samples were loaded onto 0.05-micron MCE filters (MF-Millipore™, ref. VMWP01300) and incubated in 2% OsO_4 for two hours. Then the osmium was removed, and the filter was taken out from the holder and further treated in a 24-well plate by changing the bath solution. After washing three times in distilled water, the samples were dehydrated in a graded series of ethanol (30%, 50%, 70%, 80%, 90%, absolute), treated with hexamethyldisilazane (30%, 50% mixtures with absolute ethanol, followed by pure hexamethyldisilazane), and air-dried. Samples were sputtered with Au/Pd (PECS Gatan 682) and examined with a JSM-6500F Field Emission Scanning Electron Microscope (JEOL Ltd., Tokyo, Japan).

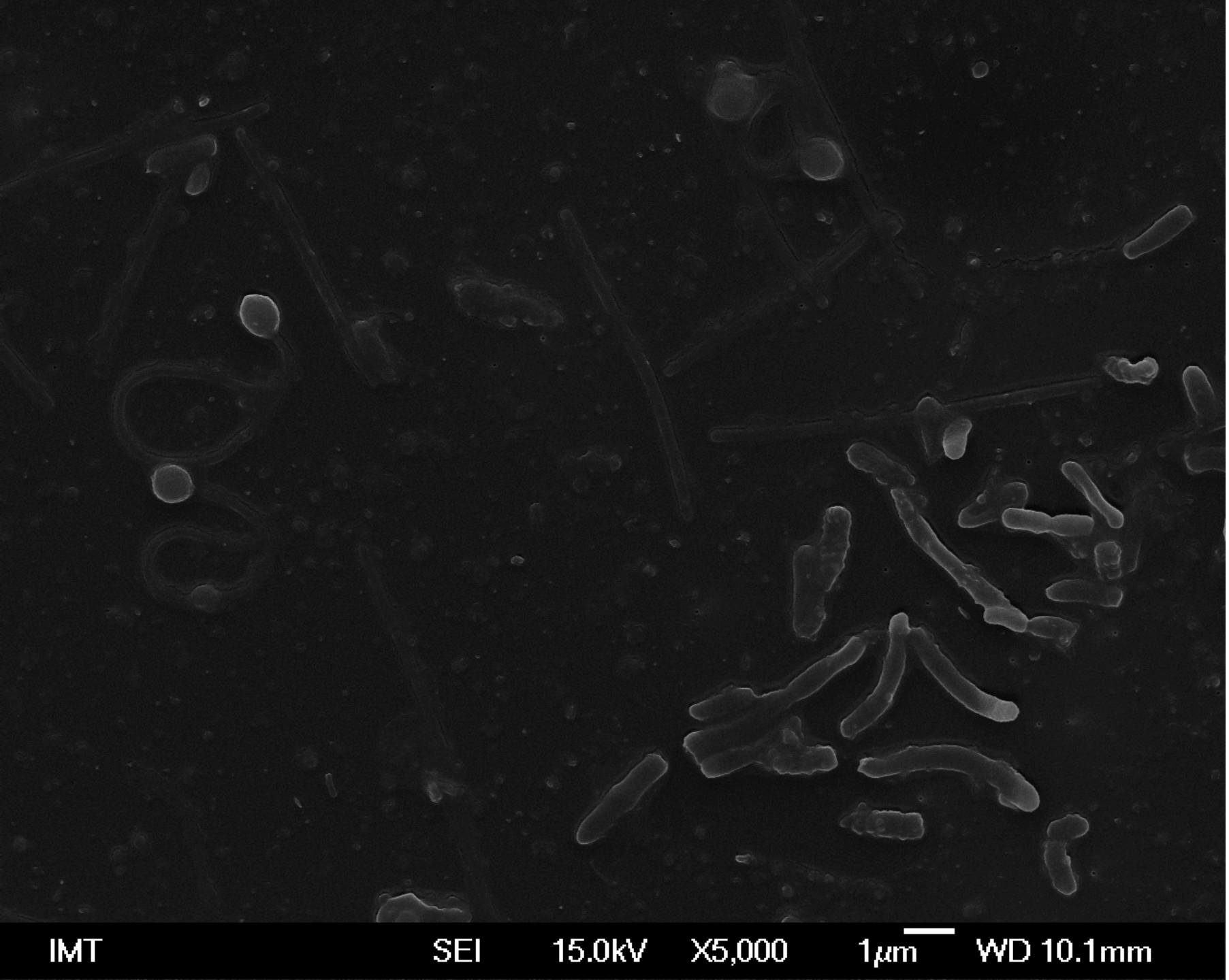

Figure *Dunaliela tertiolecta* isolate 2.000g SEM 11.

Isolation of nanoparticles (NPs)
NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. The isolate presents the pellet of the second centrifugation.

Scanning Electron Microscopy (SEM)
Samples were loaded onto 0.05-micron MCE filters (MF-Millipore™, ref. VMWP01300) and incubated in 2% OsO_4 for two hours. Then the osmium was removed, and the filter was taken out from the holder and further treated in a 24-well plate by changing the bath solution. After washing three times in distilled water, the samples were dehydrated in a graded series of ethanol (30%, 50%, 70%, 80%, 90%, absolute), treated with hexamethyldisilazane (30%, 50% mixtures with absolute ethanol, followed by pure hexamethyldisilazane), and air-dried. Samples were sputtered with Au/Pd (PECS Gatan 682) and examined with a JSM-6500F Field Emission Scanning Electron Microscope (JEOL Ltd., Tokyo, Japan).

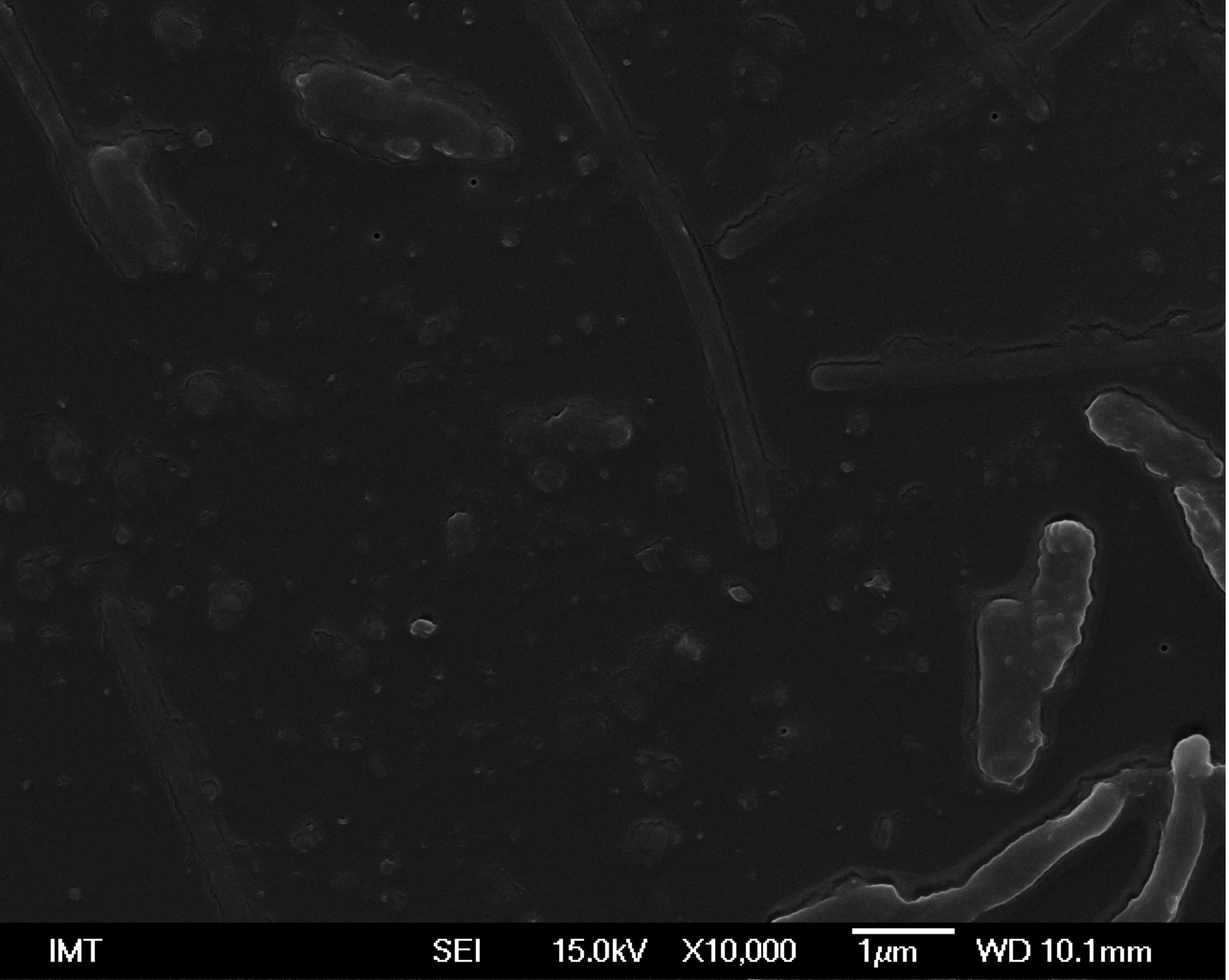

Figure *Dunaliela tertiolecta* isolate 2.000g SEM 12.

Isolation of nanoparticles (NPs)

NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. The isolate presents the pellet of the second centrifugation.

Scanning Electron Microscopy (SEM)

Samples were loaded onto 0.05-micron MCE filters (MF-Millipore™, ref. VMWP01300) and incubated in 2% OsO_4 for two hours. Then the osmium was removed, and the filter was taken out from the holder and further treated in a 24-well plate by changing the bath solution. After washing three times in distilled water, the samples were dehydrated in a graded series of ethanol (30%, 50%, 70%, 80%, 90%, absolute), treated with hexamethyldisilazane (30%, 50% mixtures with absolute ethanol, followed by pure hexamethyldisilazane), and air-dried. Samples were sputtered with Au/Pd (PECS Gatan 682) and examined with a JSM-6500F Field Emission Scanning Electron Microscope (JEOL Ltd., Tokyo, Japan).

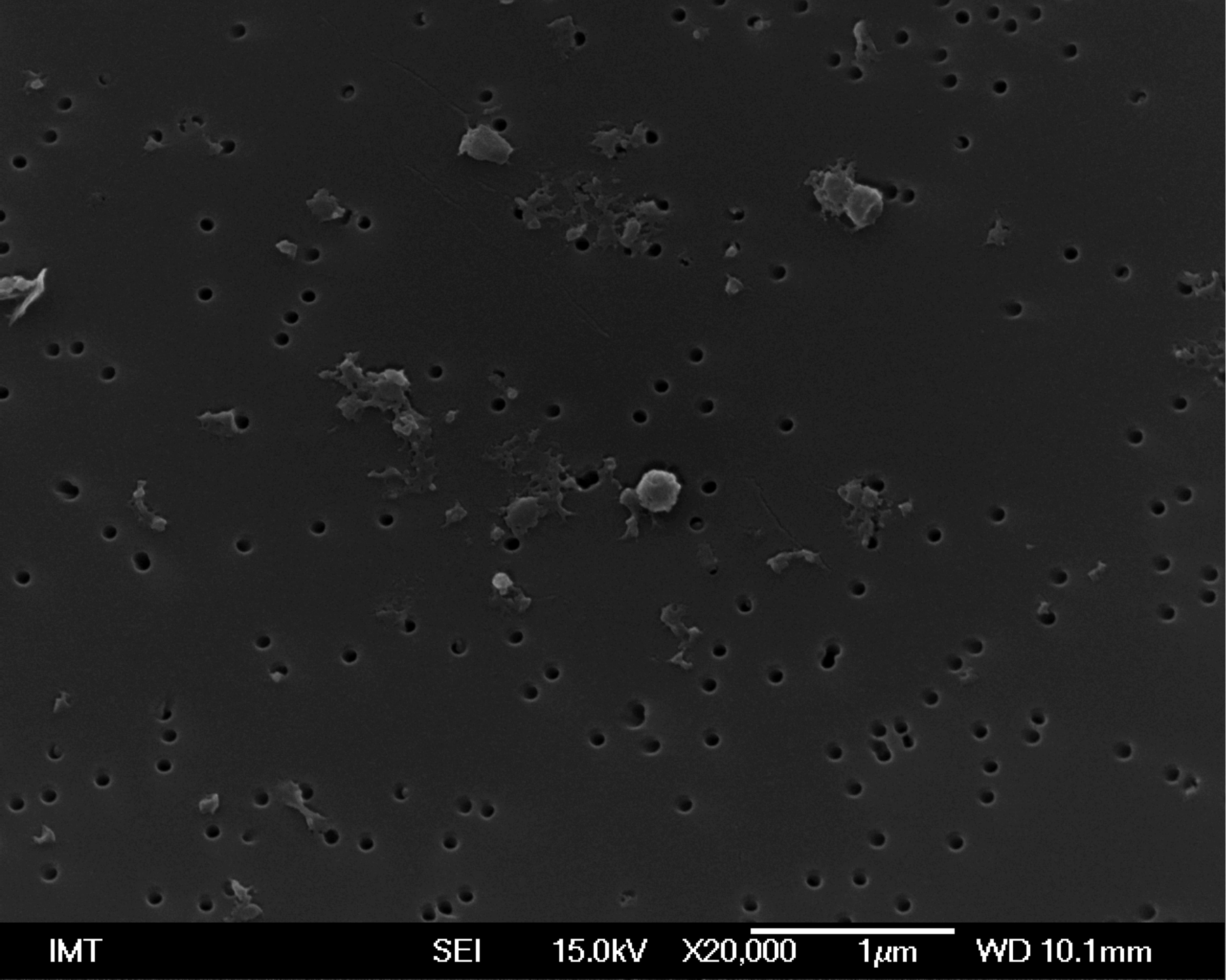

Figure *Dunaliela tertiolecta* isolate 2.000g SEM 13.

Isolation of nanoparticles (NPs)
NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. The isolate presents the pellet of the second centrifugation.
Scanning Electron Microscopy (SEM)
Samples were loaded onto 0.05-micron MCE filters (MF-Millipore™, ref. VMWP01300) and incubated in 2% OsO_4 for two hours. Then the osmium was removed, and the filter was taken out from the holder and further treated in a 24-well plate by changing the bath solution. After washing three times in distilled water, the samples were dehydrated in a graded series of ethanol (30%, 50%, 70%, 80%, 90%, absolute), treated with hexamethyldisilazane (30%, 50% mixtures with absolute ethanol, followed by pure hexamethyldisilazane), and air-dried. Samples were sputtered with Au/Pd (PECS Gatan 682) and examined with a JSM-6500F Field Emission Scanning Electron Microscope (JEOL Ltd., Tokyo, Japan).

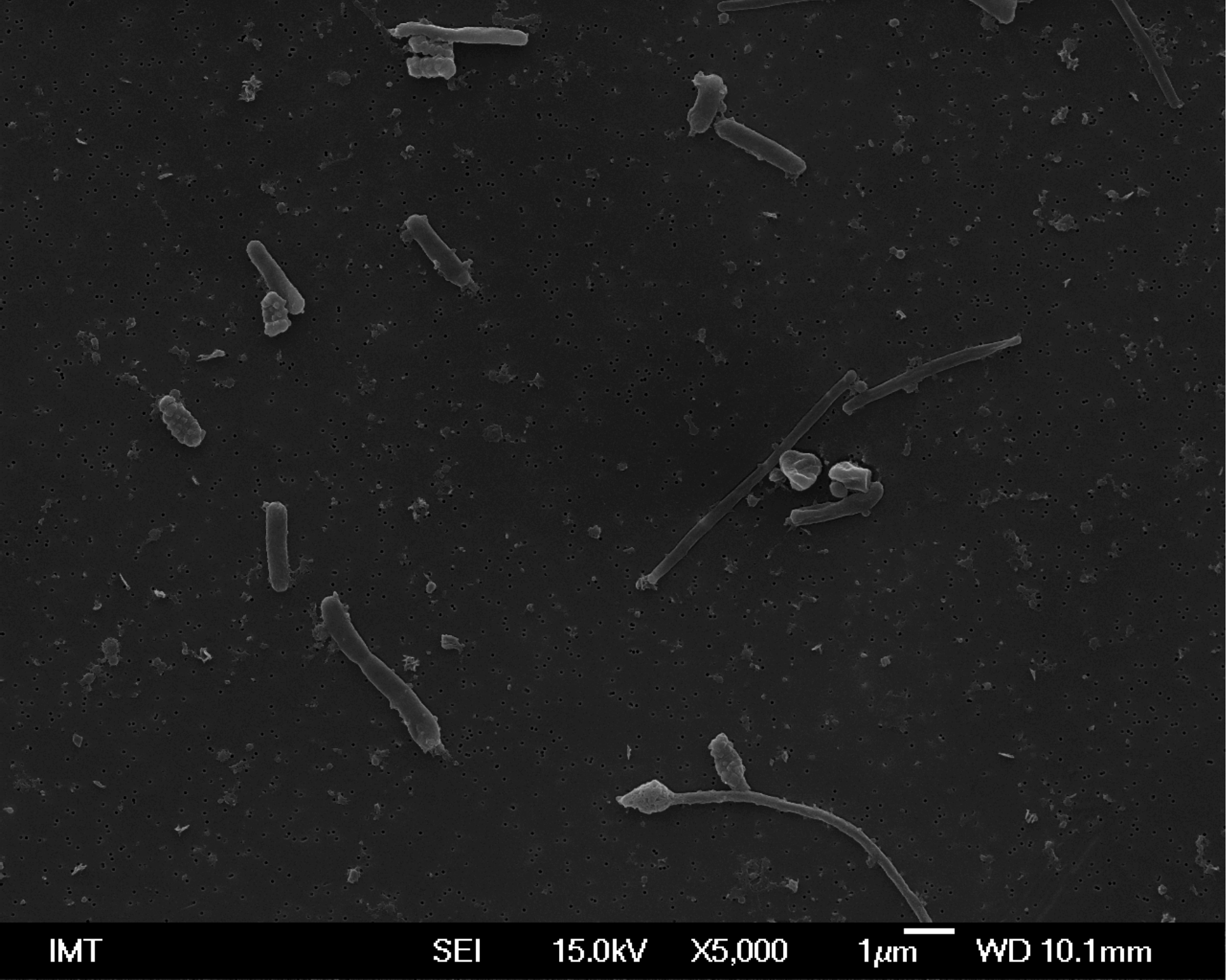

Figure *Dunaliela tertiolecta* isolate 2.000g SEM 14.

Isolation of nanoparticles (NPs)
NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. The isolate presents the pellet of the second centrifugation.
Scanning Electron Microscopy (SEM)
Samples were loaded onto 0.05-micron MCE filters (MF-Millipore™, ref. VMWP01300) and incubated in 2% OsO_4 for two hours. Then the osmium was removed, and the filter was taken out from the holder and further treated in a 24-well plate by changing the bath solution. After washing three times in distilled water, the samples were dehydrated in a graded series of ethanol (30%, 50%, 70%, 80%, 90%, absolute), treated with hexamethyldisilazane (30%, 50% mixtures with absolute ethanol, followed by pure hexamethyldisilazane), and air-dried. Samples were sputtered with Au/Pd (PECS Gatan 682) and examined with a JSM-6500F Field Emission Scanning Electron Microscope (JEOL Ltd., Tokyo, Japan).

Figure *Dunaliela tertiolecta* isolate 2.000g SEM 15.

Isolation of nanoparticles (NPs)
NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. The isolate presents the pellet of the second centrifugation.

Scanning Electron Microscopy (SEM)
Samples were loaded onto 0.05-micron MCE filters (MF-Millipore™, ref. VMWP01300) and incubated in 2% OsO_4 for two hours. Then the osmium was removed, and the filter was taken out from the holder and further treated in a 24-well plate by changing the bath solution. After washing three times in distilled water, the samples were dehydrated in a graded series of ethanol (30%, 50%, 70%, 80%, 90%, absolute), treated with hexamethyldisilazane (30%, 50% mixtures with absolute ethanol, followed by pure hexamethyldisilazane), and air-dried. Samples were sputtered with Au/Pd (PECS Gatan 682) and examined with a JSM-6500F Field Emission Scanning Electron Microscope (JEOL Ltd., Tokyo, Japan).

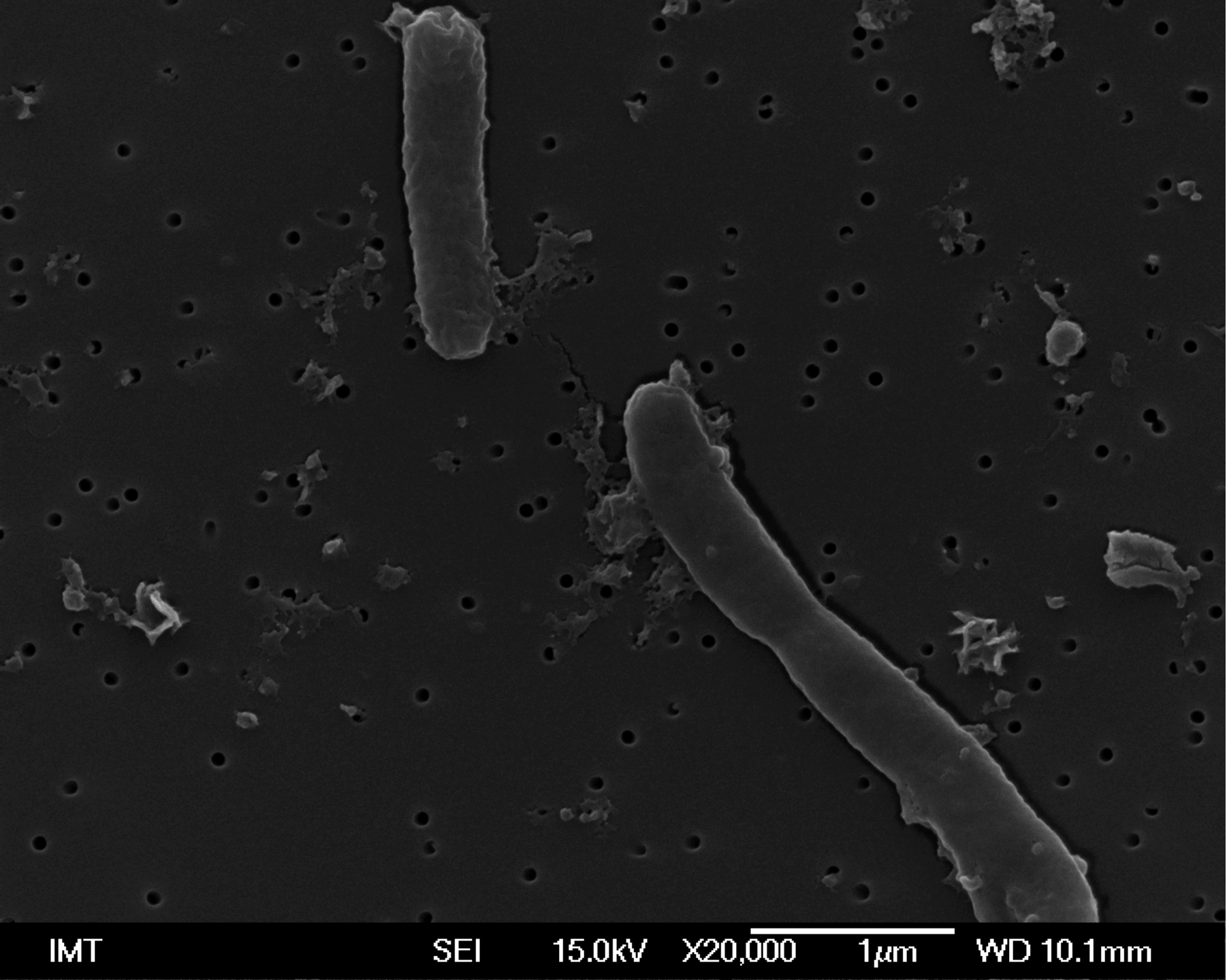

Figure *Dunaliela tertiolecta* isolate 2.000g SEM 16.

Isolation of nanoparticles (NPs)
NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. The isolate presents the pellet of the second centrifugation.

Scanning Electron Microscopy (SEM)
Samples were loaded onto 0.05-micron MCE filters (MF-Millipore™, ref. VMWP01300) and incubated in 2% OsO_4 for two hours. Then the osmium was removed, and the filter was taken out from the holder and further treated in a 24-well plate by changing the bath solution. After washing three times in distilled water, the samples were dehydrated in a graded series of ethanol (30%, 50%, 70%, 80%, 90%, absolute), treated with hexamethyldisilazane (30%, 50% mixtures with absolute ethanol, followed by pure hexamethyldisilazane), and air-dried. Samples were sputtered with Au/Pd (PECS Gatan 682) and examined with a JSM-6500F Field Emission Scanning Electron Microscope (JEOL Ltd., Tokyo, Japan).

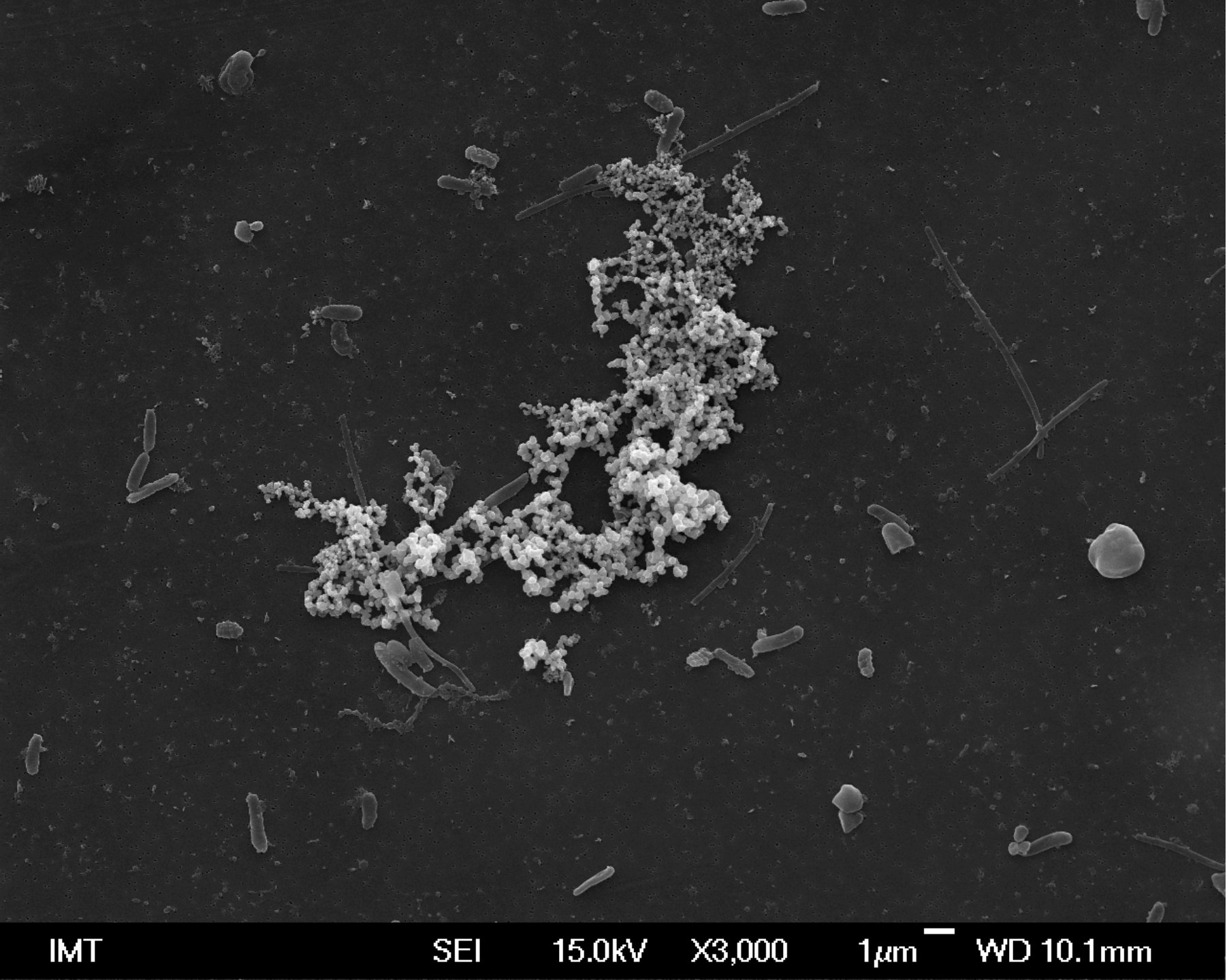

Figure *Dunaliela tertiolecta* isolate 2.000g SEM 17.

Isolation of nanoparticles (NPs)
NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. The isolate presents the pellet of the second centrifugation.

Scanning Electron Microscopy (SEM)
Samples were loaded onto 0.05-micron MCE filters (MF-Millipore™, ref. VMWP01300) and incubated in 2% OsO_4 for two hours. Then the osmium was removed, and the filter was taken out from the holder and further treated in a 24-well plate by changing the bath solution. After washing three times in distilled water, the samples were dehydrated in a graded series of ethanol (30%, 50%, 70%, 80%, 90%, absolute), treated with hexamethyldisilazane (30%, 50% mixtures with absolute ethanol, followed by pure hexamethyldisilazane), and air-dried. Samples were sputtered with Au/Pd (PECS Gatan 682) and examined with a JSM-6500F Field Emission Scanning Electron Microscope (JEOL Ltd., Tokyo, Japan).

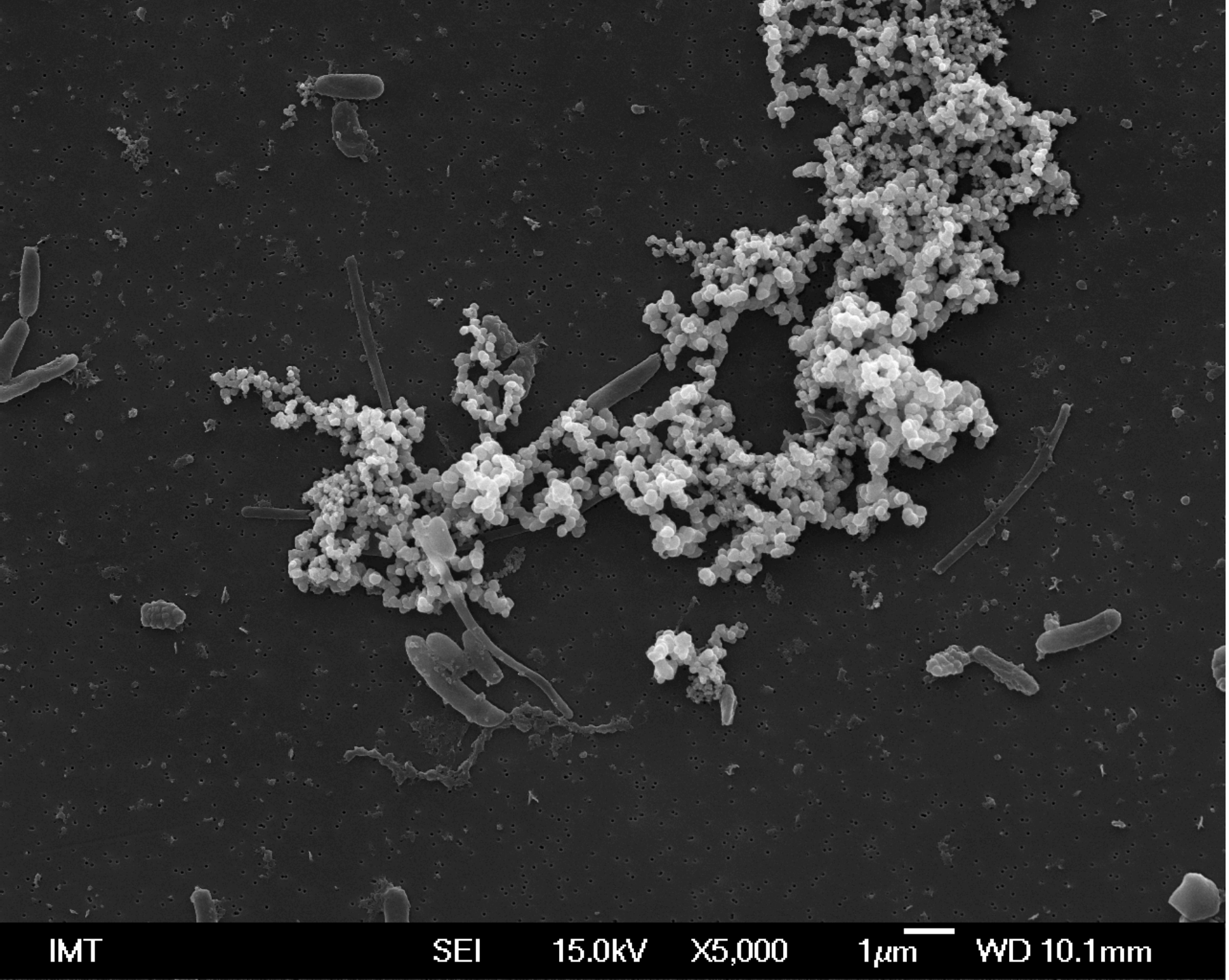

Figure *Dunaliela tertiolecta* isolate 2.000g SEM 18.

Isolation of nanoparticles (NPs)
NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. The isolate presents the pellet of the second centrifugation.

Scanning Electron Microscopy (SEM)
Samples were loaded onto 0.05-micron MCE filters (MF-Millipore™, ref. VMWP01300) and incubated in 2% OsO_4 for two hours. Then the osmium was removed, and the filter was taken out from the holder and further treated in a 24-well plate by changing the bath solution. After washing three times in distilled water, the samples were dehydrated in a graded series of ethanol (30%, 50%, 70%, 80%, 90%, absolute), treated with hexamethyldisilazane (30%, 50% mixtures with absolute ethanol, followed by pure hexamethyldisilazane), and air-dried. Samples were sputtered with Au/Pd (PECS Gatan 682) and examined with a JSM-6500F Field Emission Scanning Electron Microscope (JEOL Ltd., Tokyo, Japan).

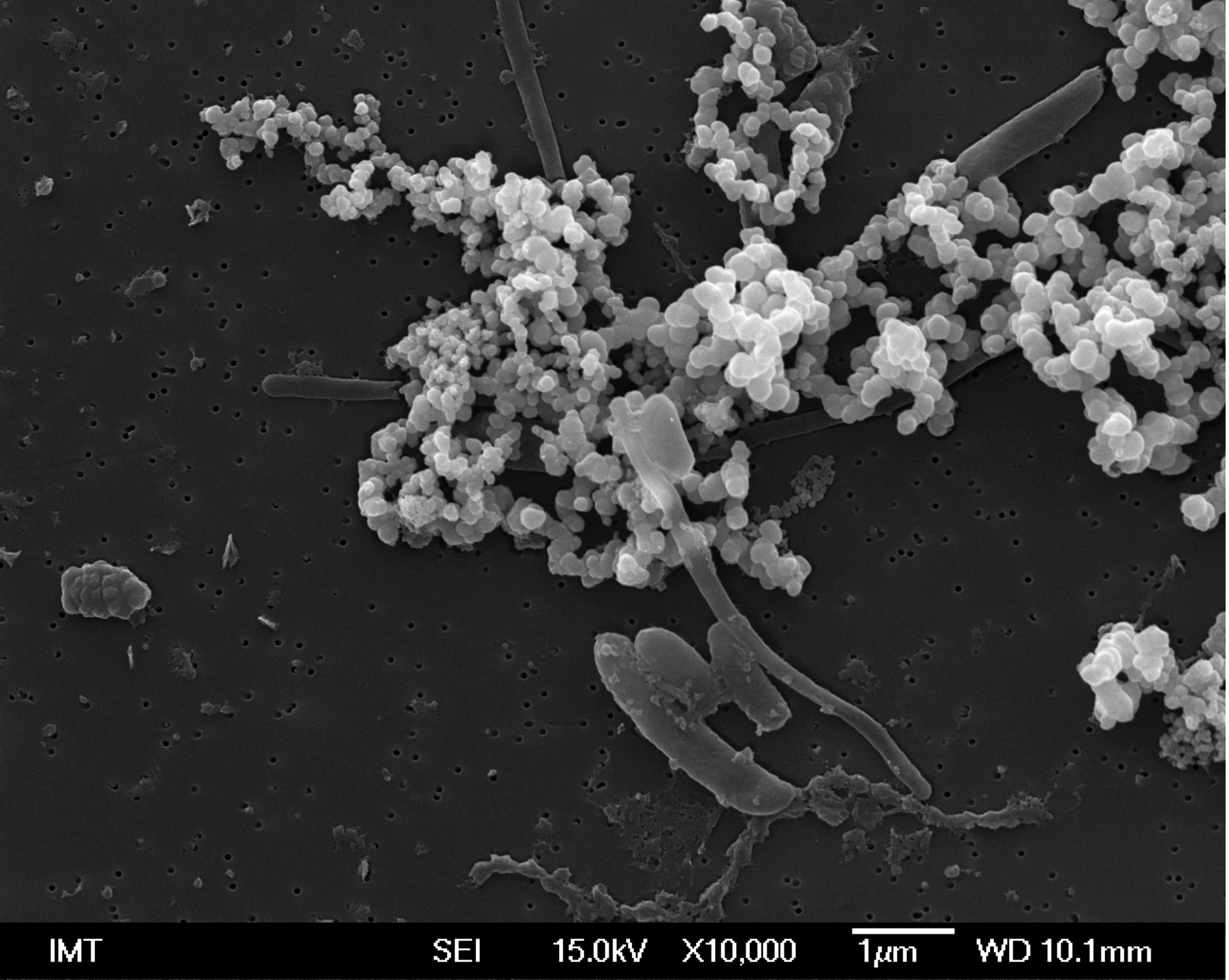

Figure *Dunaliela tertiolecta* isolate 2.000g SEM 19.

Isolation of nanoparticles (NPs)

NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. The isolate presents the pellet of the second centrifugation.

Scanning Electron Microscopy (SEM)

Samples were loaded onto 0.05-micron MCE filters (MF-Millipore™, ref. VMWP01300) and incubated in 2% OsO_4 for two hours. Then the osmium was removed, and the filter was taken out from the holder and further treated in a 24-well plate by changing the bath solution. After washing three times in distilled water, the samples were dehydrated in a graded series of ethanol (30%, 50%, 70%, 80%, 90%, absolute), treated with hexamethyldisilazane (30%, 50% mixtures with absolute ethanol, followed by pure hexamethyldisilazane), and air-dried. Samples were sputtered with Au/Pd (PECS Gatan 682) and examined with a JSM-6500F Field Emission Scanning Electron Microscope (JEOL Ltd., Tokyo, Japan).

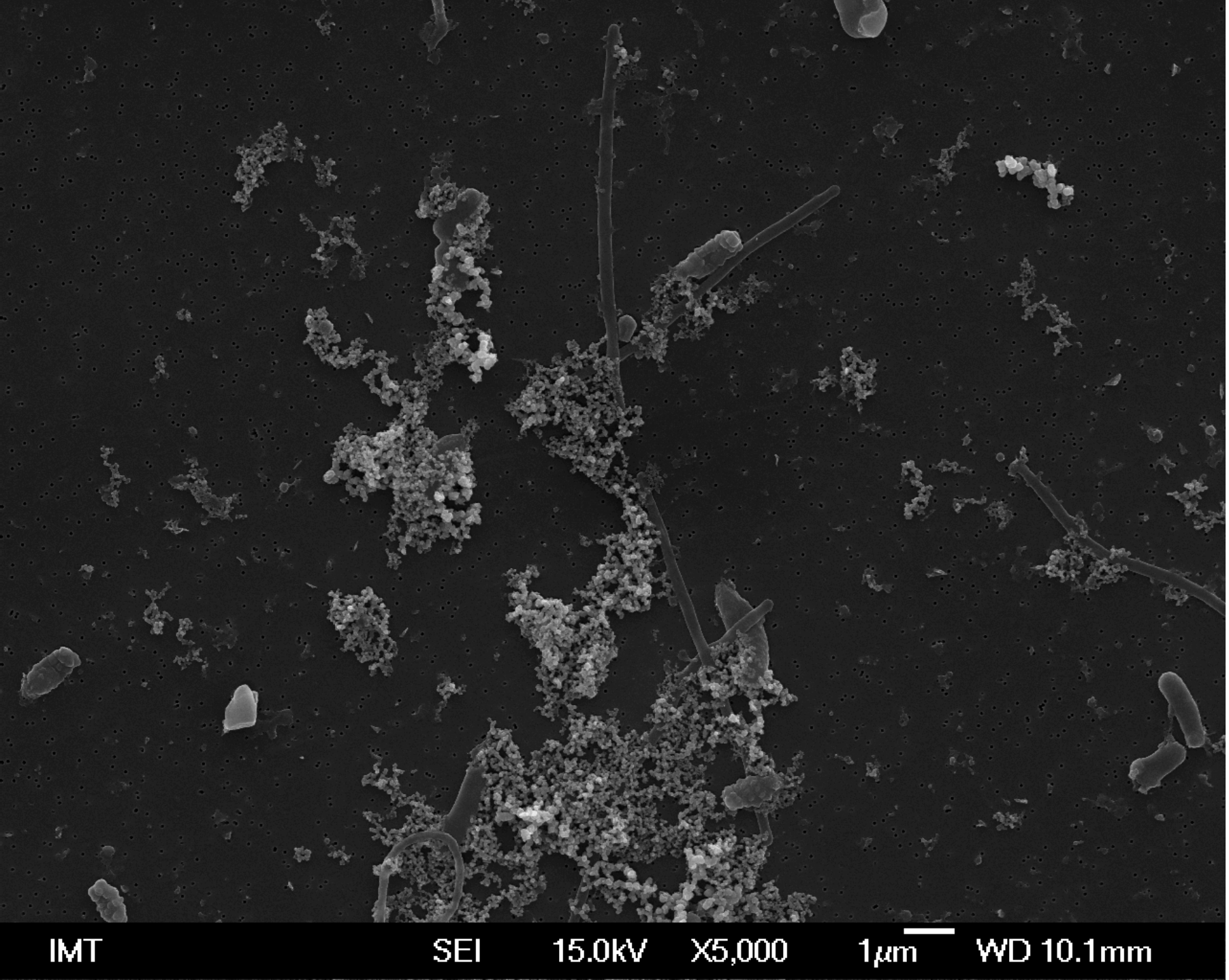

Figure *Dunaliela tertiolecta* isolate 2.000g SEM 20.

Isolation of nanoparticles (NPs)
NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. The isolate presents the pellet of the second centrifugation.

Scanning Electron Microscopy (SEM)
Samples were loaded onto 0.05-micron MCE filters (MF-Millipore™, ref. VMWP01300) and incubated in 2% OsO_4 for two hours. Then the osmium was removed, and the filter was taken out from the holder and further treated in a 24-well plate by changing the bath solution. After washing three times in distilled water, the samples were dehydrated in a graded series of ethanol (30%, 50%, 70%, 80%, 90%, absolute), treated with hexamethyldisilazane (30%, 50% mixtures with absolute ethanol, followed by pure hexamethyldisilazane), and air-dried. Samples were sputtered with Au/Pd (PECS Gatan 682) and examined with a JSM-6500F Field Emission Scanning Electron Microscope (JEOL Ltd., Tokyo, Japan).

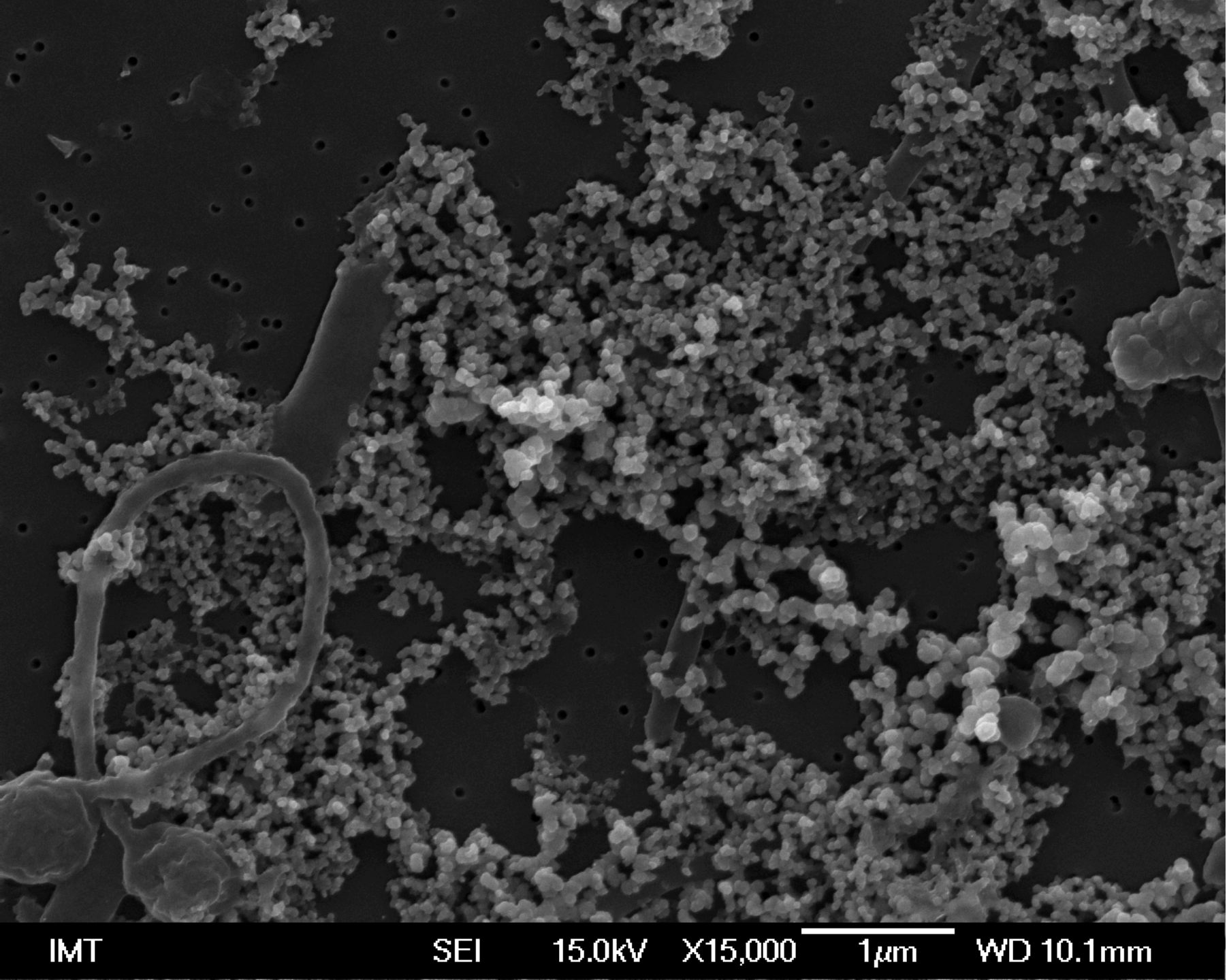

Figure *Dunaliela tertiolecta* isolate 2.000g SEM 21.

Isolation of nanoparticles (NPs)
NPs were isolated by differential centrifugation. The cells from algal culture were removed by low-speed centrifugation (300 g, 10 min, 4°C, centrifuge Centric 260R with rotor RA 6/50 (Domel, Slovenia)), using 50 mL conical centrifuge tubes (ref. S.078.02.008.050, Isolab Laborgeräte GmbH, Germany); and 2000 g, 10 min, 4°C (Centric 400R centrifuge with rotor RS4/100 (Domel, Slovenia)), using 15 mL conical centrifuge tubes (ref. S.078.02.001.050, Isolab Laborgeräte GmbH, Germany). Each step was repeated twice. The isolate presents the pellet of the second centrifugation.
Scanning Electron Microscopy (SEM)
Samples were loaded onto 0.05-micron MCE filters (MF-Millipore™, ref. VMWP01300) and incubated in 2% OsO_4 for two hours. Then the osmium was removed, and the filter was taken out from the holder and further treated in a 24-well plate by changing the bath solution. After washing three times in distilled water, the samples were dehydrated in a graded series of ethanol (30%, 50%, 70%, 80%, 90%, absolute), treated with hexamethyldisilazane (30%, 50% mixtures with absolute ethanol, followed by pure hexamethyldisilazane), and air-dried. Samples were sputtered with Au/Pd (PECS Gatan 682) and examined with a JSM-6500F Field Emission Scanning Electron Microscope (JEOL Ltd., Tokyo, Japan).