

Supplementary materials

Natural products biosynthesis by *Streptomyces netropsis* IMV Ac-5025 under exogenous sterol action

Mariia Loboda¹, Liudmyla Biliavska¹, Halyna Iutynska¹, Jake Newitt² and Ruslan Mariychuk^{3,*}

¹ Department of General and Soil Microbiology, D.K. Zabolotny Institute of Microbiology and Virology, National Academy of Sciences of Ukraine, Akademika Zabolotnoho str., 154, Kyiv, Ukraine; marichka20loboda@gmail.com (M.L.), bilyuvskal@ukr.net (L.B.), galyna.iutynska@gmail.com (H.I.)

² Department of Molecular Microbiology, John Innes Centre, Norwich NR4 7UH, United Kingdom; jake.newitt@jic.ac.uk

³ University of Presov, Presov, Slovakia; ruslan.mariychuk@unipo.sk

* Correspondence: ruslan.mariychuk@unipo.sk

Table S1. Polyene antibiotics' accumulation by *S. netropsis* IMV Ac-5025 grown in synthetic and organic nutrient media in culture liquid supplemented with exogenous β-sitosterol (ADB – absolutely dry biomass).

Table S2. Biosynthesis of squalene and sterols in *S. netropsis* IMV Ac-5025 biomass grown in synthetic nutrient medium supplemented with exogenous β-sitosterol.

Table S3. Biosynthesis of auxins in *S. netropsis* IMV Ac-5025 biomass grown in synthetic nutrient medium supplemented with exogenous β-sitosterol.

Table S4. Cytokinins' biosynthesis in *S. netropsis* IMV Ac-5025 biomass grown in synthetic nutrient medium supplemented with exogenous β-sitosterol.

Table S1. Polyene antibiotics' accumulation by *S. netropsis* IMV Ac-5025 grown in synthetic and organic nutrient media in culture liquid supplemented with exogenous β -sitosterol (ADB – absolutely dry biomass).

Syntetic nutrient medium							
β -Sitosterol, $\mu\text{g/mL}$	Culture liquid			Biomass, g/L	Biomass extract		
	Tetraene, $\mu\text{g/mL}$	Candidine, $\mu\text{g/mL}$	Sum of antibiotics, $\mu\text{g/mL}$		Tetraene, mg/g ADB	Candidine, mg/g ADB	Sum of antibiotics, mg/g ADB
0	2.68 \pm 0.09	0.32 \pm 0.07	3 \pm 0.16	2.65 \pm 0.2	1.399 \pm 0.11	0.53 \pm 0.01	1.929 \pm 0.1
0.1	7.3 \pm 0.41	0.5 \pm 0.06	7.8 \pm 0.46	3.7 \pm 0.23	1.543 \pm 0.04	0.772 \pm 0.01	2.315 \pm 0.04
0.25	7.12 \pm 0.1	3.1 \pm 0.41	10.22 \pm 0.36	3.29 \pm 0.23	1.556 \pm 0.13	0.809 \pm 0.03	2.365 \pm 0.11
2.5	7.5 \pm 0.06	3.05 \pm 0.16	10.55 \pm 0.12	4.44 \pm 0.24	2.405 \pm 0.23	0.907 \pm 0.11	3.312 \pm 0.3
5	7.66 \pm 0.25	3.9 \pm 0.57	11.56 \pm 0.32	5.2 \pm 0.2	2.442 \pm 0.26	1.326 \pm 0.02	3.768 \pm 0.29
10	8.4 \pm 0.41	4.98 \pm 0.07	13.38 \pm 0.47	4.56 \pm 0.2	3.218 \pm 0.12	1.699 \pm 0.1	4.919 \pm 0.22
Organic nutrient medium							
β -Sitosterol, $\mu\text{g/mL}$	Culture liquid			Biomass, g/L	Biomass extract		
	Tetraene, $\mu\text{g/mL}$	Candidine, $\mu\text{g/mL}$	Sum of antibiotics, $\mu\text{g/mL}$		Tetraene, mg/g ADB	Candidine, mg/g ADB	Sum of antibiotics, mg/g ADB
0	67 \pm 1.2	41 \pm 1.49	108 \pm 2.54	7.15 \pm 0.25	6.5 \pm 0.29	2.71 \pm 0.11	9.21 \pm 0.4
0.1	73 \pm 1.13	44 \pm 1.15	117 \pm 0.8	7.34 \pm 0.34	8.3 \pm 0.53	3.8 \pm 0.15	12.1 \pm 0.66
0.25	83 \pm 0.57	43 \pm 1.49	126 \pm 0.96	8.58 \pm 0.4	9.3 \pm 0.38	4 \pm 0.26	13.3 \pm 0.63
2.5	87 \pm 1.7	30 \pm 1.41	120 \pm 2.89	9.54 \pm 0.32	4.8 \pm 0.16	2.87 \pm 0.12	7.67 \pm 0.27
5	90 \pm 1.76	31 \pm 1.28	121 \pm 2	10.16 \pm 0.25	10 \pm 0.41	7.7 \pm 0.11	17.7 \pm 0.45
10	106 \pm 1.18	39 \pm 1.08	145 \pm 1.93	10.1 \pm 0.3	12.4 \pm 0.5	8.4 \pm 0.33	20.8 \pm 0.26

Table S2. Biosynthesis of squalene and sterols in *S. netropsis* IMV Ac-5025 biomass grown in synthetic nutrient medium supplemented with exogenous β -sitosterol.

β -Sitosterol, μg/mL	Compounds, mg/g of absolutely dry biomass						
	Squalene	Ergosterol	Cholesterol	Stigmasterol	β -sitosterol	24-Epi-brassinolide	Total squalen and sterols
0	0.10 ± 0.01	0.05 ± 0.01	0.03± 0.01	0.91 ± 0.32	0.51 ± 0.24	0.15 ± 0.01	1.65 ± 0.43
0.1	0.32 ± 0.18	0.08 ± 0.01	0.05 ± 0.01	1.82 ± 0.45	0.71 ± 0.28	0.21 ± 0.01	3.18 ± 0.59
0.25	0.22 ± 0.15	0.05 ± 0.01	0.11 ± 0.01	2.73 ± 0.55	1.12 ± 0.235	0.52 ± 0.24	4.75 ± 0.72
2.5	0.13 ± 0.12	0.08 ± 0.01	0.08 ± 0.01	2.25 ± 0.5	0.83 ± 0.30	0.56 ± 0.25	3.92 ± 0.66
5	0.24 ± 0.16	0.12 ± 0.01	0.13 ± 0.02	2.42 ± 0.52	0.63 ± 0.26	0.36 ± 0.20	3.90 ± 0.66
10	0.55 ± 0.24	0.36 ± 0.20	0.23 ± 0.16	0.74 ± 0.29	0.96 ± 0.32	0.26 ± 0.01	3.10 ± 0.59

Table S3. Biosynthesis of auxins in *S. netropsis* IMV Ac-5025 biomass grown in synthetic nutrient medium supplemented with exogenous β -sitosterol.

β -Sitosterol, μg/mL	Auxins, μg/g of absolutely dry biomass						
	Indole-3-acetic acid	Indole-3-acetic acid hydrazide	Indole-3-butiric acid	Indole-3-carbinol	Indole-3-carboxaldehyde	Indole-3-carboxylic acid	Sum of auxins
0	2.320±0.51	3.168±0.59	16.336±1.34	14.100±1.25	4.320±0.69	34.000±1.94	74.244±2.83
0.1	18.860±1.45	2.208±0.50	86.480±3.10	26.220±1.71	12.000±1.15	61.140±2.61	206.908±4.79
0.25	15.700±1.32	2.001±0.47	158.009±4.19	64.000±2.67	14.000±1.25	38.000±2.05	291.710±5.69
2.5	8.750±0.99	4.500±0.7	42.000±2.16	165.000±4.28	24.000±1.63	77.500±2.93	321.750±5.98
5	8.120±0.95	13.668±1.23	92.280±3.20	212.013±4.85	28.600±1.78	100.120±3.34	454.801±7.11
10	5.841±1.95	3.000±0.58	99.120±3.32	70.001±2.79	64.310±2.67	117.999±3.62	360.271±6.33

Table S4. Cytokinins' biosynthesis in *S. netropsis* IMV Ac-5025 biomass grown in synthetic nutrient medium supplemented with exogenous β -sitosterol.

β -Sitosterol, $\mu\text{g/mL}$	Cytokinins, $\mu\text{g/g}$ of absolutely dry biomass				
	Zeatin	Zeatin-riboside	Isopentyladenine	Isopentenyladenosine	Cytokinins' sum
0	48.093\pm0.58	61.612\pm2.45	86.121\pm2.26	58.007\pm2.21	253.833\pm4.51
0.1	237.016\pm2.38	187.044\pm2.4	375.211\pm16.58	303.783\pm4.28	1103.054\pm19.94
0.25	277.126\pm0.54	213.211\pm4.37	410.222\pm7.85	372.393\pm0.99	1272.952\pm14.22
2.5	67.215\pm0.34	63.211\pm0.96	535.041\pm4.65	489.522\pm0.66	1154.989\pm4.06
5	251.412\pm0.32	130.523\pm0.85	702.018\pm9.77	451.899\pm1.66	1535.852\pm13.47
10	660.336\pm1.08	396.225\pm6.39	980.135\pm21.26	660.319\pm16.72	2697.015\pm38.6