

Supplementary materials

Scientific Reports

Horizontal gene transfer and silver nanoparticles production in a new *Marinomonas* strain isolated from the Antarctic psychrophilic ciliate *Euplotes focardii*.

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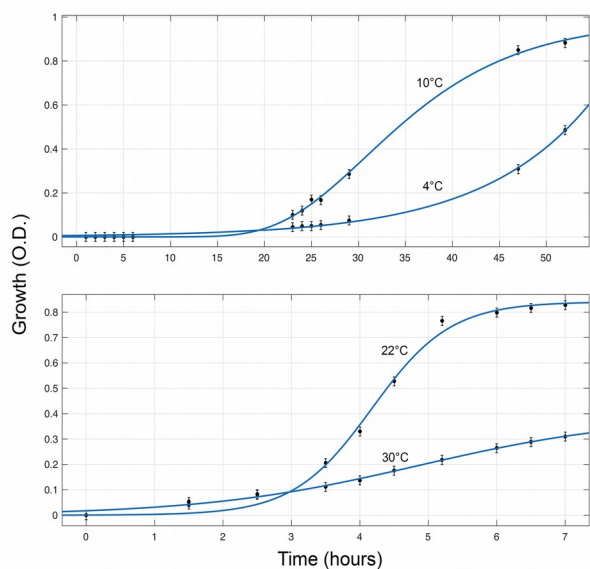


Fig. S1 - Growth curves of *Marinomonas sp. ef1* in LB medium at 4°C, 10°C, 22°C, and 30 °C. Data represent mean \pm standard error of three replicates.

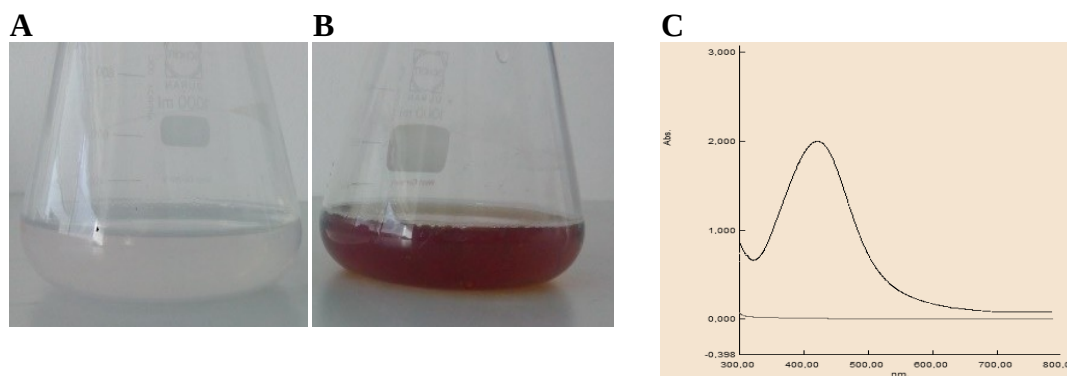
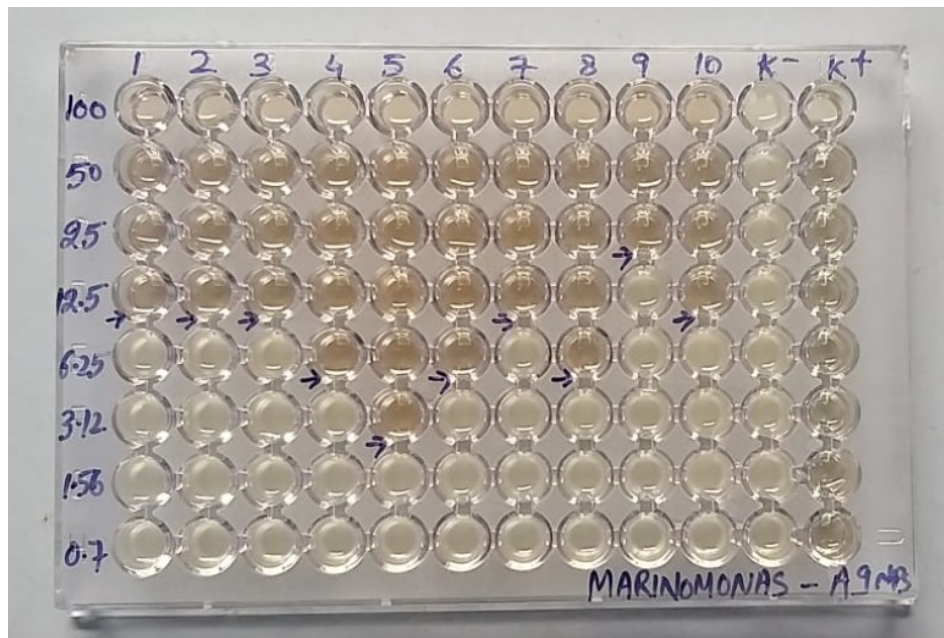


Figure S2: AgNP synthesis by *Marinomonas sp. ef1*. A and B: Visual observation of the AgNP synthesis. Medium colour change by the cultured *Marinomonas sp. ef1* from white (A) to dark brown (B) during incubation (24 hrs.) with 1mM of AgNO_3 . (C) UV-vis spectrum of *Marinomonas sp. ef1* AgNPs. A small aliquot (0.1 ml) of the culture was diluted with ddH₂O and UV-visible spectra was recorded from 300 to 800 nm wavelength at room temperature. The peak corresponds to 430 nm.

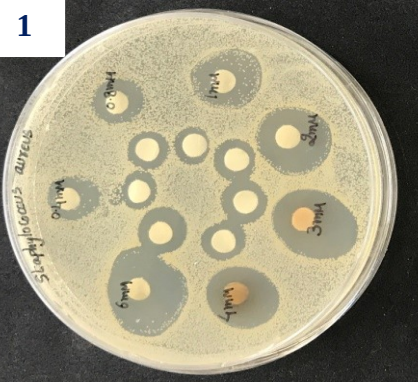


MIC and MBC of AgNPs against various pathogenic bacteria

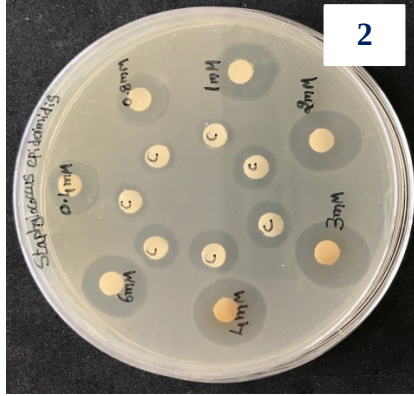
Pathogenic bacterial isolate	Reference isolate	MIC µg/ml	MBC µg/ml
<i>Staphylococcus aureus</i>	ATCC®25923	12.5±0.3	25±0.2
<i>Escherichia coli</i>	ATCC®25922	12.5±0.2	25±0.1
<i>Klebsiella pneumoniae</i>	ATCC®13883	12.5±0.3	25±0.3
<i>Pseudomonas aeruginosa</i>	ATCC®27853	6.25±0.1	12.5±0.2
<i>Proteus mirabilis</i>	ATCC®35659	3.12±0.1	6.25±0.1
<i>Citrobacter koseri</i>	ATCC®25408	6.25±0.2	6.25±0.1
<i>Acinetobacter baumannii</i>	ATCC®19606	12.5±0.2	12.5±0.3
<i>Serratia marcescens</i>	ATCC®13880	6.25±0.3	12.5±0.2
<i>Candida albicans</i>	ATCC®90028	25±0.5	25±0.2
<i>Candida parapsilosis</i>	ATCC®22019	12.5±0.3	25±0.3

Fig S3. Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration (MBC) assay of *Marinomonas* sp ef1 AgNPs by Broth microdilution method. 1. *Staphylococcus aureus* 2. *Escherichia coli* 3. *Klebsiella pneumoniae* 4. *Pseudomonas* sp 5. *Proteus mirabilis* 6. *Citrobacter koseri* 7. *Acinetobacter baumannii* 8. *Serratia marcescens* 9. *Candida albicans* 10. *Candida parapsilosis*

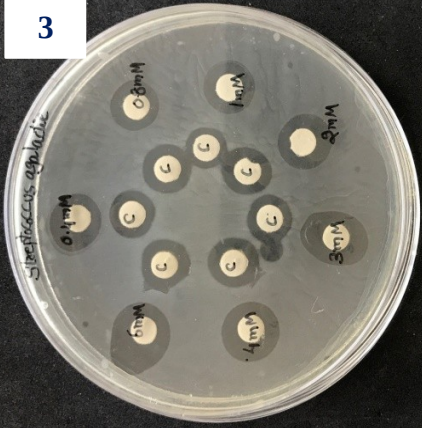
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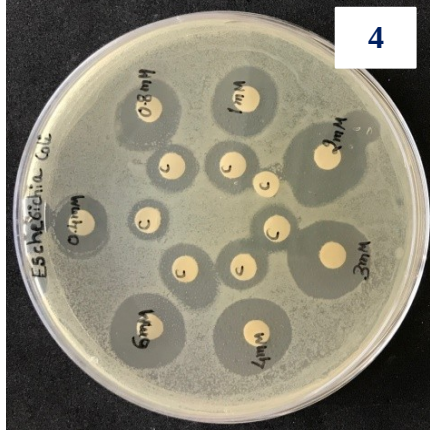
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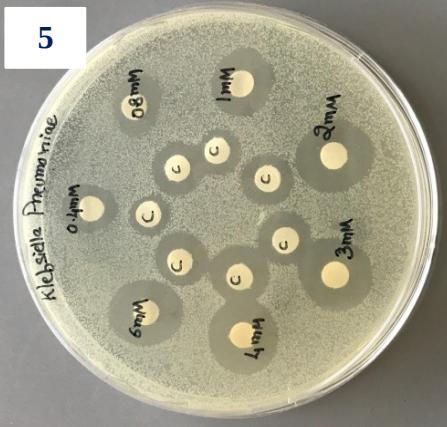
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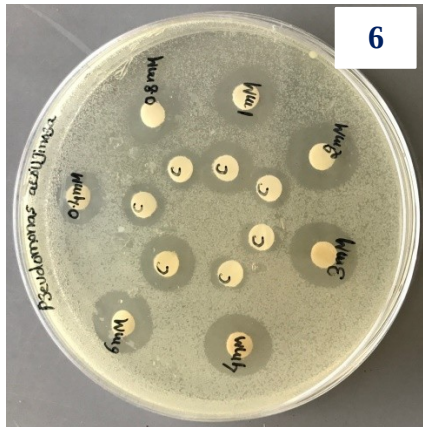
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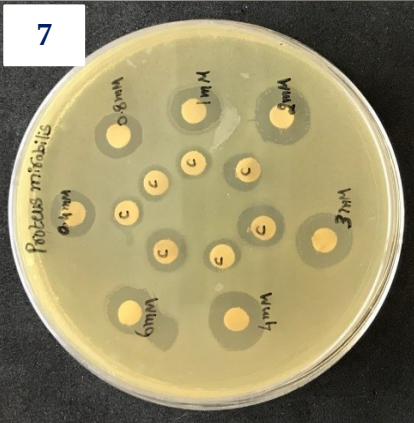
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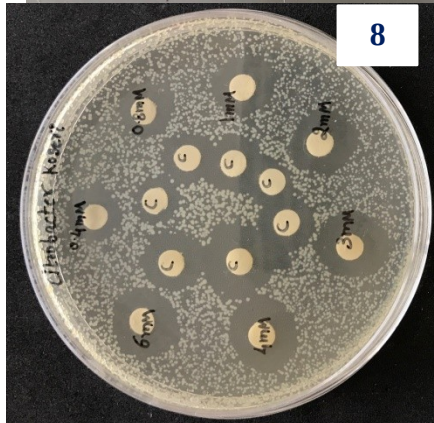
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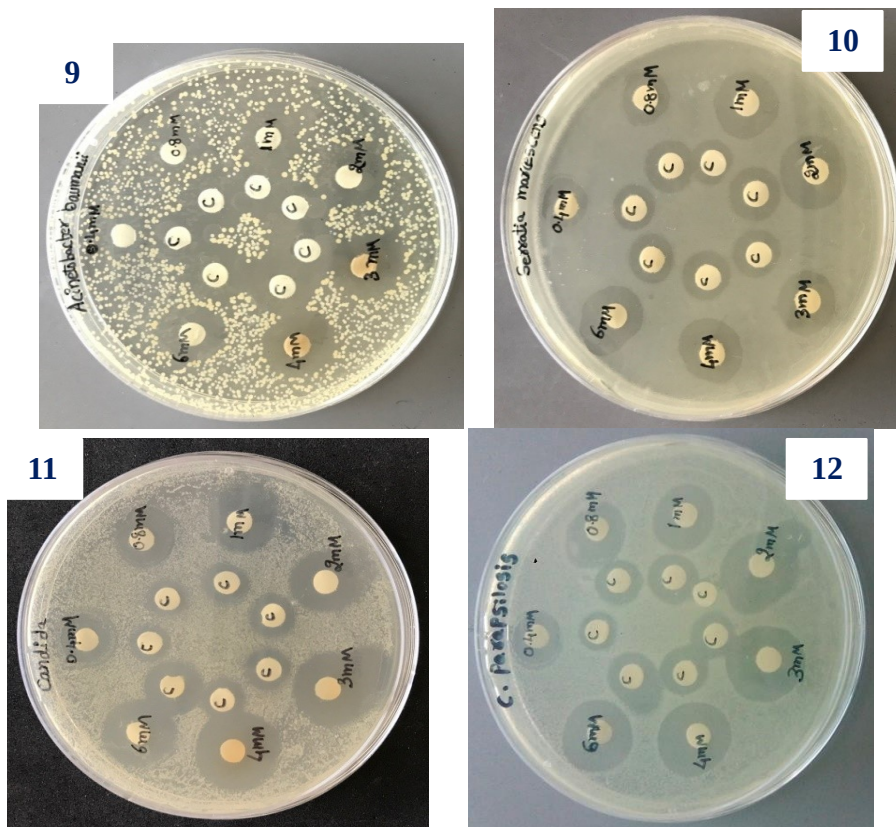


Fig. S3: Antimicrobial activity of increasing concentration (0.4mM, 0.8mM, 1mM, 2mM, 3mM, 4mM and 6mM) of AgNPs tested against twelve human pathogens and compared with increasing concentration (0.4mM, 0.8mM, 1mM, 2mM, 3mM, 4mM and 6mM) of AgNO₃. The human pathogens are: 1. *Staphylococcus aureus*, 2. *Staphylococcus epidermidis*, 3. *Streptococcus agalactiae*, 4. *Escherichia coli*, 5. *Klebsiella pneumoniae*, 6. *Pseudomonas aeruginosa*, 7. *Proteus mirabilis*, 8. *Citrobacter koseri*, 9. *Acinetobacter baumannii*, 10. *Serratia marcescens*, 11. *Candida albicans*, 12. *Candida parapsilosis*.

Supplementary tables

Table S1: List of the transposase genes present in *Marinomonas polaris* and *Marinomonas sp. ef1*

Marinomonas polaris transposases

Sequence Name	Sequence Description
EJ71DRAFT_00421	IS110 family transposase
EJ71DRAFT_01666	IS110 family transposase
EJ71DRAFT_02125	YKGN_ECOLIPUTATIVE PSEUDOGENE: ame: Full= transposase
EJ71DRAFT_02804	- Streptomyces griseus
EJ71DRAFT_02931	IS110 family transposase
EJ71DRAFT_03199	INH13_ECOLI ame: Full=Transposase for insertion sequence element IS5-13
EJ71DRAFT_03200	INSH5_ECOLI ame: Full=Transposase for insertion sequence element IS5Y
EJ71DRAFT_03752	INSH6_ECOLI ame: Full=Transposase for insertion sequence element IS5H
EJ71DRAFT_03753	IS110 family transposase
EJ71DRAFT_03856	INSN_SHIDY ame: Full=Transposase for insertion sequence element IS911
EJ71DRAFT_03944	YKGN_ECOLIPUTATIVE PSEUDOGENE: ame: Full= transposase
EJ71DRAFT_04184	INSN_SHIDY ame: Full=Transposase for insertion sequence element IS911
EJ71DRAFT_04367	INSN_SHIDY ame: Full=Transposase for insertion sequence element IS911
EJ71DRAFT_04466	INSH5_ECOLI ame: Full=Transposase for insertion sequence element IS5Y
EJ71DRAFT_04469	transposase
EJ71DRAFT_04490	INSH5_ECOLI ame: Full=Transposase for insertion sequence element IS5Y
EJ71DRAFT_04507	INSH5_ECOLI ame: Full=Transposase for insertion sequence element IS5Y
EJ71DRAFT_04544	INSN_SHIDY ame: Full=Transposase for insertion sequence element IS911
EJ71DRAFT_04547	ISSfl4 ORF1
EJ71DRAFT_04592	Transposase DDE domain

Marinomonas sp ef1 transposases

Sequence Name	Sequence Description
M_PROKKA_04338	INSH6_ECOLI ame: Full=Transposase for insertion sequence element IS5H
M_PROKKA_04339	INSH5_ECOLI ame: Full=Transposase for insertion sequence element IS5Y

Table S2: list of ORFs predicted from the *Marinomonas* sp. ef1 genomic islands

GI n.	Gene name	Percentage identity with more similar proteins							
1	Putative prophage CPS-53 integrase	<i>Marinomonas</i> sp ef1	100.00	49.63	50.49	51.84	22.93	24.32	26.01
		<i>Pantoea_agglomerans</i> _WP_033785581.1	49.63	100.00	51.73	50.82	22.28	25.59	23.98
		<i>Shewanella</i> _sp._Sh95_KPN78873.1	50.49	51.73	100.00	53.86	23.48	24.94	21.83
		<i>Vibrio_paraohaemolyticus</i> _WP_029807421.1	51.84	50.82	53.86	100.00	21.16	26.37	24.49
		<i>Marinomonas_atlantica</i> _WP_067098003.1	22.93	22.28	23.48	21.16	100.00	24.14	23.62
		<i>Marinomonas_spartinae</i> _WP_067011558.1	24.32	25.59	24.94	26.37	24.14	100.00	49.75
		<i>Marinomonas_polaris</i> _WP_072839418.1	26.01	23.98	21.83	24.49	23.62	49.75	100.00
2	Bacterial regulatory proteins, tetR family	<i>Marinomonas</i> sp ef1	100.00	52.68	51.47	52.68	18.72	22.50	22.50
		<i>Pseudomonas_amygdali</i> _KPY77198.1	52.68	100.00	59.31	95.77	22.17	21.50	21.50
		<i>Methylobacterium_versatilis</i> _301_ADI29544.1	51.47	59.31	100.00	59.31	22.28	22.50	22.50
		<i>Pseudomonas_syringae_coryli</i> _KPW95816.1	52.68	95.77	59.31	100.00	21.67	21.00	21.00
		<i>Marinomonas_gallaica</i> _WP_067219220.1	18.72	22.17	22.28	21.67	100.00	58.88	58.88
		<i>Marinomonas</i> _sp._MED121_EAQ65290.1	22.50	21.50	22.50	21.00	58.88	100.00	96.80
		<i>Marinomonas</i> _sp._S3726_KJZ10839.1	22.50	21.50	22.50	21.00	58.88	96.80	100.00
2	NAD(P)H nitroreductase YfkO	<i>Marinomonas</i> sp ef1	100.00	60.33	58.68	60.33	61.16	61.16	
		<i>Uliginosibacterium_gangwonense</i> _WP_018607979.1	60.33	100.00	78.51	65.29	65.29	66.94	
		<i>Pseudomonas_batumiensis</i> _WP_040067214.1	58.68	78.51	100.00	67.77	67.77	68.60	
		<i>Pectobacterium</i> _WP_116166044.1	60.33	65.29	67.77	100.00	72.31	73.55	
		<i>Dickeya</i> _sp._NCPBP	61.16	65.29	67.77	72.31	100.00	96.69	
		<i>Dickeya</i> _sp._Secpp	61.16	66.94	68.60	73.55	96.69	100.00	
2	Drug resistance protein MdtC	<i>Marinomonas</i> sp ef1	100.00	65.20	26.82				
		<i>Ralstonia_solanacearum</i> _WP_071093467.1	65.20	100.00	28.29				
		<i>Marinomonas_spartinae</i> _SBS25146.1	26.82	28.29	100.00				
3	Xylose operon regulatory protein	<i>Marinomonas</i> sp ef1	100.00	46.54	45.69	44.65	32.78	31.05	27.65
		<i>Verrucomicrobium_BVORR034</i> _WP_050027669.1	46.54	100.00	63.69	65.12	35.43	31.05	29.58
		<i>Sorangium_cellulosum</i> _KYG04766.1	45.69	63.69	100.00	70.61	37.87	32.79	31.94
		<i>bacterium_Ellin514_EEF60640</i> .1	44.65	65.12	70.61	100.00	35.43	33.33	29.90
		<i>Marinomonas_mediterranea</i> _MMB-1_ADZ90985.1	32.78	35.43	37.87	35.43	100.00	52.94	49.21
		<i>Marinomonas</i> _sp._MED121_EAQ63940.1	31.05	31.05	32.79	33.33	52.94	100.00	45.62
		<i>Marinomonas_atlantica</i> _WP_067094549.1	27.65	29.58	31.94	29.90	49.21	45.62	100.00
3	putative malonic semialdehyde reductase RutE	<i>Marinomonas</i> sp ef1	100.00	67.35	67.35	55.61	47.96	44.90	45.41
		<i>Cystobacter_ferrugineus</i> _OJH41342.1	67.35	100.00	96.94	55.10	50.51	44.90	43.88
		<i>Cystobacter_fuscus</i> _DSM_2262_EPX60408.1	67.35	96.94	100.00	55.61	49.49	43.88	44.39
		<i>Vogesella</i> _sp._	55.61	55.10	55.61	100.00	50.00	46.94	56.12
		<i>Marinomonas_ushuaiensis</i> _WP_051436225.1	47.96	50.51	49.49	50.00	100.00	75.71	39.50
		<i>Marinomonas</i> _sp._MED121_EAQ64291.1	44.90	44.90	43.88	46.94	75.71	100.00	37.00
		<i>Marinomonas</i> _sp._S3726_KJZ08849.1	45.41	43.88	44.39	56.12	39.50	37.00	100.00
4	2-keto-4-pentenoate hydratase	<i>Marinomonas</i> sp ef1	100.00	54.26	53.49	52.33			
		<i>Nitrospira_atlantica</i> _SEQ97558.1	54.26	100.00	61.78	58.69			
		<i>Marinobacterium_lutimaris</i> _SEG65920.1	53.49	61.78	100.00	61.78			
		<i>Nitrospira_nitratireducens</i> _EXJ13042.1	52.33	58.69	61.78	100.00			
4	Benzene 1,2-dioxygenase subunit beta	<i>Marinomonas</i> sp ef1		100.00	65.92	67.04	64.80		
		<i>Marinobacterium</i>		65.92	100.00	79.33	70.95		
		<i>Marinobacterium_georgiense</i> _DSM_11526_SEA66237.1		67.04	79.33	100.00	70.95		
		<i>Amphritea_atlantica</i> _SEQ97596.1		64.80	70.95	70.95	100.00		
4	Benzene 1,2-dioxygenase subunit alpha	<i>Marinomonas</i> sp ef1	100.00	81.92	82.37	82.18	24.82	25.53	25.29
		<i>Nitrospira_nitratireducens</i> _EXJ13038.1	81.92	100.00	85.71	84.60	23.71	23.42	24.12
		<i>Marinobacterium_georgiense</i>	82.37	85.71	100.00	85.94	25.82	24.59	25.29
		<i>Amphritea_atlantica</i> _SEQ97617.1	82.18	84.60	85.94	100.00	25.12	24.12	24.82
		<i>Marinobacterium_litorale</i> _WP_027852416.1	24.18	23.71	25.82	25.12	100.00	78.30	79.19
5	Sulfoacetaldehyde reductase	<i>Marinomonas</i> sp ef1	100.00	73.31	72.91	72.27	59.76	58.17	58.96
		<i>Aliagarivorans_marinus</i> _WP_026970871.1	73.31	100.00	98.02	68.92	58.50	57.71	54.18
		<i>Aliagarivorans_taiwanensis</i> _WP_026958872.1	72.91	98.02	100.00	68.53	57.71	56.52	54.58
		<i>Psychromonas_arctica</i> _WP_037071234.1	72.27	68.92	68.53	100.00	55.78	56.18	57.37
		<i>Marinomonas</i> _sp._MED121_EAQ66375.1	59.76	58.50	57.71	55.78	100.00	94.19	75.00
		<i>Marinomonas</i> _sp._SBI8L_KZM42849.1	58.17	57.71	56.52	56.18	94.19	100.00	75.40
		<i>Marinomonas_posidonica</i> _AEF55070.1	58.96	54.18	54.58	57.37	75.00	75.40	100.00
5	Taurine--pyruvate aminotransferase	<i>Marinomonas</i> sp ef1	100.00	79.56	76.70	78.02	64.54	64.54	62.78
		<i>Agarivorans_albus</i> _MKT_106_GAD02535.1	79.56	100.00	78.95	79.34	62.42	62.64	61.76
		<i>Psychromonas_ingrahamii</i> _37_ABM01925.1	76.70	78.95	100.00	83.74	61.57	62.34	61.57
		<i>Psychromonas_arctica</i> _WP_028868217.1	78.02	79.34	83.74	100.00	64.32	63.44	62.33
		<i>Marinomonas</i> _sp._TW1_KZN13315.1	64.54	62.42	61.57	64.32	100.00	93.26	81.96
		<i>Marinomonas_posidonica</i> _IVIA-Po-181_AEF55071.1	64.54	62.64	62.34	63.44	93.26	100.00	82.61
		<i>Marinomonas</i> _sp._SBI8L_KZM42848.1	62.78	61.76	61.57	62.33	81.96	82.61	100.00

Table S3: Comparison of *Marinomonas* sp ef1 AgNPs antimicrobial activity with that of chemical synthesized AgNPs and silver nitrate. 25 µl of 1mM of each solution were tested. Double distilled water was used as a negative control. R*= Resistance

S.No	Microorganism	Bio-AgNps (mm)	Chemical AgNps (mm)	AgNO ₃ (mm)	Dist.Water (mm)
Gram positive bacteria					
1	<i>Staphylococcus aureus</i>	15±0.4	13±0.5	8±0.3	R
2	<i>Staphylococcus epidermidis</i>	13±0.3	11±0.2	9±0.4	R
3	<i>Streptococcus agalactie</i>	14±0.2	12±0.4	8±0.2	R
Gram negative bacteria					
4	<i>Escherichia coli</i>	17±0.3	14±0.3	11±0.2	R
5	<i>Klebsiella pneumoniae</i>	16±0.1	13±0.2	8±0.4	R
6	<i>Pseudomonas sp</i>	15±0.2	12±0.3	9±0.1	R
7	<i>Proteus mirabilis</i>	14±0.3	12±0.2	8±0.2	R
8	<i>Citrobacter koseri</i>	15±0.1	13±0.3	10±0.4	R
9	<i>Acinetobacter baumannii</i>	14±0.4	11±0.2	10±0.3	R
10	<i>Serratia marcescens</i>	14±0.3	11±0.3	9±0.2	R
Fungi					
11	<i>Candida albicans</i>	14±0.4	11±0.4	8±0.2	R
12	<i>Candida parapsilosis</i>	12±0.2	10±0.2	8±0.2	R

Table S4: Antimicrobial activity of ampicillin and anphoteracin B against pathogenic microorganisms. R*= Resistance

S.No	Microorganism	6 mM (mm)	12 mM (mm)	15 mM (mm)	25 mM (mm)
Gram positive bacteria					
1	<i>Staphylococcus aureus</i>	18±0.2	21±0.3	25±0.2	27±0.3
2	<i>Staphylococcus epidermidis</i>	22±0.1	24±0.2	26±0.3	29±0.2
3	<i>Streptococcus agalactie</i>	13±0.2	16±0.4	18±0.2	22±0.2
Gram negative bacteria					
4	<i>Escherichia coli</i>	10±0.1	14±0.3	16±0.2	20±0.2
5	<i>Klebsiella pneumoniae</i>	12±0.1	16±0.2	18±0.2	24±0.1
6	<i>Pseudomonas sp</i>	18±0.2	22±0.3	24±0.1	28±0.1
7	<i>Proteus mirabilis</i>	14±0.3	16±0.2	19±0.2	24±0.2
8	<i>Citrobacter koseri</i>	R	R	R	12±0.2
9	<i>Acinetobacter baumannii</i>	R	R	R	R

10	<i>Serratia marcescens</i>	15±0.3	17±0.3	19±0.2	26±0.2
Fungi					
S.No	Microorganism	2 mM (mm)	4 mM (mm)	6 mM (mm)	10 mM (mm)
11	<i>Candida albicans</i>	R	R	12±0.2	22±0.1
12	<i>Candida parapsilosis</i>	R	13±0.2	16±0.1	24±0.1