

Supplementary Table S7. Variable proteins in *N. gonorrhoeae* biofilms listed by functional roles^a: Biofilm/Planktonic ratios expressed as log₂ values, 1.5-fold cutoff threshold (log₂ values either ≥ 0.585 or ≤ -0.585), p-values <0.05

| Accession | Name | JCVI sub-role | Extract 1 Avg ^b (N) ^c | Extract 2 Avg ^b (N) ^c | Extract 3 Avg ^b (N) ^c |
|---|--|---|--|--|--|
| Amino acid biosynthesis | | | | | |
| NGO1358 | glutamate dehydrogenase | Glutamate family | 0.888 (3) | -0.947 (2) | -1.726 (2) |
| Cell envelope | | | | | |
| NGO0332 | putative reductase; dTDP-4-dehydrorhamnose reductase | Biosynthesis and degradation of surface polysaccharides and lipopolysaccharides | -0.807 (1) | 0.856 (1) | |
| NGO1056 | hypothetical protein; lipoprotein NlpD | Other | | -1.151 (1) | 0.954 (3) |
| Cellular processes | | | | | |
| NGO0328 ^e | hypothetical protein; peroxiredoxin Q/BCP | Detoxification | -0.681 (1) | 0.614 (1) | |
| NGO0926 | putative peroxiredoxin family protein/glutaredoxin | Detoxification | | 0.610 (1) | -0.655 (1) |
| Energy metabolism | | | | | |
| NGO0617 | Eno; phosphopyruvate hydratase; enolase | Glycolysis/gluconeogenesis | 1.120 (3) | -0.900 (2) | -1.812 (1) |
| NGO0562 | putative dihydrolipoamide dehydrogenase | Pyruvate dehydrogenase | 0.996 (3) | -0.731 (3) | -1.142 (2) |
| NGO0915 | dihydrolipoamide dehydrogenase | Pyruvate dehydrogenase | 0.841 (3) | -0.967 (2) | |
| NGO2150 | F ₀ F ₁ ATP synthase subunit beta; F-type H ⁺ -transporting ATPase subunit beta | ATP-proton motive force interconversion | 0.826 (1) | -1.543 (2) | -1.719 (1) |
| NGO0916 | dihydrolipoamide succinyltransferase | TCA cycle | 0.754 (1) | | -2.314 (1) |
| NGO0217 | ABC transporter, periplasmic binding protein, iron related; iron(III) transport system substrate-binding protein | Electron transport | 0.618 (2) | -1.657 (3) | -0.958 (3) |
| NGO0200 | phosphoenolpyruvate synthase | Glycolysis/gluconeogenesis | 0.598 (3) | -1.351 (1) | -1.403 (1) |
| Protein synthesis | | | | | |
| NGO1853 | RplJ; 50S ribosomal protein L10 | Ribosomal proteins: synthesis and modification | 0.648 (1) | | -1.213 (1) |
| NGO2134 | RpsU; 30S ribosomal protein S21 | Ribosomal proteins: synthesis and modification | 0.592 (1) | | -0.852 (1) |
| NGO1825 ^f | RplF; 50S ribosomal protein L6 | Ribosomal proteins: synthesis and modification | | -0.701 (1) | 0.799 (2) |
| NGO1824 ^g | RpsE; 30S ribosomal protein S5 | Ribosomal proteins: synthesis and modification | | -1.000 (1) | 0.586 (1) |
| Purines, pyrimidines, nucleosides, and nucleotides | | | | | |
| NGO0799 | inosine 5'-monophosphate dehydrogenase | Purine ribonucleotide biosynthesis | 1.180 (3) | -0.716 (2) | |
| NGO0614 | ribonucleotide-diphosphate reductase subunit alpha | 2'-Deoxyribonucleotide metabolism | -0.656 (1) | 1.086 (1) | |
| Regulatory functions | | | | | |
| NGO0460 ^d | succinyldiaminopimelate transaminase; acetylornithine/N-succinyldiaminopimelate aminotransferase | Other | -0.622 (1) | 0.681 (1) | |
| Transcription | | | | | |
| NGO0335 | polynucleotide phosphorylase/polyadenylase; polyribonucleotide nucleotidyltransferase | Degradation of RNA | 0.981 (3) | -1.093 (3) | |
| NGO1850 | DNA-directed RNA polymerase subunit beta' | DNA-dependent RNA polymerase | 0.805 (3) | -0.770 (1) | -0.595 (2) |
| NGO1851 | RpoB; DNA-directed RNA polymerase subunit beta | DNA-dependent RNA polymerase | 0.735 (3) | -1.045 (2) | -0.892 (2) |
| NGO1818 | DNA-directed RNA polymerase subunit alpha | DNA-dependent RNA polymerase | 0.585 (3) | -1.063 (3) | |
| Transport and binding proteins | | | | | |
| NGO1494 | putative ABC transporter, periplasmic binding protein, polyamine | Amino acids, peptides and amines | 1.287 (3) | | -0.970 (1) |
| NGO1656 ^h | hypothetical protein | Unknown substrate | 0.760 (1) | -1.094 (1) | |

^a Functional role assignments (both main and JCVI sub-roles) for the *N. gonorrhoeae* proteins were downloaded from the JCVI-CMR website (<http://cmr.icvi.org/tigr-scripts/CMR/CmrHomePage.cgi>).

^b Average log₂ values for biofilm/planktonic protein ratios. Values given in red meet the 2-fold cutoff threshold (log₂ values either ≥ 1.000 or ≤ -1.000).

^c (N) represents the number of measurements (biological replicates) averaged.

^d Proteins with more than one functional role assignment provided (alternate Main role/JCVI sub-role): NGO0460, Unknown function/Enzymes of unknown specificity.

^e NGO0328 tentatively reclassified based on orthology to *N. meningitidis* NMCC_0714 (97.9% identity).

^f NGO1825 tentatively reclassified based on orthology to *N. meningitidis* NMCC_1992 (97.7% identity), NMC0147 (97.7% identity), and NMB0157 (97.7% identity).

^g NGO1824 tentatively reclassified based on orthology to *N. meningitidis* NMB0159 (100% identity) and NMA0112 (100% identity).

^h NGO1656 tentatively reclassified based on orthology to *N. meningitidis* NMCC_1797 (98.6% identity). Possible alternate Main role/JCVI sub-role assignments for this protein include: Transport and binding proteins/Amino acids, peptides and amines (NMCC_1797) and Cell envelope/Other (NMB0345, 98.6% identity).