TransAAP: A Web-based Transporter Annotation Tool

User Guide

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TransAAP is a web-based transporter annotation tool. It allows users to submit their favorite genomes for transporter annotation utilizing our high-thoughput transporter analysis pipeline, view automatic annotation and supporting evidence, as well as curate the transporter annotation, all of which are through a user-friendly web interface. This manual summarizes the process and illustrates the procedures step by step.

To maximize the possibility of finding out all the potential candidates of transporters, we employed four different kinds of searches with a query genome (Figure 1): blastp search against our internal curated transporter database and a general non-redundant protein database; HMM search against Pfam and TIGRfam; rpsblast seach against NCBI COG; and transmembrane segment predictions by tmhmm. These searches are mutual inclusive, which means any positive hits above the cutoff from any of the four searches will be collected as potential transporters. In the meantime, there are a set of negative rules enforced to remove the false positives. Each predicted transporter will be shown on individual web pages where users can view annotation, supporting evidence, and curate the annotation.





Users can go to the TransportDB web site, click on the link to TransAAP (<u>http://www.membranetransport.org/transaap/TransAAP login.html</u>), and login as a registered user. To submit a request for transporter annotation, users only need to fill in simple information like Taxonomy ID for genomes deposited in the Genebank, TIGR database name for a TIGR/JCVI sequenced genome, or the FASTA format protein sequences/links to FTP protein sequence download site for unfinished genomes (Figure 2).

| TransAAP: Transporter Automatic Annotation Pipeline | | | | | | | | | |
|---|--|--|--|--|--|--|--|--|--|
| Submit Your Genome | | | | | | | | | |
| User Name: | test | | | | | | | | |
| Genus Name: | | | | | | | | | |
| Species Name: | | | | | | | | | |
| Strain Name: | | | | | | | | | |
| Genome Source: | Complete Genome deposited to GeneBank (No need for FASTA sequences) NCBI Taxonomy ID: TIGR Genome (No need for FASTA sequences) Database Name Unfinished Genome (Please paste peptide FASTA file below) | | | | | | | | |
| | | | | | | | | | |
| | Submit Reset | | | | | | | | |

Figure 2: TransAAP: submit a query genome

Once we received the request, we will run all the required searches, predict the complete transporter contents, and load all the results to a database. Users can check the status of the annotation on their submitted genomes (Figure 3)

| TransAAP: Transporter Automatic Annotation Pipeline | | | | | | | | | | |
|---|-----------|-------------|---------|------------|---------|--------------------|-------------------------------|--|--|--|
| List of genomes you submitted for annnotation (Click on <u>GenomeID</u> or <u>Status</u> to see the full list of transporters) | | | | | | | | | | |
| GenomelD | Genus | Species | Strain | TaxonomyID | TIGR_db | Date of Submission | Status | | | |
| N/A | Nautilia | sp. | Am-H | | gamh | 2007-03-13 | Submitted | | | |
| ngon1 | Neisseria | gonorrhoeae | FA 1090 | 242231 | | 2007-03-09 | Automatic Annotation Finished | | | |
| | | | | | | | | | | |

Figure 3: TransAAP: check the status of submitted genomes

After all the searches and automatic annotation are finished, uses can access a list of predicted transporters (Figure 4), or individual transporter annotation pages (Figure 5). Users can view annotation, supporting evidence, and curate the annotation on each individual transporter annotation page.

| TransAAP: Transporter Automatic Annotation Pipeline | | | | | | | | | |
|---|----------|---------------------|---|-----------------|------------------------------|----------------------|--------------------|--|--|
| User Name: tes Genome ID: ngo | t in1 | | | | | | | | |
| Complete List of Membrane Transporter in the Genome Total transporter genes predicted: 141 | | | | | | | | | |
| ORF Name <mark>order</mark> | GenomelD | FID <u>order</u> | Transporter Family | Subfamily | Substrate/Function order | Auto- annotation? | Curation? order | | |
| NG00023 | ngon1 | ABC | The ATP-binding Cassette (ABC) Superfamily | | iron compounds | | | | |
| NG00088 | ngon1 | МОР | The Muttidrug/Oligosaccharidyl-lipid/Polysaccharide (MOP) Flippase Superfamily | PST | polysaccharide export | | × | | |
| NG00110 | ngon1 | CitMHS | The Citrate-Mg2+:H+ (CitM) Citrate-Ca2+:H+ (CitH) Symporter (CitMHS) Family | | proton:citrate symporter | | × | | |
| NG00136 | ngon1 | NhaC | The NhaC Na+:H+ Antiporter (NhaC) Family | | sodium ion:proton antiporter | | | | |
| NG00142 | ngon1 | MFS | The Major Facilitator Superfamily (MFS) | | glucose/galactose | | | | |
| NG00143 | ngon1 | NhaC | The NhaC Na+:H+ Antiporter (NhaC) Family | | sodium ion:proton antiporter | | × | | |
| NG00168 | ngon1 | ABC | The ATP-binding Cassette (ABC) Superfamily | binding protein | manganese/zinc ion | | × | | |
| NG00169 | ngon1 | ABC | The ATP-binding Cassette (ABC) Superfamily | membrane | manganese/zinc ion | | × | | |
| NG00170 | ngon1 | ABC | The ATP-binding Cassette (ABC) Superfamily | ABC | manganese/zinc ion | | × | | |
| NG00181 | ngon1 | Tat | The Twin Arginine Targeting (Tat) Family | | protein export | | × | | |
| NG00182 | ngon1 | Tat | The Twin Arginine Targeting (Tat) Family | | protein export | | × | | |
| NG00183 | ngon1 | Tat | The Twin Arginine Targeting (Tat) Family | | protein export | | × | | |
| NG00189 | ngon1 | RND | The Resistance-Nodulation-Cell Division (RND) Superfamily | SecDF | protein export (SecDF) | | × | | |
| NG00190 | ngon1 | RND | The Resistance-Nodulation-Cell Division (RND) Superfamily | SecDF | protein export (SecDF) | | × | | |

Figure 4: TransAAP: list of predicted transporters

TransAAP: Transporter Automatic Annotation Pipeline

| <u>Username:</u> te | st <u>Ge</u> | nomelD: ngon1 | Return to the T | ransporter List in ngon1 | | | | | | | |
|---|--|--|--|---|--|---|---|----------------------------------|---|--|--|
| NGO0136 The NhaC Na+:H+ Antiporter (NhaC) Family sodium ion:proton antiporter | | | | | | | | Genome: ngon1 ORF Name: New Gene | | | |
| Transmont | or list t | ndav | | | | | | | | | |
| iransport | | ndex NCON | 188 | NCO8118 | | NCO0436 | NG00142 | | NC00143 | | |
| Fa | mily | MO | P CitMHS | | | NhaC | MFS | | NhaC | | |
| < Subfa | mily | PST | 1 | | | | | | >>> | | |
| Subst | Substrate /Function polysaccharide export proton:citrate symporter sodium ion:proton antiporter glucose/galactose sodium | | | | | | | | ium ion:proton antiporter | | |
| Transport | Annot | ation: | | | | | | | | | |
| Transporte | er Predio | tion #1: | | | | | | | | | |
| FID | TC# | Transport | er Farnily | Subfamily | Subst | trate/Function | | | | | |
| NhaC | 2.A.3 | 5 The NhaC (NhaC) Far | Na+:H+ Antiport nilv | er | sodi | um ion:proton antiporter | | | | | |
| Comments | 8: | (| | | | | | | | | |
| annotat | ed | | | | | | | | elete this annotation? Submit Change | | |
| Transmen | nbrane | Segment Pr | ediction (Trr | HMM): | | | | | | | |
| Protein Len 506 | igth: | | Topology: | | | | | | | | |
| Total: 13 TMS 1: TMS 2: TMS 2: TMS 3: TMS 4: TMS 6: TMS 6: TMS 6: TMS 8: TMS 8: TMS 10: TMS 10: TMS 11: TMS 12: | Yrdicted TMS: >NGO0136 hypothetical protein [Neisseria gonorrhoeae FA 1090] Tota: 13 MOLIDYSHSFFSVYPPFLALALAVI TRRVIDSIGIG LIVGVAPLVGONFVDGLTHLKDNVYGLANADGDM TMS1: 4-26 SLGKPK ILVVFFLLLGTFJLLTYSSGNOAFADMAKENIKTRCGAKAVGTACUPPYTFIDDYFLAVGANAVGATA TMS2: 28-50 PPVTDKFKVSRAKLAYILDSTASPMCVLMPVSSMGASILAUTTACULPYTFIDDYFLAVGANIVYGA TMS3: 77-96 FALIMPYVVAMFSPDIGSMAFFCOALNEADATKRHCGAKASDATKGKVYALLISTVAMINTGAOA TMS4: 174-139 SETFSILGAPENTDVNTSLVFGGTCGVLAVVLCTFGTIKTADDYFKAVMQGAKSMFGAIALLILAULISTV TMS6: 167-169 VGEMITGD VLSTLVÅGNINPGGTPVLIFFLASVMAFATGTSMOTFGIME FLAAMAVKVEFALIIPCMSA TMS6: 201-223 VGEMITGD VLSTLVÅGNINPGGTPVLIFFLASVMAFATGTSMOTFGIME FLAAMAVKVEFALIIPCMSA TMS7: 256-276 LAVLIFFL TMS9: 393-318 SETSILGAFENTDV TMS1: 392-414 SETSIGA TMS1: 392-414 SETSIGA | | | | | | | | | | |
| ITCH SPOTU TDB (NMB053 TDB (HD1227 TDB (HD1227 TDB (HD1227 TDB (HD1227 TDB (MS1321 CDB (MS1321 CDB (MS1321) | 15 & IC 31 1567 7 33152 1 15602 5 16273 1 52425 | DE Bldst Sec 6438 NhaCl2. 323 NhaCl2. 736 NhaCl2. 480 NhaCl2. 376 NhaCl2. | A.35 sodium A.35 sodium A.35 sodium A.35 sodium A.35 sodium A.35 sodium | ion/proton conserv ion/proton conserv /proton unknown [P ion/proton? conserv ion/proton NhaC pr | ved hyp ved put asteure rved hy rotein | othetical protein [Neisser: ative integral membrane pro lla multocida Pm70] pothetical integral membran [Mannheimia succiniciproduc | ia meningitidis MC58] otein [Haemophilus ducre ne protein [Haemophilus cens MBEL55E] | yi 35000H influenza | 2514 5.7e-263 P] 1813 1.1e-18 1543 4.5e-155 E KW20] 1540 9.4e-155 1514 5.3e-156 | | |
| | phyloge | | Transporter II | | | | | | | | |
| COG Sear | rch | | | | | | | | | | |
| COG1757 | Nha | C, Na+/H+ a | ntiporter [E | nergy production a | and conv | version]. | | | 292 3e-80 | | |
| Pfam Soar | ch | | | | | | | | | | |
| 2.A.35 | S. | | | NhaC | | The NhaC Na+:H+ Antipor | ter (NhaC) Family | | | | |
| PF03553 | 3. | 8e-102 | Na+/H+ antipo | rter family | | | | 1 31 | 9 165 476 | | |
| NRAA Blas | st Sear | sh | | | | | | | | | |
| RFIYP_207 | 306.1 | h r | ypothetical f_status=pro Neisseria go | protein (Neisseria ovisional;)^ ^GB AA onorrhoeae FA -TRUM | a gonor: AW88894 NCA | rhoeae FA 1090;} (exp=0; wg .1 59717489 AE004969 conse | <pre>gp=1; cg=1; closed=1; pu wed hypothetical protei;</pre> | b=0; n 2553 | 5 1.2e-264 | | |
| RFINP_273 | 576.1 | h | ypothetical f_status=pro | protein (Neisseria ovisional;)^ ^GB AA | a menin AF40961 | gitidis MC58;} (exp=0; wgp= .1 7225756 AE002098 conserv | =1; cg=1; closed=1; pub= red hypothetical protein | 1; 2514 | 4 2.6e-260 | | |
| GB CAM097 | 70.1 | p | utative inte losed=0; pub | egral membrane prot ==0; rf status=;) | tein (N | eisseria meningitidis FAMI8 | 3;) (exp=0; wgp=0; cg=1; | 2500 | 0 8.0e-259 | | |
| RFINP_283 | 509.1 | i p p | ntegral memb ub=l; rf_sta rotein {Neis | orane protein (Neis atus=provisional;)/ sseria mening-TRUNO | sseria : ^ ^GB C. C A | meningitidis Z2491;} (exp=0 AM07964.1 121051661 AL15795 |); wgp=1; cg=1; closed=1 59 putative integral mem | ; brane 2490 |) 9.1e-258 | | |
| Conse RFINP_873676.1 cg=1. puta | | onserved put g=l; closed- utative inte | <pre>served putative integral membrane protein (Haemophilus ducreyi 35000HP;) (exp=0; wgp=1; l; closed=1; pub=0; rf_status=provisional;)^ ^GB AAP96065.1 33148546 AE017143 conserved ative integral membran=TRUNCA</pre> | | | | | 3 5.0e-186 | | | |
| hypothetical protein (Pasteurella cg=1; closed=1; pub=1; rf_status= (Pasteurella multocida subsTF | | | | protein (Pasteure) 1; pub=1; rf_statu multocida subsp. | lla mul us=prov -TRUNCA | <pre>multocida subsp. multocida str. Pm70;) (exp=0; wgp=1; provisional;)^\^GB AAK02955.1 12721182 AE004439 unknow; UNCA</pre> | | | 3 2.0e-157 | | |
| SP P44263 | | H P (| ypothetical ub=0; rf_sta Haemophilus | protein HI1586. (H atus= ;)^ ^RF[NP_43 influenzae Rd KW20 | Haemoph 39731.1 D;} (e-' | ilus influenzae;) (exp=0; u 16273480 NC_000907 isoleud TRUNCA | ogp=-1; cg=-1; closed=-1 cyl-tRNA synthetas | ; 1540 | 0 4.3e-157 | | |
| OMNI (HI_1 | 586 | N P | <pre>ha+/H+ antipo hub=-1; rf_st</pre> | orter family (Haemo tatus= ;) | ophilus | influenzae Rd KW20;) (exp | -l; wgp=-l; cg=l; close | d=-1; 1540 | 0 4.3e-157 | | |
| RF YP_248 | 941.1 | p c a | redicted Na- losed=1; pub ntiporter (H | +/H+ antiporter (Ha b=1; rf_status=prov Haemophilus influer | aemophi visiona n-TRUNC, | lus influenzae 86-028NP;) 1;)^ ^GB AAX88281.1 6805802 A | (exp=0; wgp=1; cg=1; 28 CP000057 predicted Na | +/H+ 1539 | 9 5.4e-157 | | |
| RF[YP_068513.1 rf_status=provisional;}/*/GB[AM379263.1]52307428[AE016627 NhaC protein (Mannheimia 1514 succiniciproducens MDELSSE;) (ex-TRUNCA | | | | | | | 4 2.4e-154 | | | | |

Figure 5: TransAAP: individual transporter annotation page