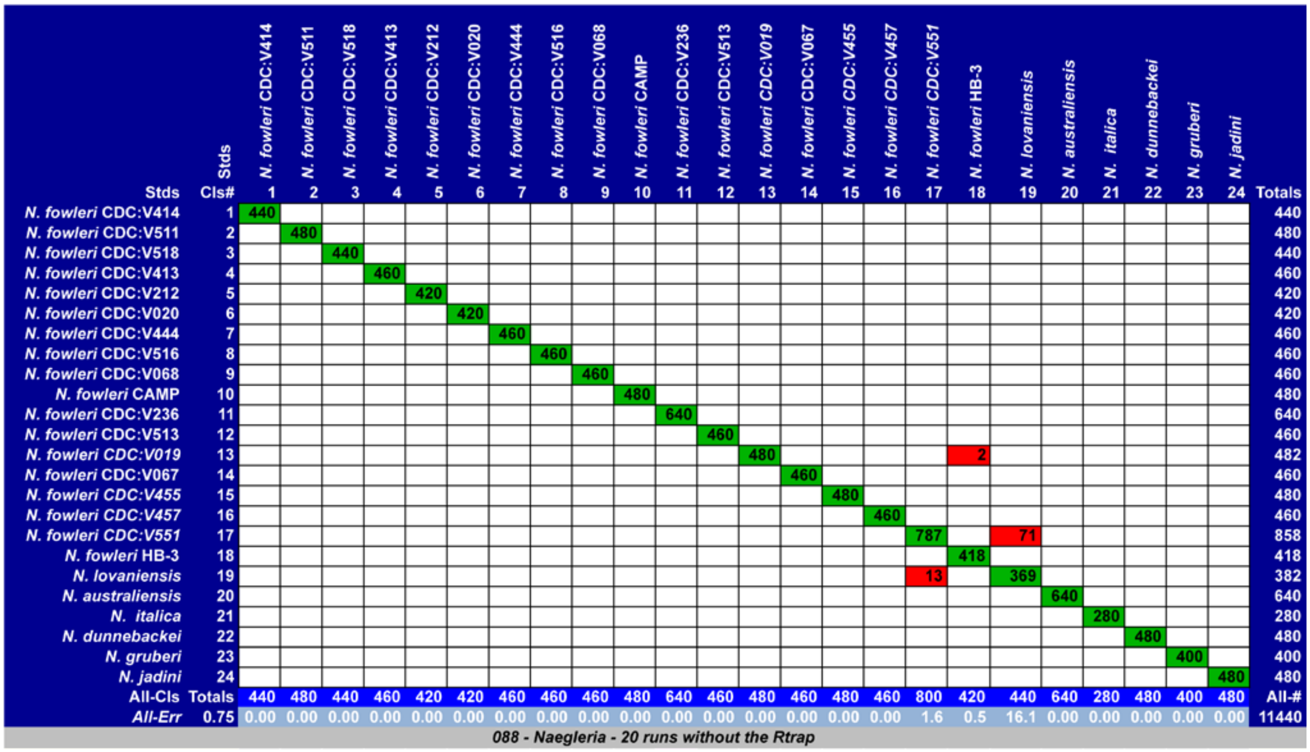
## SUPPORTING INFORMATION

**Detection of Biomarkers of Pathogenic *Naegleria fowleri* through Mass Spectrometry and Proteomics** by Hercules Moura, Fernando Izquierdo, Adrian R. Woolfitt, Glauber Wagner, Tatiana Pinto, Carmen del Aguila & John R. Barr



| Stds | Stds Cls# | *N. fowleri* CDC:V414 1 | *N. fowleri* CDC:V511 2 | *N. fowleri* CDC:V518 3 | *N. fowleri* CDC:V413 4 | *N. fowleri* CDC:V212 5 | *N. fowleri* CDC:V020 6 | *N. fowleri* CDC:V444 7 | *N. fowleri* CDC:V516 8 | *N. fowleri* CDC:V068 9 | *N. fowleri* CAMP 10 | *N. fowleri* CDC:V236 11 | *N. fowleri* CDC:V513 12 | *N. fowleri CDC:V019* 13 | *N. fowleri* CDC:V067 14 | *N. fowleri CDC:V455* 15 | *N. fowleri CDC:V457* 16 | *N. fowleri CDC:V551* 17 | *N. fowleri* HB-3 18 | *N. lovaniensis* 19 | *N. australiensis* 20 | *N. italica* 21 | *N. dunnebackei* 22 | *N. gruberi* 23 | *N. jadini* 24 | Totals |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| *N. fowleri* CDC:V414 | 1 | 440 | | | | | | | | | | | | | | | | | | | | | | | | 440 |
| *N. fowleri* CDC:V511 | 2 | | 480 | | | | | | | | | | | | | | | | | | | | | | | 480 |
| *N. fowleri* CDC:V518 | 3 | | | 440 | | | | | | | | | | | | | | | | | | | | | | 440 |
| *N. fowleri* CDC:V413 | 4 | | | | 460 | | | | | | | | | | | | | | | | | | | | | 460 |
| *N. fowleri* CDC:V212 | 5 | | | | | 420 | | | | | | | | | | | | | | | | | | | | 420 |
| *N. fowleri* CDC:V020 | 6 | | | | | | 420 | | | | | | | | | | | | | | | | | | | 420 |
| *N. fowleri* CDC:V444 | 7 | | | | | | | 460 | | | | | | | | | | | | | | | | | | 460 |
| *N. fowleri* CDC:V516 | 8 | | | | | | | | 460 | | | | | | | | | | | | | | | | | 460 |
| *N. fowleri* CDC:V068 | 9 | | | | | | | | | 460 | | | | | | | | | | | | | | | | 460 |
| *N. fowleri* CAMP | 10 | | | | | | | | | | 480 | | | | | | | | | | | | | | | 480 |
| *N. fowleri* CDC:V236 | 11 | | | | | | | | | | | 640 | | | | | | | | | | | | | | 640 |
| *N. fowleri* CDC:V513 | 12 | | | | | | | | | | | | 460 | | | | | | | | | | | | | 460 |
| *N. fowleri CDC:V019* | 13 | | | | | | | | | | | | | 480 | | | | | 2 | | | | | | | 482 |
| *N. fowleri* CDC:V067 | 14 | | | | | | | | | | | | | | 460 | | | | | | | | | | | 460 |
| *N. fowleri CDC:V455* | 15 | | | | | | | | | | | | | | | 480 | | | | | | | | | | 480 |
| *N. fowleri CDC:V457* | 16 | | | | | | | | | | | | | | | | 460 | | | | | | | | | 460 |
| *N. fowleri CDC:V551* | 17 | | | | | | | | | | | | | | | | | 787 | | 71 | | | | | | 858 |
| *N. fowleri* HB-3 | 18 | | | | | | | | | | | | | | | | | | 418 | | | | | | | 418 |
| *N. lovaniensis* | 19 | | | | | | | | | | | | | | | | | 13 | | 369 | | | | | | 382 |
| *N. australiensis* | 20 | | | | | | | | | | | | | | | | | | | | 640 | | | | | 640 |
| *N. italica* | 21 | | | | | | | | | | | | | | | | | | | | | 280 | | | | 280 |
| *N. dunnebackei* | 22 | | | | | | | | | | | | | | | | | | | | | | 480 | | | 480 |
| *N. gruberi* | 23 | | | | | | | | | | | | | | | | | | | | | | | 400 | | 400 |
| *N. jadini* | 24 | | | | | | | | | | | | | | | | | | | | | | | | 480 | 480 |
| All-Cls | Totals | 440 | 480 | 440 | 460 | 420 | 420 | 460 | 460 | 460 | 480 | 640 | 460 | 480 | 460 | 480 | 460 | 800 | 420 | 440 | 640 | 280 | 480 | 400 | 480 | All-# |
| *All-Err* | 0.75 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 1.6 | 0.5 | 16.1 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 11440 |

*088 - Naegleria - 20 runs without the Rtrap*

**Fig.S1**. Random Forest analysis of the studied *Naegleria* species and isolates. Datasets consisted of summed spectra from three harvestings of each organism as a training set (columns) and ~3 summed spectra as unknowns (rows). The estimated classification error was 0.75%, and the RF algorithm successfully classified all the *Naegleria* species and the majority of the isolates in the training set.

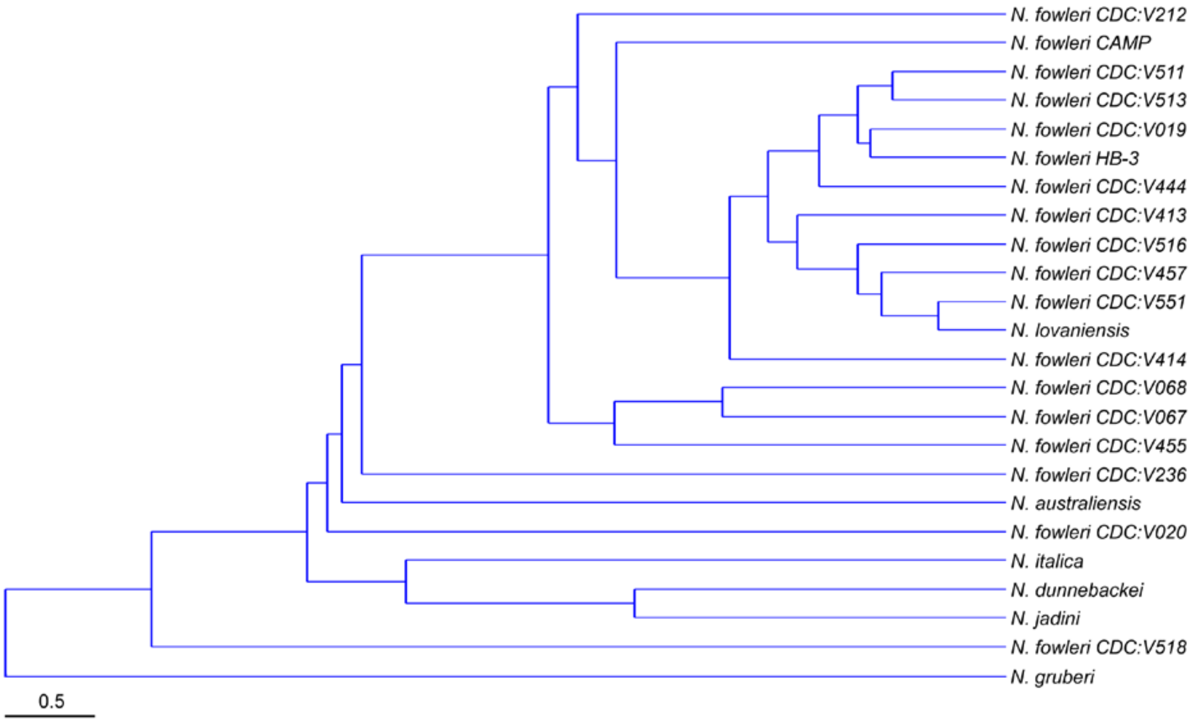


**Fig.S2.** Dendogram obtained with PAST (http://folk.uio.no/ohammer/past/index.html). For this analysis, all spectra for each strain were summed to give one representative spectrum per organism.