**Supplemental information**

**Table S1. Proteome data set from** *G. lamblia* **trophozoites, strains WBC6, WBA1, and GS/M-H7.** Trophozoites of the resistant strains were grown and subjected to MS shotgun analysis as described in Materials and Methods. [See Excel spreadsheet PAR\_2020\_0020\_assemblage\_A\_B\_Full\_dataset\_PRIDE]

**Table S2. Single peptide dataset set from** *G. lamblia* **trophozoites, strains WBC6, WBA1, and GS/M-H7.** Trophozoites of the resistant strains were grown and subjected to MS shotgun analysis as described in Materials and Methods. [See Excel spreadsheet PAR\_2020\_0020\_assemblage\_A\_B\_peptides]

**Table S3. Major surface antigens** **expressed in** *G. lamblia* **WBC6 trophozoites grown on two different media.** Trophozoites of the resistant strains were either grown as described in Materials and Methods (this work; referred to as “old medium”), or on a culture medium containing heat-inactivated fetal bovine serum (Biochrome, Berlin, Germany) instead of adult bovine serum (Biofluids, Rockville MD) and casein peptone from a different provider (Merck, Darmstadt, Germany instead of Becton Dickinson, Cockeysville, MD; referred to as “new medium”). The complete dataset corresponding to the trophozoites grown on “new medium” has been published elsewhere. Mean values ± standard deviations of LFQ values (x106) are given for three biological replicates. Nd, not detected. VSPs with mean LFQ levels > 108 are highlighted.

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| **Accession N°** | **Old annotation** | **“old medium”** | **“new medium”** |
| GL50803\_137740 | VSP3 | 5.9 ± 0.3 | 45.0 ± 13.8 |
| GL50803\_101074 | VSP88 | **427.3 ± 16.8** | **185.5 ± 7.0** |
| GL50803\_112801 | VSP169 | 2.6 ± 0.8 | nd |
| GL50803\_113450 | VSP44 (VSP417-4) | **273.3 ± 11.0** | 44.5 ± 10.8 |
| GL50803\_113797 | VSP5 (TSA417 partial) | **3847.3 ± 86.0** | 11.0 ± 2.1 |
| GL50803\_116477 | VSP149 | **491.4 ± 15.0** | nd |
| GL50803\_13194 | VSP38 | 18.5 ± 2.1 | 30.1 ± 3.1 |
| GL50803\_137608 | VSP175 | 1.0 ± 0.2 | nd |
| GL50803\_137612 | VSP160 | **152.4 ± 4.2** | **147.1 ± 8.5** |
| GL50803\_137613 | VSP188 | **835.6 ± 39.2** | **702.7 ± 46.1** |
| GL50803\_137617 | VSP77 | 79.3 ± 0.2 | **107.0 ± 4.4** |
| GL50803\_137618 | VSP8 | **1998.4 ± 119.5** | **1027.6 ± 70.9** |
| GL50803\_14586 | VSP186 | **437.9 ± 9.2** | 97.1 ± 2.0 |
| GL50803\_221693 | VSPA6 (CRP170) | 5.0 ± 3.6 | **646.9 ± 31.1** |
| GL50803\_33279 | VSP100 | 49.9 ± 2.1 | 29.7 ± 3.4 |
| GL50803\_41472 | VSP49 | 41.4 ± 1.9 | 7.7 ± 1.9 |
| GL50803\_5812 | VSP121 | **107.6 ± 20.6** | nd |

**Figure S1. CRP170 aligns to the hypothetical protein GL50803\_221693.** The CRP170 partial) sequence (P15799.1; red bar in Fig. S1 C) was BLASTed against non-redundant proteins. A, maximal and total scores of the three proteins sharing E value 0.0, query cover and identity 100% with CRP170. B, total scores of closest homologues (E= 0.0 for all proteins) found in the GiardiaDB to the three proteins listed in A. C, graphical alignment of the four sequences. VSP, variant-specific surface protein.



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| **Figure S2. Mapping of identified peptides to the coding sequence of GL50803\_221693.** The identified sequences are highlighted in bold red letters. The highly repetitive region TNPSDPTGTCVSAVDCQGSAGYYTDDSVSDAKECKKCNAPCTACAGTADKCTKCDANGAAPYLKK is highlighted in yellow. The GAAPYLKK motif recognized by mAb6E7 printed in blue. | **AA #** |
| MCPRFRWPLCSIEFRDMKQDPVRLLMQTRAVLCQLLASMLVGIGLIFAVALADACSPSIDGCAECDSTGKKCTK**CDANGNTPYLKKTNPSDPTGTCVSAVDCQGSAGYYTDDSVSDAKECKKCNAPCTACAGTADKCTKCDANGAAPYLKK**TNPSDPTGTCVSAVDCQGSAGYYTDDSVSDAKECKKCNAPCTACAGTADKCTKCDANGAAPYLKKTNPSDPTGTCVSAVDCQGSAGYYTDDSVSDAKECKKCNAPCTACAGTADKCTKCDANGAAPYLKKTNPSDPTGTCVSAVDCQGSAGYYTDDSVSDAKECKKCNAPCTACAGTADKCTKCDANGAAPYLKKTNPSDPTGTCVSAVDCQGSAGYYTDDSVSDAKECKKCNAPCTACAGTADKCTKCDANGAAPYLKKTNPSDPTGTCVSAVDCQGSAGYYTDDSVSDAKECKKCNAPCTACAGTADKCTKCDANGAAPYLKKTNPSDPTGTCVSAVDCQGSAGYYTDDSVSDAKECKKCNAPCTACAGTADKCTKCDANGAAPYLKKTNPSDPTGTCVSAVDCQGSAGYYTDDSVSDAKECKKCNAPCTACAGTADKCTKCDANGAAPYLKKTNPSDPTGTCVSAVDCQGSAGYYTDDSVSDAKECKKCNAPCTACAGTADKCTKCDANGAAPYLKKTNPSDPTGTCVSAVDCQGSAGYYTDDSVSDAKECKKCNAPCTACAGTADKCTKCDANGAAPYLKKTNPSDPTGTCVSAVDCQGSAGYYTDDSVSDAKECKKCNAPCTACAGTADKCTKCDANGAAPYLKKTNPSDPTGTCVSAVDCQGSAGYYTDDSVSDAKECKKCNAPCTACAGTADKCTKCDANGAAPYLKKTNPSDPTGTCVSAVDCQGSAGYYTDDSVSDAKECKKCNAPCTACAGTADKCTKCDANGAAPYLKKTNPSDPTGTCVSAVDCQGSAGYYTDDSVSDAKECKKCNAPCTACAGTADKCTKCDANGAAPYLKKTNPSDPTGTCVSAVDCQGSAGYYTDDSVSDAKECKKCNAPCTACAGTADKCTKCDANGAAPYLKKTNPSDPTGTCVSAVDCQGSAGYYTDDSVSDAKECKKCNAPCTACAGTADKCTKCDANGAAPYLKKTNPSDPTGTCVSAVDCQGSAGYYTDDSVSDAKECKKCNAPCTACAGTADKCTKCDANGAAPYLKKTNPSDPTGTCVSAVDCQGSAGYYTDDSVSDAKECKKCNAPCTACAGTADKCTKCDANGAAPYLKKTNPSDPTGTCVSAVDCQGSAGYYTDDSVSDAKECKKCNAPCTACAGTADKCTKCDANGAAPYLKKTNPSDPTGTCVSAVDCQGSAGYYTDDSVSDAKECKKCNAPCTACAGTADKCTKCDANGAAPYLKKTNPSDPTGTCVSAVDCQGSAGYYTDDSVSDAKECKKCNAPCTACAGTADKCTKCDANGAAPYLKKTNPSDPTGTCVSAVDCQGSAGYYTDDSVSDAKECKKCNAPCTACAGTADKCTKCDANGAAPYLKKTNPSDPTGTCVSAVDCQGSAGYYTDDSVSDAKECKKCNAPCTACAGTADKCTKCDANGAAPYLKKTNPSDPTGTCVSAVDCQGSAGYYTDDSVSDAKECKKCNAPCTACAGTADKCTKCDANGAAPYLKKTNPSDPTGTCVSAVDCQGSAGYYTDDSVSDAKECKKCNAPCTACAGTADKCTKCDANGAAPYLKKTNPSDPTGTCVSAVDCQGSAGYYTDDSVSDAKECKKCNAPCTACAGTADKCTKCDANGAAPYLKKTNPSDPTGTCVSAVDCQGSAGYYTDDSVSDAKECK**KCAEGQKPNTAGTQCFSCSDANCERCDQNDVCARCSTGAPPENGK**CPAATPGCHSSCDGCTENAMTNQADKCTGCKEGRYLKPESAAGQSGACLTAEECTSDKTHFTREKAGDSK**GMCLSCSDATHGITGCKK**CALK**TLSGEAESTVVCSECTDKR**LTPSGNACLDNCPAGTYADNINGVSVCASCHATCAECNGDANAASCTACYPGYSLLYGSGTAGTCVK**ECTGAFITNCADGQCTANVGGAKYCAQCKDGYAPIDGICTTVAAAGRDASVCTAADGK**CTK**CAGEYTLMSGGCYGVAK**LPGK**SVCTLASNGKCTMCAANNQAPVQEKCPECSEGCAKCNDSNACTECLPGYYKGAGDK**CFK**CTASSGNNNQITGVANCVSCAPPAGGNGGPVTCYIKTDGDNTGGSVNK**SGLSTGAIAGISVAVVVVVGGLVGFLCWWFICRGKA |  60 120 180 240 300 360 420 480 540 600 660 720 780 840 900 9601020108011401200126013201380144015001560162016801740180018601920198020402100216022202259 |