

Table S1. Summary of the results obtained using real-time PCR, assemblage-specific PCR, and *tpi* gene analysis of *Giardia*-positive samples collected from two separate geographical areas in Algeria, previously tested positive for intestinal parasites by microscopy of fecal concentrates and preserved in either potassium dichromate 2.5% or ethanol 70% prior to analysis.

Sample ID	Date of sampling	Location	Preserving medium	Sex	Age group	Microscopy for <i>Giardia duodenalis</i>	Real-time PCR results (Ct value)	Assemblage-specific PCR result	<i>tpi</i> result
ALG-001	25/03/2018	Doctor Saadane Hospital - Biskra	Potassium dichromate 2.5%	Female	Child	Positive	16.75	Assemblage B + other band(s)	Assemblage A
ALG-002	05/03/2018	Doctor Saadane Hospital - Biskra	Potassium dichromate 2.5%	Female	Adult	Positive	19.27	Negative	Assemblage A
ALG-003	03/09/2013	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Female	Undetermined	Positive	19.87	Assemblage B	Assemblage B
ALG-004	01/02/2016	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Male	Undetermined	Positive	20.16	Assemblage A	Assemblage A
ALG-005	14/11/2018	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Male	Child	Positive	20.93	Negative	Assemblage A
ALG-006	19/01/2017	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Undetermined	Undetermined	Positive	20.96	Negative	Assemblage A
ALG-007	10/09/2017	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Female	Undetermined	Positive	21.1	Negative	Assemblage B
ALG-008	27/08/2013	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Female	Undetermined	Positive	21.11	Assemblage B	Assemblage B
ALG-009	19/12/2016	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Female	Undetermined	Positive	21.11	Negative	Assemblage A
ALG-0010	11/03/2018	Doctor Saadane Hospital - Biskra	Potassium dichromate 2.5%	Female	Adult	Positive	21.55	Negative	Assemblage A
ALG-0011	02/04/2013	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Female	Child	Positive	22.16	Assemblage B	Not performed
ALG-0012	18/09/2013	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Female	Child	Positive	22.17	Assemblage B	Not performed
ALG-0013	28/01/2013	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Male	Undetermined	Positive	22.49	Negative	Assemblage A
ALG-0014	08/10/2013	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Male	Adult	Positive	22.54	Assemblage B	Not performed
ALG-0015	06/11/2017	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Male	Undetermined	Positive	22.56	Negative	Assemblage A
ALG-0016	02/04/2018	Doctor Saadane Hospital - Biskra	Potassium dichromate 2.5%	Male	Child	Positive	22.78	Assemblage B + other band(s)	Assemblage A
ALG-0017	31/05/2018	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Male	Child	Positive	22.79	Assemblage B + other band(s)	Assemblage B
ALG-0018	19/09/2017	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Male	Child	Positive	22.79	Negative	Assemblage A
ALG-0019	07/01/2016	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Female	Undetermined	Positive	22.97	Assemblage B	Assemblage B
ALG-0020	03/10/2013	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Male	Undetermined	Positive	23.07	Assemblage B	Assemblage B
ALG-0021	25/02/2018	Doctor Saadane Hospital - Biskra	Potassium dichromate 2.5%	Male	Child	Positive	23.25	Assemblage B + other band(s)	Assemblage A
ALG-0022	26/03/2018	Tolga Hospital - Biskra	Potassium dichromate 2.5%	Male	Child	Negative	23.42	Assemblage B	Assemblage B
ALG-0023	05/03/2018	Doctor Saadane Hospital - Biskra	Potassium dichromate 2.5%	Female	Child	Positive	23.56	Assemblage B	Assemblage B
ALG-0024	27/03/2018	Doctor Saadane Hospital - Biskra	Potassium dichromate 2.5%	Female	Child	Positive	23.6	Negative	Not performed
ALG-0025	24/03/2018	Tolga Hospital - Biskra	Potassium dichromate 2.5%	Female	Child	Negative	23.61	Assemblage B	Assemblage B
ALG-0026	23/04/2013	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Male	Child	Positive	23.71	Assemblage B	Assemblage B
ALG-0027	03/12/2017	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Male	Child	Positive	23.8	Negative	Assemblage A
ALG-0028	26/02/2018	Doctor Saadane Hospital - Biskra	Potassium dichromate 2.5%	Male	Adult	Positive	23.91	Assemblage A	Assemblage A
ALG-0029	13/02/2018	Doctor Saadane Hospital - Biskra	Potassium dichromate 2.5%	Female	Child	Positive	24.24	Negative	Assemblage A
ALG-0030	25/01/2016	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Male	Undetermined	Positive	24.26	Assemblage B	Assemblage B
ALG-0031	24/03/2018	Tolga Hospital - Biskra	Potassium dichromate 2.5%	Female	Child	Negative	24.32	Assemblage B	Assemblage B
ALG-0032	27/04/2018	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Male	Child	Positive	24.81	Assemblage B	Assemblage B
ALG-0033	27/03/2013	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Female	Child	Positive	25.02	Negative	Assemblage A
ALG-0034	25/03/2018	Tolga Hospital - Biskra	Potassium dichromate 2.5%	Male	Child	Negative	25.12	Assemblage B	Assemblage B
ALG-0035	31/03/2013	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Undetermined	Undetermined	Positive	25.5	Assemblage B + other band(s)	Assemblage B
ALG-0036	21/08/2013	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Male	Undetermined	Positive	25.5	Assemblage B	Assemblage B
ALG-0037	24/03/2018	Tolga Hospital - Biskra	Potassium dichromate 2.5%	Male	Child	Negative	25.59	Assemblage B	Assemblage B
ALG-0038	10/04/2018	Tolga Hospital - Biskra	Potassium dichromate 2.5%	Male	Child	Positive	25.84	Negative	Assemblage B
ALG-0039	01/10/2013	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Female	Child	Positive	26.24	Negative	Assemblage A
ALG-0040	22/05/2013	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Female	Child	Positive	26.42	Assemblage B	Assemblage B
ALG-0041	27/03/2018	Tolga Hospital - Biskra	Potassium dichromate 2.5%	Female	Adult	Negative	26.7	Negative	Assemblage B

ALG-0042	27/03/2018	Doctor Saadane Hospital - Biskra	Potassium dichromate 2.5%	Male	Adult	Positive	26.73	Assemblage A	Assemblage A
ALG-0043	21/08/2013	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Male	Child	Negative	26.75	Assemblage B	Assemblage B
ALG-0044	03/09/2013	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Female	Child	Positive	26.76	Assemblage B + other band(s)	Assemblage B
ALG-0045	02/09/2013	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Female	Adult	Positive	27.4	Assemblage B + other band(s)	Assemblage A
ALG-0046	27/03/2018	Doctor Saadane Hospital - Biskra	Potassium dichromate 2.5%	Male	Adult	Positive	27.92	Assemblage A	Assemblage A
ALG-0047	05/04/2016	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Male	Undetermined	Positive	28	Assemblage B	Assemblage B
ALG-0048	27/03/2018	Doctor Saadane Hospital - Biskra	Potassium dichromate 2.5%	Female	Child	Positive	28.36	Assemblage B	Assemblage B
ALG-0049	16/09/2013	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Female	Child	Positive	28.41	Negative	Assemblage A
ALG-0050	06/10/2013	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Male	Child	Positive	29.5	Negative	Assemblage B
ALG-0051	01/08/2016	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Female	Undetermined	Positive	29.76	Negative	Assemblage B
ALG-0052	14/04/2013	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Female	Child	Positive	30.03	Negative	Not performed
ALG-0053	28/12/2016	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Male	Undetermined	Negative	30.44	Negative	Assemblage A
ALG-0054	Undetermined	CHU Nafissa Hamoud, Hussein Dey - Algiers	Ethanol 70%	Male	Undetermined	Positive	30.49	Negative	Not performed
ALG-0055	Undetermined	CHU Nafissa Hamoud, Hussein Dey - Algiers	Ethanol 70%	Male	Child	Positive	30.86	Negative	Not performed
ALG-0056	14/02/2018	Doctor Saadane Hospital - Biskra	Potassium dichromate 2.5%	Male	Adult	Positive	30.98	Negative	Not performed
ALG-0057	10/04/2013	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Female	Child	Positive	31.1	Negative	Not performed
ALG-0058	Undetermined	CHU Nafissa Hamoud, Hussein Dey - Algiers	Ethanol 70%	Male	Undetermined	Positive	31.76	Negative	Not performed
ALG-0059	Undetermined	CHU Nafissa Hamoud, Hussein Dey - Algiers	Ethanol 70%	Female	Undetermined	Positive	32.18	Negative	Not performed
ALG-0060	Undetermined	CHU Nafissa Hamoud, Hussein Dey - Algiers	Ethanol 70%	Male	Undetermined	Positive	32.21	Negative	Not performed
ALG-0061	22/09/2013	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Female	Child	Positive	32.23	Other bands	Not performed
ALG-0062	08/04/2018	Doctor Saadane Hospital - Biskra	Potassium dichromate 2.5%	Male	Child	Positive	32.27	Negative	Not performed
ALG-0063	Undetermined	CHU Nafissa Hamoud, Hussein Dey - Algiers	Ethanol 70%	Female	Undetermined	Negative	33.54	Negative	Not performed
ALG-0064	03/10/2018	CHU Nafissa Hamoud, Hussein Dey - Algiers	Ethanol 70%	Female	Adult	Negative	33.84	Negative	Not performed
ALG-0065	Undetermined	CHU Nafissa Hamoud, Hussein Dey - Algiers	Ethanol 70%	Male	Undetermined	Positive	34.15	Negative	Not performed
ALG-0066	Undetermined	CHU Nafissa Hamoud, Hussein Dey - Algiers	Ethanol 70%	Female	Undetermined	Negative	34.22	Negative	Not performed
ALG-0067	15/05/2018	CHU Nafissa Hamoud, Hussein Dey - Algiers	Ethanol 70%	Male	Child	Positive	35.88	Negative	Not performed
ALG-0068	26/02/2018	Tolga Hospital - Biskra	Potassium dichromate 2.5%	Female	Adult	Positive	35.93	Negative	Not performed
ALG-0069	23/07/2018	CHU Nafissa Hamoud, Hussein Dey - Algiers	Ethanol 70%	Male	Child	Positive	36.68	Negative	Not performed
ALG-0070	Undetermined	CHU Nafissa Hamoud, Hussein Dey - Algiers	Ethanol 70%	Undetermined	Undetermined	Positive	36.71	Negative	Not performed
ALG-0071	08/10/2018	CHU Nafissa Hamoud, Hussein Dey - Algiers	Ethanol 70%	Male	Child	Positive	36.89	Negative	Not performed
ALG-0072	10/10/2018	CHU Nafissa Hamoud, Hussein Dey - Algiers	Ethanol 70%	Male	Child	Negative	37.15	Negative	Not performed
ALG-0073	27/05/2013	CHU Issad Hassani, Beni Messous - Algiers	Potassium dichromate 2.5%	Female	Child	Positive	37.43	Negative	Not performed
ALG-0074	21/10/2018	CHU Nafissa Hamoud, Hussein Dey - Algiers	Ethanol 70%	Male	Child	Positive	37.47	Negative	Not performed
ALG-0075	04/04/2018	Doctor Saadane Hospital - Biskra	Potassium dichromate 2.5%	Female	Child	Negative	38	Negative	Not performed
ALG-0076	03/07/2018	CHU Nafissa Hamoud, Hussein Dey - Algiers	Ethanol 70%	Male	Child	Positive	38.39	Negative	Not performed
ALG-0077	24/07/2018	CHU Nafissa Hamoud, Hussein Dey - Algiers	Ethanol 70%	Female	Child	Positive	39.1	Negative	Not performed
ALG-0078	02/05/2018	CHU Nafissa Hamoud, Hussein Dey - Algiers	Ethanol 70%	Male	Child	Positive	39.93	Negative	Not performed
ALG-0079	10/10/2018	CHU Nafissa Hamoud, Hussein Dey - Algiers	Ethanol 70%	Male	Child	Negative	39.97	Negative	Not performed
ALG-0080	26/06/2018	CHU Nafissa Hamoud, Hussein Dey - Algiers	Ethanol 70%	Female	Child	Positive	41.51	Negative	Not performed

Table S2. Comparative analysis of the extent of allelic sequence heterozygosity in the *tpi* gene of *Giardia duodenalis* Assemblage B identified in the study.

Assemblage/Subtype	Material	Nucleotide position from the beginning of the gene																				
		77	91*	105	111	121	132	141	162	165*	168*	205	216	261	271	280	297	312	333	407	429	448
BIII/ 2924 (AY228628)		C	C	G	G	G	C	C	G	C	C	C	C	G	C	A	A	C	T	A	G	G
BIV/Ad-19 (AF069560)		C	T	G	G	G	C	C	G	T	T	C	C	G	C	A	A	C	T	A	A	G
ALG-003	Stool sample	C	C	G	G	G	C	Y	R	C	Y	C	C	G	Y	R	R	C	Y	A	G	G
ALG-007	Stool sample	C	T	G	G	G	C	C	G	T	T	C	C	G	C	A	A	C	T	A	A	G
ALG-008	Stool sample	C	C	G	G	G	C	Y	R	Y	Y	C	C	G	Y	R	R	C	Y	A	G	G
ALG-0017	Stool sample	C	T	G	G	G	C	C	G	T	T	C	C	G	C	A	A	C	T	A	A	G
ALG-0019	Stool sample	C	T	G	G	G	C	C	G	T	T	C	C	G	C	A	A	C	T	A	A	G
ALG-0020	Stool sample	C	Y	G	R	R	C	C	G	C	C	C	C	G	C	A	A	C	T	A	G	G
ALG-0022	Stool sample	Y	C	G	G	G	C	C	R	Y	Y	C	C	G	C	A	A	C	T	A	G	G
ALG-0023	Stool sample	C	Y	R	G	G	C	C	R	C	C	C	C	G	C	A	A	Y	T	A	G	R
ALG-0025	Stool sample	Y	C	G	G	G	C	C	R	Y	Y	C	C	G	C	A	A	C	T	A	G	G
ALG-0026	Stool sample	C	T	G	G	G	C	C	G	Y	Y	C	C	G	C	A	A	C	T	A	G	G
ALG-0030	Stool sample	C	C	G	G	G	C	C	R	Y	Y	C	C	G	C	A	A	C	T	A	G	G
ALG-0031	Stool sample	Y	C	G	G	G	C	C	R	Y	Y	C	C	G	C	A	A	C	T	A	G	G
ALG-0032	Stool sample	C	Y	G	G	G	C	C	R	C	C	Y	C	G	C	A	R	C	Y	A	G	G
ALG-0034	Stool sample	T	C	G	G	G	C	C	A	C	C	C	C	G	C	A	A	C	T	A	G	G
ALG-0035	Stool sample	C	C	G	G	G	C	C	R	C	C	C	C	G	C	A	A	C	T	A	G	G
ALG-0036	Stool sample	C	T	G	G	G	C	C	A	C	C	C	C	G	C	A	A	C	T	A	G	G
ALG-0037	Stool sample	Y	C	G	G	G	C	C	R	C	C	C	C	G	C	A	A	C	T	A	G	G
ALG-0038	Stool sample	C	C	G	G	G	C	C	G	C	C	C	Y	G	C	A	A	C	T	A	G	G
ALG-0040	Stool sample	C	T	G	G	G	C	C	G	T	T	C	C	G	C	A	A	C	T	A	A	G
ALG-0041	Stool sample	C	C	G	G	G	Y	C	G	C	C	C	C	G	C	A	A	C	T	A	G	G
ALG-0043	Stool sample	C	C	G	G	G	C	C	R	C	C	C	C	G	Y	R	R	C	Y	A	G	G
ALG-0044	Stool sample	C	C	G	G	G	C	C	R	Y	C	C	C	G	C	A	A	C	T	W	G	G
ALG-0047	Stool sample	C	C	G	G	G	C	C	R	C	C	C	C	R	C	A	A	Y	T	A	G	G
ALG-0048	Stool sample	C	C	G	G	G	C	C	G	Y	Y	C	C	G	C	A	A	C	T	A	G	G
ALG-0050	Stool sample	C	C	G	G	G	C	C	G	C	C	C	C	G	C	A	A	C	T	A	G	G
ALG-0051	Stool sample	C	T	G	G	G	C	C	G	Y	Y	C	C	G	C	A	A	C	T	A	R	G

Keys: Asterisks (*) represent a substitution patterns suggested as markers for distinct *Giardia* assemblages B subtypes [10,39].

Table S3. Comparative sequence analysis of expressed amino acid substitutions in the *tpi* gene of *Giardia duodenalis* Assemblage B samples identified in the study. Only samples with double peaks resulting in the existence of non-synonymous substitutions were included.

Amino acid position from the beginning of the <i>tpi</i> gene								
Amino acid sequences	26	31	41	87	91	94	136	150
<i>tpi</i> reference sequence (GenBank Accession no. KP687784)	A	Y	V	M	H	I	K	A
ALG-003	A	H	V	M	(H ↔ Y)	(I ↔ V)	K	A
ALG-008	A	H	V	M	(H ↔ Y)	(I ↔ V)	K	A
ALG-0020	A	(H ↔ Y)	(I ↔ V)	M	H	I	K	A
ALG-0022	(A ↔ V)	H	V	M	H	I	K	A
ALG-0023	A	(H ↔ Y)	V	M	H	I	K	(T ↔ A)
ALG-0025	(A ↔ V)	H	V	M	H	I	K	A
ALG-0031	(A ↔ V)	H	V	M	H	I	K	A
ALG-0032	A	(H ↔ Y)	V	M	H	I	K	A
ALG-0037	(A ↔ V)	H	V	M	H	I	K	A
ALG-0043	A	H	V	M	(H ↔ Y)	(I ↔ V)	K	A
ALG-0044	A	H	V	M	H	I	(K ↔ M)	A
ALG-0047	A	H	V	(I ↔ M)	H	I	K	A

Alignment analysis of the translated nucleotides sequences into amino acid with the complete *tpi* gene reference sequence (GenBank accession no: KP687784.1) indicated that the samples ALG-003, ALG-008, and ALG-0043 had two expressed substitutions of amino acid at positions 91 (H ↔ Y) and 94 (I ↔ V) resulting in replacement of amino acid Histidine to Tyrosine, and Isoleucine to Valine, respectively. Sample ALG-0020 had two expressed substitutions at positions 31 of amino acid (H ↔ Y) and 41 (I ↔ V) inducing replacement of amino acid Histidine to Tyrosine and Isoleucine to Valine, respectively. Samples ALG-0022, ALG-0025, ALG-0031, and ALG-0037 indicated one expressed substitution at position 26 (A ↔ V) triggering a change of amino acid Alanine to Valine. Sample ALG-0023 exhibited two expressed substitutions at positions 31 (H ↔ Y) and 150 (T ↔ A) causing a change of amino acid Histidine to Tyrosine and Threonine to Alanine. Samples ALG-0032, ALG-0044, and ALG-0047 had one expressed substitution at positions 31 (H ↔ Y), 136 (K ↔ M), and 87 (I ↔ M) leading change of amino acid Histidine to Tyrosine, Lysine to Methionine, and Isoleucine to Methionine, respectively. All the replacements of amino acids are reversible.