Using two gene finders Aragorn (ARA) and tRNAScan-SE (TSE), 4381 genes were predicted. Genes with tse score of less than 50 and aragorn score of less than 107 were dismissed. The cutoff score is set based on the ARA and TSE score distribution shown in figure 1a,b.

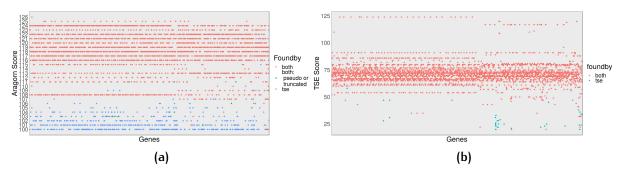


Figure 1: a) ARA score of genes found by both genefinders TSE and ARA, and genes found by only ARA. b) TSE score of genes found by both genefinders and genes found by only TSE

3588 genes were left from which two genes marked as truncated by TSE were also removed. 22 genes in the remained gene set had different identity between TSE and ARA shown in table 1 and were partially removed.

Table 1: 22 genes with different anticodon assignment by two gene finders TSE and ARA

tse identity ara identity	(g w)	(m l)	(y n)
frequency	3	9	10

Each of these three ambiguties have been analysed and compared to the other genes of the same identity. Genes $(y \mid n)$ were manually compare to the other identified genes with identity n and y found by both gene finders. In all cases these 10 genes were considerably more similar to the genes marked with identity Y than genes identified as N. Also, by looking at genes as clusters (a group of two or more genes found within a genome located within a thousand base pairs of each other) they were observed in clusters "LSMEM $(y \mid n)V$ ", " $V(y \mid n)MEMSL$ ", "LSMEMSM $(y \mid n)V$ ", and $V(y \mid n)V$. Further we observed that there was no occurrence of genes NY or YN within any cluster, however there were 6 occurrences of YV or VY (one of them in cluster EMYV). Genes with ambiguity $V(y \mid n)V$ were disregarded as they all appear as singleton genes, and not in any of the clusters. Genes with ambiguity $V(y \mid n)V$ have also been removed since we were not able to resolve the ambiguity.

The final gene set has 3574 genes from which 36 genes are found only by aragorn and the rest are predicted by both gene finders.

Table 2: nucleotide composition of 46 TriTryp genomes

organism	A%	T%	<u>C</u> %	G%	AT%	GC%	Seq#	Gene#
TbruceigambienseDAL972	26	26	24	24	53	47	11	63
TevansiSTIB805	27	27	23	23	53	47	13	66
CfasciculataCfCl	21	22	29	28	43	4 7 57	31	105
TbruceiLister427	26	26	22	23	4 3 51		32	66
LpanamensisMHOMPA94PSC1	21	21	28	28		45 56		
LdonovaniBHU1220					41		35 26	74
LdonovaniBPK282A1	19	20	29	29	39	57	36 26	84
	19	20	29	29	39	57 60	36 26	85
LinfantumJPCM5 LmajorFriedlin	20	20	30	30	40		36 26	84
LmajorSD75.1	20	20	30	30	40	60 5 0	36 26	84 82
TcruziCLBrenerEsmeraldo-like	20	20	30	30	40	59	36	
	20	20	20	20	39	40	41	57
TcruziCLBrenerNon-Esmeraldo-like	21	21	22	22	42	43	41	57
TcruziSylvioX10-1	24	23	25	25	47	50	47	66
LpyrrhocorisH10	21	22	28	28	43	56	60	104
TbruceiTREU927	27	27	23	23	54	45	131	72
LbraziliensisMHOMBR75M2904	21	21	29	29	42	58	139	83
LgerbilliLEM452	20	20	30	29	40	59	142	81
LaethiopicaL147	19	20	30	29	39	59	160	83
LtropicaL590	19	19	29	28	38	57	160	87
LarabicaLEM1108	20	20	29	29	40	58	168	85
LturanicaLEM423	19	20	30	29	39	59	219	86
LspMARLEM2494	20	20	30	29	40	59	251	8o
TtheileriEdinburgh	26	26	17	17	52	35	253	155
LenriettiiLEM3045	20	20	29	29	40	59	495	82
BayalaiBo8-376	22	22	27	27	45	55	546	69
LmexicanaMHOMGT2001U1103	20	20	30	30	40	60	588	84
LbraziliensisMHOMBR75M2903	19	20	27	27	39	53	745	86
LmajorLV39c5	20	20	29	29	40	59	809	84
LpanamensisMHOMCOL81L13	21	21	29	28	42	57	856	88
TcruzicruziDm28c	24	24	26	26	48	52	1029	95
TcruziDm28c	25	25	26	25	49	50	1210	50
LseymouriATCC30220	22	22	28	28	44	55	1222	94
LtarentolaeParrotTarII	21	21	27	27	42	55	1351	78
EmonterogeiiLV88	23	23	26	26	46	52	1961	103
PconfusumCUL13	18	18	28	28	35	57	2188	61
LamazonensisMHOMBR71973M2269	20	20	30	30	41	59	2627	65
TcongolenseIL3000	21	21	20	20	43	40	2839	67
TgrayiANR4	23	23	27	27	46	54	2871	94
TrangeliSC58	24	23	27	26	47	53	7433	6
TvivaxY486	21	2 1	-/ 23	23	42	46	8 2 90	79
TcruziJRcl4	24	23	26	24	4 7 47	4 0 50	15312	79 69
TcruziEsmeraldo	23	22	24	23	47 45	47	15803	74
TcruzimarinkelleiB7	22	22	23	23	43	47 45	16783	56
TcruziSylvioX10-1-2012	24	24	26	26	43 49	43 51	27019	68
TcruziCLBrener	23	24 23	27	27			29407	14
TcruziTulacl2	23	23 21	-	-	47 42	53 46		-
TCT UZI TUIUCIZ		41	23	23	43	46	45711	119

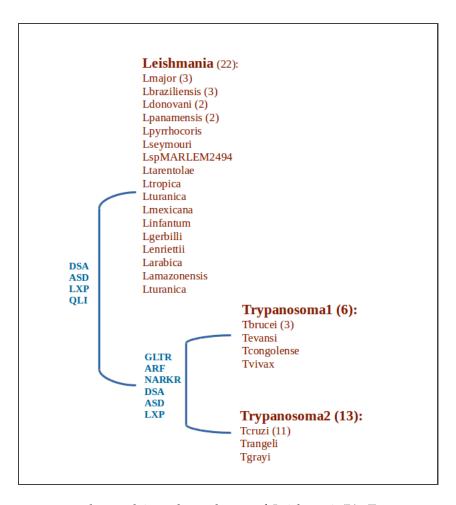


Figure 2: TriTryp genomes clustered into three classes of Leishmania(L), Trypanosoma1(T1), and Trypanosoma2(T2)

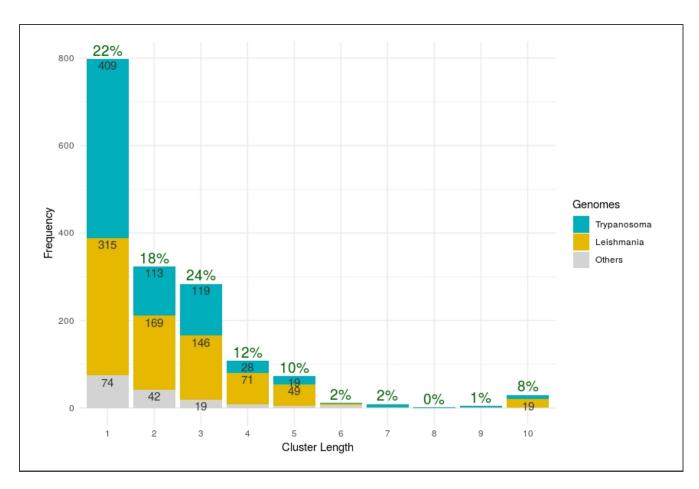


Figure 3: Cluster size distribution for three categories of TryTryp genomes. Labels in green on top of each bar show the percentage of total number of genes as cluster of a specific length. Each color refers to one category of TriTryp genomes. Numbers within each color section of the bar shows the counts of clusters with a specific length.

Table 3: frequecty of gene clusters of length > 3 with frequency of atleast 2. T% and L% refer to the percentage of Trypanosoma and Leishmania genomes, in order, that contain the cluster

Tryp	anosoma	Leishmania					
Cluster	dir	T%	L%				
EVRH	-++	91	О	Cluster	dir	L%	T%
IVQRLTRKGW	+++-+	82	О	NARKR	+-+++	55	0
VIMLS	-++-+	82	O	IOVKGLTRKR	-++++-++-	<i>35</i>	0
CLXP	+-+	73	О	GLTR	++-+	<i>33</i>	0
NYTPN	++	64	O	EMSV	-+-+	20	0
GTGP	+-+	55	О	IQVK	-+++	20	0
NPTYN	-+++	36	О	ILQQI	+-++		0
YTTY	-++	36	О	LSMEMYV		15	-
PGTTG	-++-	18	O	RKRTLGKVQI	-+-++-+	15	0
SLMIV	-+-+	14	O	LSMEMYV	+-+++	15	0
EARR	-++	9	О	LSIVIEIVI I V	-+-+-+	10	О
YTTT	+	9	0				

Table 4: frequecty of gene clusters of length > 2. T% and L% refer to the percentage of Trypanosoma and Leishmania genomes, in order, that contain the cluster

	na (20 geno:		Leishmania (22 genomes)					
71-4110301	(801.0.			Cluster	dir	L%	T%	
				DSA	-++	55	95	
				NARKR	+-+++	55	0	
				ARF	-+	50	0	
				IQL	-++	35	o	
				IQVKGLTRKR	-+++-+-	35	o	
C1 .	1.	TTO/	T 0/	PTN	-++	35	О	
Cluster	dir	T%	L%	ASD	-+	30	5	
DSA	-++	95	55	GLTR	++-+	30	0	
EVRH	-++	91	0	LQI	-+	30	О	
PVK	+++	91	0	LXP	-+	30	23	
QLI	+-+	86	0	PXL	-++	30	0	
RRA	-+	86	0	VEF	-++	30	0	
IVQRLTRKGW	+++-+	82	0	FRA	-++	25	0	
VIMLS	-++-+	82	0	HEF	+++	25	0	
CLXP	+-+	73	0	LXP	+-+	25	0	
VHF	++-	68	0	VLM	_	25	0	
NYTPN	-++	64	0	EMSV	-+-+	20	0	
KGN	+-	59	0	IQVK	-+++	20	О	
GTGP LAG	+-+	55	0	SLS	+-	20	О	
NPTYN	-++	45 26	0	FEV	-+	15	O	
YTTY	-+++	36 26	0	ILQQI	+-++	15	О	
GAL	-++ -+	36 27	0	LSMEMYV	-+-++-+	15	О	
LXP	-+	27	0	RKRTLGKVQI	+-+++	15	О	
PGTTG	-+ -++-	23	30	LSMEMYV	-+-+-+	10	0	
SLMIV	-+-+	18	0	NTP	-+	10	О	
EARR	-+-+ -++	14	0	ARFARF	-+-+	5	O	
KVP		9		ARKR	-+++	5	О	
MRR	-++	9	0	ASDD	-++	5	O	
TGP	-++	9	0	DSV	-++	5	О	
YTTT	 +	9	0	EML	-++	5	O	
ARR	-++	9 5	0	EMYV	++-+	5	O	
ASD	-+	<i>5</i>	30	FIQ	_	5	0	
DEDE	+-+-	<i>5</i>	0	FIQFIQ		5	O	
DRTY	++++	<i>5</i>	0	ILQ	+-	5	O	
EFHV	-+-	<i>5</i>	0	IQI	+++	5	0	
FHVRH	+-++	<i>5</i>	0	IQQL	-++	5	O	
GEE	+-	5	0	IQQLI	-++-	5	0	
GKWRTL	-+-++	5	0	IQV	-++	5	0	
GTGP	++-+	5	0	IQVKGLTR	-++++-+	5	0	
GTGPVK	+-+++	5	0	KGL	+++	5	0	
GTP	+-+	5	0	KVQI	+	5	О	
HRE	-+	5	0	LSM	-+-	5	0	
ILIQ	-++-	<i>5</i>	0	LSMEMSMYV	-+-++-+-+	5	О	
IVQRLTKGW	++	<i>5</i>	0	LTP	-+	5	О	
IVQRLTRWKG	+++-+	5	0	MEM	-+	5	О	
LAA	-++	5	0	MEMSL	-+-+	5	О	
NGK	++-	5	0	MEMSL	-++-+	5	О	
PTG	_	5	0	MMAP		5	О	
PXLC	-++-	5	0	NARK	+-++	5	О	
QILI	+-+	5	0	NWW	+++	5	О	
QLQ	-+-	5	0	PKP	+-	5	О	
QVI	++-	5	0	PTYN	-+-+	5	О	
RQVI	+++-	5	0	QFIQ		5	О	
VHFE	++-+	5	0	QLI	-+-	5	О	
VYIML	-+++-	5	0	QVK	+++	5	О	
VYIMLS	-+++-+	5	0	RHLP	++++	5	0	
WGKRTL	-+-++	5	0	RKKK	+-+	5	0	
YTRD		5	0	RKR		5	0	
YTY	-+	5	0	RKRAN	—+-	5	0	
)		RKRRA	+	5	О	
				RKRTLGKVQ	+-++	5	О	
				SLS	++-	5	О	
				VHEF	-+++	5	О	
				VYMEMSL	-+-+-+	5	О	
				VYMEMSL	-+-++-+	5	О	
				WWNN		5	О	

Table 5: frequecty of gene clusters of length > 2. T1% and T2% refer to the percentage of Trypanosoma1 and Trypanosoma2 genomes, in order, that contain the cluster

Trypano				Trypanos	oma2 (13 gen	omes)	
				Cluster	dir	T1%	T2%
				IQL	-++	54	O
				IQVKGLTRKR	-+++-+-	54	О
				PTN	-++	54	O
				DSA	-++	46	83
				LQI	-+	46	0
				NARKR	+-+++	46	83
Cluster	dir	T1%	$T_2\%$	VEF	-++	46	0
ARF	-+	83	38	ARF	-+	38	83
DSA	-++	83	46	LXP	-+	38	17
GLTR	++-+	83	8	PXL	-++	38	0
HEF	+++	83	O	FRA	-++	31	О
LXP	+-+	83	О	FEV	-+	23	O
NARKR	+-+++	83	46	LSMEMYV	-+-++-+	23	О
VLM		83	0	RKRTLGKVQI	+-+++	23	O
ASD	-+	67	15	ASD	-+	15	67
EMSV	-+-+	67	0	LSMEMYV	-+-+-+	15	0
IQVK	-+++	67	O	NTP	-+	15	О
SLS	+-	67	O	ARFARF	-+-+	8	О
ILQQI	+-++	50	O	ASDD	-++	8	О
EML	-++	17	O	DSV	-++	8	О
FIQ	_	17	O	EMYV	++-+	8	О
FIQFIQ		17	O	GLTR	++-+	8	83
ILQ	+-	17	O	IQQLI	-++-	8	O
IQI	+++	17	O	IQVKGLTR	-++++-+	8	O
IQV	-++	17	O	LSM	-+-	8	O
KGL	+++	17	O	LSMEMSMYV	-+-++-+-+	8	O
LXP	-+	17	38	MEM	-+	8	O
QFIQ		17	O	MEMSL	-+-+	8	O
QLI	-+-	17	O	MMAP		8	O
QVK	+++	17	О	NARK	+-++	8	О
RHLP	++++	17	О	PTYN	-+-+	8	O
SLS	++-	17	О	RKKK	++	8	О
				RKR		8	О
				RKRAN	+-	8	O
				RKRRA	+	8	О
				RKRTLGKVQ	+-++	8	О
				VYMEMSL	-++-+	8	О
				VYMEMSL	-+-++-+	8	О
				WWNN		8	О

Table 6: Sets of clusters with potential variations (duplication, deletion, inversion). There are three genome classes T, L and O which refer to Trypanosoma, Leishmania and Other genomes in order.

clusters	set	Genomeclass	Tdirs	Ldirs	Odirs			8			
GEE	0	L	Tulis	-++	Cuits						
LAA	0	L		+-+-							
MEM	0	L		++++							
		L L									
PKP	0			-++		clusters	set	Genomeclass	Tdirs	Ldirs	Odirs
RKKK	О	L		-+-		EVRH	13	LT	+-+ -+	-+	
YTY	О	L		+-++		HRE	13	O			-+-+
III	О	LT	-+	-+		RVHE	13	T	-+		
LSSS	О	O			+++	FHVRH	14	T	-+-+ -++-+		
QLQ	0	O			-+-+	QFIQ		L		1.1	
WWNN	0	O			-++-+		15			-++	
KKN	0	OL		+-+	+-+	FIQ	15	O			+
YTTT	0	OL		-++	-++	FIQFIQ	15	T			
MRR	0	OLT	-++	-++	-++	FSI	16	T	+-++		
ARR	0	T	-+			KLAG	17	L		++-	
DEDE		T				LAG	17	L		-+++	
	0		-+-+			GAL	17	T	+-+++		
GAA	О	T	-+++			ILQ	18	L		-++-	
IQI	О	T	-++			IQL	18	L		_	
NWW	О	T	-++			IQVKGLTR	18	L		-++-	
RKR	О	T	-++			IVQRLTKGW	18	L		-+-	
RRA	0	T	-+-+			IVQRLTRWKG	18	Ĺ		++-	
SLS	О	T	++-+								
YTTY	0	T	-+			RKRTLGKVQI	18	L		+++-	
ARF	1	T	_			IVQRLTRKGW		0			+++
ARFARF	1	T				LQI	18	О			++-
FRA	1	T	-++			VGWRTQI	18	O			-++
ENKRRA	2	L	-11	-+		WGKRTL	18	O			-++-
		L				ILIQ	18	OL		++	++
NARK	2			+-		IQV	18	OL		+++	+++
NARKR	2	L		-+-++		ITRLQVGWR	18	OL		+-+	+-+
RKRAN	2	L		+-+ ++-+		RQVI	18	OL		-+	-+
RKRRA	2	L		+-+++		IVQ	18	OLT	-+-	+-+	+-+
RRAE	2	L		+-+		GKWRTL	18	T		1-1	1-1
ARKR	2	O			+-	GLTR			-+		
EARR	2	O			_		18	T	+++		
RKAREN	2	T	++-+			ILQQI	18	T	+-		
ASDD	3	L		-+		IQQL	18	T	-++		
DSA	3	Ö		·	-+	IQQLI	18	T	-+-+		
ASD		T				IQVK	18	T	-++		
	3		+++			IQVKGLTRKR	18	T			
CLXP	4	L		-++-		KGL	18	T	+++		
LXC	4	T	+-			KVQI	18	T	++++		
LXP	4	T	+-++			LTRKGW	18	T	+-+		
LXPXP	4	T	+++			QILI	18	T			
PXL	4	T	-++			QLI	18	T	_		
PXLC	4	T	-++						+-		
DNE	5	T	-++-			QVI	18	T	+		
DRTY	6	T	-++			QVK	18	T	+-++		
YTRD	6	T	-+++			RKRTLGKVQ	18	T	+-+++		
DSV	7	T	-+++-+			GTGP	19	L		-+-+	
HEF	-	L	-1111-1			TGP	19	L		++-+	
	9			++-+		PGTTG	19	O			+-+-+-
VEF	9	L		+++-+		GTP	19	OL		-+	-+
EFHV	9	0			-+++-+-	PVK	19	OL		++-	+++
FEV	9	О			+-	GTGPVK	19	T	+- ++-		
VHEF	9	OL		+++-+	++-++	KVP	19	T	-++		
VHFE	9	OL		+-	+-	PTG	-	T	-+++		
EFFVH	9	T	-+++-+-				19		-+++		
VHF	9	T	+++			KGN	22	L		-++-+	
EMYV	10	L		_		NGK	22	T	_		
LSMEMSMYV	10	L		-++		LTP	24	L		-+++-	
LSMEMYV	10	Ĺ		-++		MMAP	25	OL		-+++-+	-+++-+
EML		O		-11		NTP	26	L		-+-++	
	10				++-	YTN	26	L		+	
EMSV	10	0			-++	PTN	26	O			_
VIMLS	10	0			-++	PTYN	26	OL			
VLM	10	О			_	NPTYN	26	T	-+-++-+ -+-+-+		
VYIMLS	10	O			-++-+	NYTPN	26	T	1-11-71-7-7		
VYMEMSL	10	O			_						
LSM	10	T	-+			QPL	27	L		-++	
LSMIVY	10	T	-+			RHLP	28	L		-+	
MEMSL	10	T	-+-								
MEYSL	10	T	-+-++-+-								
SLMIV		T	-+-++-+								
	10	T									
VYIML	10	1	-+								