Ασφάλεια και κίνδυνοι από τα παράγωγα αίματος: Κίνδυνοι για μετάδοση παρασιτικών λοιμώξεων

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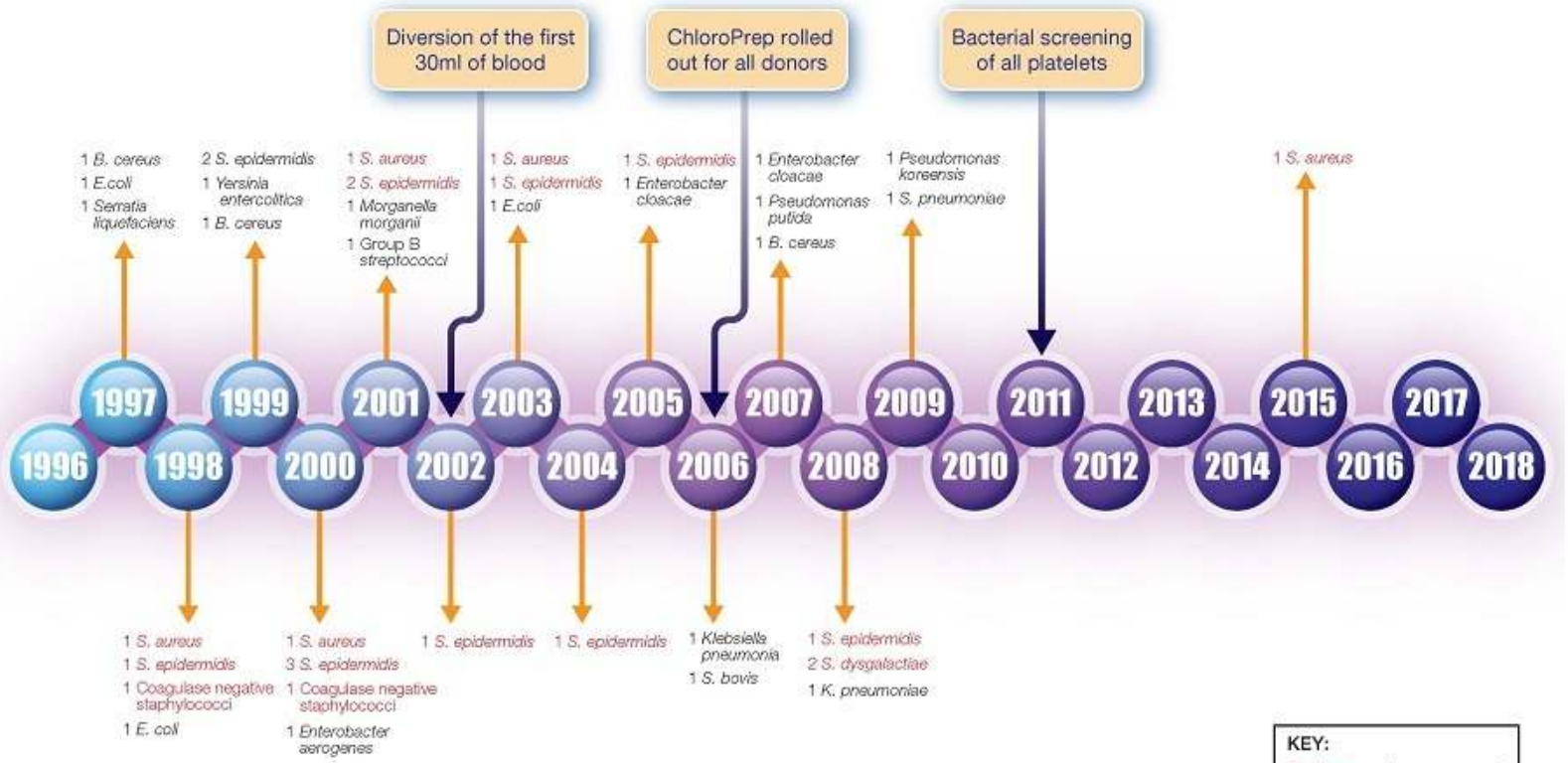
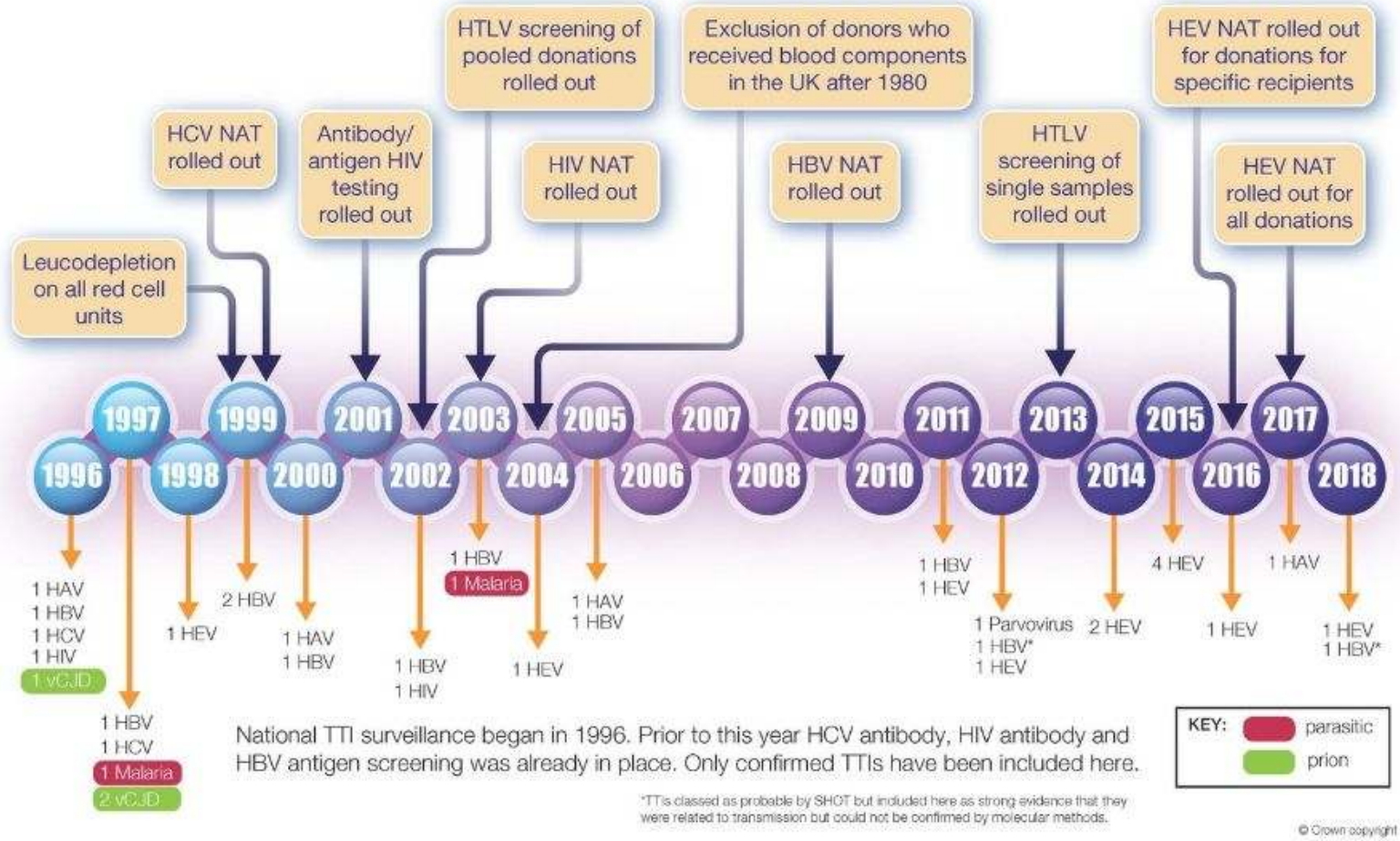
Bacterial Risk Reduction Strategies vs Transfusion Transmitted Infections



Viral Risk Reduction Strategies vs Viral Transfusion Transmitted Infections



National TTI surveillance began in 1996. Prior to this year HCV antibody, HIV antibody and HBV antigen screening was already in place. Only confirmed TTIs have been included here.

*TTIs classed as probable by SHOT but included here as strong evidence that they were related to transmission but could not be confirmed by molecular methods.

EUROPEAN COMMISSION
DIRECTORATE GENERAL FOR HEALTH AND FOOD SAFETY

Directorate B - Health systems, medical products and innovation
B4 – Medical products: quality, safety, innovation

SUMMARY OF THE 2018 ANNUAL REPORTING OF SERIOUS ADVERSE REACTIONS AND EVENTS FOR BLOOD AND BLOOD COMPONENTS

(Data collected from 01/01/2017 to 31/12/2017 and submitted to the European Commission in 2018)

EXECUTIVE SUMMARY

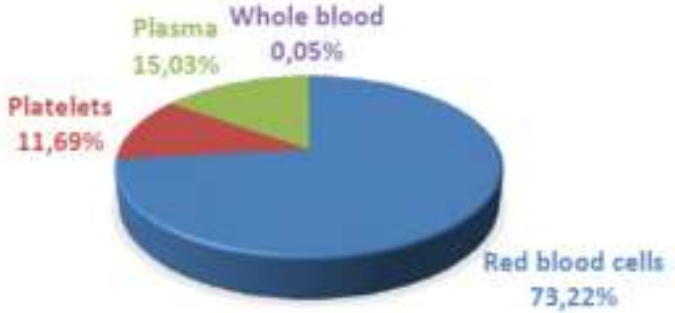


Figure 1: Units issued[6] (per blood component); data 2017.

Component type	Units issued
Red Blood Cells	18,374,971
Platelets[7]	2,934,371
Plasma	3,772,062
Whole blood	12,502
Total	**25,093,906**

Table 1. Number of units issued per blood component; data 2017.

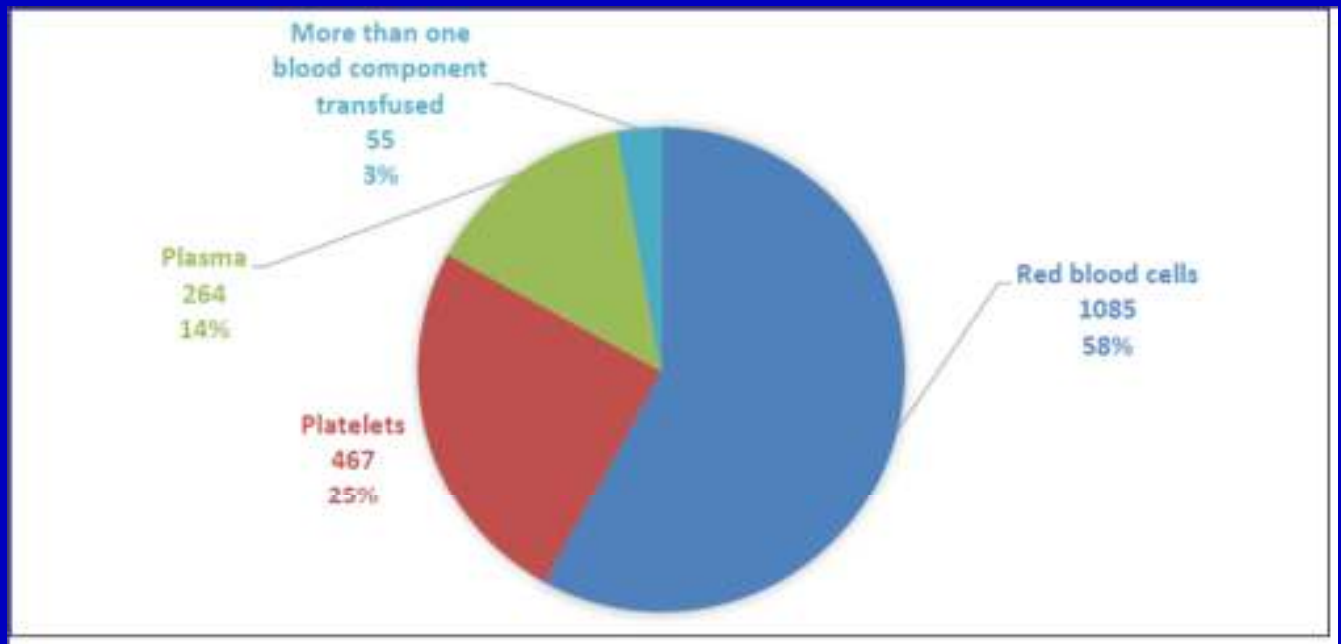


Figure 4. Percentage of SAR (imputability 2-3) per blood component; data 2017.

Component type	Units transfused per SAR
Red Blood Cells	14,285
Platelets	4,885
Plasma	10,080

Table 4. Units per component type transfused per SAR; data 2017.

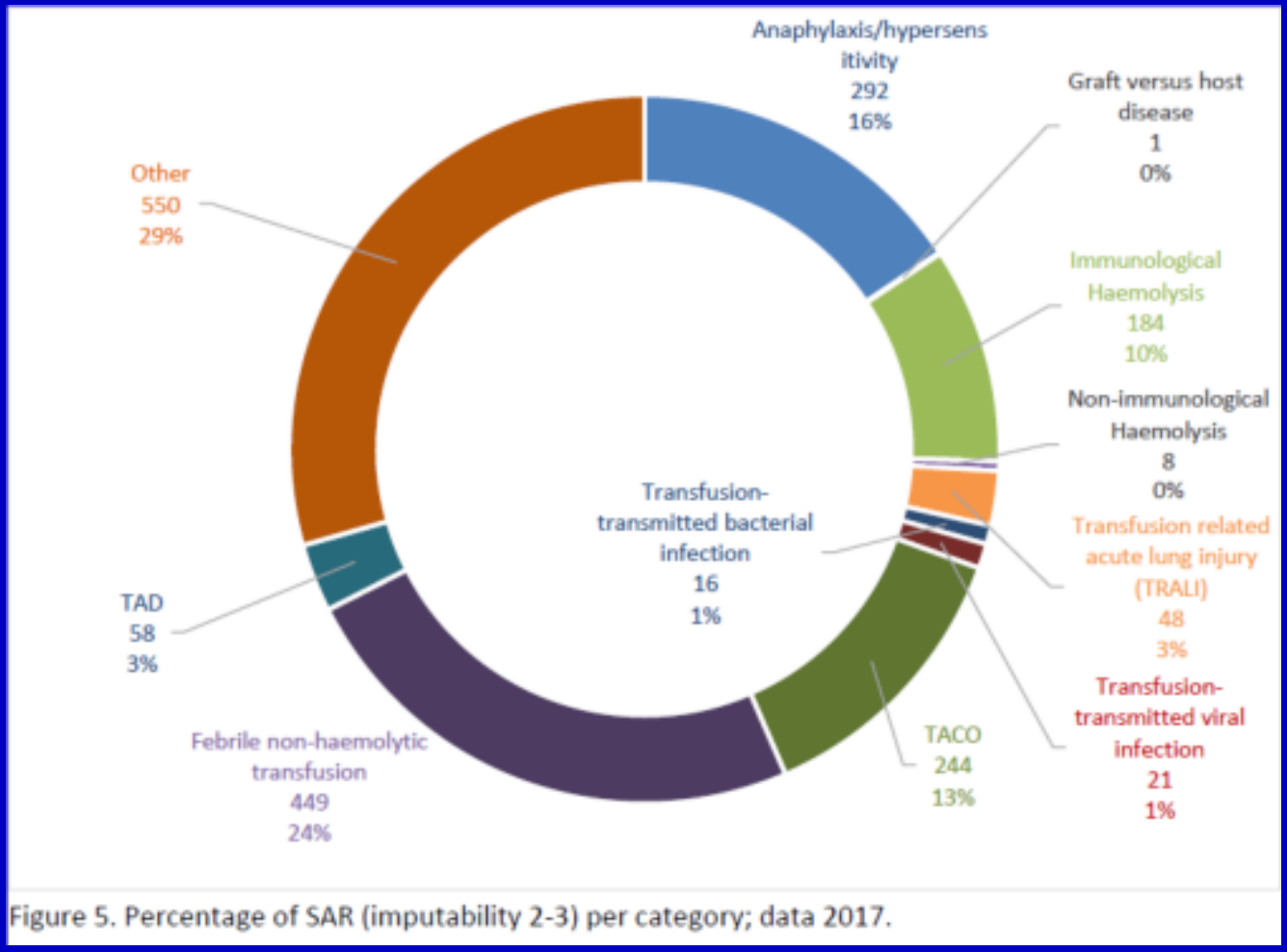


Figure 5. Percentage of SAR (imputability 2-3) per category; data 2017.

The 1,871 SAR (imputability level 2 or 3) reported were classified as follows:

- Febrile non-haemolytic transfusion reaction (FNHTR): 449 cases
- Anaphylaxis/hypersensitivity: 292 cases
- Transfusion-associated circulatory overload (TACO): 244 cases
- Immunological haemolysis: 184 cases, of which
  - 67 cases due to ABO incompatibility and
  - 117 cases due to other alloantibodies
- Transfusion-associated dyspnoea (TAD): 58 cases
- Transfusion-related acute lung injury (TRALI): 48 cases
- Transfusion-transmitted viral infection: 21 cases (3 hepatitis A, 4 hepatitis B, 12 hepatitis E, 1 hepatitis C and 1 Parvovirus B19)
- Transfusion-transmitted bacterial infection: 16 cases
- Non-immunological haemolysis: 8 cases
- Graft versus host disease: 1 case
- Other: 550.

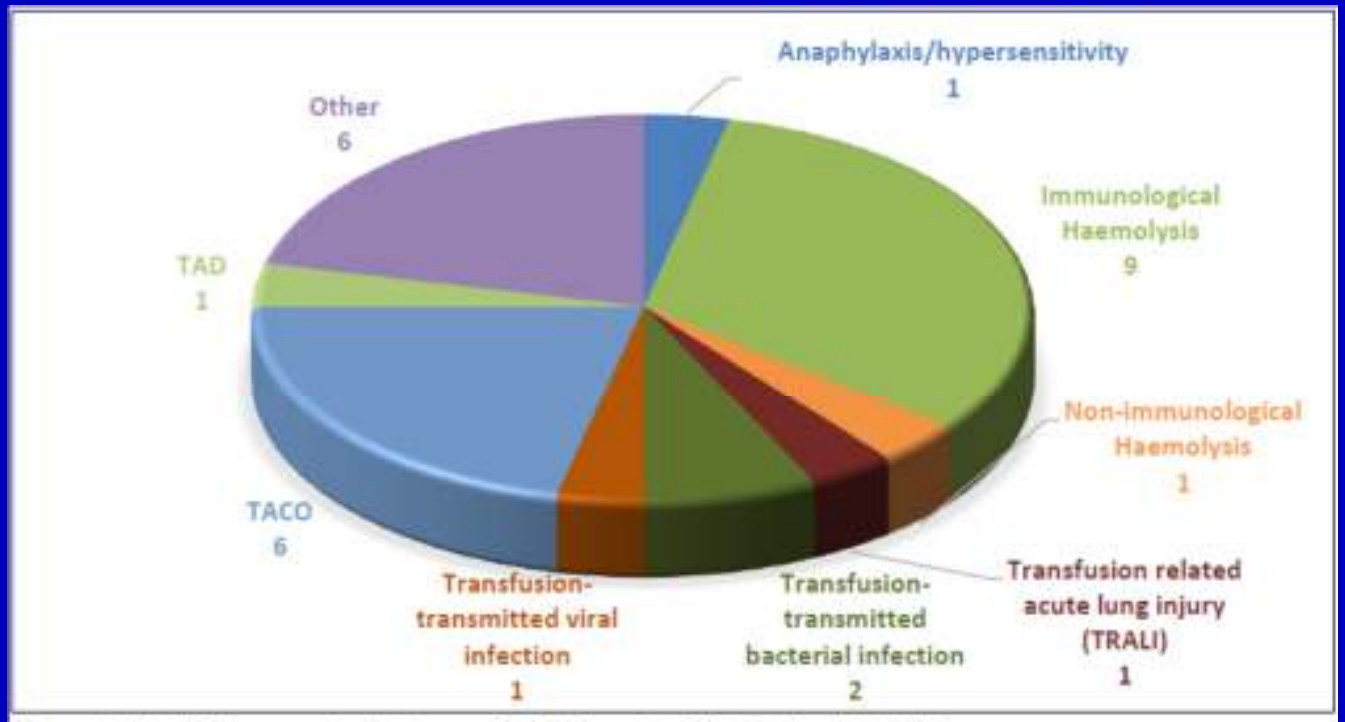


Figure 6. Fatalities reported by type of SAR (imputability 2-3); data 2017.

	2011		2012		2013		2014		2015		2016		2017		2018	
	Countries reporting	Number	Countries reporting	Number	Countries reporting	Number	Countries reporting	Number	Countries reporting	Number	Countries reporting	Number	Countries reporting	Number	Countries reporting	Number
Units issued	26	22,817,166	29	24,821,809	27	25,129,344	27	24,043,766	27	25,717,028	26	25,324,888	29[11]	24,827,516	29[11]	25,093,906
Units transfused	19	16,718,258	17	12,311,691	20	13,351,948	22	16,564,817	25	21,425,047	25	21,443,125	25	20,910,579	24	20,674,603
Recipients transfused	11	2,290,304	16	2,964,839	19	3,595,155	20	3,216,938	18	4,190,835	18	4,246,978	20	3,134,944	19	3,522,623
SAR (1-3)	30	2,449	30	3,133	30	3,519	30	2,831	30	2,441	31	2,587	30	2,950	29	3,114
SAR (2-3)	30	1,259	30	1,574	30	1,831	30	1,739	30	1,410	31	1,349	30	1,737	29	1,871
SAR death (2-3)	30	20	30	14	30	22	28	22	30	27	31	25	30	16	29	28
SAE	28	16,360	25	4,115	28	2,955	30	2,972	30	4,460	24	2,358	30	2,599	29	2,920
SAR in donors					18	2,494	23	2,470	20	3,723	23	7,769	23	7,658	23	4,655

Ασφάλεια και κίνδυνοι από τα παράγωγα αίματος:

Κίνδυνοι για μετάδοση παρασιτικών λοιμώξεων

Ευρωπαϊκές Στρατηγικές κατά του κινδύνου μετάγγισης με παράσιτα καταστρώνονται σε συνάρτηση με οδηγίες από τον ΠΟΥ και το FDA:

Λοιμώξεις με πρωτόζωα είναι ενδημικές σε τροπικές χώρες κυρίως χαμηλού εισοδήματος και επηρεάζουν εκατομμύρια ανθρώπους.

Οι παρασιτικές ή λοιμώξεις από πρωτόζωα που μεταδίδονται εύκολα από μεταγγίσεις κυτταρικών στοιχείων του αίματος είναι:

- Η **Ελονοσία**
- Η Αμερικανική τρυπανοσωμίαση (**Trypanosoma cruzi**/N. Chagas)
- **Babesiosis** (Babesia), πρωτόζωα που συνήθως μεταδίδονται από τσιμπούρια
- **Λεϊσμανίαση**

- Τυχόν μολυσμένη με παράσιτα μετάγγιση αίματος ή παραγώγων αυτού σε ανοσοκατασταλμένους ασθενείς σε Δυτικές μη ενδημικές χώρες καθιστά δύσκολη την έγκαιρη διάγνωση ενώ η εξέλιξη της παρασιτικής νόσου μπορεί να εξελιχθεί σε βαριά μορφή με δυσκολία για εφαρμογή αποτελεσματικής θεραπείας.
- Σε πρόσφατο Διεθνές Φόρουμ 2017 στην ειδικότητα Τράπεζας αίματος / αιμοδοσίας / μεταγγίσεων, τα συμπεράσματα δείχνουν ότι στην Ευρώπη και ΗΠΑ η πρόληψη για λοιμώξεις από πρωτόζωα σε μεταγγίσεις εξαρτάται κυρίως από κατευθυνόμενα ερωτηματολόγια που πρέπει να απαντούν σωστά οι δότες πρό αιμοληψίας.

(Transfusion Clinique et Biologique Volume 12, Issue 1, February 2005, Pages 1-4)

(Brouwer et al. Malaria Journal 2013, 12:439)

PUBLIC HEALTH ENGLAND
MALARIA REFERENCE LABORATORY
PATIENT REPORT FORM

MRL No. __ __ / __ __ __ __

In confidence

Family name

All other names

Home post code

Primary care team

Address in UK

Date of birth: __ __ / __ __ / __ __ __ __ Age Gender: M / F

Country of birth Country of usual residence

Ethnicity (mark one)	Reason for travel (mark one)	Malaria prophylaxis taken (mark as relevant)
☐ White British	☐ New arrivals to UK	☐ None
☐ Other White background	☐ Visiting family in country of origin	☐ Mefloquine (Lariam)
☐ Black African	☐ UK citizen living abroad	☐ Malarone
☐ Black Caribbean	☐ Civilian aviation crew	☐ Doxycycline
☐ Other Black background	☐ British armed forces	☐ Chloroquine (Nivaquine/Avloclor)
☐ Indian Sub-Continent	☐ Business/Professional travel	☐ Proguanil (Paludrine)
☐ South East Asian	☐ Foreign student studying in UK	☐ Unknown
☐ Other Asian background	☐ Holiday travel to malarious country	☐ Other (please specify)
☐ Mixed Ethnicity	☐ Foreign visitor / tourist to UK	
☐ Other (please specify)	☐ Children visiting parents living abroad	Prophylaxis taken regularly? Y / N
	☐ Other (please specify)	Continued on return for ____ weeks

Date of onset of illness: __ __ / __ __ / __ __ __ __ Date of starting treatment: __ __ / __ __ / __ __ __ __

Date of arrival in UK from malarious country __ __ / __ __ / __ __ __ __

Duration of stay abroad

Country(ies) where infection acquired

For trivia, please specify areas visited

G.P. Name & Address

Tel. No.

Name and contact details of person completing this form if not G.P.

Please complete other side ⟶

Fig. 1. Public Health England malaria reference laboratory patient report form.

UK

British Journal of Haematology, 2012, 158, 113–121

ΕΛΟΝΟΣΙΑ

- Οι δότες μοιράζονται σε 2 ομάδες:
- (ι) Άτομα που έχουν ζήσει τα πρώτα 5 χρόνια της ζωής τους σε ενδημική περιοχή για ελονοσία
- (ιι) Άτομα που έχουν γεννηθεί και ζούν σε μη-ενδημικές περιοχές αλλά επισκέφθηκαν ενδημικές περιοχές.
- Στην (ι), οι δότες απορρίπτονται από αιμοδοσία για τα πρώτα 3 χρόνια μετά την τελευταία επίσκεψη τους σε ενδημική περιοχή. [Μία χώρα από τις συμμετέχουσες τους απορρίπτει συνολικά].
- Σε κάποιες χώρες, οι αιμοδότες μπορούν να συμμετέχουν σε αιμοδοσία μετά από 4 μήνες – 3 χρόνια μετά την επιστροφή τους εφ’ όσον ο εργαστηριακός έλεγχος για ελονοσία αποβεί αρνητικός.

- Στην (ιι), άτομα από μη ενδημικές περιοχές που επισκέφθηκαν ενδημικές για ελονοσία περιοχές, απορρίπτονται για αιμοδοσία για 4-12 μήνες μετά την επιστροφή τους και ελέγχονται πρό αιμοδοσίας για ελονοσία.
- Αριθμός χωρών, απορρίπτουν αιμοδότες για 3 έτη ή για πάντα εφ’ όσον οι δότες έχουν ζήσει σε ενδημικές περιοχές για διάστημα >6 μήνες.
- Η απόρριψη αιμοδοτών για τον κίνδυνο μετάδοσης ελονοσίας κυμαινόταν από 0.003-0.43% όλων των αιμοδοσιών για μετάγγιση και που βάσει ερωτηματολογίου ελέγχθηκαν.
- Συγκριτικά, την περασμένη 10-ετία μόνο μερικές περιπτώσεις εντοπίσθηκαν προφανώς λόγω καλύτερης εφαρμογής προ-ελέγχου με ερωτηματολόγια και εφαρμογής διαγνωστικών εξετάσεων σε επιλεγμένες περιπτώσεις πρίν την αιμοδοσία και μετάγγιση.

- Στις ΗΠΑ από το 1972-1988: 0.25 περιστατικά ανά 1 εκατ. αιμοδοσίες
- Και από το 1993-1998: 0-0.18 περιστατικά ανά 1 εκατ. αιμοδοσίες

- Ταξίδι στην Αφρική παρουσιάζει αύξηση του κινδύνου μόλυνσης με μετάγγιση κατά > 1000 φορές σε σύγκριση με ταξίδι σε ενδημικές περιοχές του Μεξικό.
- Οπότε οι ΗΠΑ τροποποίησαν την (ι) σε (ιι) για το Μεξικό κερδίζοντας έτσι 57000 αιμοληψίες κατ’ έτος.

Table 1 Overview of most relevant events

Date	Donor/Recipient	Event
August 2005	Donor	Visit to Kenya, Tanzania and Zanzibar
3 April 2006	Donor	First visit to blood bank for examination and giving blood samples for testing
4 May 2006	Donor	Blood donation #1
2 August 2006	Donor	Blood donation #2
Aug – Sept 2006	Donor	Visit to Thailand (only to low risk areas for malaria, no precautions required)
9 November 2006	Donor	Blood donation #3
27 February 2007	Donor	Blood donation #4 at a thrombocyte count of 141×10^9/L
22 May 2007	Donor	Blood donation #5 at a thrombocyte count of 94×10^9/L
29 May 2007	Donor	Informed about thrombocytopenia, referred to GP
14 August 2007	Donor	Blood donation #6
September 2007	Donor	Visit to Costa Rica
26 May 2008	Donor	Blood donation #7
25 August 2008	Donor	Blood donation #8
17 November 2008	Donor	Blood donation #9
11 February 2009	Donor	Blood donation #10
8 June 2009	Donor	Blood donation #11
3 December 2009	Donor	Blood donation #12
1 March 2010	Donor	Blood donation #13
10 May 2010	Donor	Blood donation #14
9 February 2011	Donor	Blood donation #15
14 February 2011	Recipient	Coronary-artery bypass surgery and transfusion of red blood cell concentrate from blood donation #15
14 April 2011	Recipient	Diagnosis *P. malariae* infection
14 April 2011	Donor	Informed about post-transfusion malaria infection in recipient
7 June 2011	Donor	Blood collection for malaria tests
23 June 2011	Donor	First laboratory evidence for subclinical *P. malariae* infection in specimen collected on 7 June 2011
17 August 2011	Donor	First visit to Harbour hospital and initial examination
8 November 2011	Donor	Second visit to Harbour hospital, confirmation of on-going, subclinical *P. malariae* infection
10 November 2011	Donor	Start oral chloroquine treatment

Table 2 Overview of relevant laboratory test results of donor

Collection date	Laboratory test	Result	Specimen
27 February 2007	Thrombocyte count	141 × 10^9/L	Fresh blood
22 May 2007	Thrombocyte count	94 × 10^9/L	Fresh blood
9 February 2011	*Plasmodium* species PCR *	Negative	Stored plasma
	Malaria serology by ELISA *	Negative (0.36, cut off <1.0)	Stored plasma
	Malaria serology by immunofluorescence assay (IFA) *	Positive (1:40), cut-off <1:40)	Stored plasma
7 June 2011	*Plasmodium* genus PCR	Positive for *Plasmodium* spp.	EDTA-blood
	Malaria serology by immunofluorescence assay (IFA)	Positive (1:160, cut-off <1:40)	Serum
	Plasmodium species PCR	Positive for *P. malariae*	EDTA-blood
17 August 2011	Thrombocyte count	126 × 10^9/L	Fresh blood
	Thick blood smear, QBC, malaria antigen test	Negative	Fresh blood
8 November 2011	Thrombocyte count	128 × 10^9/L	Fresh blood
	QBC, malaria antigen test, thin blood smear, malaria serology (ELISA)	Negative	Fresh blood
	Thick blood smear	One structure suspected for *P. malariae*	Fresh blood
	Plasmodium species PCR	Positive for *P. malariae*	Fresh blood
	Plasmodium species PCR	Positive for *P. malariae*	Bone marrow biopsy
	Leishmania spp. PCR	Negative	Bone marrow biopsy
26 November 2011	Thrombocyte count	144 × 10^9/L	Fresh blood
1 December 2011	Thrombocyte count	157 × 10^9/L	Fresh blood
12 July 2012	Thrombocyte count	139 × 10^9/L	Fresh blood
	Thick and thin blood smears, QBC, malaria antigen test, malaria serology (ELISA), *Plasmodium* species PCR	Negative	Fresh blood
6 March 2013	Thrombocyte count	209 × 10^9/L	Fresh blood
	Thick and thin blood smears, QBC, malaria antigen test, malaria serology (ELISA), *Plasmodium* species PCR	Negative	Fresh blood

The tests were performed in the following laboratories: Diagnostic Services, Sanquin Blood Supply Foundation, Amsterdam (2007 and malaria serology by ELISA first half 2011); Dept. Medical Microbiology, University Medical Centre St Radboud, Nijmegen, and/or Dept. Parasitology, Leiden University Medical Centre, Leiden (first half 2011); the Harbour Hospital Laboratory, Dept. Medical Microbiology and Infectious Diseases and Clinical Chemistry, Erasmus University Medical Centre and Harbour Hospital, Rotterdam (from August 2011 onwards). Tests marked by an *indicate results of tests performed retrospectively in May to July 2011 on materials collected on 9 February 2011.

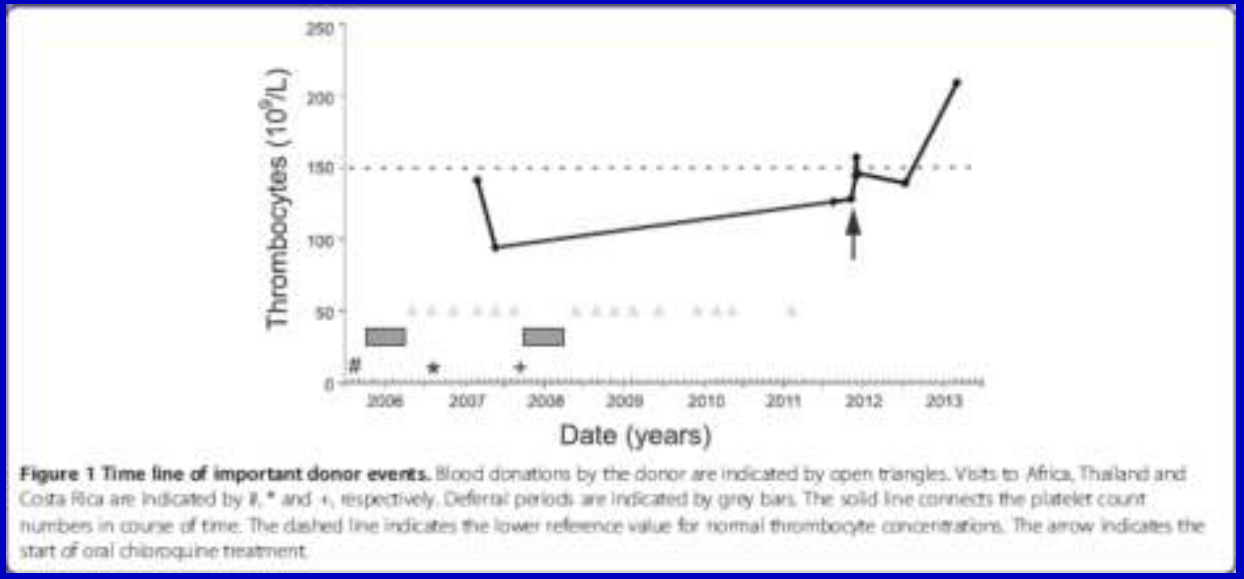


Figure 1 Time line of important donor events. Blood donations by the donor are indicated by open triangles. Visits to Africa, Thailand and Costa Rica are indicated by #, * and +, respectively. Deferral periods are indicated by grey bars. The solid line connects the platelet count numbers in course of time. The dashed line indicates the lower reference value for normal thrombocyte concentrations. The arrow indicates the start of oral chloroquine treatment.

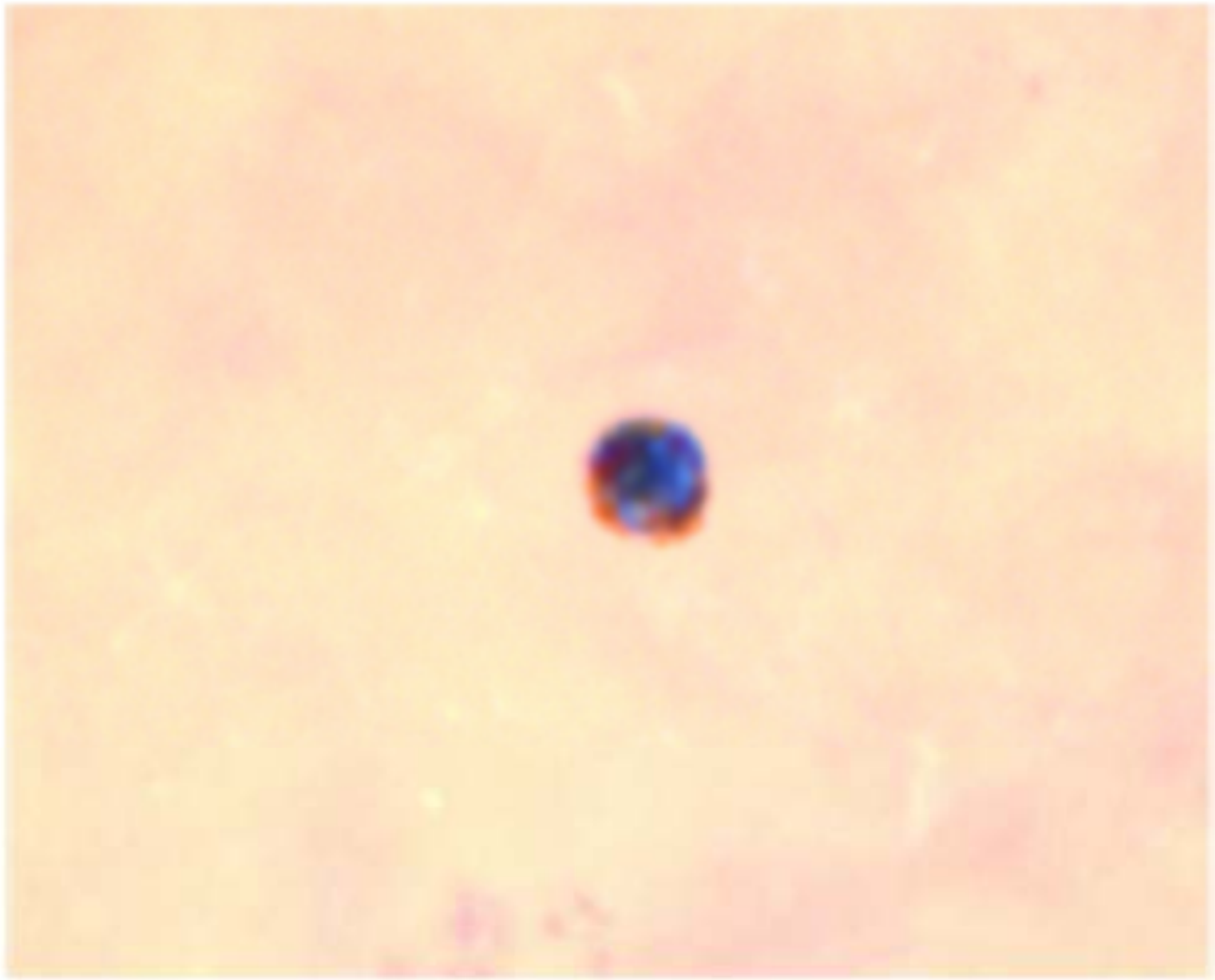


Figure 2 Giemsa-stained thick blood smear from the donor with a structure suspected for a malaria parasite.

Trypanosoma cruzi (Αμερικανική τρυπανοσωμίαση):

- Σε αριθμό χωρών, με ερωτηματολόγια διερευνάται η κινδυνότητα μετάδοσης του T. Cruzi.

- Σε μερικές χώρες οι αιμοδότες απορρίπτονται όταν αυτοί ή οι μητέρες τους, γεννήθηκαν στην Νότια ή Κεντρική Αμερική ή αν έχουν λάβει μετάγγιση στις χώρες αυτές και όταν έζησαν εκεί σε αγροτικές περιοχές για >4 μήνες.

(Transfus Med. 2009 Feb;19(1):16-23. doi: 10.1111/j.1365-3148.2009.00915.x.)

(Transfus Clin Biol. 2011 Apr;18(2):286-91)

- N. Chagas (Αμερικανική τρυπανοσωμίαση)
- Προκαλείται από το πρωτόζωο Trypanosoma cruzi
- Είναι ενδημικό στο Μεξικό μέχρι την Αργεντινή
- Η «εισαγώμενη» N. Chagas στην Ευρώπη φαίνεται να αποτελεί σήμερα μία νέα απειλή σε μη-ενδημικές χώρες κυρίως λόγω μετανάστευσης από την Λατινική Αμερική και του τουρισμού προς τις χώρες της Λ. Αμερικής.
- Σε μη-ενδημικές χώρες ή μετάγγιση αποτελεί την πιο σημαντική οδό μετάδοσης της νόσου.
- Στρατηγικές για μείωση της μετάδοσης αφορούν την επιλογή αλλά και την απόρριψη δοτών με ιστορικό και βάσει ερωτηματολογίων, με συμπληρωματικούς ελέγχους αίματος, μείωση λευκών αιμοσφαιρίων (leukoreduction), χρήση φίλτρων και συστήματα απενεργοποίησης παθογόνων (inactivation systems).

- Σε ενδημικές περιοχές, γίνετε έλεγχος αίματος για αντισώματα T. cruzi.
- Σε μη ενδημικές περιοχές υπάρχουν 2 στρατηγικές:
- (ι) Απόρριψη αιμοδοτών με πιθανότητα για T. cruzi
- (ιι) Αποδοχή αιμοδοσίας εφ’ όσον ειδικές εργαστηριακές εξετάσεις αποβούν αρνητικές για T. Cruzi (πχ. Όπως στην Γαλλία από το 2007)

Η (ιι) προσέγγιση εισάγεται για χώρες με σημαντικό πληθυσμό από την Λ. Αμερική όπως ΗΠΑ, Ισπανία, Γαλλία

* Στη Γαλλία (Transfus Clin Biol 2011 18(2):286-91) από 05/2007-12/2008 και από 4637479 αιμοδοσίες ελέγχθηκαν οι 163740 (3.5%). Ο επιπολασμός T. cruzi βρέθηκε 1 σε 32800 αιμοδοσίες. Δηλαδή εντοπίσθηκαν 5 θετικά για T. cruzi όλα από άτομα που προέρχονταν από ενδημική περιοχή.

Table II - Different control strategies for the prevention of transfusion-transmitted Chagas disease (TT-CD) in non-endemic countries.

Non -endemic country	Estimated n. of people affected by CD	Strategy for TT-CD control	Implemented since year	Infected donations/ donors	Transfusion-acquired cases
United States of America	38,777-339,954[1]	Universal donor screening/selective one time testing of donors	1989-2009/ 2010 →	1/27,500	Yes
Canada	Fewer than 100,000[2]	Selective donor screening (questionnaire)	2010	3/1,000	Yes
Spain	12,533-25,728[1]	Selective donor screening (questionnaire)	2005	1/218	Yes
France	1,311-1,712[1]	Selective donor screening (questionnaire)	2007	1/32,800	No
United Kingdom	1,006-1,324[1]	Selective donor screening (questionnaire)	1998-2005 (donors), 2005 → (donations)	1/12,861	No
Italy	6,000-12,000[3]	Deferral period after exposure (no testing): under revision	2005	3.9/100	No
Sweden	1,118[1]	Permanent deferral of at-risk donors	-	-	No
Switzerland	3,000[2]	Selective donor screening (questionnaire)	2013	-	No
Australia	1,928[4]	Selective donor screening (questionnaire)	-	-	Yes
China	-	No strategies	-	-	No
Japan	3,000[4]	Permanent deferral of affected donors	-	-	No

-: data not known;

Other European countries are currently following the European Commission's 35 directives, 2004/33/CE and 2006/17/CE.

1) Guerri-Guttenberg RA, Grana DR, Ambrosio G, Milei J. Chagas cardiomyopathy: Europe is not spared! Eur Heart J 2008; **29**: 2587-91.
2) Hotez PJ, Dumonteil E, Betancourt Cravioto M, et al. An unfolding tragedy of Chagas disease in North America. PLoS Negl Trop Dis 2013; 7: e2300.
3) World Health Organization. Control and prevention of Chagas disease in Europe. Report of a WHO Informal Consultation (jointly organized by WHO headquarters and the WHO Regional Office for Europe) 2009;WHO/HTM/NTD/IDM/2010.1.
4) Jackson Y, Pinto A, Pett S. Chagas disease in Australia and New Zealand: risks and needs for public health interventions. Trop Med Int Health 2014; **19**: 212-8.

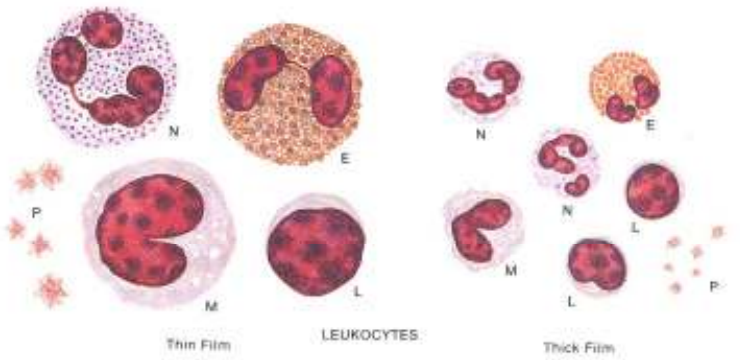
Table 28.4 Prevalence of infectious disease markers in blood donors in the region of the Americas during 2001.

Country	anti-HIV	HBsAg	anti-HCV	anti-syphilis	Anti-*T. cruzi*	TOTAL
Argentina	0.18	0.60	0.66	0.92	4.50	6.86
Bolivia	0.04	0.57	0.28	1.54	9.91	12.34
Brazil	0.33	0.88	0.78	1.08	0.65	3.72
Chile	0.04	0.16	0.13	0.88	0.61	1.82
Columbia	0.25	0.48	0.56	1.56	0.67	3.52
Costa Rica	0.05	0.16	0.21	0.38	0.58	1.38
Cuba	0.01	0.70	0.74	0.55	Not required	2.00
Ecuador	0.24	0.27	0.20	0.52	0.08	1.31
El Salvador	0.20	0.43	0.25	0.93	3.70	5.51
Guatemala	3.71	0.93	1.08	2.38	1.48	9.58
Honduras	0.38	0.43	0.65	0.85	1.40	3.71
Mexico	0.20	0.05	0.07	0.02	0.05	0.39
Nicaragua	0.17	0.52	0.28	1.06	0.06	2.09
Panama	0.10	1.70	0.60	0.40	0.90	3.70
Paraguay	0.20	0.52	0.59	4.42	4.46	10.19
Peru	0.10	0.82	0.50	0.89	0.29	2.60
Dominican Rep.	0.42	1.20	0.64	0.86	Not required	3.12
Uruguay 1998	0.07	0.24	0.33	0.57	0.45	1.66
Venezuela	0.25	0.94	0.56	1.40	0.67	3.82
Average	0.37	0.61	0.48	1.12	1.79	4.17

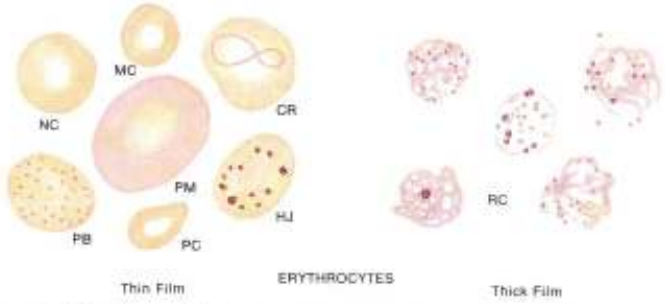
- **Babesia και Leishmania:**
- Σε καμμία από τις χώρες που συμμετείχαν στο σχετικό Φόρουμ δεν χρησημοποιείτο ερωτηματολόγιο σχετικά με λοίμωξη από Babesia ή Leishmaniasis.

- **Εργαστηριακή Διάγνωση**:
- **Ι. Ελονοσία**
- * Δεν χρησημοποιείται RDT
- Ορολογικός έλεγχος για Plasmodium IgG Abs, Malaria Total Ab EIA kit (Lab21, Healthcare Ltd), ΔΕΝ αποτελούν κατάλληλες δοκιμασίες για διάγνωση P. malariae
- MONO:
- Sensitive serological immune fluorescence assay (IFA)
- Real Time PCR analysis
- Αποτελούν αποδοτικές μέθοδοι για τη διάγνωση πλασμωδίων και δή ασυμπτωματικής λοίμωξης P. Malariae (εξ' ορισμού με πολύ μικρή παρασιταιμία)
- Συμπληρωματικά: Μικροσκοπία με χρώση Giemsa (λεπτή ή παχεία)

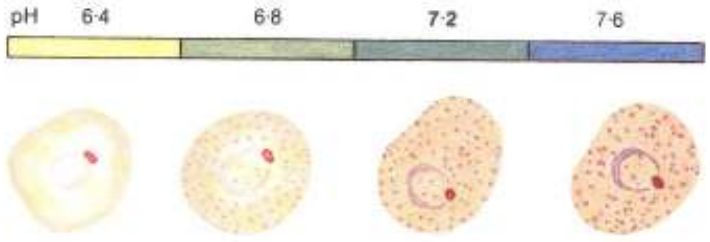
Fig. 13. Appearance of cellular elements in Giemsa-stained thin and thick blood films: effect of pH on Giemsa staining of malaria parasites



N = Neutrophil, E = Eosinophil, M = Monocyte, L = Lymphocyte, P = Platelets



NC = Normocyte, MC = Microcyte, PM = Polychromatic macrocyte, PC = Poikilocyte, PB = Punctate basophilia, CR = Cabot's ring, HJ = Howell-Jolly bodies, RC = Reticular 'clouds' and chromatoid bodies in severe anaemia



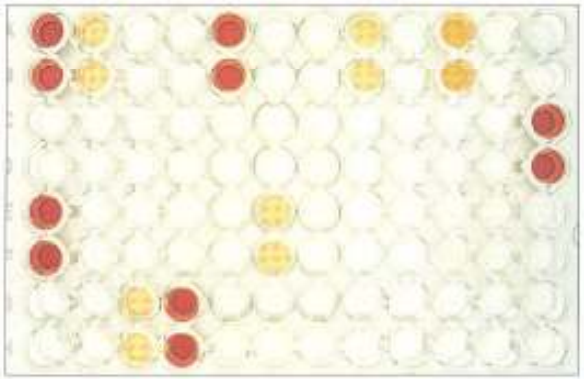
MALARIA STAINING AND pH

- **II. Trypanosoma cruzi**
- T. cruzi Ab
- Χρήση T. cruzi Ag homogenate και Immunologic Assays
- Ή
- Recombinant Antigens Assays

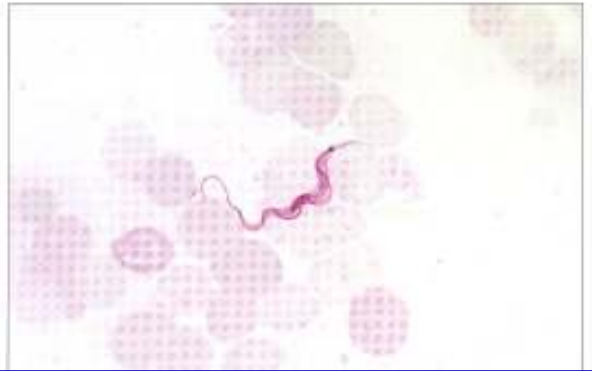
- Και συμπληρωματικά με:

- Indirect Immunofluorescence (IIF)
- Ή
- Immunoblot τεχνικές

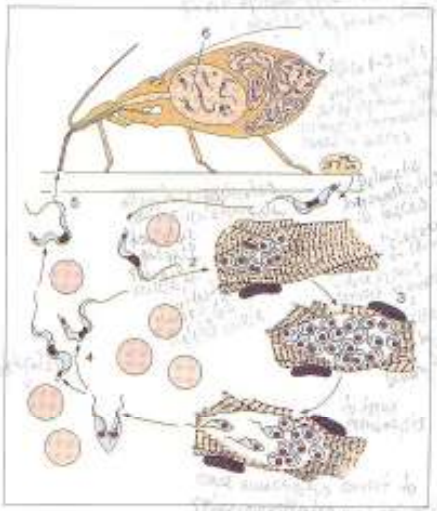
189 Xenodiagnosis Absolute confirmation of active infection is obtained by demonstrating that the patient can infect the vector (xenodiagnosis). Laboratory-bred, clean reduviid bugs are fed on patients suspected of having trypanosomiasis. Two weeks later the hindgut is dissected out and is examined for metacyclic trypanosomes.



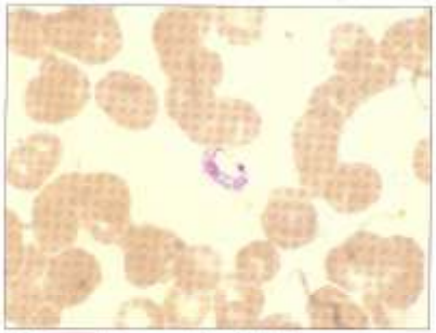
190 ELISA test An ELISA test employing antigen from epimastigotes of *T. cruzi* cultivated *in vitro* is widely used, and is one of the most sensitive means of diagnosis. It can indicate past or present infection and does not necessarily imply the presence of parasites. Five positive reactions are shown in this microtitre plate in which an immunoperoxidase procedure was followed. Fluorescent antibody tests may also be employed, using whole cultured epimastigotes as the antigen. The exquisitely sensitive PCR technique, which is capable of detecting the DNA from a single parasite, will be invaluable in the control of blood for transfusion in endemic countries, as well as for the diagnosis and follow-up of individual patients in whom few parasites may be accessible for detection by other methods. (The Machado–Guerreiro test is is no longer in general use.)



191 *Trypanosoma rangeli* *T. rangeli* is a long, slender trypanosome also transmitted by reduviid bugs from wild animals to man. It is readily distinguished by its shape from *T. cruzi* in blood films, and appears to be non-pathogenic to man. (Giemsa × 1 200)



173 Cycle of *T. cruzi* in man and triatomid bugs (1) Infective, metacyclic trypomastigotes passed with the faeces enter the body either through the site of the bite or directly penetrate mucosal surfaces such as that of the eye; some enter reticuloendothelial cells of the subcutaneous tissues near the site of entry to produce a local swelling known as a 'chagoma'. (2) Other trypomastigotes reach the circulatory system from which they penetrate smooth muscle in organs such as the heart, the walls of the gastrointestinal tract, or skeletal muscle. Associated autonomic-system ganglion cells may also be invaded. (3) In these organs, they convert to amastigotes which reproduce by binary fission to form pseudocysts. (4) Some amastigotes later revert to sphaeromastigotes and epimastigotes which form trypomastigotes that re-enter the circulation. (5) Circulating trypomastigotes, which are taken up when another triatomid feeds, enter the bug's midgut. (6) In the gut, the flagellates change to short epimastigote and rounded sphaeromastigote forms in which they multiply profusely by binary fission. (7) After one to two weeks, longer epimastigotes enter the rectum where they form metacyclic trypomastigotes that are passed when the bug again defaecates (*adapted from Geigy and Herbig, 1955*). (*See also* **174**, **177** *and* **181**.)



174 *T. cruzi* in human blood film The causative agent occurs in blood films characteristically as short 'C'-shaped or 'S'-shaped trypomastigotes with a prominent kinetoplast. It is otherwise monomorphic. (Giemsa × 950)

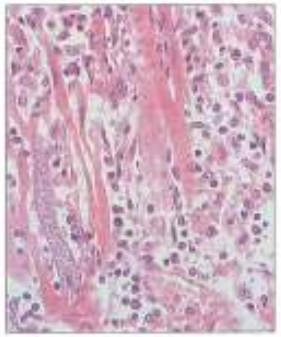
175 Ecology of triatomid vectors The favourite habitats of the reduviid bugs are cracks in the walls of mud huts in poor rural areas; here, the insects shelter and breed. Transmission occurs predominantly at night. Adults and clusters of eggs can be seen here.

176 Male *Triatoma infestans* feeding on a human arm Note the position of the proboscis. Reduviid bugs (also known as 'assassin' or 'kissing' bugs), particularly in the genera *Triatoma*, *Rhodnius* and *Panstrongylus*, transmit *T. cruzi* while feeding, not by inoculation but by faecal contamination. Both sexes are similar and feed on blood, with the male having a small, ventral abdominal swelling. The insects normally hide in the walls during the daytime and emerge after dark to feed on the sleeping inhabitants. The bite is remarkably painless even though the bug may take from 10–20 minutes to become fully engorged. (× 1.6)

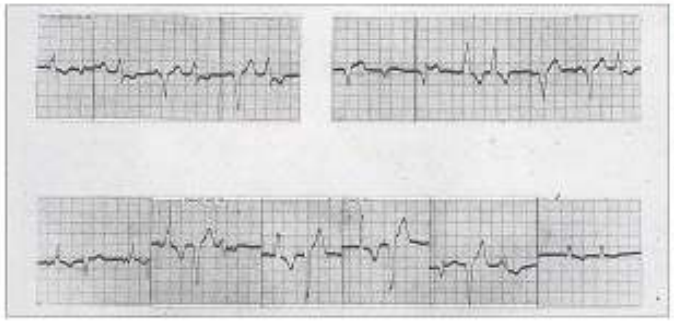
177 Metacyclic stage in faeces Infection is through contamination by parasites in bug faeces produced on the skin. These may invade the site of the bite or adjacent mucosa (e.g. the conjunctiva). (*Giemsa × 1 500*)

178 & 179 Reservoir hosts of *T. cruzi* Chagas' disease is a zoonosis *T. cruzi* has an extensive mammalian reservoir in both wild hosts (especially armadillos, such as the *Dasypus novencinctus* seen here – **178**, top) and opossums (*Didelphis* species – **179**, bottom), as well as some domestic animals.

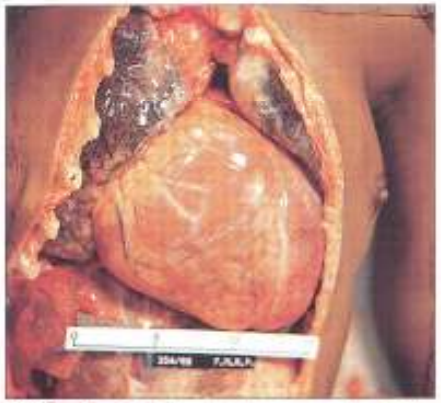
180 Romaña's sign The infection often begins with a local lesion, the chagoma. It causes marked local oedema which, should it occur in the region of the eye or within the conjunctival sac, is accompanied by swelling of the lids and chemosis. These unilateral periorbital changes constitute Romaña's sign.



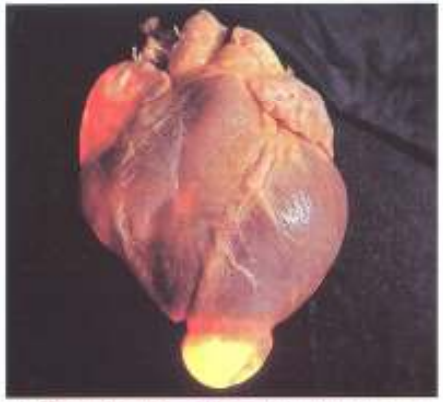
181 Amastigotes in heart muscle After a stage of initial parasitaemia associated with fever (often unrecognised), trypomastigotes pass to the cardiac muscle and the smooth muscle lining the intestinal tract. Here, they transform to the amastigote stage (Leishman–Donovan bodies) in which they multiply to form pseudocysts. In the heart, this is associated with severe myocarditis, especially in the early stages of the infection. The severity of the acute myocarditis seems to seal the eventual fate of the sufferer from chronic cardiac changes. These can be reactivated in patients with AIDS. (Giemsa × 350)



182 ECG showing heart block Dysrhythmias of various types and degrees are a characteristic feature of Chagas' disease; complete heart block with Stokes–Adams attacks can occur and may result in sudden death.



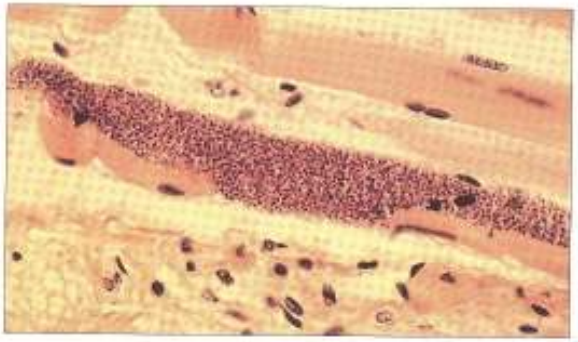
183 Cardiomegaly The heart shows gross enlargement and dilatation. The dilatation of the right atrium and both ventricles is marked in this specimen. The pathogenesis seems to be associated with a loss of autonomic control due to destruction of the ganglionic plexuses. Auto-antibodies are probably involved in this process. (See also **185**.)



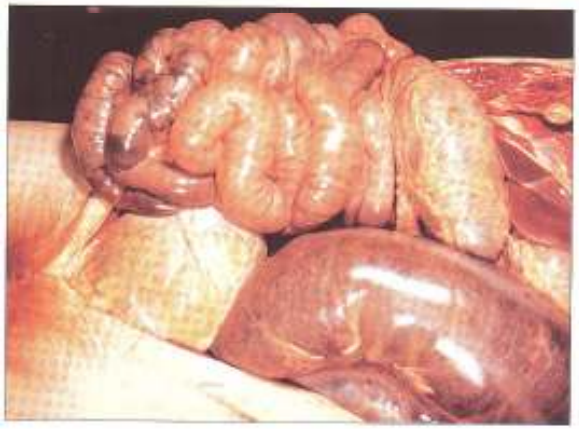
184 Apical aneurysm of heart Mural thrombi may be present at the apex of the left ventricle, with marked thinning of both ventricular walls. Apical aneurysmal formation is commonly seen.

185 Sympathetic ganglion in wall of atrium Degenerative changes in neuronal cells from a ganglion in the heart of a patient with Chagas' disease who died of sudden cardiac failure. Mononuclear cellular infiltration is conspicuous, especially round the capsule of the ganglion. CNS involvement may present as diffuse meningoencephalitis with necrosis in individuals who develop AIDS. (*H&E × 130*)

186 X-ray of megaoesophagus Muscular degeneration and denervation of segments of the alimentary tract through destruction of the cells of Auerbach's plexus cause megaoesophagus, megastomach and megacolon, etc., which can be detected radiologically.



187 Amastigotes in oesophageal muscle Pseudocysts containing amastigotes of *T. cruzi* can rarely be demonstrated in ganglion cells of the intestinal tract, although the smooth muscle is often invaded. (*× 480*)



188 Post-mortem of patient with megacolon Gross megacolon is shown here in a woman who died of chronic Chagas' disease.

- **Babesiosis**
- Νόσος από το πρωτόζωο Babesia με εύρος κλινικών εκδηλώσεων που κυμαίνεται από ασυμπτωματική νόσο μέχρι σε απειλητική για τη ζωή κατάσταση.
- Διαφορετικά είδη βρίσκονται σε διάφορες γεωγραφικές κατανομές (πχ. Στις ΗΠΑ στα Β.Α και Μεσοδυτικά).

- Περισσότερα από 150 περιπτώσεις έχουν αναφερθεί σε σχέση με μεταγγίσεις στις ΗΠΑ.
- Ελάχιστες φορές γίνονται ερωτήσεις στους αιμοδότες σχετικά με αυτή τη νόσο ή σε σχέση με δήγμα από τσιμπούρια.
- "Serosurveys" στη Γαλλία, Ελβετία & Γερμανία από άτομα που εκτέθηκαν σε τσιμπούρια έδειξαν σε αιμοληψίες για αιμοδοσία Αντισώματα έναντι Babesia από 1.0% to 11.5%. Αυτά τα ευρήματα δείχνουν ότι η συχνότητα της λοίμωξης από Babesia είναι συχνότερη από ότι εθεωρείτο και μπορεί να επηρεάζει υγιή άτομα, άτομα με σπληνεκτομή/ ανοσοκατασταλμένους.

- **Εργαστηριακή διάγνωση:**
- Δύσκολη διάγνωση
- Ab έναντι- Babesia (Ορολογικές εξετάσεις)
- Fluorescent microscopy
- Real Time PCR
- Μικροσκοπία:
- Χρώση Giemsa (πλακάκι)
- Χρώση acridine orange (πλακάκι)

- Οι εξετάσεις αυτές δεν εφαρμόζονται θεσπισμένα σε αιμοδοσίες παγκοσμίως!!!

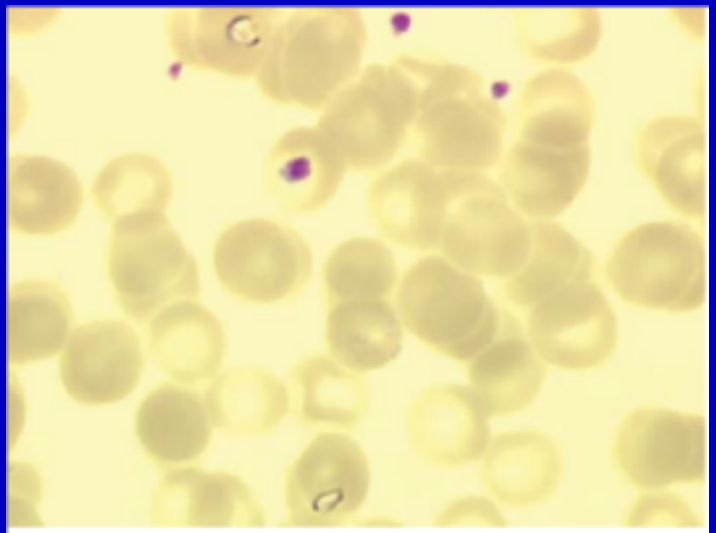


Figure 1. Two trophozoites, pear-shaped, of *Babesia divergens* in erythrocytes from case-patient 1 (original magnification ×1,000, May-Grünwald-Giemsa stain).

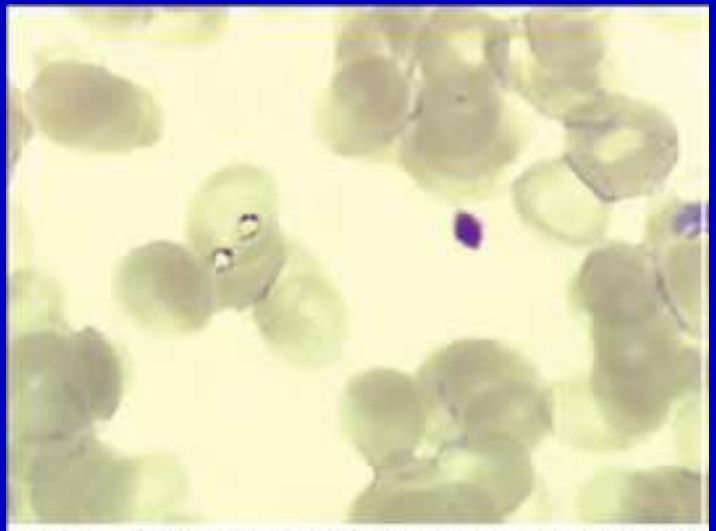


Figure 2. Two trophozoites of *Babesia* spp. in 1 erythrocyte from case-patient 2 (original magnification ×1,000, May-Grünwald-Giemsa stain).

(*See* **Table 5**.)

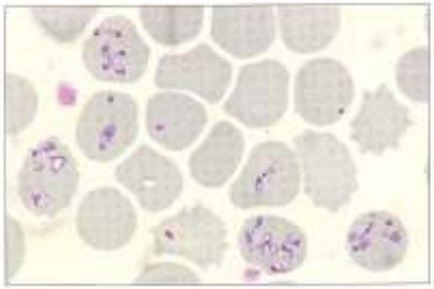
Table 5 The protozoa (phylum Apicomplexa) of medical importance

Phylum	Subclass	Order	Family	Genus and Species
SARCOMASTIGOPHORA (see Tables 16 and 17)				
CILIOPHORA (see Table 16)				
MICROSPORA (see Table 19)				
APICOMPLEXA	PIROPLASMASINA		Theileridae [1]	*Theileria microti*
			Babesiidae [1]	*Microbabesia bovis*
				M. divergens
				Babesia spp.
				Entoxylopoides sp.
	COCCIDIASINA			
		EUCOCCIDIORIDA		
	Subordo	HAEMOSPORORINA	Plasmodiidae	*Plasmodium* (*Plasmodium*) *vivax*
				P.(P.) ovale
				P.(P.) malariae
				P.(Laverania) falciparum
	Subordo	EIMERIORINA	Sarcocystidae	*Toxoplasma gondii*
				Sarcocystis hominis [2]
				S. suihominis [3]
				Sarcocystis spp. [4]
			Eimeridae	*Isospora belli*
				Cyclospora cayetanensis
			Cryptosporidiidae	*Cryptosporidium* spp.

[1] All rare in man
[2] Man definitive, cattle intermediate host
[3] Man definitive, pig intermediate host
[4] 'Sarcocystis lindemanni' consists of a number of unidentified species for which man is an intermediate host.

PIROPLASMOSIS IN MAN

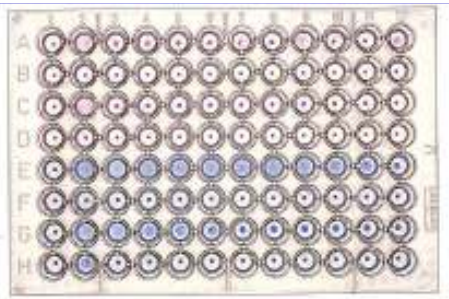
The piroplasms causing human infection have been reclassified into several genera (*see* **Table 5**). While most piroplasmosis in man is rare and usually occurs in immunocompromised (e.g. splenectomised) individuals, zoonotic infection with *Theileria microti* is relatively common, especially in parts of North America. The infections are transmitted by various species of ticks from animal reservoir hosts.



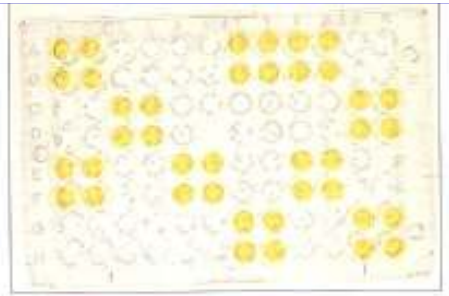
77 *Babesia divergens* in human blood Human infection with species of *Microbabesia* from cattle is a rare occurrence. Infection in normal people may give rise to a self-limiting fever and parasitaemia – e.g. the rodent parasite *Th. microti* on the Eastern seaboard of the United States. Heavy red-cell infection may develop, however, in splenectomised patients, leading to fatal haemolytic anaemia. This patient died from an infection acquired from the cattle parasite, *M. divergens*, in Scotland. (Giemsa × 700)

Leishmaniasis

- Direct Agglutination Test detecting circulating Abs
- Real Time PCR
- Μυελική βιοψία
- Σε Ινδούς ΜΟΝΟ: Έλεγχος Buffy coat φυγοκεντριμένου αίματος/ορού για παρουσία Leishmania (Σπλαχνική λεϊσμανίαση)

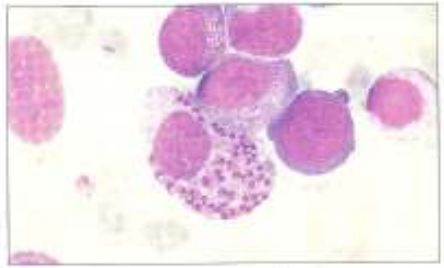


212 The direct agglutination test (DAT) This procedure, which has largely replaced the FAT, is now considered to give the most reliable and specific diagnosis of kala-azar, and works by the detection of circulating antibodies. The plasma samples in rows E and G of this picture are strongly positive. (*See also* **190**.)



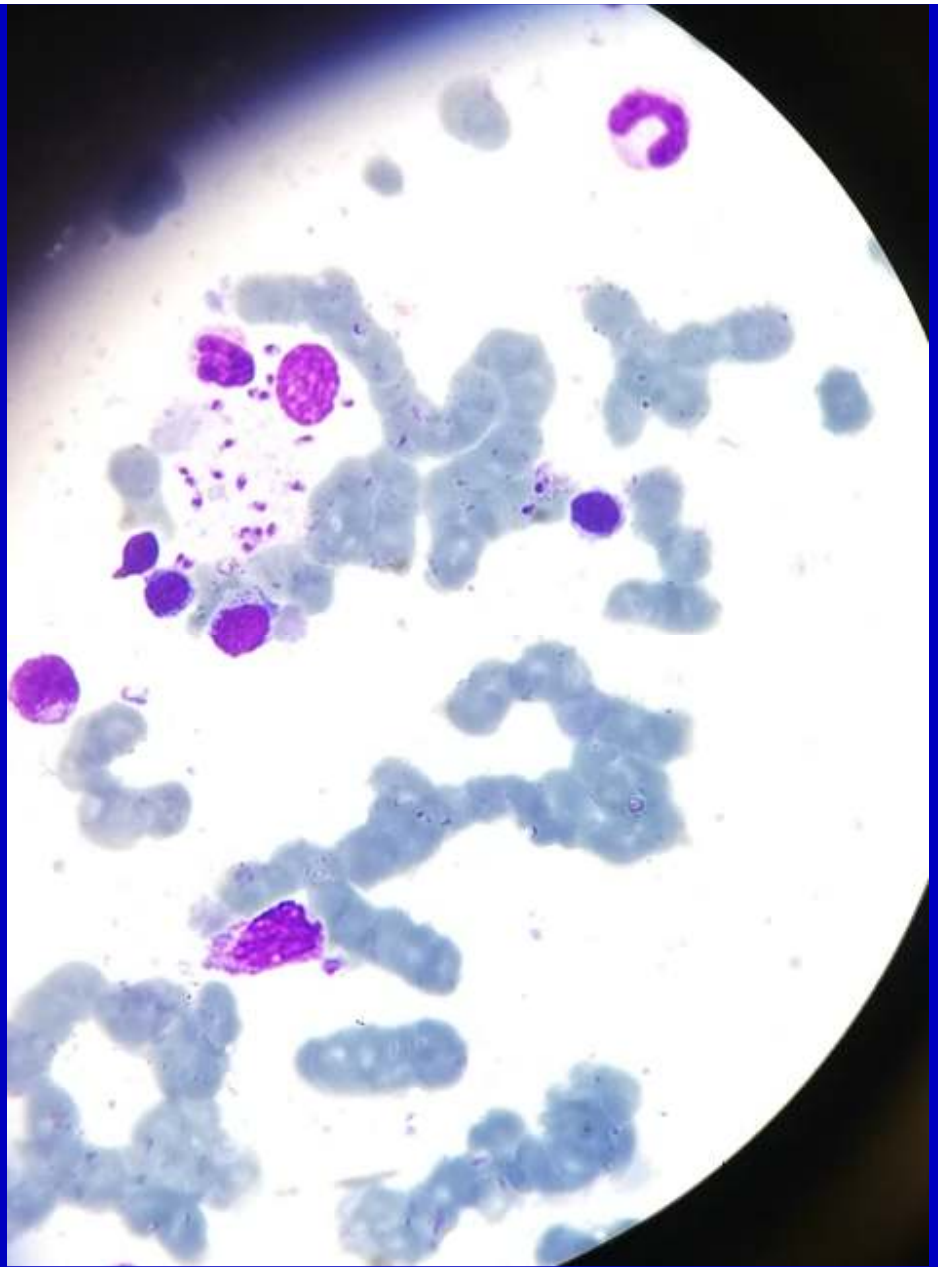
213 Polymerase chain reaction (PCR) in the diagnosis of leishmanial infection This exquisitely sensitive technique – it is capable of detecting the DNA from a single parasite – is invaluable in the diagnosis of subclinical or cryptic infection from a simple blood sample. This slide illustrates the technique as used with *L. donovani* in experimentally infected mice. Each block of four yellow wells indicates the presence of parasites in a single infected animal, samples from negative control animals remaining uncoloured.

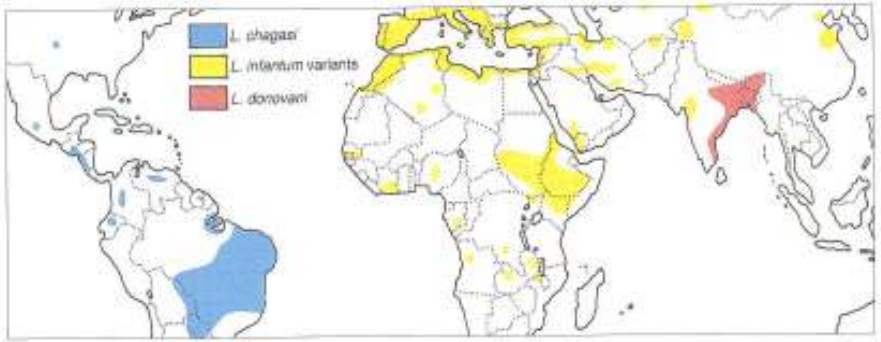
214 Iliac crest puncture of bone marrow The most direct means of diagnosis of kala-azar is the detection of amastigotes in bone-marrow, spleen or blood; the organisms are recognised in dried smears of material stained with a Romanowsky stain by their characteristic morphology. The newer serological techniques shown above, however, avoid the necessity for this more invasive procedure which is not always necessary.



215 *L. infantum* in macrophage from bone marrow While typically found in macrophages as shown here, isolated extracellular amastigotes from disrupted host cells are commonly seen in such preparations. (*Giemsa* × 810)

216 Promastigotes in NNN culture After inoculation of aspirated material into appropriate media such as blood agar [NNN (Novy–Nicolle–MacNeal) medium] and incubation at 28°C for 1–4 weeks, promastigotes may appear in the fluid overlay. *L. donovani* is more readily isolated in culture than *L. infantum*, which sometimes grows better in Schneider's insect tissue culture medium than in NNN. The parasites may also be isolated by intrasplenic inoculation into hamsters; after 4–6 weeks, characteristic visceral lesions are seen macroscopically and amastigotes are found in large numbers in smears of the liver and spleen. (*Giemsa* × 950)



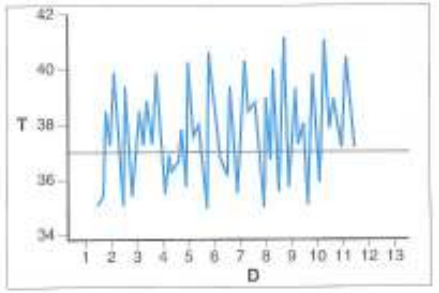


202 Distribution Visceral leishmaniasis caused by parasites of the *L. donovani–L. infantum* complex occurs in the Mediterranean littorals, the Middle East and adjacent parts of the former USSR, The Sudan, East Africa, the Indian subcontinent and China, and South America ('*L. chagasi*') (see **Table 7**). An arid, warm environment provides ideal ecological conditions for the breeding of many species of sandflies. Zoonotic kala-azar due to *L. infantum* and *L. chagasi* is commonly associated with dry, rocky, hill country where cases are typically scattered. In India, *L. donovani* is essentially an anthroponosis. This type of kala-azar may occur in severe epidemic fashion as can kala-azar in The Sudan.

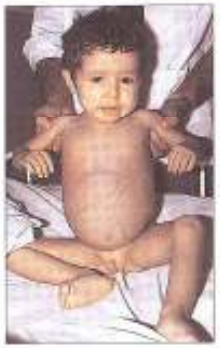
203 Ecology of kala-azar in India In parts of India (e.g. North Bihar) new epidemics of kala-azar have occurred in recent years. The close association between man and his domestic animals in this Bihari village favours the growth of populations of the local vector of *L. donovani*, *Phlebotomus argentipes*.

204 Reservoirs of zoonotic *L. infantum* The massive loss of hair, overgrowth of nails and generally poor condition of this dog in Colombia are typical of chronic infection with *L. chagasi* and *L. inantum*. Several varieties of this parasite that occur in the Old World are distinguishable by isoenzyme typing; these varieties can give rise to different clinical syndromes in man – e.g. cutaneous or mucocutaneous lesions rather than kala-azar. In addition to dogs, wild carnivores and various species of rodents may serve as reservoirs for the different varieties (zymodemes) (see **Tables 7** and **8**).

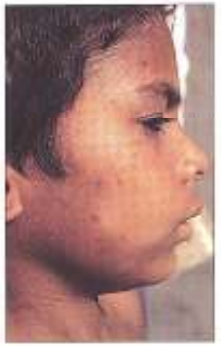
205 Temperature chart in kala-azar The temperature chart shows a double peak every 24 hours. Despite the high temperature, the patient often looks remarkably well and has a good appetite. A leucopenia with a relative lymphocytosis is often present. Kala-azar should be suspected if this picture is seen in an HIV-positive patient. [D – day of illness; T – temperature [°C].]



206 Clinical picture of kala-azar in Kenya
Increasing enlargement of the spleen and liver is a characteristic feature while, in dark-complexioned subjects, deepening skin pigmentation is seen – hence the synonym kala-azar, the 'black sickness'. A generalised lymphadenopathy is common in African kala-azar; the parasite in this area is considered to be in the *L. donovani* complex.

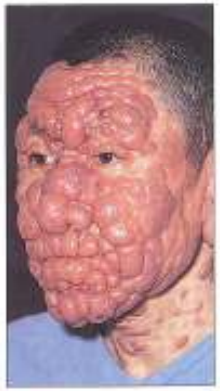


207 Infantile kala-azar
Children present with chronic, irregular fever, anaemia, leucopenia and thrombocytopenia, a moderately enlarged, non-tender liver, and a greatly enlarged firm spleen. If left untreated, the infection is often fatal, commonly from secondary infections. This infant was infected in the northeast of Brazil with *L. chagasi* (regarded as some to be synonymous with *L. infantum* of the Old World).

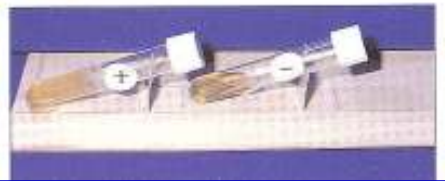


208 Purpura in kala-azar The condition may present as a petechial rash if the thrombocytopenia is severe as in this Indian boy with *L. donovani, sensu stricto*. Petechial haemorrhages of the retina also occur not infrequently.

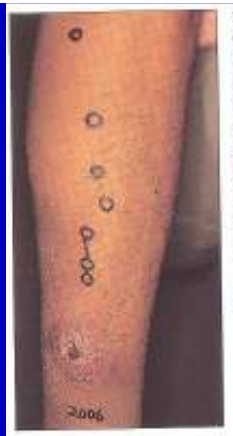
209 Post kala-azar dermal leishmaniasis (PKDL) This syndrome is a sequel to visceral leishmaniasis that may arise several years following successful treatment of the primary *L. donovani* infection, as in this Indian woman. Dermal lesions vary greatly in appearance and may contain amastigotes in large numbers. Some patients with hypopigmented macules may serve as reservoirs for fresh epidemics if conditions occur that favour sandfly breeding and transmission.



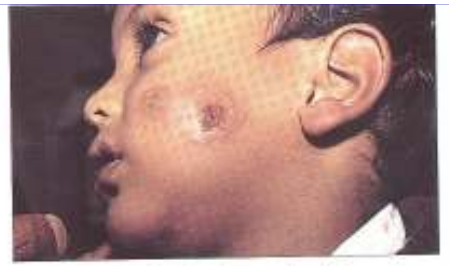
210 PKDL in a Chinese patient The patient was completely cured by chemotherapy. This response differentiates the condition from the anergic, 'diffusa' type of leishmaniasis (see **244**).



211 Formol-gel test Large quantities of IgG are produced by patients with kala-azar and the A/G ratio is reversed. This may be demonstrated by electrophoresis or by a simple test such as the addition to serum of a drop of 30% formalin. The formation of a gel after standing at room temperature for about 20 minutes indicates the presence of a high proportion of globulin in the sample – the formal-gel test. CFT becomes positive later than the FAT (*v. I 250*).



223 Lymphatic spread of *L. major* The ink marks indicate a line of subcutaneous nodules along the lymphatic, passing proximally from the lesion on the lower part of this man's arm. Readily palpated, the lymphatic is sometimes referred to as a 'beaded cord'. The nodules usually resolve without complications when the primary lesion heals with or without specific therapy.



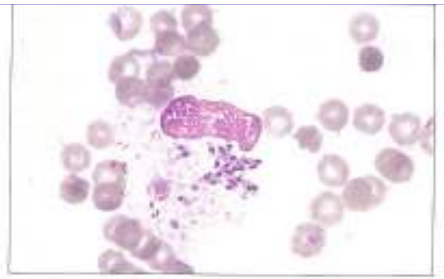
224 Simple 'dry' lesion of *L. tropica* This parasite often produces dry, usually self-healing lesions which, unlike the infection seen here, are generally single. This form is commonly seen in and around towns from North Africa and the Middle East to the former USSR, Afghanistan and western states of India, especially in mountainous areas. The lesions frequently contain very large numbers of parasites.

225 Leishmaniasis recidivans Infection with *L. tropica* may become very chronic, with a hyperallergic reaction leading to lupus-like lesions such as seen in this child in Israel. Amastigotes may be very difficult to find in the lesions.

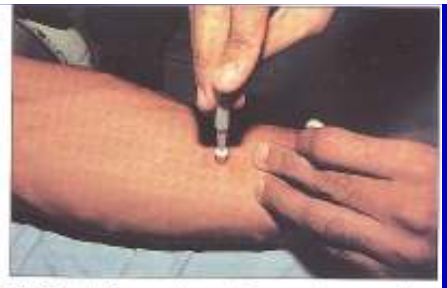
226 Mucocutaneous leishmaniasis in Ethiopia In addition to simple cutaneous lesions due to infection with *L. aethiopica*, other forms are seen in the Ethiopian highlands where rock hyraxes are the reservoirs. It has not yet been ascertained whether the mucocutaneous condition seen here is due to this or another species of *Leishmania* (see also **221**). Mucocutaneous lesions in Sudanese patients are probably associated with *L. major*.

227 Dry lesion on nose of woman in Southern France This lesion proved to be due to a zymodeme of the *L. infantum* complex.

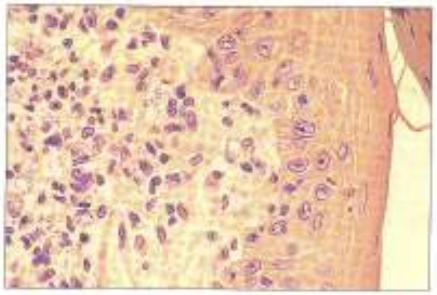
228 Typical healed cutaneous lesion An atrophic, papery, slightly depressed scar results when healing occurs. This man was probably infected with *L. tropica* which healed spontaneously.



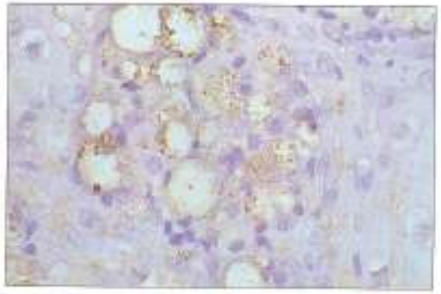
229 Amastigotes in macrophages from skin The diagnosis is confirmed by demonstrating amastigotes in smears made from the cutaneous lesions (see captions for **224** and **225**). Various simple techniques such as biopsy by dental broach or skin-slit and scrape can be used. (Giemsa × 950)



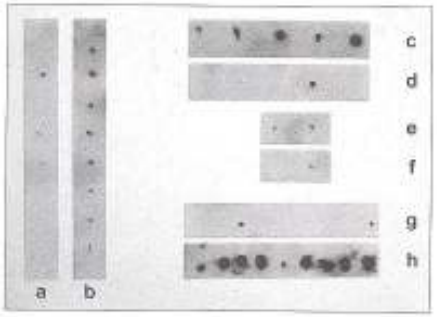
230 Punch biopsy A piece of tissue may be removed under local anaesthetic with a disposable skin punch for histology, culture and the direct demonstration of amastigotes.



231 Parasitised macrophages in skin section Many parasitised macrophages can be seen in this section from an acute lesion caused by *L. major*. (H&E × 220)



232 Immunoperoxidase staining of *Leishmania* This procedure is very useful to demonstrate small numbers of amastigotes in tissue sections in which H&E staining is often unsatisfactory. The section shown here has many brown-staining amastigotes in parasitophorous vacuoles within mauve-staining macrophages. (× 260)



233 DNA probe for the identification of *L. major* in *Ph. papatasi* Highly specific DNA probes have been developed not only for *Leishmania*, but also for sandflies; their combined application can be invaluable in epidemiological studies. Sandflies collected in the field are squashed onto special papers and are then hybridised sequentially with probes against the parasite and suspected vector species. This permits both the numbers of the vectors in the collection and the proportion of those vectors that are infected to be determined in parallel in a short time, thus avoiding the need for laborious and time-consuming dissections and subsequent cultivation and identification of the parasites. The figure shows autoradiographs of *Ph. papatasi*, some of which are infected with *L. major*. (a – positive hybridisation to an *L. major* probe in 3/8 flies found positive on dissection; b – positive responses to a *Ph. papatasi* probe in 8/8 flies; d, e, f, g – positive responses to *L. major*; c, h – positive responses to both probes.)

Συμπέρασμα:

- Ι. Επι του παρόντος τα μέτρα για πρόληψη μετάδοσης της ελονοσίας με μετάγγιση θεωρούνται ικανοποιητικά και η διαδικασία εντοπισμού και διάγνωσης των πλασμωδίων είναι αποδοτική με ερωτηματολόγια και διαγνωστικές εξετάσεις βάσει αξιολόγησης.

- ΙΙ. Για την Αμερικανική τρυπανοσωμίαση εφαρμόζεται επιλεκτικά η λήψη ιστορικού και η αξιολόγηση ερωτηματολογίου κυρίως σε χώρες με μεγάλο πληθυσμό από και προς την Λατινική Αμερική.

- ΙΙΙ. Περισσότερη έρευνα και καταγραφή χρειάζεται προκειμένου να αξιολογηθεί η μετάδοση με μετάγγιση της Babesia και Leishmania στην Ευρώπη και τη Βόρεια Αμερική.

ΕΥΧΑΡΙΣΤΩ