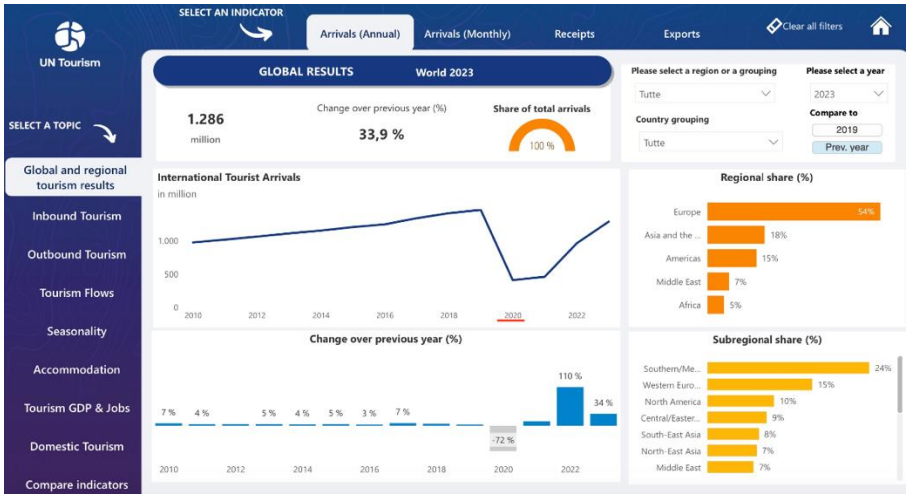
TROPICAL DISEASES

The topics treated are the most relevant ones in terms of morbidity and mortality, including:

1. Malaria
2. Arboviruses
3. Bacterial infections
4. Hemorrhagic fever

In the end, we'll go through a brief introduction to rare diseases.

There is an increasing number of people going abroad for tourism and other reasons, indeed the international tourist arrivals reported in the graphic in *Fig.1* reports that since 2010 people's mobility has increased, with the only exception of 2020-2021 due to COVID-19.



TRAVELER'S HEALTH RISK

The increase in incidence can be caused by:

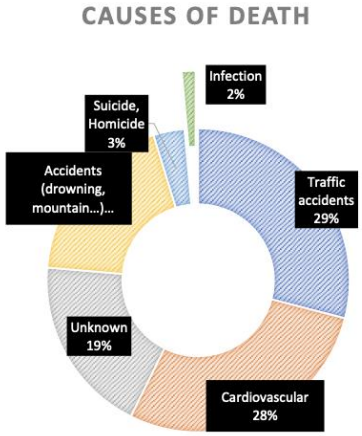
1. Frequent flights: the number of travelers has increased massively in recent years.
2. VRFs (Visiting relatives and friends) are defined as the traveling of people born in tropical countries that then went living somewhere else and went back to their own country for the purpose of VRFs. These people are at higher risk because for two main reasons and the first one is malaria. Malaria does not give a permanent immunity, you can get it a thousand times in your life, but if you get malaria a lot of times before the age of 5, you get a sort of immunity, called premunition, that allows you to have almost no symptoms in case you get it again in your life. However, if then they go away from their origin country and you T cells are not used to react against malaria anymore, you lose the premunition in about 3 years. The second reason is that when they go back to their country, they stay in areas of the city in which the risk for contracting a disease is higher, whereas travelers usually stick to visit touristic/commercial areas.
3. Medical tourism increased in certain countries, such as India for the purpose of plastic surgery and Turkey for hair transplant.

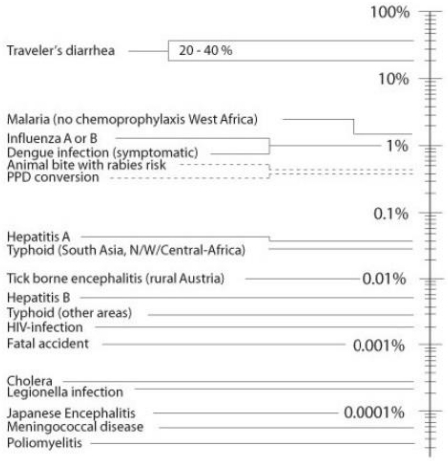
Out of 100.000 travelers to resourceless settings for one month, half of them will develop some health problems and one will die. Calcagno's advice is "try not to be that one".

Bear in mind that infections make up only 2% among the causes of death in travelers (*Fig. 2*). They cause a great number of comorbidities but have low mortality. More common causes of death are traffic accidents, cardiovascular issues, suicides, psychiatric problems and violence.
The most important infection people can have when traveling to tropical countries is traveler's diarrhea (20-40%), it is not so severe because of hospitalization, but people can get dihydrated. Other risks are respiratory viruses, influenza etc (see *Fig. 2* on the right). The message is, when you have a patient that has just came

back from tropics, do not think just about malaria, but also to other diseases.
There are other risks to the traveler:

- Accidental injury





- Environmental hazards
- Animal bites, stings and envenomation. Dangerous animals are jellyfish, snakes and spiders. For example, the most dangerous animal in Australia is a special kind of jellyfish that can induce the death of the people that has been stung.
  If a snake bites someone, you have to follow the "RIGHT" criterion: R=reassure the patient, I= immobilize, GH= go to hospital, T= tell the doctor. It is also useful to take a picture of the snake, if possible, in order to allow the doctors to recognize it and know immediately which treatment is better to use. Indeed, some snakes have a neurotoxic venom that must be removed from the body as soon as possible. Anyway, only a small number of snakes have neurotoxic venom and just a little part of them can inject enough venom to be dangerous.
- Crime and assault
- Psychiatric problems
- Dermatologic disorders
- Mountain sickness (>3000 mt).

RISK STRATIFICATION AND PRE-TRAVEL VISIT

- Reason for travel: it is usually associated to certain risks.
- Duration of travel: certain vaccinations are suggested only for more than one month; expatriates that were born there but then changed their habits by moving to other countries and lose their immunity. It is important also to pay attention to the season, if it's dry, wet.
- Budget: to understand the type of vaccination the traveler has (complete= high budget, incomplete= low budget)

Table 3 Examples of diversity among international travelers

Characteristic	Classification
Reason for travel	• Tourism • Visiting friends and relatives (travelers with migrational background visiting their country of origin) • Missionary/volunteer/researcher/aid worker • Business • Student/education • Military
Duration of travel	• Very short trip (eg, less than a week) • Short travel—usually reflecting normal vacation time (eg, less than a month) • Intermediate travel (eg, less than 3 months) • Long-term travel (eg, 3 months and longer) • Expatriate (permanent residence in a destination country mostly using the infrastructure used by local residents—independent of travel duration, may interrupt stay in destination country for regular visits in the country of origin)
Budget	• Low budget (incomplete vaccinations, no/incomplete/insufficient malaria prophylaxis, no/limited exposure prophylaxis including mosquito protection, back-packer style accommodation, low hygiene standards) • High budget (complete vaccination, continuous malaria prophylaxis, air-conditioned hotels/cars, high hygiene standards)

IMMUNIZATION TO CONSIDER FOR ADULT TRAVELERS

Usually, doctors suggest being vaccinated for Diphtheria/Tetanus/Pertussis Measles/Mumps/Rubella/Polio but also for:

- Hepatitis A which is usually suggest for most of travelers
- Hepatitis B for all the non immune patients
- Typhoid fever, the vaccine is not that effective, it covers between 50-70% of strains. There are two vaccines, an intramuscular one and an oral one.
- Yellow fever, it is a live vaccine, nasty, that gives a lot of fever, and it is suggest just for selected countries
- Cholera, usually done if there is an epidemic in the country. The vaccine we have now works nicely, covers also for a certain percentage of other pathogens that cause traveler's diarrhea such as E. Coli.
- Japanese Encephalitis, suggested for people that stay for more than one month in South-East Asia, South of China and Japan and that stay outdoor or if they work with animals. This disease affects ticks, mosquitoes, animals and humans.

Vaccine	Route	Doses	Schedule	Notes
Hepatitis A	i.m.	2 doses (at least 1)	6 months apart	Most travellers
Hepatitis B	i.m.	3 doses	0, 1, 6 months	All non immune
Typhoid fever	i.m.	1 dose (2 weeks before)	Every 2 years	Most travellers
	oral	3-4 doses (10 days before)	Every 5 years	
Yellow fever	i.m.	1 dose (10 days before)	no	Selected countries – live vaccine!
Cholera	Oral (i.m.)	2-3 doses (1 week before)	Every 2 years	Selected countries

Vaccine	Route	Schedule	Notes
Pneumococcus	i.m.	Usually 1 dose	Comorbidities or age >65 years
Influenza	i.m.	Every year	Comorbidities or age >65 years
Rabies	i.m.	0,7,21 days Repeated if low immunity	High-risk countries and outdoor/animal contacts
Japanese Encephalitis	i.m.	0 and 28 days	SEA. Long-term, outdoors, veterinarians.
Meningococcus	ACYW B	2-3 doses	Meningitis belt countries
Varicella	i.m.	0 and 28 days	No exposure to chickenpox (VZV)

Fig. SEQ

The graph in Fig. 5 shows the international tourists' arrival. It can be seen how many tourists travel from rich resource countries to low resource countries every year, which is approximately 1 billion. According to some studies, it can be concluded that among the diseases that are not present in Europe, 92% are linked to tourism and 8% to migration.

The second important point in tropical disease, apart from the increase of travels to endemic countries, is the

*presence of **vectors**.*

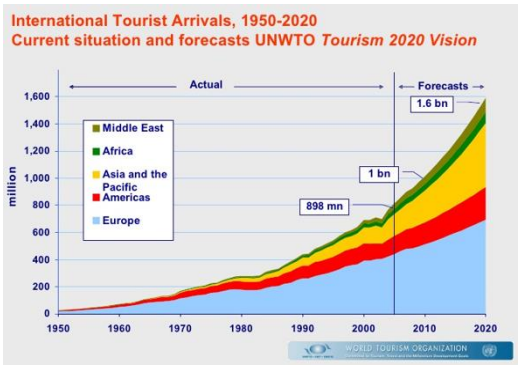


Fig.

Indeed, tropical diseases spread mainly through vectors. Vector changes can affect the spread of such diseases, for example Fig. 5 shows how changes in the presence of Aedes Albopictus in Europe, accompanied with climate changes such as temperature and humidity, has affected the spread of Chikungunya (this lead to small outbreaks of Chikungunya in Italy, a few years ago).

*The third important point is that these **diseases travel with humans**. Change of location due to traveling will also change the clinical presentation according to genetic background and climate changes.*
For example, the Zika virus was discovered in Uganda. Then it moved to the East resulting in outbreaks in the Pacific Islands. Afterward, the virus reached South America which led to the discovery of the association of Zika with CNS conditions such as microcephaly and some other brain abnormalities in children born from an infected mother.
The other discovery was that this virus is associated with Guillain-Barré syndrome.

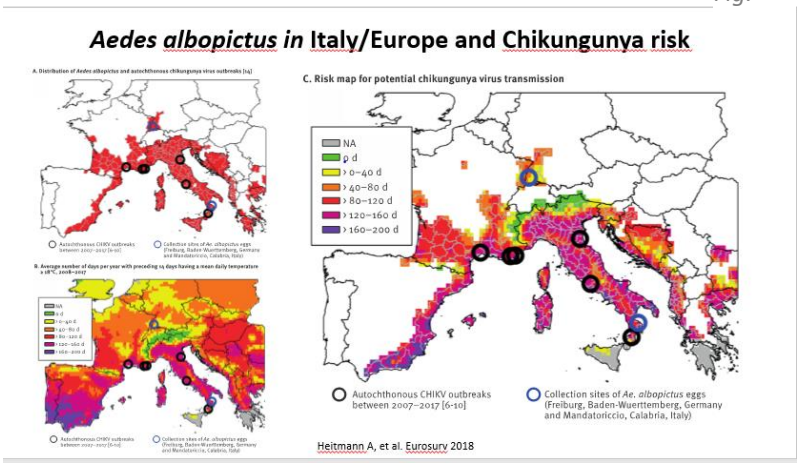


Fig.

It's not known exactly why this difference in clinical presentation occurs, but it's related to genetic differences between populations. In addition, last month in France a case of Zika virus was reported in which the affected man hadn't been traveling outside Europe which suggests the presence of the virus in Europe.

Another example is the Crimean Congo Hemorrhagic fever, a severe disease, mainly transmitted by tick bites and droplets. In 2016, a Spanish man was reported to be affected: he presented fever, altered mental status then shock followed by death. The nurse that was taking care of him also got infected and survived.
Furthermore, 5 years before this reported case, scientists discovered the presence of ticks vectors and infected birds in West Spain. The virus can also be transmitted by birds, which migrate from the Middle East to Spain, carrying the virus and transmitting it.

*Another aspect is the possibility of developing **epidemics**. Every tropical country is at risk of an epidemic. For instance, the outbreak of plague in Madagascar in 2017; the plague is mainly spread by infected fleas and presents firstly in a bubonic form, which can, however, evolve into a pneumonic one. The change in the plague form indicates a human to human transmission of the disease. Therefore, in the second part of the epidemic, there was an increase in pneumonic plague cases, due to transmission between humans. This epidemic was controlled by isolation.*

The Ebola epidemic in Congo was much harder to control. This epidemic caused more than 3000 cases with around 2000 deaths. The number of cases was much lower than those in West Africa but it was much harder to control due to the highly populated and smaller area as well as political aspects (war episodes nearby). This epidemic was finally managed thanks to the large efforts made, such as isolation and widespread use of vaccination.

FEVER IN RETURNING TRAVELERS

Fever is the most frequent condition reported in travelers coming back from tropical areas. It's very important to investigate and rule out potentially deadly contagious diseases and treat curable illnesses.

This indicates the importance of past and recent medical history, that helps in:

1. **Determining the source** of infections, exposure and incubation period. For example, malaria has an incubation time that goes from 6 days to 6 months maximum, so you need to understand when the symptoms started, the duration of the fever, if this person was exposed to certain sources of infection such as fresh water.
2. **Signs and symptoms** (such as fever duration)
3. **Vaccination and prophylaxis** (ask about compliance!)
4. **Ask also if some other people in the group had the same symptoms**

In the last 15 years in Turin we had only 2 suspected cases of hemorrhagic fever, one due to the epidemic in West Africa and the other one was a very strange story. Some researchers went to Nepal to collect ticks and one of them came back with very very high levels of AST, ALT and fever; two days after he started bleeding from the GUT.

For example, if fever begins after more than 21 days from returning, it's unlikely that the patient got infected with dengue, rickettsial infections, viral hemorrhagic fevers such as yellow fever and Lassa fever.

DIAGNOSTIC FLOW

It is the usual investigation for travelers.

1. Thick and thin smear (useful to rule out malaria because it's a deadly disease)
2. Complete blood count
3. Liver function test
4. Urine analysis
5. Hemoculture, stool (it can tell something about parasites and Salmonella) and urine cultures (rule out UTI, Traveler's diarrhea...)
6. Chest X-ray to exclude upper and lower respiratory tract infections
7. Serologies (Dengue, Rickettsia, Schistosoma, Leptospira, and acute HIV infection)

INCUBATION PERIOD

Different diseases have different incubation periods, and this helps physicians to rule out certain conditions.

- Incubation period can be **less than 2 weeks** and present with fever associated with (*Fig. 7*):

1. Fever with no other signs
2. Hemorrhagic symptoms
3. CNS symptoms
4. Respiratory symptoms (take into account influenza and respiratory viruses!)

- Incubation period **between 2 and 6 weeks**
- **Longer than 6 weeks**

Fever +	Aetiologies
...	Malaria, Dengue, Rickettsiosis, Typhoid Fever, HIV
Haemorragic symptoms	Meningococcal sepsis, Malaria, Yellow fever, Ebola
CNS symptoms	Malaria, Typhoid Fever, Rickettsiosi, Rabies, Japanese Encephalitis, African Tuypanosomiasis
Respiratory sympotms	Influenza, Legionellosis, SARS

Fig. SEQ

14 days to 6 weeks	Malaria Typhoid Fever HAV and HEV Schistosomiasis (Katayama's fever)
>6 weeks	Malaria Tuberculosis HBV Visceral Leishmaniosis Filariasis Schistosomiasis Amoebic abscesses (liver)

Fig. SEQ

MALARIA

Malaria's highest incidence is in Sub-Saharan Africa. Other tropical countries affected by malaria are South and Central America and Southeast Asia (Indonesia and Papua New Guinea, in which resistance is common). Malaria is very common in terms of morbidity. On the good side, there is a decreasing trend in mortality of malaria, thanks to various prevention methods that have been employed in recent years.

Table 3.1.
Global estimated malaria cases and deaths, 2000–2022[a] *Source: WHO estimates.*

Year	Number of cases (000)				Number of deaths		
	Point	Lower bound	Upper bound	% *P. vivax*	Point	Lower bound	Upper bound
2000	**243 000**	227 000	263 000	8.3%	**864 000**	835 000	905 000
2001	**248 000**	230 000	271 000	8.3%	**873 000**	841 000	918 000
2002	**245 000**	227 000	267 000	7.7%	**841 000**	811 000	885 000
2003	**249 000**	232 000	271 000	8.0%	**813 000**	783 000	856 000
2004	**250 000**	232 000	277 000	7.9%	**808 000**	774 000	866 000
2005	**249 000**	232 000	273 000	8.0%	**770 000**	738 000	819 000
2006	**244 000**	226 000	268 000	6.9%	**776 000**	745 000	826 000
2007	**240 000**	223 000	262 000	6.6%	**754 000**	723 000	796 000
2008	**239 000**	223 000	259 000	6.4%	**716 000**	686 000	757 000
2009	**245 000**	227 000	267 000	6.4%	**726 000**	692 000	775 000
2010	**247 000**	229 000	272 000	6.6%	**703 000**	668 000	755 000
2011	**241 000**	225 000	263 000	7.0%	**665 000**	633 000	707 000
2012	**237 000**	221 000	257 000	7.0%	**619 000**	590 000	660 000
2013	**232 000**	215 000	251 000	6.0%	**591 000**	560 000	633 000
2014	**230 000**	209 000	253 000	5.5%	**588 000**	551 000	643 000
2015	**231 000**	211 000	254 000	4.9%	**586 000**	548 000	645 000
2016	**232 000**	214 000	253 000	4.6%	**582 000**	546 000	645 000
2017	**237 000**	219 000	258 000	3.7%	**580 000**	545 000	644 000
2018	**232 000**	215 000	253 000	3.0%	**581 000**	545 000	656 000
2019	**233 000**	213 000	255 000	2.7%	**576 000**	537 000	660 000
2020	**244 000**	221 000	271 000	1.9%	**631 000**	587 000	747 000
2021	**244 000**	220 000	272 000	2.1%	**610 000**	568 000	726 000
2022	**249 000**	225 000	278 000	2.8%	**608 000**	566 000	738 000

Fig.

Every year the WHO presents data (*Fig. 9*) about how many people presumably had malaria, it is an estimate, and also how many people died for it.
About 250 000 people every year experience malaria, and if compared to the year 2000, the number has not changed so much.
On the contrary we have reduction in mortality, from 860 000 in 2000 and 608 000 in 2022.

Looking at the countries in which we have malaria (*Fig. 10*), all the red ones are in Africa, but we also have cases in Pacific regions, America, Mediterranean regions. Talking about Africa, the incidence is so high because Plasmodium Falciparum is mostly based there.
The prevention efforts for malaria actually work. Indeed, between 2000 and 2010, malaria incidence rates and mortality fell to 50%.

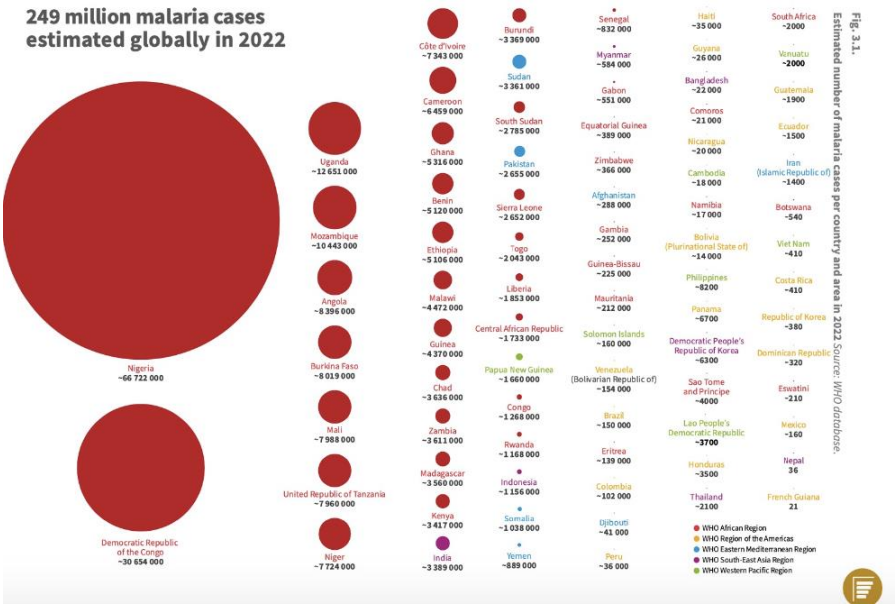
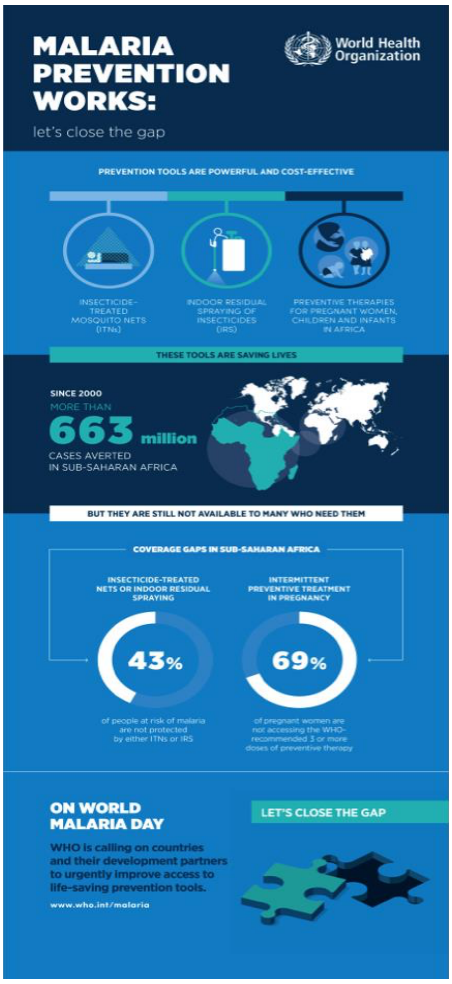


Fig. 3.1. Estimated number of malaria cases per country and area in 2022 *Source: WHO database.*

These prevention methods include **indoor spraying** to help in reducing the number of mosquitoes bites; the effect can be enhanced by spraying just before the wet weather season. Another prevention tool is **impregnated bed nets** containing a toxin that kills mosquitoes. These bed nets have a life span and they need to be replaced because they get damaged and the effect of the toxin (pyrethroids) is reduced over time. However, these nets are still effective in reducing the number of mosquito bites per person, which is also due to the fact that often mosquitoes bite at a specific time of the night (dusk and dawn).

The third prevention tool is the use of **artemisinin-based combination therapy**, a drug combination given orally. One of the drugs is artesunate, a quick and effective drug acting against both parasites and gametocytes, the sexual form of malaria plasmodia. Since gametocytes are the ones picked up by the mosquito, their death reduces the transmission of the disease.

These preventive methods require lots of effort and money, but they are important as they actually do prevent malaria, their impact is huge.

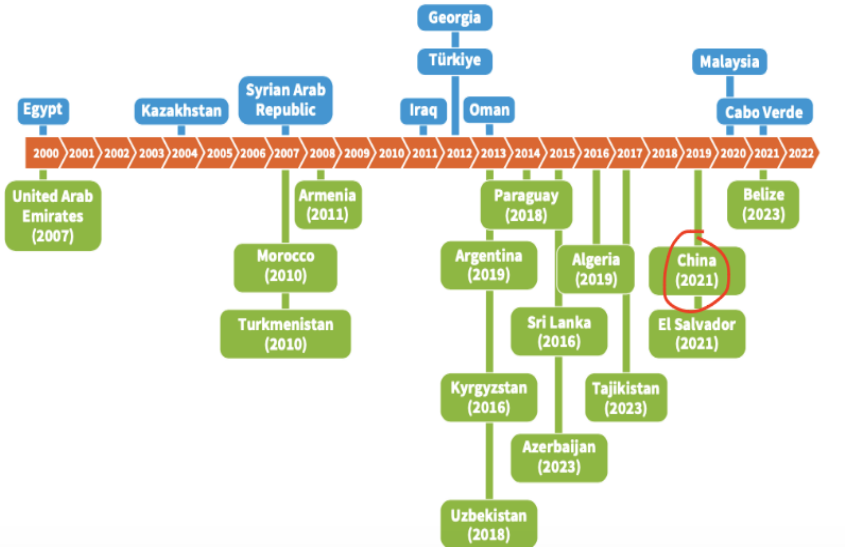


EPIDEMIOLOGY

Malaria is a disease of tropical countries.
Malaria's most severe form is caused by *Plasmodium falciparum*.
The highest incidence is found in Africa, some cases in South America and Asia.

Outside these areas, other Plasmodium types are found, such as *Plasmodium vivax*, *Plasmodium malariae* and *Plasmodium ovale*, which cause a less severe form of the disease and with lower mortality.

These (*Fig. 12*) are the countries which are certified malaria-free, and we can notice that in 2021 also China has been certified malaria-free.



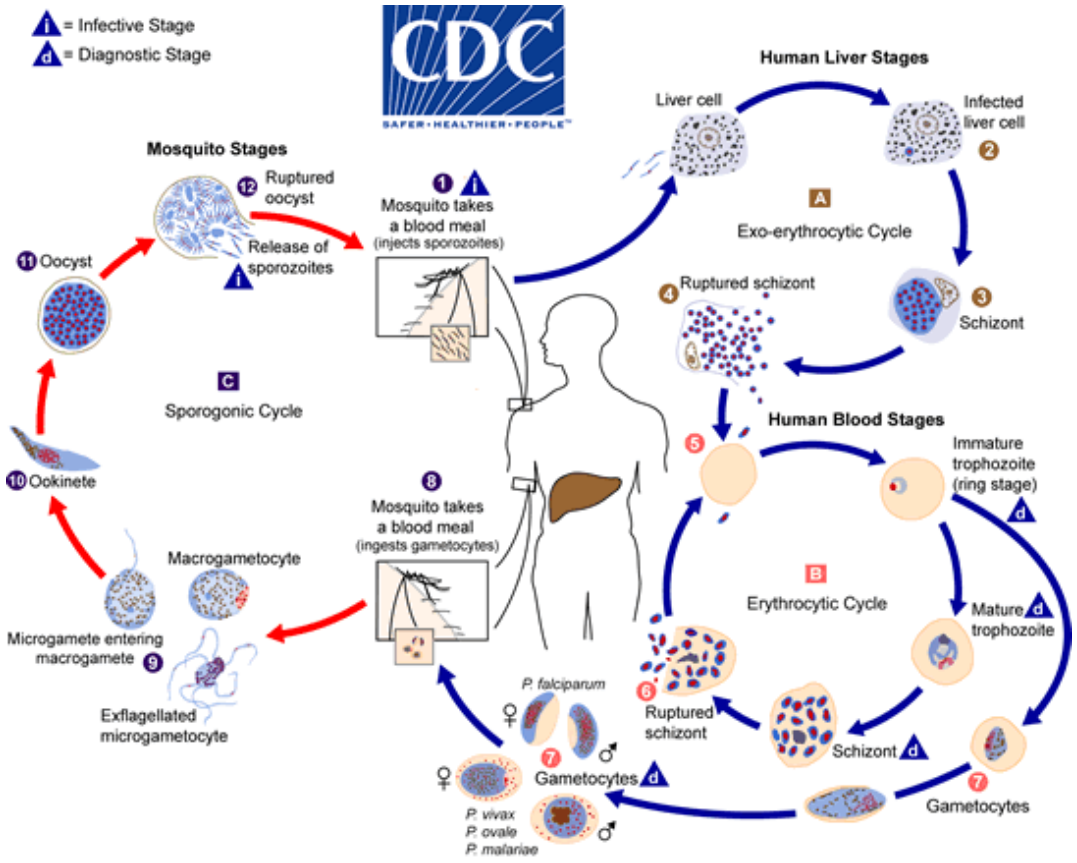
PLASMODIUM, VECTOR, HOST

Malaria transmission requires 3 elements: a Plasmodium, a vector, and a host. Each of these might change over time affecting the transmission of the disease.

- **Plasmodium**:
  Plasmodia are protozoa and there are five known species that might affect humans, which can be morphologically distinguished with thin and thick blood smears (discussed later in the lecture)

  1. *Plasmodium falciparum*, that causes severe malaria.
  2. *Plasmodium vivax*
  3. *Plasmodium malariae*, less relevant from a clinical point of view
  4. *Plasmodium ovale*, possibility of having hepatic forms that can stay in the liver forever causing relapses over time, every 6 to 12 months.
  5. *Plasmodium knowlesi*, which can also cause severe malaria, it was thought to be just for the chimpanzee, but now we know that it can affect humans too. In the last 3 years we saw an increase in Asia, Malesia and Panama and probably we'll see more cases in the next future.

The **plasmodium life cycle** is pretty complex. Malaria infection starts when the mosquito bites a human injecting the sporozoites in the bloodstream. Those sporozoites migrate into the liver where they replicate and infect liver cells and develop into schizont which, after 7 days, when they are all synchronized at the same stage of maturity, rupture the hepatocytes and reach the bloodstream infecting red blood cells.
P. falciparum can infect RBCs in all their forms, while *P. vivax* and *P. ovale* can infect only erythroblasts causing lower parasitemia.
What can be seen in the blood of patients infected with malaria is the presence of trophozoites (ring stage: ring near the cell border) after RBCs infection. Here the symptoms start, when trophozoites break RBCs causing fever, anemia etc. Trophozoites can develop into mature trophozoite, and this cycle is repeated as the mature trophozoite can form schizont.
In certain cases, schizonts migrate to the liver and result in liver stages again.
P. vivax and *P.ovale* have liver stages that can persist up to months and years.
In addition, the mature trophozoite can form gametocytes (sexual forms of plasmodium). During the mosquito blood meal, when the mosquito bites an infected person, it ingests the gametocytes, resulting in sporogonic cycle inside the mosquito, mostly in salivary glands.
The cycle is repeated when the mosquito bites another human.



- **Vector**
  Mainly **Anopheles** mosquitoes, existent in 20 different species that usually bite from dawn to dusk. These mosquitoes are anthropophilic and zoophilic, meaning that they can bite humans but also other animals. Those mosquitoes are endophilic, they prefer to inhabit and rest indoors (they usually prefer rural environments). Transmission is influenced by several factors including the longevity of the vector, temperature, humidity and altitude, indeed they do not fly above 1100 mt.
  Anopheles stephensi is an effective vector of P. Falciparum and P. vivax, we recently known that it is relevant for humans and it comes from Asia and Arabian Peninsula. It is able to resist very high temperatures and has also high resistance to insecticides.

- **Host**
  Host resistance is related to genetics, as certain genotypes can favor resistance to malaria. For example, having sickle cell anemia, thalassemia or G6PD deficiency, which are basically alterations in hemoglobin and other enzymes within RBCs, is protective against malaria as this condition affects RBCs. Newborns are protected against malaria up to 6 months of age due to the presence of fetal hemoglobin, maternal antibodies, breastmilk, and lack of primed T cells (for this reasons malaria in newborns is absent/not severe).

IMMUNITY

!!!<u>DO NOT ASK FOR SEROLOGY</u> FOR MALARIA AND TUBERCOLOSIS!!! The only case in which we can ask for serology in case of malaria is when people want to donate blood and they are just came back from an endemic country.
Immunity is not based on antibodies, but on T cell priming by plasmodia antigens.

1. **Clinical immunity**: no or mild symptoms despite circulating parasites (parasitemia). It develops after several infections and within the first years of life in high endemic areas. Patients can come to clinics

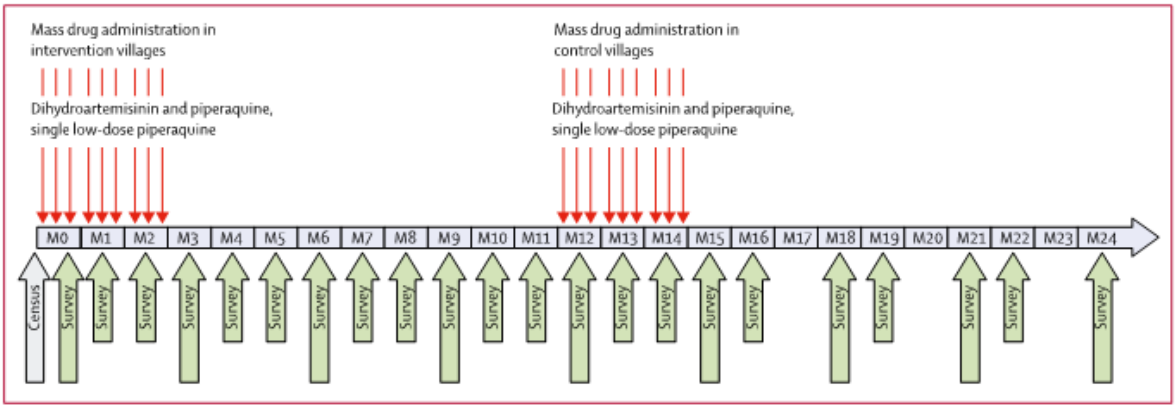
only with headaches and they turn out to be actually infected with malaria.

2. **Anti-parasite immunity** develops later in life (more than 10 years of age) and requires a high parasite rate. Typical of adults in highly endemic areas having low parasitemia and no symptoms.

These two types of immunity might be lost if the patient migrates and moves to an area with low endemicity. If the patient comes back to his original area he may develop a severe form of malaria.

Fig.14 is taken from a study published on The Lancet, about whether massive drug administration was effective or not. In this study, people from 4 villages in Vietnam participated, then ultrasensitive PCR was performed in all subjects in order to understand whether there has been a low level of replication during the massive drug administration. The uPCR results indicated lots of low levels of plasmodium replication, some of *P. falciparum*, others of *P. vivax* , and in 25% there were co-infections with both *P. falciparum* and *P. vivax*. Specifically, a persistent low level of replication with some peaks was observed.
These peaks were not associated with symptoms and what could be concluded is that persistent largely asymptomatic plasmodium infections are common in some areas, which proves the presence of clinical and anti-parasite immunity in such places.

In addition, it's important to tackle malaria infections, as it was thought that Vietnam had low seasonal malaria transmission although some subjects have a persistent malaria infection for a duration of almost one year.



TRANSMISSION

Malaria has two transmission patterns:

1. ***stable transmission pattern** is found in high endemic areas where the transmission is stable during the year. Children and pregnant women are at a higher risk. This transmission pattern is harder to control as it requires to control many hosts all year round.*
2. ***unstable transmission pattern** is found in low endemic countries. Malaria infection can occur at every age. This pattern is a seasonal transmission found in South East Asia, in which there is low malaria immunity and a potential epidemics may develop because there is no widespread protection against malaria. This transmission can be controlled, as happened in Italy before malaria was eradicated in the XX century, it was a summer transmission malaria rather than a stable one.*

INCUBATION TIME

*It ranges from **7 to 30 days** and varies among species, shorter for P. falciparum and longer for P. malariae. Prophylaxis can delay the onset of symptoms. In case of P. vivax and P. ovale infections, symptoms may appear after months especially with prophylaxis, due to hypozonite forms of these species. This means that prophylaxis might be able to tackle the first infection, but it may appear months later.*
*The individual is considered to be at risk of developing malaria up to **one year** after travelling to an endemic country, hence in order to rule out malaria the patient must be tested for it.*

CLINICAL PRESENTATION

- **Non-complicated malaria**
  The classic malaria presentation is rarely observed.
  The malaria attack lasts 6-10 hours and presents initially as cold and chills, subsequently with high fever, headache, vomiting, massive sweating with defervescence and deep asthenia.
  The malaria attack occurs every two days for *P.falciparum, P.ovale* and *P.vivax* (tertian fever) and every 3 days for *P.malariae* (quartan fever). Both are rarely observed nowadays, and the fever timing is due to the synchronized breakdown of the schizonts with merozoites and trophozoites release into the bloodstream, followed by RBCs attack.
  Usually, malaria clinical presentation is a combination of fever with chills, sweating, headache, general malaise, nausea, vomiting and widespread pain (much less classic). Anyway, do not expect to see always fever because people take drugs to lower it down.

  Signs and symptoms
  1. High fever
  2. Sweating
  3. Weakness
  4. Splenomegaly can be a chronic condition referred to as tropical splenomegaly that develops after several malaria infections.

  Fig.15 shows an 11-year old child with massive splenomegaly (more than 25 cm, it gets to the iliac spine) in Burundi.
  This causes chronic anemia and thrombocytopenia, the spleen is extremely big making it able to destroy lots of circulating RBCs and platelets

  5. Hepatomegaly
  6. Jaundice, if there is a massive destruction of RBCs
  7. Tachypnea, in case of acute respiratory distress syndrome, a compilation of malaria.

  To sum, the presentation of malaria includes headache (present in 95% of cases), with fever and chills.

  Lab tests will show anemia, thrombocytopenia, elevated CRP and ESR with normal WBCs (this is how we differentiate with Dengue that usually shows leukopenia). Liver function test will show increased AST, and creatinine as well. There won't be lymphopenia, that is an important distinction between malaria and Dengue.

- **Severe malaria**
  Severe malaria is due to ***P. falciparum*** infection in 99% of cases, and to *P.vivax and P. knowlesi* in 1% of them. In high endemic areas it affects children below the age of 5, while in low endemic ones it mostly affects adults.

  Criteria for severe malaria:
  1. Cerebral malaria
  2. Severe anemia
  3. Haemoglobinuria
  4. Acute respiratory distress syndrome ARDS
  5. Spontaneous bleeding and/or DIC
  6. Shock
  7. Acute tubular necrosis (microvascular obstruction)
  8. Hyperparasitemia (>10% of infected RBCs)

9. Metabolic acidosis
10. Hypoglycemia (can be due to malaria itself or to drugs such as quinine), especially in children because Plasmodia use sugars leading to coma

In severe malaria, the therapy moves from oral to **intravenous** administration.

CEREBRAL MALARIA

It's not common and usually affects children. Cerebral malaria is defined as an **altered state of consciousness** with very common seizures and coma in a patient with **parasitemia** and no other cause of brain involvement (including meningitis, sepsis, hypoglycemia, rabies, hypertensive encephalitis, trauma, etc). Cerebral malaria has a high mortality rate, around 20-50%, and leaves neurological sequelae which are very common in children (more than 40% of cases) and quite rare in adults (<10% in adults).

Cerebral malaria is judged according to Glasgow common scale or the Blantyre coma scale.

Glasgow Coma Scale

Response	Score
Eye opening	
Opens eyes spontaneously	4
Opens eyes in response to speech	3
Open eyes in response to painful stimulation (eg, endotracheal suctioning)	2
Does not open eyes in response to any stimulation	1
Motor response	
Follows commands	6
Makes localized movement in response to painful stimulation	5
Makes nonpurposeful movement in response to noxious stimulation	4
Flexes upper extremities/extends lower extremities in response to pain	3
Extends all extremities in response to pain	2
Makes no response to noxious stimuli	1
Verbal response	
Is oriented to person, place, and time	5
Converses, may be confused	4
Replies with inappropriate words	3
Makes incomprehensible sounds	2
Makes no response	1

BLANTYRE COMA SCALE

A) Best motor response	Score
• Localizes painful stimulus	2
• Withdraws limb from pain	1
• Non-specific or absent response	0
B) Verbal response	
• Appropriate cry	2
• Moan or inappropriate cry	1
• None	0
C) Eye movement	
• Directed (e.g., follows mother's face)	1
• Not directed	0

*Fig. SEQ Fig. * ARABIC 16*

The **pathogenesis** of cerebral malaria is not completely clear. It's not encephalitis, nor a parasitic infection of the brain, we won't find an infection in the brain, there are no plasmodia there, but rather it is a vascular problem caused by several mechanisms such as obstruction of microvessels in the brain and plasma leakage outside the vessels. Basically, there is a combination of altered blood flow with increased vascular leakage causing brain damage, it is a kind of diffused cerebral ischemia because RBCs with plasmodia inside obstruct the local circulation. The resulting damage is partially reversible which is why neurological sequelae are higher in children.
There is nothing we can do; the only thing is to treat it as soon as possible in order to limit the damage.

Patients affected with cerebral malaria is seen in *Fig. 17*, as shown he is suffering from neurological complications and coma and by looking at the patient's conjunctiva the presence of severe anemia can be concluded without running lab tests.

PATHOPHYSIOLOGY

Pathophysiology consists of destruction of RBCs thus developing anemia.
This causes the parasite and RBC products to be released into the bloodstream triggering the host reaction, meaning that immunity has a big role and may lead to complications. Indeed, patients having a kind of degree of immunity don't overreact and thus don't develop severe malaria, hence the risk is higher for children and non-immune hosts *(travellers ed.).*
Being filled with *P. falciparum,* RBCs are deformed, tend to stick to each other forming a peculiar structure named *rosette* (aggregation of RBCs and platelets), which may occlude some vessels. This is another mechanism that causes microvessel damage, thus interfering with blood flow and also result in metabolic interference (like in the liver).

Another complication of malaria associated with ***P. falciparum*** infection is **blackwater fever**.
It's an **intravascular hemolysis** with immune/allergic pathogenesis.
Patients who are already on treatment may present sudden onset of fever (>40 degrees), chills, lumbar pain, jaundice, oliguria and anuria, and **dark urine** (coke color).
Lab tests will show **hemoglobinuria** due to tubular and persistent renal dysfunction.
In addition, blackwater fever is facilitated by quinine, so in this case it's suggested to switch to another treatment.

Fig.

Nephrosyc syndrome is another rare but severe complication of malaria with late diagnosis. It's associated more with both ***P.malariae*** **and** ***P.falciparum*** than the other Plasmodia. This renal complication is characterized by high loss of proteins in urine and edema in the face. It's even more severe in malnourished children who already have low protein concentration in their blood.

MALARIA DURING PREGNANCY

Mostly P. falciparum → severe forms in mothers (severe anemia) and causing negative pregnancy outcomes (premature delivery, abortion, low birth weight and neonatal death)

Malaria during pregnancy is a special case that can occur in two settings:

- **high transmission setting** with partial immunity, usually mothers are asymptomatic, with/without parasitemia and with anemia. Parasite presence in the placenta leads to low birth weight. It's higher in first pregnancies, except for *P.vivax* that has higher risk in successive pregnancies.
- **low transmission setting**, anemia is very frequent, increased risk of severe malaria and may lead to spontaneous abortion, stillbirth and other negative pregnancy outcomes like prematurity and low birth weight.

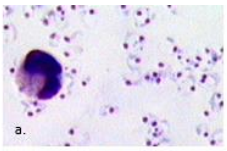
The WHO recommends a package of interventions for prevention and treatment of malaria during pregnancy, consisting of:

1. the use of long-lasting insecticidal nets while sleeping (**LLINs**)
2. in all areas with moderate to high malaria transmission in Africa, intermittent preventive treatment in pregnancy with sulfadoxine pyrimethamine (**SP),** this prophylaxis is suggested every trimester.
3. prompt **diagnosis** of malaria and effective **treatment** of potential episodes.

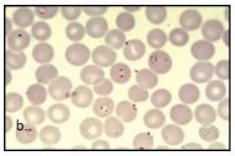
DIAGNOSIS

1. Think of malaria in patients who have recently travelled to endemic countries and presenting with seizures, fever and headache (*7 days to 6 months P.vivax*)
2. Examination (hepatomegaly, splenomegaly, jaundice, paleness).
3. **Thick smear** detects the presence of the plasmodium and provides a quantitative assessment, i.e. plasmodia number in comparison to RBCs (*giemsa stain)*.
4. **Thin smear** is more accurate in number calculation and to differentiate between species of plasmodium *(P.Falciparum, P.ovale, P.vivax*). The differentiation of species is not easy for trophozoites, it's easier in the presence of schizonts and gametocytes instead (*May-Grunvald or Giemsa*).
5. Antigen rapid test with chromatographic methods, it is useful, quick and cheap.
6. PCR, for certain settings and very rich countries, very sensitive.
7. Lab test (haemocrome, LDH, AST, ALT, BUN, creatinine, glycemia, haemogas analysis with lactate).

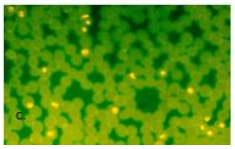
a. Thick smear



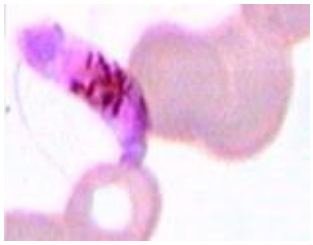
b. Thin smear



c. Fluoroscent dye



Thick blood smear: one drop of capillary blood is placed on a glass slide, using the corner of another slide spread it and stain with Giemsa. The thick smear shows granulocytes and ring-shaped trophozoites.



A **thin blood smear** is a drop of blood that is spread across a large area of the slide, it allows to see the RBCs with the trophozoite inside. Thin blood smear helps to count precisely and differentiate the species.

Another technique is using **fluorescent dye**, more expensive and hence less employed.

Rapid Diagnostic tests RDTs, referred to as **cassettes**, are lateral flow immunochromatographic antigen detection tests. There is one common antigen shared by all four species and one falciparum-specific antigen, they can even be dosed in combination to detect the species.
It is cheap, quick and sensitive, but it stays positive for a few weeks, so if a patient gets reinfected with malaria, you're not able to differentiate. Moreover, now we have a mutation in the genome of the plasmodium and so this test gives a lot of false negative results (20%) which do not recognizes this mutation.

TREATMENT

There are different drugs with different effects and specific for different stages.

- **Artemisinin and derivatives**
  This group of drugs derive from a plant known as *Artemisia annua*. It has been used in China for

hundreds of years.
These drugs include artemisinin, artemether, artesunate and dihydroartemisinin (effective moiety). Some of these are administered orally, other intramuscularly and other IV. All of these drugs can have rectal administration, which is used especially in children before getting to the hospital as an attempt to reduce the time to the first dose of antimalarial drug in high endemic countries.
These drugs are very quick in killing schizonts and trophozoites resulting in rapid reduction in the parasitic load, the estimated reduction is about 10.000 parasites/48 hours thus reducing the mortality in severe malaria.
These drugs are also able to kill gametocytes reducing the number of plasmodia and inhibiting the establishment of a second cycle in mosquitoes preventing them from infecting other subjects.
The duration of this treatment is **7 days** if used alone or **3 days** if given with long half-life compounds (such as lumefantrine, amodiaquine, piperaquine, mefloquine, pyronaridine).
The combination of these drugs (artemisinin and derivatives) with a long half-life compound is known as Artemisinin based Combination Therapy (**ACT**), which is the standard therapy for non-severe malaria everywhere in the world. It may change according to local availability, resistance patterns of companion drugs etc., yet still remains the gold standard treatment for malaria.

Side effects: generally, these drugs are well tolerated, but GI intolerance, QT prolongation, type 1 hypersensitivity reactions may happen, and there are no safety studies in the first trimester of pregnancy, hence others are employed.

AQUAMAT study: children with severe malaria were randomized and received artesunate vs quinine. A decrease in mortality in artesunate was shown with the administration of artesunate. A meta-analysis of RCTs confirmed this, hence this study changed the WHO policy regarding which drug should be used, and if WHO guidelines changes, everywhere in the world should be able to provide ACTs.

- **Quinine**
  Not used much nowadays, because it was mainly replaced by ACTs (which showed reduced mortality rate).
  Quinine is still used in certain conditions such as the first trimester of pregnancy. It must be administered with slow infusion to avoid risk of developing hypotension and arrhythmias.
  It has many side effects including tinnitus, nausea and hypoglycemia.

- **Chloroquine**
  Used to treat non-*P.falciparum* uncomplicated malaria, it has some side effects which are usually well tolerated.

- **Amodiaquine:**
  Similar to chloroquine. Effective on chloroquine resistant strains of *P. falciparum*, used in association with artesunate derivatives ACTs.

- **Sulfadoxine/ Pyrimethamine**

- **Mefloquine:**
  Used mainly for **prophylaxis**. It has a long half life of 21 days thus some drugs cannot be administered after mefloquine administration.
  It has side effects such as QT prolongation, as well as sleep disturbances (nightmares and vivid dreams), psychoses and depression (in almost 10%). Whoever experiences mefloquine side effects will not take it again.

- **Primaquine:**
  It is important because it seems to be the only drug effective against **hypnozoites**. Therefore, in case of *P. ovale* and *P. vivax* infections, after treating the acute episode, primaquine must be administered to reduce the risk of relapses overtime.
  It's also effective against gametocytes (*P.falciparum*).

As it regards the **prophylaxis**, we only have 2 drugs, Malarone (atovaquone and proguanil hydrochloride) which is taken every day, expensive and can give GI disturbances and Mefloquine, taken once a week, generally well tolerated by 90% of people, but for the remnant 10% severe side effects such as psychiatric symptoms (depression, anxiety, psychosis).

CRITERIA FOR CHOOSING ANTIMALARIAL TREATMENT

1. Plasmodium specie
2. Geographic area (including resistance, rate of mixed infections P.ovale P.vivax etc)
3. Severity of clinical presentation – severe malaria is usually treated with intravenous drugs or if you do not have them intramuscular drugs, non-severe malaria is treated with oral drugs (combination of Artemisine and a second drug).
4. Previous prophylaxis/treatment
5. Comorbidities and concomitant conditions (long QT, unable to swallow, etc)
6. Local availability of drugs that may change in different scenario

TREATMENT OF NON-COMPLICATED MALARIA:

1. *P. falciparum: ACTs orally, in case of severe vomiting or risk of complicated malaria, drugs can be given Intramuscular (IM) or Intravenously (IV).*
2. *P.malariae or P. knowlesi: chloroquine.*
3. *P.vivax or P.ovale: chloroquine + Primaquine.*

In countries with widespread chloroquine resistance, ACTs is indicated (such as Indonesia, Peru, Oceania)

TREATMENT OF SEVERE OR COMPLICATED MALARIA

Start with IV or IM artesunate or artemether for at least 24 hours and when the patient can tolerate oral medications complete treatment with ACTs for 3 full days.

Resistance to ACTs

This is a concerning point. Resistance has already started in Southeast Asia. Plasmodium gene mutations suggested that there may be a selection of mutants that make artemisinin less effective. The presence and development of these mutations are checked worldwide, together with the rate of failure of ACTs seen in *Fig.21*, the high rates of ACTs failure are found in Cambodia, Thailand, Vietnam. These have been exposed to several antimalarial drugs for many years and this led to the selection of plasmodia resistant to antimalarial drugs. That's why there is no effective prophylaxis for patients traveling to these countries.

Basically, resistance to Artemisinin is due to 2 proteins that allow hemoglobin to get into the hepatocytes; if Hb cannot get inside, Artemisinin is not activated and so cannot act. In this case, the mutation allows less Hb to get inside the hepatocyte and so the efficacy of the drug is much lower.

SUPPORTIVE CARE

1. Antibiotics in case of bacterial infections
2. Transfusions

3. Inotropic drugs for shock
4. Oxygen and ventilation in case of ARDS
5. Antipyretics
6. Glucose administration in case of hypoglycemia
7. Treatment of seizures in case of cerebral malaria
8. Careful fluid balance is really important. For example, as in Dengue infection, there's fluid leakage out of the vessels and the risk of edema, especially pulmonary edema, is very high. Fluid balance must be kept at minimum for the maintenance of circulation without having risk of edema.

PREVENTION OF MALARIA

1. *Vector control: environment control and indoor spraying*
2. *Personal prevention with the use of ITN (Insecticide Treated Nets) and repellent.*
3. *Chemoprophylaxis: suggested for travellers and VRFs. Prophylaxis depends on the scenario, according to the season, duration of travel and resistance in the area. Pre-travel consultation is suggested in a travel clinic in order to decide the type of prophylaxis.*

Travellers and VFRs

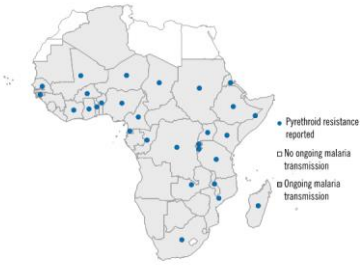
. *Mefloquine or atovaquone/ proguanil (Malarone) and doxycycline*
. *No prophylaxis if resistance is reported (go to hospital within 12 hours of fever).*

Intermittent Preventive Therapy (IPT) suggested in high endemic areas with different scenarios:

. *Second and third trimester of pregnancy in high endemic areas*
. *Children in high endemic areas (3 doses together with routine vaccination)*
. *Seasonal malaria chemoprevention in certain areas*

This is a way of reducing the number of malaria episodes and lowering the clinical consequences.

Figure 4.9 **Malaria-endemic countries in Africa reporting resistance to pyrethroids in at least one malaria vector in at least one monitoring site, 2011.**



Source: Reports from WHO regional entomologists in AFRO and EMRO. A dot indicates that resistance to pyrethroids has been reported in at least one malaria vector in at least one monitoring site. Note that map provides no indication of how widespread resistance is within a country. Countries with no insecticide resistance reported may have no resistance, or no susceptibility testing may be performed or results of susceptibility test may be unavailable.

There are two main concerns worldwide:

- *resistance to antimalarial drugs and repellents that may reduce the efficacy of both insecticides-treated nets and repellents.*
- *how to control malaria.*

Resistance for malaria prevention can also happen. For example, pyrethroids resistance has been demonstrated in certain countries (repellent resistance seen in Fig. 22)

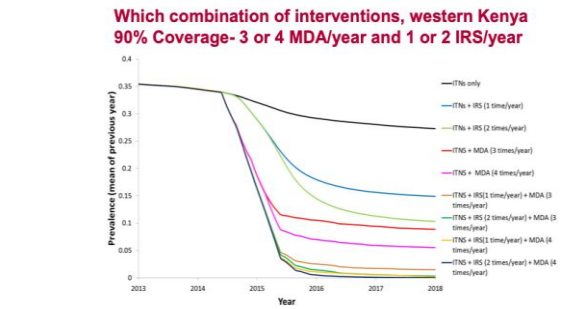


Fig. 23 A model developed by the Liverpool School of Tropical Medicine shows that the usage of indoor sprays and nets in Kenya causes a reduction in malaria. However, a plateau is reached after certain limits. In order to reduce and possibly eradicate malaria furthermore, other strategies are needed.

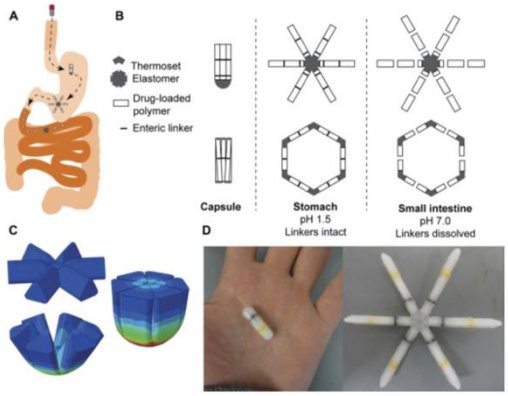
Mass drug based strategies for malaria elimination

- *Population wide drug based strategies*
- *Mass drug administration MDA*
- *Focal mass drug administration FMDA (only in local populations, in certain groups)*
- *Mass screening and treat MSAT or Mass test and treat MTAT*

These aggressive strategies might potentially lead to malaria elimination.

Currently, the use of ivermectin is being studied. ***Ivermectin*** *is an anti-parasitic drug that is used to treat strongyloidiasis and other helminthic infections. However, it's also effective in killing mosquitoes: when this drug is found in the blood, the mosquito's life expectancy is massively reduced thus lowering significantly the mosquito's opportunity to transmit malaria to other individuals.*

A study suggested the use of oral ultra-long-lasting drug delivery toward malaria elimination goal. This is a capsule that dissolves and deploys a star-shaped dosage form that releases the drug it was loaded with while assuming that geometry. The star tips are linked by pH-dependent bridges, once the capsule is in the stomach these bridges open turning the capsule into star-shaped dosage form that resides in the duodenum for several days releasing slowly ivermectin. Ivermectin is released for several days and when a mosquito bites the person in that time frame, a reduction in its life expectancy will occur thus reducing the number of vectors favoring malaria elimination.



MALARIA VACCINE

To date, an approved vaccine against malaria does not exist yet, but only an experimental vaccine (RTS, S / AS01) against P. Falciparum in phase 3. This study was published 3 years ago: 8922 children and 6537 infants were vaccinated with 3 or 4 doses. 4 doses efficacy was 36.3% for malaria, and 32.2% for severe malaria (low efficacy) and in young infants the efficacy was even lower (25.9% for malaria and 17.3% for severe malaria). The low efficacy is still better than nothing as it's more urgent in some areas and it was more effective in some areas compared to others. Three countries in Africa (Malawi and other two) launched a vaccine program which will provide a large-scale data on how protective the vaccine is for severe forms of malaria. The target is not malaria per se, but severe malaria. Results came out last year, in which the protection was around 1 life saved over 200 vaccines in children.
In conclusion, last year WHO added malaria vaccine in the suggested ones for people going to topical countries.
We will probably have more effective vaccines in the next future, such as the one published on the Lancet 3 years ago, which is shown to be more effective in terms of immunity and also protection.

ARBOVIRUSES (DENGUE, ZIKA, CHIKUNGUNYA)

These three viruses are quite similar, we can have a slight differentiation between them:

- Dengue generally develops with very high fever and all the other symptoms reported in *Fig. 25*
- Zika has high rush presentation
- Chikungunya presents especially arthralgia

Features	Dengue	Zika	Chikungunya
Fever	+++	++	+++
Rash	+	+++	++
Conjunctivitis	-	++	-
Arthralgia	+	++	+++
Myalgia	++	+	+
Headache	++	+	++
Hemorrhage	++	-	-
Shock	+	-	-

Fig. SEQ

1. DENGUE VIRUS

It is a Flavivirus (arbovirus) transmitted by *Aedes mosquitoes* that, in comparison to Anopheles mosquitoes, usually bite during daytime and prefer urban (but also rural) environments with small amounts of water e.g. small pawns or artificial containers for water.
3.9 billion people are at risk and there are 390 million cases per year.
Low mortality (2.5% for severe dengue), 4.000 -12.000 deaths per year. Hence, the overall mortality rate is low but still epidemics caused by the virus are important for healthcare systems because many patients need to be assessed.
Seasonal epidemics are found in some countries such as Rio de Janeiro, Singapore, Puerto Rico and Hawaii.
Local transmission in France and Croatia in 2010. Meaning that the virus is present in Europe and comes mainly through travelers. Thus, it is very important to report the presence of people with Dengue infection in order to prevent its spread to the vector and then from the vector to other patients.

There is a global increase in the number of cases in many countries in the world, but mostly in America.
A substantial reduction in mortality is demonstrated due to the fact that severe dengue like the hemorrhagic fever and shock are now treated. Thanks to treatment, the mortality has reduced from 20% to 0-1%. However, Dengue hemorrhagic fever has still a substantial mortality above the one reported before (0-1%) that indeed includes severe and non-severe cases.

GEOGRAPHICAL DISTRIBUTION

Dengue infections are widespread in most tropical countries such as Southeast Asia and South America [fig. 2]. Several countries in Africa have high presence of Dengue too, but infections are very limited there. This is potentially due to the presence of co-infection of dengue-malaria. The geographical distribution of dengue is similar to that of malaria.

The dengue virus has 4 different serotypes widely diffused. The massive people travelling who are infected with dengue lead to the distribution of these serotypes in the last 30 years.

CLINICAL PRESENTATION

The incubation is short (4-7 days, *ranging 3–14 days*). The clinical presentation often includes leukopenia and thrombocytopenia.

The majority of patients will just present:

- fever
- **"classic" Dengue**, which is characterized by fever, headache, arthomyalgias, lymphadenitis (50%), and there might be a rash (diffuse or maculopapular rash or even petechiae *Fig. 26*). It is important, if a patient presents these symptoms from March to November, even in Europe, to test him for Dengue.

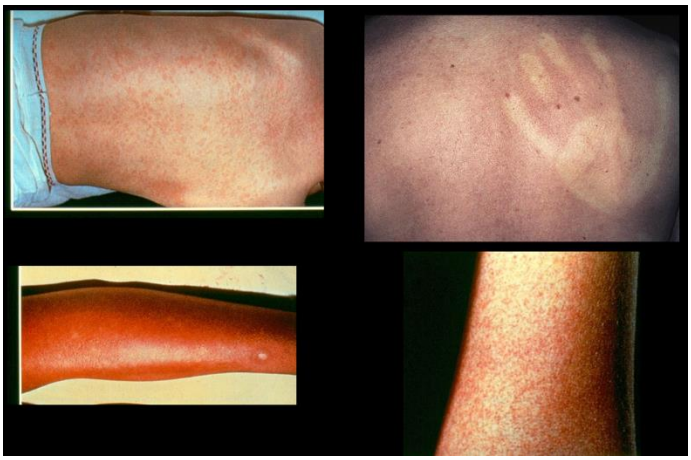


Fig.

Only a small number of patients have the severe form of Dengue that can cause the death of the patient if not treated:

- **Dengue Haemorrhagic Fever (DHF)**

DHF usually develops in 1% of patients. It is rare among travelers, but it is more common in patients with previous infections with different serotypes (so it is an immunity-based reaction) and in those living in

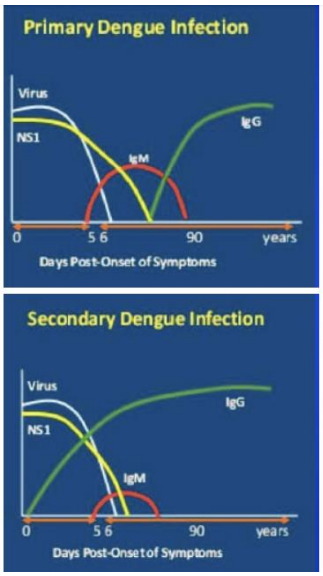
endemic areas. DHF consists of fever, hemorrhages and severe thrombocytopenia (<100k).

- **Dengue Shock Syndrome (DSS)**

MORTALITY

Mortality in classic dengue is very low <1% (hemorrhages in peptic ulcer carriers). In DHF it is 20% without treatment, which lowers to 1% when intensive supportive therapy is provided; therefore, it is a controllable situation. For this reason, it is common to see warning posters among hospitals in the affected endemic. Such posters alert the population about persistent vomiting and spontaneous bleeding, which may suggest DHF and the need to seek medical care.

DIAGNOSIS



Diagnosis is different among primary and secondary dengue infections. Diagnosis is made on antibodies.
Figs. 27 represent the "kinetic" of IgG (in green), IgM (in red), the virus i.e. Dengue RNA (in white), and the early antigen NS1 (in yellow). NS1 is really important, it is very rapid and sensitive.
The Dengue RNA is positive only during the first 5-6 days, so it is quite hard to find (and it is also expensive). Therefore, the common diagnostic path includes a combination of IgG, IgM, and antigen. The latter is quick to measure, thus providing a rapid and sensitive test used to diagnose Dengue. Secondary Dengue infection differs from the primary infection because of the IgG have a much higher titration and the very small increase in IgM.

Serological Markers

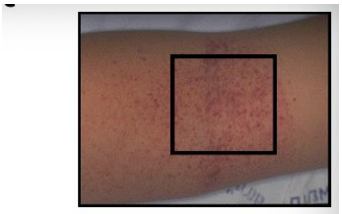
Primary dengue infection:

- *IgM (5 days after symptoms)*
- *IgG (7 days after symptoms)*
- *RNA (only for the first 5-7 days after symptoms, while it persists in urine and saliva for 14 days) = useful for serotype identification*
- Antigen NS1 (rapid diagnosis and sensitive)

Secondary dengue infection:

- *IgM (low titer and for few days)*
- *IgG (rise quickly, high titer)*
- *RNA*
- *Dengue RNA and NSA1 antigen*

Tourniquet test



The Tourniquet is part of the new WHO case definition for Dengue; it is a marker of capillary fragility and can be used as triage tool to differentiate patients with other acute gastroenteritis, for example, from those with Dengue.

How perform a tourniquet test?

- take the patient's blood pressure and record it
- inflate the cuff to an average pressure between SBP and DBP and maintain it for 5 min
- reduce and wait 2 minutes
- count the petechiae below the antecubital fossa [*fig. 28*]
- the test is positive if >10-20 petechiae per square inch.

The combination of a positive tourniquet test and lymphopenia is highly suggestive of dengue.

If you have a patient with Dengue, you have to evaluate him every 4 hours in terms of pressure, hematocrit and hemoglobin, infuse fluids and plasma or RBCs in some cases in the first 2 days.

DENGUE CONTROL

There are some objectives [fig. 8], regarding dengue control, related to the reduction of mortality, morbidity and burden of disease – all of which are hard to control since it is a viral disease and is often underreported. There have been problems with the **vaccine**: despite the effective beginning, malignant problems causing an increase in mortality arose in specific scenarios. There have also been legal problems as the Philippines decided to use the vaccine even if the WHO was contrary to it (because of the warning signs and related deaths) – indeed, some children were vaccinated and after 3 years, when they got infected with Dengue, they developed the most severe symptoms and eventually died.
Up to date, no vaccine is legally available yet.

2. ZIKA VIRUS

"I don't want you to be an expert in Zika virus". For Zika Professor wants us to remember just 2 things:

- Risk of brain malformation for the fetus if it is contracted during pregnancy
- It is associated with proximal ascending paralysis - Guillain-Barré syndrome

Just take a look at the slides.

The virus
It is the latest emerging arbovirus in the Americas:

- *single stranded RNA virus*
- *genus Flavivirus, family Flaviviridae*
- *closely related to Dengue, Yellow fever, Japanese encephalitis, and West Nile viruses*

The vector: Ades species mosquitoes

- *also transmit Dengue and Chikungunya viruses*
- *lay eggs in domestic water-holding containers*
- *live in and around households*
- *aggressive daytime biters*

Transmission

- *epidemic (urban cycle)*
- *sylvatic (jungle) cycle*

Other modalities of transmission:

- *maternal-fetal (intrauterine and perinatal)*
- *theoretical (organ or tissue transplantation and breast milk)*
- *sexual*
- *blood transfusion*
- *laboratory exposure*

Epidemiology

- *first isolated from a monkey in Uganda in 1947*
- *prior to 2007, only sporadic human disease cases reported from Africa and southeast Asia*
- *in 2007, first outbreak reported on Yap Island, Federated States of Micronesia. This was the first documented transmission outside of its traditional endemic areas in Africa and Asia*
- *in 2013-2014, >28,000 suspected cases reported from French Polynesia*

Clinical disease course and outcomes

- *It has an incubation period: 3-12 days after infection*
- *Viremic period: 3-5 days after onset of symptoms*
- *asymptomatic cases: approx. 80%*
- *clinical illness usually mild*
- *symptoms last several days to a week*
- *severe disease requiring hospitalization are uncommon*
- *fatalities are rare*

Potential complications:

- ***Microencephaly** in fetuses and newborn*
- ***Guillain-Barré syndrome***

Diagnosis

Detection of viral RNA

- *RNA, RT-PCT collected ≤7 days after illness onset (serum and saliva)*
- *RNA, RT-PCR up to 10 days after onset (urine)*
- *RNA, RT-PCR for specific investigations (amniotic and cerebrospinal fluids)*
- *Immunohistochemical (IHC) staining for viral antigens or RT-PCR on fixed tissues (e.g. placenta)*

Zika-specific antibodies

- *IgM-Ab in serum collected ≥4 days after illness onset*
- *Confirmation by plaque reduction neutralization tests*

Initial assessment and treatment

- *no specific antiviral therapy*
- *treatment is supportive (i.e. rest, fluids, analgesics, antipyretics)*
- *suspected Zika virus infecitions should be evaluated for possible Dengue or Chikungunya virus infections*

- *aspirin and NSAIDs should be avoided until Dengue can be ruled to reduce the risk of hemorrhages*

ZIKA VIRUS AND PREGNANCY

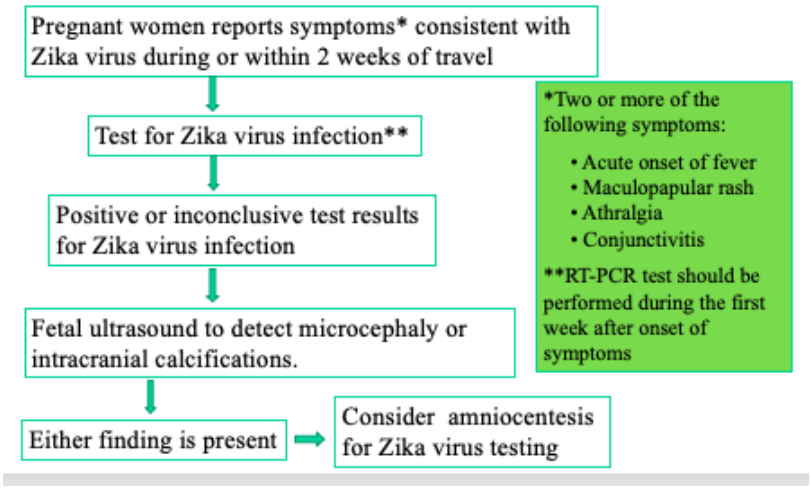
Evidence of maternal-fetal transmission (throughout pregnancy):

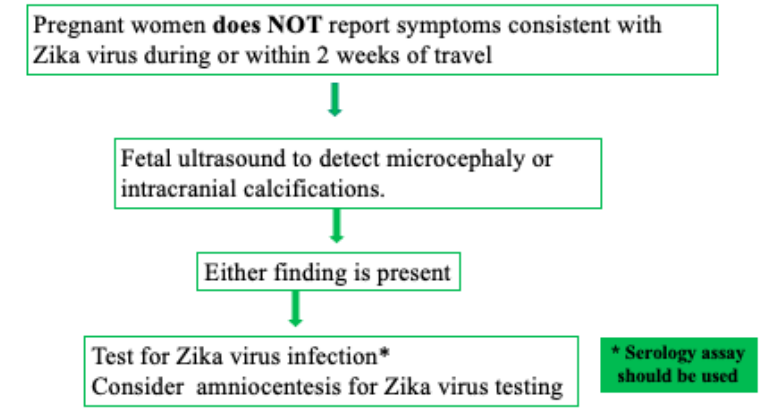
- Zika virus infection confirmed in infants with microcephaly
- Zika virus RNA identified in specimens of fetal losses
- Zika virus detected prenatally in amniotic fluid

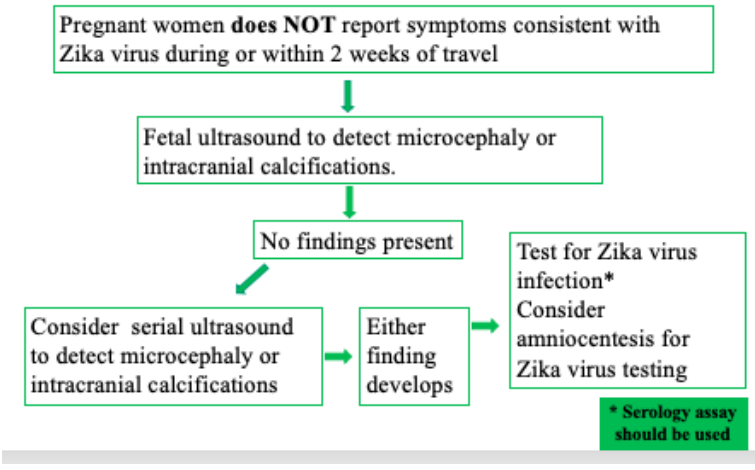
Evidence of perinatal transmission (during delivery time):

- pregnant women tested positive to Zika virus RNA by RT-PCR
- Zika virus infection confirmed in newborns, 1-3 days after delivery (it is unlikely that neonates were exposed to mosquitoes)

Testing algorithm for a pregnant woman with history of travel to an area with Zika virus transmission.







Clinical Management in case of maternal or fetal infection:

- Antepartum: consider serial US every 3-4 weeks
- Peripartum:
  - histopathologic examination of the placenta and umbilical cord
  - testing of frozen placental tissue and cord tissue for Zika virus RNA
  - testing of cord serum for Zika and Dengue virus IgM and neutralizing antibodies

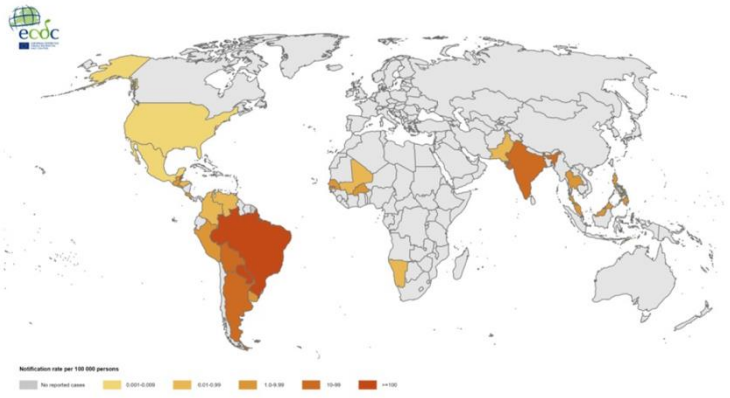
CDC recommendations on pregnant women considering travel:

- **pregnant women** in any trimester or who plan to become pregnant should consider avoiding travel to areas where Zika is present
- pregnant women who cannot avoid travelling to one of these countries should take great care to avoid mosquito bites during the trip

- **male travelers** returning from areas with local transmission of Zika virus should consider using a condom with a female partner at risk of getting pregnant or already pregnant:
    - or 28 days after their return from an active Zika transmission area if they have not had any symptoms compatible with Zika virus infection;
    - for 6 month following recovery from a laboratory-confirmed Zika virus infection

3. CHIKUNGUNYA

Have a look at the map in *fig. 30*: Chikungunya is widespread now and its largest diffusion occurred after 2013 (it is quite recent). A Chikungunya outbreak has been detected in Central Italy (in Anzio) in 2017, because of a local transmission by a carrier.



It is transmitted by *Aedes* mosquitos and has a short incubation period (4-7 days). The clinical presentation is a little different from the other arboviruses infection, as three clinical phases can be distinguished:

- ACUTE PHASE (up to 10 days): *arthritis, cutaneous rash, cardiovascular/neurological/renal/ocular manifestations*
- POST-ACUTE PHASE (4-12 weeks): symptoms (mostly arthritis) are slowly fading but chronic asthenia and chronic pain persist
- CHRONIC PHASE (after 12 weeks): chronic or long-lasting symptoms (they may last up to years), often with painful resurgence in the most commonly used joints, because of their inflammatory state after the infection

Treatment for the chronic phase includes acetaminophen, tramadol, or codeine (*no NSAIDs, no corticosteroids*), but it is still not clear how to treat it properly.

DIFFERENCES BETWEEN THE THREE ARBOVIRUSES

There are no great differences in the clinical manifestations of the three Arboviruses, but the most common reasons for medical consultation may vary:

- Dengue = fever, myalgia
- Chikungunya = joint pain, fever
- Zika = exanthema or pruritus

Referring to the evolution to chronic form: there is no chronic form in Dengue, and it is not described in Zika, but is very common in Chikungunya.

All of these characteristics are relevant for the differential diagnosis. However, at the end, the usual procedure is to rule out Malaria and Dengue and, if they are negative, proceed with a serological examination for Zika and Chikungunya.

BACTERIAL INFECTIONS (RICKETTSIAE, ENTERIC FEVER)

Bacterial infections can be the reason for a medical consultation after coming back from the tropics.

1. RICKETTSIAE

There are several species of Rickettsia:

- *African tick-bite fever (R. africae)*
- *Boutonneuse fever (R. conorii)*
- *Rocky Mountains fever (R. rickettsii)*
- *Scrub typhus (Orientia tsutsugamushi)*
- *Murine typhus (R. typhi)*
- *Epidemic typhus (R. prowazekii)*

They are all transmitted by different vectors (ticks, mites, lice, and fleas), which may also reflect the geographical differentiation.

CLINICAL PRESENTATION

In general, all species of Rickettsia manifest with fever, headache, myalgias; they have an abrupt onset. Other signs include:

- conjunctival suffusion
- regional lymphadenopathy
- generalized rash (except African form) also palmoplantar
- leuco- and thrombocytopenia
- painless escara (R. conorii “tache noire”)

When dealing with these patients, it is important to investigate if there were any recent travel in camping areas, forests, hiking, or trips in grassy areas (e.g. strict contact with nature). This should be the first warning sing. The second most important warning sign is the presence of a painless escara (called “tache noire” if caused by *R. Conorii*), which a typical lesion shown in *fig. 31*.

DIAGNOSIS AND TREATMENT

- *suspect of any recent camping site, hikers and safari in grassy areas*
- *clinical presentation*
- *serological confirmation*

The treatment of choice is usually Doxycycline.

2. ENTERIC FEVERS (Typhoid and Paratyphoid)

Salmonella typhi (and paratyphoid A and B) is transmitted by oral-fecal route through contaminated water, food, vegetables, seafood. It has an exclusively human reservoirs, with chronic carriers having it in their intestine and gallbladder. There are some hyperendemic areas (India, Philippines, Latin Americas).

CLINICAL PRESENTATION

The classic definition of typhoid fever divides the clinical course into four 7-days periods (1st, 2nd, 3rd, and 4th weeks), which are characterized by different clinical presentations. It is a kind of sepsis, because Salmonella disseminates in the blood, organs, and tissues.

The disease has an insidious onset, really abrupt, *after 10-14 days of incubation period*. Patients usually

present fever and headache with few other clinical signs, which might be:

- abdominal pain, constipation, rarely diarrhea ("pea puree" in children)
- sometimes leuko- or thrombocytopenia, *dry cough, lymphadenopathy*

The high-grade fever is accompanied by a relative bradycardia. The patient might be in a **typhoid status** (i.e. confusion, altered mental state) and present some **rose spots** [*fig. 32*], which are groups of 10-15 pink blanching papules (little bumps) on the anterior trunk *(between nipples and umbilicus), 2-8 m, last 3.5 days. Possible complications are GI bleeding, perforation, sepsis, abscesses.*

Fig.

All of these elements: exposure in tropical countries, insidious onset with fever, and other certain features may make you suspicious of enteric fevers.

DIAGNOSIS AND TREATMENT

Diagnosis is not easy because even if there is a serology (the famous Widal serology) it is not a proper diagnostic exam given the high number of false positives and false negatives. ***Cultures*** *are the preferred diagnostic examinations: hemoculture (2^{nd}-3^{rd} weeks), stool cultures (1^{st}-3^{rd} weeks), and, in certain cases, cultures of the bone marrow.*

Treatment:

- *Fluoroquinolones, 3rd generations cephalosporins (mostly ceftriaxone), azithromycin.*
- *Ceftriaxone (10-14 days).*
- *in case of severe systemic illness: dexamethasone (3 mg/Kg followed by 1 mg/Kg every 6 h for a total of 46 h).*
- *There is an orally administered vaccine that is partially protective (70-80%), therefore it reduces the risk of having a severe form of typhoid fever. This vaccine is suggested to travelers in endemic areas.*

HEMORRHAGIC FEVERS (including Yellow fever)

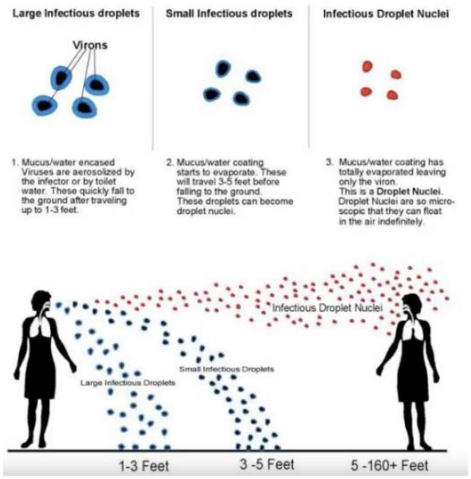
The differential diagnosis of hemorrhagic fever is with Neisseria meningitidis (Meningococcemia), malaria, leptospirosis e rickettsiosis, that can all lead to hemorrhages.
The most classical forms of hemorrhagic fevers are caused by RNA viruses:

- Filovirus (Ebola – mortality 30-50%, Marburg - mortality is 80%)
- Arenavirus (Lassa mortality 40-50%, Guanarito, Machupo, Junin, Sabia)
- Bunyavirus (Rift Valley fever, Congo-Crimea, Hantavirus)
- Flavivirus (Dengue, Yellow Fever and other Asian hemorrhagic fevers)

Transmission: there can be directly transmitted viruses and arthropod-borne viruses.

For this lecture, the focus will be on the transmission routes and the required protection for health care providers. For instance, Ebola is transmitted by droplets but sometimes patients get so sick (vomiting, diarrhea, coughing) that the transmission/exposure is difficult to avoid –they release so many infectious droplet nuclei that the infection becomes somewhat airborne [*fig. 33*]. Therefore, doctors must wear specific protections, but it is so difficult to work in those conditions that they are forced to change in less than an hour; moreover, undressing (as well as dressing) can be quite long because there are strict instructions to be followed.

EBOLAVIRUS PERSISTENCE



- Sporadic EVD cases continued to appear outside of this window and several reports strongly suggest that these unexpected re-emergences occurred due to viral transmission from persistently infected EVD survivors.
- Male survivors should continue to practice safe sex for at least 12 months after the onset of symptoms (WHO, 2016b). Viral recrudescence outside of the male genital tract (MGT) can also develop after filovirus infection, as initially observed in 1977 for a single case of Marburg virus uveitis (Kuming and Kokoris, 1977).
- During the West African outbreak, recrudescent cases were again observed within the eye, and also the CNS several months after initial infection (Jacobs et al., 2016; Varkey et al., 2015).

1. YELLOW FEVER

The frequency of yellow fever is now reducing, but it is still a cause of disease and it may cause hemorrhagic symptoms. It is caused by a Flavivirus (a small single-stranded RNA+, enveloped and antigenically conserved virus – indeed, it seems to be similar worldwide).

It is transmitted by *Aedes* and *Haemagogus* mosquitoes in three different scenarios:

- **Sylvatic (jungle) yellow fever**: in tropical rainforests, monkeys are the primary reservoirs of yellow fever. They are bitten by wild mosquitoes that pass the virus to the other monkeys. Occasionally humans working or travelling in the forest are bitten by infected mosquitoes and develop yellow fever.
- ***Intermediate yellow fever**: semi-domestic mosquitoes (those that breed both in the wild and around households) infect both monkeys and people. Increased contact between people and infected mosquitoes, leads to increased transmission and many separate villages in an area can develop outbreaks at the same time. This is the most common type of outbreak in Africa.*
- **Urban yellow fever**: large epidemics occur when infected people introduce the virus into heavily populated areas with high density of *Aedes aegypti* mosquitoes and where most people have little or no immunity, due to lack of vaccination or prior exposure to yellow fever. In these conditions, infected mosquitoes transmit virus from person to person.

Fig. 34 shows the most recent maps representing the countries at risk. They are mainly located in the tropical areas of Africa, but there are also other areas that, despite only having a partial outbreak, require vaccination prior travelling (e.g. South and Central America) – this is because they are trying to eradicate yellow fever. There has also been a yellow fever outbreak in the coastal area Brazil in 2017, so they are considered at risk.

A modelling study based on African data sources estimated the burden of yellow fever during 2013 was 84-170k severe cases and 29-60k deaths per year – i.e. there is a slight reduction, but it is still a high number.

CLINICAL PRESENTATION, DIAGNOSIS AND TREATMENT

Alike other arboviruses, the incubation period is short (3-6 days).

- **Acute phase**: fever with concomitants symptoms *(chills, headache and myalgias, nausea, vomiting, relative bradycardia)*.
- **Toxic phase** *(only 15%)*: after defervescence, fever reappears with **jaundice** (that is why it is called "yellow fever": there is a hepatic toxicity with hyperbilirubinemia), abdominal pain, and vomiting. During this phase hemorrhagic symptoms may appear e.g. "black vomit". The toxic phase has a much higher, mostly due to renal failure – which is the most common and more severe complication.
- *Complications: renal failure, myocardial dysfunction, CNS involvement, pancreatitis.*

Mortality accounts for 20-50% of cases and is usually due to its complications.

DIAGNOSIS

Diagnosis is achieved through a combination of serology and PCR (although the latter can turn out positive only during the first days, so it should be performed early).

TREATMENT

There is no specific treatment (only supportive care), while vaccine is effective – that is why a lot of strategies to control yellow fever rely on vaccinations.

Vaccination:

Live-attenuated and life-long protection from day 10 (a booster dose is not needed i.e. a single dose is protective).

- *In high-risk areas where vaccination coverage is low, prompt recognition and control of outbreaks using mass immunization is critical. It is important to vaccinate most (80% or more) of the population at risk to prevent transmission in a region with a yellow fever outbreak.*

*However, there might be severe **adverse event following immunization (AEFI).** Apart from the fact that is painful, patients could develop liver and renal toxicity, which are not that common but specific groups of patients are at higher risk, thus are excluded from the vaccination.*

TRYPANOSOMIASIS, FILARIAE, HYPEREOSINOPHILIAS AND LEISHMANIASIS – exotic tropical diseases

Fever with neurologic involvement may be caused by different pathogens, here is the differential diagnosis:

- meningitis and encephalitis
- malaria
- extrapulmonary TB
- typhoid fever
- rickettsiosis
- poliomyelitis
- **rabies**
- leptospirosis
- **African Trypanosomiasis**
- **Japanese Encephalitis**

These last two are the most typical ones (they will be discussed in the ADE - EMERGING BUGS AND

TREATMENTS IN INFECTIOUS DISEASES).

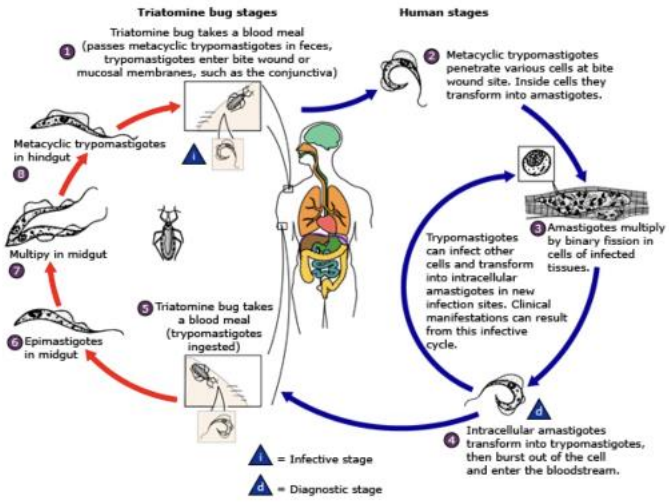
1. CHAGAS' DISEASE - AMERICAN TRYPANOSOMIASIS

It is an infection caused by *Trypanosoma Cruzi*, mostly transmitted by Triatomine bugs ("kissing bugs"), however other possible transmission routes are:

- vertical
- transfusion and transplant
- ingestion of contaminated food or drink (e.g. cane sugar contaminated with Trypanosoma feces)
- laboratory exposure.

It is found mostly in America (expanding from southern US to north of Chile, with highest burden in Bolivia, Argentina, Paraguay, Ecuador, El Salvador, and Guatemala). The estimated prevalence accounting for 6 million people (high prevalence).

LIFE CYCLE



When a bug feeds on a human (i.e. it bites to take a "blood meal", as seen in *fig. 35*) it usually defecates. The feces pass *trypomastigotes* that penetrate several cells at the bite wound site; inside the cells they transform into *amastigotes*, which multiply. Intracellular *amastigotes* develop again into *trypomastigotes*, which burst out of the cell and enter the bloodstream, potentially infecting other cells and transform into intracellular amastigotes in new infection sites. Once in the blood *trypomastigotes* can also be transmitted to another bug.

CLINICAL PRESENTATION

- **ACUTE TRYPANOSOMIASIS**

Acute trypanosomiasis is usually a mild disease, just a small percentage *(>1%)* of patients are actually diagnosed, indeed it is usually asymptomatic. The incubation period is of 1-2 weeks (longer for transfusion/transplant – up to 120 days). Patients might present signs at portal entry, such as **Chagoma sign** (*fig. 36 on the right*, a round rash around the bite site) or **Romaña sign** (*fig. 36 on the left*, monoliteral swelling of one of the eyelids). They can also have fever, systemic symptoms, hepatosplenomegaly, atypical lymphocytosis, and (rarely) meningoencephalitis and myocarditis. There usually is such a high level of parasitemia that is possible to see *Trypomastigotes* on Giemsa stained thick and thin blood smears (*Fig. 36 at the bottom*), but PCR is much more sensitive thus is preferred for diagnosis.

*Fig. SEQ Fig. **

- **CONGENITAL INFECTION**

Congenital infection is uncommon, just in South America. It is similar to acute form, affecting 5-10% of newborns from infected mothers. It is mostly mild or asymptomatic and is diagnosed by cord or neonatal blood (smear + PCR).

- **CHRONIC INFECTION**

Chronic infection is highly variable, but the two most important forms are:

- **cardiomyopathy**: presents conduction system defects, arrhythmias, *apical aneurysms*; the most common form is a **dilated cardiomyopathy with congestive heart failure**, which usually causes SCD. Some of the patients require transplantation because of heart failure.
- **GI dysfunction**: the destruction of enteric neurons during acute infection results in **severe organ enlargement**, mainly involving the esophagus and colon:
    - Megaesophagus: causes dysphagia, odynophagia, weight loss, *regurgitation, aspiration.*
    - Colon: chronic severe constipation and fecalomas formation.
    -

Treatment: benznidazole, nifurtimox

2. HELMINTHS INFECTION-HYPEREOSINOPHILIA

Among the causes of hyper-eosinophilia there are *Helminth* infections. Diagnosis requires 3 stool samples and serology for some of the helminths that can be found in the stools, such as *Strongyloides, Toxocara, Trichinella*. The latter is the most typical one; it causes muscle pain, fever, GI symptoms and hyper-eosinophilia. It usually presents in clusters, as it happened this past year with a few patients in Val di Susa who ate wild boar salami and presented with all these symptoms.

SOIL TRANSMITTED HELMINTHS

- *Ascaris lumbricoides [800 million - 1.1 billion]*
- *Whipworm (Trichuris trichiura) [600-800 million]*
- *Hookworm (Ancylostoma duodenale and Necator americanus) [570-740 million]*
- *Strongyloides stercoralis [30-100 million]*

Soils transmitted Helminths are relevant for the health of millions of children in limited resources settings – have a look at the above numbers of estimated prevalence around the world. They are transmitted from soil and can cause chronic intestinal infections with chronic anemia and may affect neurodevelopment – so they may cause substantial problems in the development of those children. There are various NGO programs that try to tackle malnourishment: one of the things that is usually done, apart from assessing the malnutrition, is deworming of children.

Some examples of parasitic infections caused by soil transmitted Helminths are:

- *Larva migrans*, caused by Hookworms, which usually presents in the feet [*fig. 37*]. You do not see it moving because it is kind of slow, it does 1 cm/day.
    - Asymptomatic but causes anemia!

- Pruritus, dry cough
-

Fig. SEQ Fig

- *Larva currens,* caused by *Strongyloides stercoralis*, which is a much quicker larva that coils under the skin [*fig. 38*].
  - *Malabsorption and abdominal pain*
  - *Hyperinfection in immunedepressed hosts*
  - *Serology!*

- *Ascaris lumbricoides (Fig. 39)*
  - *Often asymptomatic*
  - *Risk of bowel obstruction*
  - *Dry cough*

3. SCHISTOSOMIASIS

A particular case of fever with hyper-eosinophilia is Schistosoma infection, called **snail fever**. Schistosoma is a Helminth that is usually acquired when swimming in contaminated water (e.g. lakes of endemic areas), indeed it penetrates in the skin from a lesion.
They are usually very "romantic", the male lives around the female to protect her and they live in fresh water.

There are different species of Schistosomes that have different organ and tissue tropism:

- *S. mansoni*, usually causes chronic liver problem;
- *S. haematobium* localizes in the bladder giving chronic problems in the urinary tract;
- *S. japonicum*;
- *S. mekongi* (in south-east Asia).

It is a relevant infection: it is estimated that more than 200 million people have it. Areas at risk are mostly localized in Africa, but also South East Asia and South America.

CLINICAL PRESENTATION

- The most limited clinical presentation is **cercarial dermatitis** (called "swimmers' itch"), due to cercariae penetration into the skin, nothing serious [*fig. 41*]
- after that, cercaria can migrate into the gut, resulting in **intestinal schistosomiasis** *(granulomas → polyps, protein loss, malabsorption, strictures)*
- the parasites can further migrate into other locations causing chronic infections with chronic consequences (15 years after infection):
  - migration to the liver causes **hepatosplenic schistosomiasis**, which leads to portal hypertension → ascites, varices, splenomegaly, but normal hepatic function
  - migration to the bladder causes **urinary schistosomiasis**, resulting in hematuria, chronic

infection, obstruction

- *others (cardiopulmonary, CNS, etc.)*
- ACUTE FORM: **Katayama fever** is an acute inflammatory syndrome that manifests 3-8 weeks after the cercariae infection. It includes high fever, cough, general malaise, and hyper-eosinophilia. *It is a hypersensitivity reaction that heals without treatment (granulomas).*

CONTROL OF SCHISTOSOMIASIS

Suggestions for prevention are, of course, to avoid swimming in lakes in endemic areas. However, there are some problems for the control of Schistosomiasis, which is obtained through a Massive Drug Administration (MDA) to the target population *(children 6-15)* in the attempt of reducing the burden of Schistosomiasis and its long-term consequences on liver and bladder.

4. FILARIASIS

Different types of filariasis exist. Even though their incidence is lowering, they can still be found in some areas of Sub-Saharan Africa.

- **Lymphatic Filariasis**, causes massive lymphatic edema in lower limbs (where vectors i.e. mosquitoes usually bite). Vectors are Aedes, Culex, and Anopheles.
- **Loa Loa**, transmitted by flies and usually resides in the conjunctiva *(mango/deer fly: Chrysops)*
- **Onchocerciasis,** transmitted by black flies *(black fly: Simulium)* that can cause blindness or skin psoriasis (chronic lesions in the skin).