Skin and arthropods: an effective interaction used by pathogens in vector-borne diseases

In the last years, the skin has been described as a major interface in arthropod borne diseases. Although it constitutes an efficient immune and physical barrier, pathogens have developed effective strategies to thwart the host. In this process, the arthropod plays a major role. For mosquitoes, the quick blood meal is made through an efficient inoculation process directly into the blood vessel. For the long lasting blood meal of hard ticks, the sophisticated biting pieces and the tick saliva provide potent tools to help pathogen transmission. Lyme borreliosis and leishmaniases have been particularly well investigated in this context.

**Key words:** keratinocytes, fibroblasts, dendritic cell, arthropod saliva, vector-borne diseases, leishmaniasis, Lyme borreliosis

Vector-borne diseases (VBDs) rely on complex interactions between three actors: the vertebrate host, the arthropod and the pathogen (figure 1).

First, arthropods are attracted to the vertebrate host through their olfaction. Volatile products are detected such as lactic acid, sweat products modified by skin resident bacteria [1]. Then, facing the skin barrier, the arthropod punctures the skin by its biting pieces, either through a solenophage or telmophage bite. Pathogens are inoculated into the skin, where they are going to interact with different skin cells [2]. In this process, arthropod saliva plays a major role. First it targets the coagulation cascade very efficiently, then the different immune cells or immune pathways (complement pathway) allowing the pathogen to escape, hide or multiply in the skin. Besides, it is now well documented that arthropods react to pathogens they transmit by mounting an innate immunity [3-5], but within the vector, microorganisms seem to be not affected by this immune response. In this review, we detailed some of these processes that make VBDs an example of fascinating interactions for pathogen survival. Lyme borreliosis and leishmaniases are VBDs that illustrate the best this triad of interactions.

**The arthropod: a major actor**

Two major groups of arthropods are involved in VBDs: insects and ticks. Insects and soft ticks (Argasidae) have a short blood meal, while hard ticks have a long lasting blood meal of several days. Vector saliva is a critical parameter for the efficient transmission of the pathogen in VBDs. It contains anti-hemostatic molecules, anti-inflammatory and immunomodulatory factors to facilitate the uptake of the blood meal, essential to their survival [10, 11]. In VBDs, some pathogens use arthropod saliva to facilitate their transmission. Studies of Lyme borreliosis and leishmaniasis revealed a major contribution from the saliva of *Ixodes* ticks and sandflies in the transmission of the bacteria [12] and the parasite [13] respectively. The vectors are not simple syringes that inoculate pathogens; they play an essential role as reservoir for certain pathogens, as virulence factor...
and facilitating agent for pathogen transmission. Indeed, within the arthropod the pathogens are subject to major antigenic modifications that increase their virulence and in the presence of vector saliva, fewer pathogens are necessary to induce an infection in the vertebrate host. Transcriptomics and proteomics analyses of arthropod salivary glands, infected and uninfected, have shown that certain pathogens increase arthropod proteins to facilitate their transmission to the vertebrate host [14]. Some proteins have been well characterized and have been the subject of vaccine trials in laboratory animals. For example, a peptide called maxadilan has been identified as a facilitating molecule in the sandfly saliva [10, 15]. Similar studies have been made in tick vectors. Because of their long blood meal (3-10 days), the hard tick must develop different strategies to remain attached efficiently. This probably explains the high number of molecules with pharmacological and immunomodulatory properties found in hard ticks [16, 17]. In Lyme borreliosis, bacteria specifically upregulate a tick salivary protein, Salp15. Its allows *Borrelia* to escape the immune system in the vertebrate host [18] thereby improving the transmission of the bacterium. Arthropod saliva is therefore a potential target for vaccine development [10, 15, 19]. For some VBDs including malaria, analyzing the effects of saliva deserves more study since its initial role in pathogenesis is unclear or controversial [20].

The pathogens

Some pathogens have evolved strategies to efficiently infect vertebrate hosts by taking advantage of the injury produced in the skin barrier via the bite of the arthropods, insects or mites. These VBDs have a major impact on public health by their morbidity and mortality. During the infectious bite, pathogens use arthropod saliva to circumvent the pharmacology and immunity of the vertebrate host. Consequently, the vector transmission becomes an extremely efficient process and requires very little inoculation of pathogens [2]. Once in the skin, pathogens use this immune-privileged organ as an amplification and/or persistence site [21]. Some host-pathogen interactions are particularly well studied (e.g. leishmaniasis and Lyme borreliosis). Recent technologies such as intravital imaging and the development of fluorescent pathogens (gfp: green fluorescent protein) showed new developmental mechanisms in the skin for some VBDs.

Insect borne diseases

Insects are major vectors of VBDs. Mosquitoes are the most important as they transmit a large variety of pathogens, mainly parasites (malaria, filariasis) and viruses (dengue, yellow fever, West Nile virus…). The bite is very short in time. According to the insect, the long proboscis catheterizes the blood vessel (mosquitoes) or the small cutting mouthparts lacerate the skin tissues, inducing hemorrhages of surface capillaries (sandfly, blackfly). The physiopathological processes involved in the transmission of *Leishmania* has been the most studied so far.

Leishmaniases

This parasitic infection is caused by a trypanosomidae parasite, *Leishmania*, transmitted by a small diptera, the sandfly *Phlebotomus* (Old World) or *Lutzomyia* (New World). The parasite can remain localized in the skin or diffuse in the body according to the parasite species involved. The first clinical manifestation is a skin lesion that may persist for several weeks. For decades, it was thought that the promastigote forms only infected macrophages where it turned into amastigote forms. By altering the pH of the phagolysosome, they were able to escape the host immune response. Recently, it was shown that the cell first targeted by the parasite is in fact the neutrophil. These phagocytic cells are among the first to migrate at the site of infection. A study by fluorescence microscopy showed that *Leishmania* first locates in neutrophils and then only after within macrophages [22]. Parasitized neutrophils become apoptotic and are then preferentially internalized by macrophages [23].

Malaria

It is a parasitic infection transmitted by *Anopheles* mosquito. *Plasmodium* is a protist Apicomplexa causing approximately 1 million deaths per year in tropical countries. The mosquito inoculates sporozoites after a probing time without causing visible damage to the skin. The