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Impact of Mosquito Gut Microbiota on Propagating Pathogenic Infections

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

The mosquito vectors of various human diseases host a diversified microbial community. These microbiomes seem to be beneficial in several aspects of mosquito biology. They can influence mosquitoes' susceptibility to various pathogenic infections, therefore affecting the vectorial capacity of mosquitoes through different direct or indirect mechanisms. These microbes act as natural barriers against several mosquito-transmitted infectious diseases. They may be considered as a new transmission-blocking strategy to limit the transmission of pathogens like Plasmodium, Trypanosome, Zika, Dengue and Chikungunya viruses, and filarial parasites. It is through an understanding of the interaction between the mosquito, its microbiota, and the transmitted pathogens that some promising approaches may be developed for limiting the transmission of pathogenic diseases.In this review, we investigate the role of mosquito's gut microbiome in the propagation of pathogenic infections. It is summarized here in a brief manner how the current knowledge is used for the purpose of limiting the transmission of mosquito-borne diseases through the alteration of mosquitoes' vector capacities.

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1. INTRODUCTION

Malaria, filarial disease, dengue fever, chikungunya, and zika are mosquito-transmitted diseases that pose a major health threat worldwide. Mosquitoes in endemic areas of these diseases are continuously facing these pathogens. In their habitat,they remain exposed todiversified microorganisms. Some of these microorganisms have prospered symbiotic relationships with these insects. Some are intake from their nectar sources or breeding places and are adapted to persist within the host commensally. Several lines of evidence suggest that these symbiotic bacteria nutritionally benefit the insect [1,2]. Many studies have documented the mosquito microbiota's significant role in maintaining the host's basal immune activity [1,3,4,5]. A mosquito's immune response can be modulated by its microbiota, affecting its ability to transmit human pathogens. For example, the antibiotic treatment promotes *A. gambiae* and *A. aegypti* to become more susceptible to *Plasmodium falciparum* and infection with the dengue virus, respectively [1,6,7]. These endobacteria can influence mosquito's vector competence by impairing pathogen infections through three mechanisms. They compete for resources, secret anti-pathogen molecules and ultimately activate mosquito's responses of the immune system [1,8,9].

Insect vectors, human hosts, and pathogens interact to determine the fate of infectious diseases transmitted by vectors. The majority of research targeting control of the spread of diseases transmitted by mosquitoes has been run so far largely focused on the human-pathogen or human-vector interactions [1]. A major area of research today focuses on the interaction of insect vectors with pathogens to determine the impact of this tripartite interaction, pathogens and insect microbiota in controlling the fate of disease transmission. This review aims to introduce and review the latest knowledge about mosquito microbiota, as well as the current understanding of how microbiota may modulate infections transmitted by mosquitoes. This approach may become a new promising alternative strategy for controlling infectious diseases.

Mosquitoes' gut, genital organs and salivary glands are colonized primarily by bacteria, viruses, and fungi. The bacterial part of this microbial community is best characterized [10,11,12,13]. This microbiota interferes with disease transmission by inhibiting pathogen colonization and development and affecting a wide range of physiological aspects of mosquito physiology. Thus, micro-organisms colonizing mosquitoes are a potential target in eliminatingthe mosquito-transmitted pathogenic infection. Destruction of these microorganisms results in the shortened lifespan of the mosquito or also causes decreased rates of pathogen infection, either via natural competition mechanisms [10,14,7] or through the production of anti-pathogenic factors [10,15,16,17]. This strategy's main advantage is that it simultaneously targets mosquitoes and pathogens. The mosquitoes acquire their microbiomes partly from mother's genitalia at the time of embryonic development and partly from their breeding places. Some are trans-steadily shifted to the adults. Adults acquire some other micro-organisms during feeding or mating [10,18,19,20]. The ingested blood is stored in the mid-gut over 2 days during digestion. The gut microbiota shows drastic proliferation after a blood meal. During its early development, *Plasmodium* undergoes several stages in the middle of the gut [10]. The current review elaborates on comprehending the tripartite interactions between mosquitoes, their microbiomes, and transmitted pathogens. It also discusses how microbes and some environmental inputs maintain these groups' composition andt their diversity.

2. INTERACTION BETWEEN MOSQUITO AND INVADING PATHOGENS

Mosquitoes mainly rely on innate immune systems to escape from invading pathogens. Like vertebrates, different immune signaling pathways also shape and control their immune responses. Unlike higher organisms, physical barriers, such as hard exoskeletons and adaptive immune systems, are established by mosquitoes to protect themselves. Viral, bacterial, fungal, and parasitic infections are handled completely by mosquitoes' innate immunity. A mosquito's body responds to pathogens by forming physiological barriers. Various responses occur to clear pathogens from the system depending on the location and degree of infection. In order to successfully introduce infection, a pathogen must dismantle both the physiological and physical barriers. Their tightly closed, hydrophobic outer cuticles protect them against external environmental factors and pathogen

access. In the event that pathogens penetrate the mosquito through an adventitious break in the outer cuticle, coagulation, scar formation and hemocyte degranulation are triggered, preventing pathogen entry. There are two components to insect immunity: a cellular component as well as a humoral component, which allows mosquitoes defeat any pathogens that have penetrated the body once they have been ingested. In the cellular arm of immunity, hemocytes play a major role. Innate immunity comprises humoral elements, such as “antimicrobial peptides (AMPs)”, “pattern-recognition receptors (PRRs)” and “phenoloxidase cascade components”. In response to pathogen recognition by PRRs, immune pathways are activated. Thus, AMPs and anti-pathogenic molecules are produced [21,1,22], stimulating other defense mechanisms. Current knowledge about the pathways involved in the immune signaling of insects is primarily based on the findings of research done so far in mosquitoes and *Drosophila* [23]. In response to diverse pathogens, the immune pathways in insects [JAK/STAT signaling pathways and immune deficiency signaling pathways (Toll, Janus kinase/STAT)] are activated. The Imd pathway, the NF-kB signaling route in insects, regulates the body's immune response against viruses, bacteria and *P. falciparum* [24,25]. Another NF-kB pathway in insects is the Toll route. Gram-positive bacteria, viruses, and fungi are the main players in activating this enzyme [26,27]. As an interferon-induced signaling route, evidence suggests that JAK/STAT is involved in the immunity to insects to viruses, bacteria and malaria parasites [28]. A mosquito's microbiota can determine the vector's susceptibility to human pathogen infection by influencing the immune system, which simultaneously shows antibacterial, antiparasitic, and antiviral responses [1,29].

The gut bacteria multiply rapidly after a blood meal, stimulating the immune system to combat micro-organisms. With the help of the Peptidoglycan Recognition Protein, the midgut epithelium detects the peptidoglycan of the bacterial cell wall, triggering an immune response using the Imd pathway.Currently, it is not clear how Imd immune signaling affects parasite colonization.This immune signaling, however, increases the expression of TEP1, which plays an important role in microbiota-dependent pathogen control [28]. In *Plasmodium*-infected mosquitoes, ookinete numbers are decreased with the gut epithelium's decrease in microbiota load. Thus the microbiota-influenced immune response can defend against the invading pathogens before the action of host’s complement system [28]. Mosquitoes are also influenced by their microbiota composition regarding vector competence. A dynamic community of intestinal bacteria, viruses, fungi, and archaea, known as the gut microbiota, has been found to affect humoral immunity and vaccine effectiveness, which opens up the possibility of engineering microbiomes for optimum immune responses. In future research, to gain a deeper understanding of the role of gut microbiota in the growth of host immunity, we will decode the relationship between microbiome variations and pathogen-specific systemic and mucosal immune responses by studying gut microbiota variation. Mosquitoes use many pathways and molecules in response to infectious agents. In their gut, female mosquitoes transmit various bacteria, predominantly Gram-negative [10].

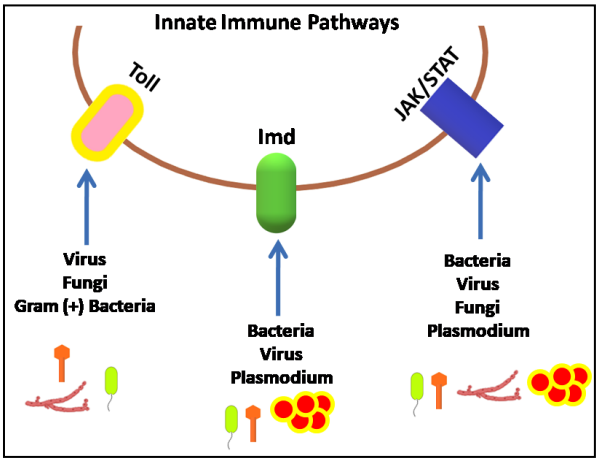

Fig. 1. Schematic diagram of mosquito’s innate immune pathways activated by various pathogens

3. MOSQUITO GUT MICROBIOTA

The microbiomes that live in the mosquito's midgut have a proven ability to prevent the spread of pathogens. Using the sequencing of the “16S rRNA gene” and the use of the MiSeq technology, [30] described field collected 12 wild bacterial species of mosquitoes from Champaign, Illinois. The gut microbiota population varied noticeably among the members of the same species. But certain mosquito species showed similar gut microbiota makeup in their individual members. The remaining sequences underwent quality screening and rarefaction before being allocated to 181 (one hundred eighty one) operational taxonomic units (OTUs). 80% of these OTUs were found two (02) different mosquito species, according to the survey. On the other hand, all 12 species of mosquito had the three “OTUs Gluconobacter (OTU 1), Propionibacterium (OTU 9), and Staphylococcus (OTU 31)” [30].

Gram-negative bacteria dominate the mosquito gut microbiota. An earlierexperimental study recognizedabout 98 (ninety-eight) bacteria genera in*Anopheles* mosquito.Among them, the most common bacteria genus includes *Aeromonas*, *Pseudomonas*, *Comamonas*, *Asaia*, *Elizabethkingia*,*Klebsiella*, *Enterobacter*, *Serratia*and *Pantoea* [30]. On the other hand, gram-negative bacteria dominate the *Aedes* mosquito [31]. Apart from the pro-bacteria, the abundance of“18S rRNA”within the mosquito host indicates the presence of the eukaryotic microbiota, which will be confirmedby 18S rRNA gene sequencing. But the community of eukaryotic microbes in mosquitoes keeps poorly studied. Individuals from the same species and study site usually harbour different gut microbes. On the other hand, the microbiota of *Ae. albopictus* was significantly least diverse compared to *An. quadrimaculatus*, *An. crucians*, *Ae. Ae. triseriatus*, *Culex resturans* and *japonicas* [32].

The microbial community of “*Ae. Albopictus”* and “*Cx. Pipiens”* was markedly different from those of other mosquito species and was dominated by *Wolbachia* [32]. Several researchers have studied the composition and structure of the gut microbiota of various mosquito species. Findings from their studies may provide the variation in microbial composition and the role of individuals in vector competence for diverse pathogens and its potential utility in preventing and treating illnesses brought on by these infections.

Anopheles mosquitoes host diverse bacterial and non-bacterial communities in their midgut and develop a complex ecosystem. It has been reported that *Comamonas*, *Acinetobacter*, and *Pseudomonas* showed dominancy in bacterial ecosystems inhibiting salivary and female reproductive organs of *Anopheles* mosquitoes [13]. Several evidence shows that *Anopheles* midgut microbiota plays a significant role in parasitic malarial disease transmission. Tchioffo et al. (2016) reported that the abundance of *Serratia* in salivary and midguts exhibited a considerable difference between *P. falciparum* mosquitoes that are both infected and uninfected. *Furthermore,* the presence of *Serratia* in those tissues in mosquitoes with malaria suggests that the interaction between micro-organisms and parasites may facilitate malaria infection through some unidentified mechanism. Thus, targeting this bacterium may be a promising tactic for the management of falciparum malaria.

The bacterial community structure of *Culex* mosquito was best studied in *C. tarsalis* by Duguma et al. (2015). They performed beta diversity objectivity and indicated that the bacterial structure altered among different developmental stages of *C. tarsalis*. *Thorsellia*, a gammaproteobacterium, was found to be most abundant among the three stages of life (larvae, pupae and adults) of field *C. tarsalis*, but was infrequent in laboratory-cultured mosquito colonies. It has been found that the proportions of *Thorsellia* were highest in Pupae and lowest in newly emerged adults [33]. Thus the role of *Thorsellia* in *Culex* transmitted diseases have to be evaluated and the distribution of *Thorsellia* across different species of *Culex* warrant further investigation.

4. EFFECT OF MICROBIOTA ON PATHOGEN TRANSMISSION

4.1 Wolbachia (WLBC)

The most elaborately studied microbial approach to diminish the vector competence of various mosquito species utilizes *Wolbachia* (*WLBC*). Approximately 60% of insects are infected with this endosymbiont [34]. It has been repeatedly reported that this endosymbiont can manipulate host reproduction and transmit vertically within the insect community [35]. *WLBC* mainly relies on cytoplasmic incompatibility (CI) to facilitate its spread within insect populations. When an infected male *WLBC* mates with a female who is

not infected or infected with an incompatible strain, CI occurs and results in embryonic death. When an infected female mates in the population, it transmits *Wolbachia* within insect populations [35]. *WLBC*-mediated CI is an incompatible insect technique (IIT) that may reduce mosquito populations [36,37]. The ability of *WLBC* to confer pathogen interference, induce CI, and transmit vertically may make it an attractive candidate for controlling mosquito-transmitted diseases.

WLBC can block the maturation of variouspathogens. *Ae. aegypti* is generally uninfected by *WLBC.* The anti-pathogenic effect is particularly enhanced when *WLBC*is artificially transferred into that vector [38]. An infection with *w*AlbA and *w*AlbB strains of *WLBC* was recently reported in *Ae. albopictus* mosquitoes collected in Florida, USA [39]. Transinfected *Ae. Aegypti* have diminished vector competence to arboviruses such as dengue virus [40,41,42], yellow fever virus [43] zika virus [44,45] and chikungunya virus [8,44]. In addition, *WLBC*-based control strategies are also under examination for Japanese encephalitis vectored by *C. tritaeniorhynchus* [46]. *Ae. albopictus* infected with *w*AlbA and *w*AlbB exhibited antiviral activity after transinfection with novel strains [47]. Prior to the introduction of the novel *w*Mel strain, resident strains would be removed from *Drosophila* by antibiotic treatment. *Ae. albopictus* with *w*Mel-infection have lower vector competence for dengue virus than uninfected and double infected (with *w*AlbA and *w*AlbB) mosquitoes [47]. Native *WLBC* infections reduce the West Nile virus load in *Cx. Quinquefasciatus* and dengue and chikungunya virus load in *Ae. albopictus* [48,49,50]. But infection with natural *WLBC* strains makes the vector more competent. Compared to the management of arboviral infections, implementing WLBC-based control techniques against human malaria continues to be more difficult. The *w*MelPop strain can interfere with developing a murine malaria model, *P. berghei* [51]. Infection with the *w*AlbB strain causes CI in *An. Stephensi*and thus markedly blocked *P. falciparum* [52]. The infection with this bacterium exerted a substantial fitness cost on the host and may also contribute to malaria control strategies. *WLBC* infections have been discovered in some *Anopheles* populations [53,54].

4.2 Gut Microbiota

The midgut of a mosquito represents the first and key obstruction for developing various pathogens. The microbiota in its lumen is the major cause of such a bottleneck. Experimentation has been used in many different ways to study the effect of *Anopheles*' midgut microbiome on the spread of the *Plasmodium* infection.The findings obtained from these studies reveal an in the total inhibitory action of the microbial community on the pathogens. The inhibitory effects are independent of the vector and pathogen species, and are mostly specific for bacterial strains [55,28,56,57]. Mainly gram-negative bacteria show anti-parasitic effects [56,57].

The *Anopheles* microbiota can impede *Plasmodium* colonization in the mosquito gut mainly by activating the mosquito's immune responses and producing anti-Plasmodium metabolites, which directly reduce parasite survival. Blood sucking stimulates the very fast multiplication of mosquito gut bacteria and triggers antimicrobial immune responses. Within the midgut epithelium, the Imd pathway mainly triggers these immune responses. The Imd pathway is stimulated by the binding of bacterial cell wall peptidoglycan with "Peptidoglycan Recognition Protein (PGRP)" of the midgut epithelium of the mosquito vector [58,59,60]. The Imd pathway enhances theexpression of TEP1. It has been reported that TEP1 is associated with the microbiota-mediated control of *Plasmodium* infection [61,62]. Gao et al. (2019) found that the ookinete numbers were reduced in the mosquito gut epithelium,which was dependent on microbiota load. It suggests microbiota-induced immunity may quickly affect *Pasmodium* infection even before the complement system action [63]. Metabolites of some micro-organisms locating within the mosquito gut can directly impair the colonization of *Plasmodium* and suppress its infective potential to the mosquito.

Gram-negative enterobacteria are harbouring in *An. arabiensis* mosquito gut was found to diminish a load of various stages of *P. falciparum* in *An. gambiae* through the generation of reactive oxygen species (ROS) and inhibition of the antioxidant parameters of the parasite [28,64]. Antioxidant enzymes in the mosquito gut have been documented to be able to be inhibited by *Plasmodium* infection, and their interaction has been proven to be mutual. This incidence reduces the mosquito microbiota and consequently enhances pathogenic infection [65]. *Serratia marcescens* and *Chromobacterium spp* are reported to reduce the *P. falciparum* infection in *Anopheles* mosquito and exhibited *in*

vitro anti-parasitic activity through secreting metabolites [59,66]. In addition, the yeast *Wickerhamomyces anomalus* from the gut of *An. stephensi* produces a lethal toxin with β-1,3-glucanase activity. Thus it can inhibit *in vitro* development of *P. berghei* ookinetes [67].

Besides, the effects of *Anopheles* microbiota on *Plasmodium* and some other consequences are indirectly examined in several literatures. Moreover, the microbiota plays a role in the production of the peritrophic matrix that is found in the gut epithelium. Peritrophic matrix is a chitin and protein-composed layer that has been synthesized after blood-feeding surrounding the midgut epithelium. This barrier prevents malaria parasites from entering the body cavity and prevents germs from the midgut from entering the body cavity [35]. *Plasmodium* ookinetes synthesize and secrete chitinase enzyme to cross this peritrophic matrix [21]. With this stance, one can say that this mechanism may be a microbiota-induced colonization resistance operation against *Plasmodium*. Moreover, the microbiota can also nutritionally affect the malaria parasite in the mosquito gut. The sequencing of the entire genomes of the bacteria that make up the mosquito microbiome led to the discovery that these bacteria have genes that are involved in the digestion of macromolecules found in the host [68]. However, it is yet to be known whether the symbiont with this digestive potential results in an advantage for the mosquito host, invading pathogens, or nutrient competition between the host and parasite. As for example, the endosymbionts in *Drosophila* cause resistance to virus infection partially due to competition for cholesterol [69]. It has been reported that *Plasmodium* manipulates its nutritional requirement at early developmental stages by overproducing digestive enzymes. So, nutrient availability is a limiting factor for *Plasmodium* colonization [70].

Some species of gut bacteria can directly alter the transmission of pathogens in mosquitoes while not affecting the vector's immune response. Bacteria such as *Enterobacter ludwigii*, *Pseudomonas rhodesiae*, and *Vagococcus salmoninarium* were isolated from the midgut of *Ae. albopictus* all block La Crosse virus infection simultaneously, which suggests that these bacteria may secrete anti-viral compounds into the environment in which they live [71]. Panama strain of *Chromobacterium* sp.produces an aminopeptidase that can break down the dengue virus envelope, thus decreasing dengue infection in *Ae. aegypti* [72]. *Chromobacterium sp.* also synthesizes rhomidepsin, an anti-parasitic protein that inhibits the infection of *P. falciparium* in *An. gambiae* [73]. An *Enterobacter* from wild *An. arabiensis* populations in Zambia were reported to produce ROS and can intervene with *P. falciparum* growth before the midgut epithelium invasion [9]. Mosquito gut bacteria may promote the infection rate of vectored pathogenic microbes. *Serratia odorifera* secretes a polypeptide, P40, which interacts with a cysteine-rich protein, prohibitin, which is needed for mosquito infection. Thus, this bacteria species inhibits the host's immune response and increases the host's susceptibility to virus infection [74,75,76]. *S. marcescens* secretes a protein named smEnhancin that digests peritrophic matrix-associated mucins and thus makes the vector more prone to an infectious virus [77]. The interplay between the mosquitoes and their gut-associated microbiota and transmitted pathogenic organisms is not only a one-way relationship, but it is obvious that vectored pathogens can shape the composition of bacterial population and the microbial load in the mosquito midgut. For example, *P.vivax* significantly reduces the microbial load during the pre-invasive phase when ookinetes and oocysts infect the host gut [78]. Thus *Plasmodium* can control bacterial growth before ookinete invasion by reducing the microbiota-influenced immune response. Besides, the Zika virus can change the composition of the microbial community in *Ae. aegypti* [79] and the Chikungunya virus enhances the multiplication of Enterobacteriaceae in *Ae. Albopictus* [80].

5. PARATRANSGENESIS: MICROBIOTA-MEDIATED CONTROL OF VECTOR-BORNE DISEASES

Bacteria, viruses or parasites colonize and replicate in various mosquito organs, mostly midgut and rarely salivary glands, hemolymph, male accessory glands and female ovaries [81]. Though, most research done so farhasfocused on gut microbiota. The midgut and ovary of adult mosquitoes share several prevailing bacteria classes and some other bacteria which are specific for tissues or developmental stages [82]. The accumulated knowledge about the insects and resident microbiota interaction promotes the progression of some microbiota-dependent interruption techniques for controlling mosquito-borne diseases. The principal of these microbiota-dependent interruptions of vector-borne diseases is the use of genetically

manipulated symbionts for the production and secretion of effector molecules to decrease the vector competence for that parasite [83,84,21].

In Paratransgenesis, selecting a symbiont, which has to be manipulated genetically, is crucial [85]. A perfect candidate symbiont must have a stable relationship with its insect vector, be able to transmit vertically or horizontally, and persist for an extended period to create effectors [86]. Second, the symbiotic bacteria should be simple to cultivate and genetically modifiable [84]. Thirdly, the designed symbiont should not affect the host's fitness and should be as suitable as the wild type one [83]. Finally, the symbiont must create and secrete some hostile chemicals to interact with the virus more effectively [84]. Various projects regarding paratrangenesis have been explored as a parasitic disease control technique. Riehle et al. (2007) reported that engineered *E. coli* expresses the two antimalarial molecules, phospholipase, salivary gland, and midgut peptide 1. But the bacterium could not persist long in the *Anopheles* gut, and the expression of functional phospholipase was toxic to the bacterium [87].

The symbiotic bacteria of the mosquito belonging to *Asaia, Pantoea* and *Serratia* have been scrutinized as promising for the paratransgenesis technique. *Pantoea agglomerans* are distributed in the gut of *Anopheles*. This non-pathogenic bacterium has been engineered for the production and secretion of a few anti-*Plasmodium* factors. [87,17]. *Serratia* colonizes males and females of *An. stephensi* mosquitoes which have a very low fitness cost for that insect [88]. *Asaia,* a symbiotic bacterium of *Anopheles* and *Aedes* mosquitoes, has been engineered to produce effector molecules showing inhibitory efficacy against *P. berghei* [89,90]. Recently, a modified strain of *Asaia*, is reported as proficient in stimulating the host immune system against the heartworm (*Dirofilaria immitis*) infection [91]. Paratransgenic technique had also been examined on trypanosomiasis and leishmaniases. The symbiont *Sodalis* has been identified as a promising micro-organism to block trypanosome transmission in the tsetse flies. An effector molecule from the engineered *Sodalis* 'attacin' has been identified as a potential inducible immune peptide showing efficacy against protozoans and some gram-negative bacteria [92].

Some non-bacterial micro-organisms have been studied for their prospective use in paratransgenesis. *Metarhizium robertsii*, a fungus [21], densonucleosis viruses [93,94] are found to be a promising agent for paratransgenesis.

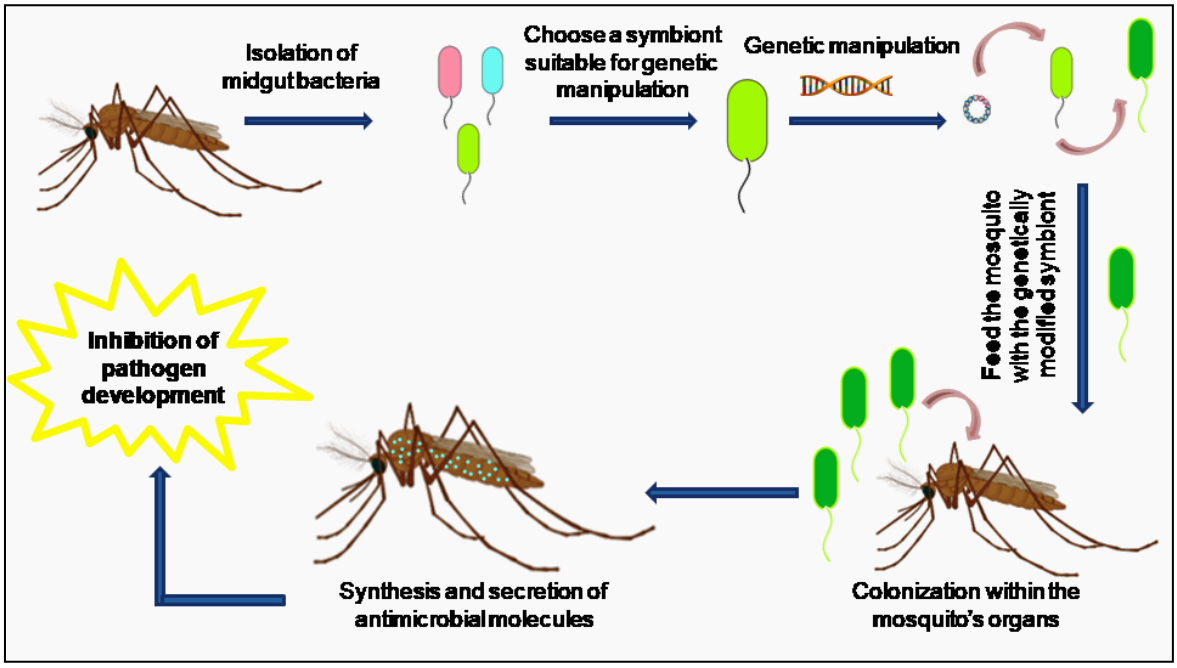

Fig. 2. Application of paratransgenesis to control mosquito-borne diseases by inhibiting pathogen development within the mosquito vector

6. FUTURE PERSPECTIVE ON THE ROLE OF GUT MICROBIOTA IN ERADICATING MOSQUITO-TRANSMITTED INFECTIOUS DISEASES

Mosquito midgut hosts a variable and dynamic microbiota. The composition of microbial communities differs significantly between species or individuals of the same species. This variation mostly depends on environmental factors (seasonality, larval breeding site), individual history (diet, blood-feeding history), and also on host genetic identity, which can strongly influence the content of midgut bacteria [11]. Several studies have reported the high diversity in the microbial composition between individuals of the same habitat independently of the mosquito species [59]. Moreover, it has also been reported that the mosquito's microbial composition is not random, some bacterial genera are routinely found in individuals of the same species. These Gram-negative bacteria are mostly aerobic or facultative aerobic, and belong to the families Acetobacteraceae, Enterobacteriaceae and Flavobacteriaceae.

Anopheles mosquitoes mainly host several typical microorganisms or enterotypes rather than a core microbiota obtained from their environment, individual history and host genetics. Remarkably, laboratory-reared mosquitoes exhibit a lower diversity in the midgut bacterial community than field-collected ones, which share most of the bacteria species in common [33,95]. But most of the research on the interactions between infected pathogens and the microbiota harbouring the vector has been done on laboratory-reared mosquitoes. In the case of laboratory-reared mosquitoes, the composition of microbiota differs between insectaries due to the variations in husbandry and habitat [95]. Thus this variability can explain the discrepancies in results obtained from various vector biology laboratories. Boissiere et al. (2017) and Tchioffo et al. (2016) reported that the abundance of *Enterobacteriaceae* or *S. marcescens*, a gram-negative bacterium in *An. gambiae* adults derived from field-collected larvae showed a positive correlation with parasite infection. Furthermore, suitable microbiota models are urgently needed to more elaborately understand the tripartite relationship between the microbiota, various pathogens and their mosquito host. Selecting the most relevant model to answer the scientific queries and merging the use of such relevant models may propagate the current understanding of vector biology. It may also help control parasitic disease transmission [10].

7. CONCLUSION

In the last two decades, the crucial role of the microbial flora of mosquitoes in the invasion of pathogen, development of infection and transmission of pathogenic infection has gradually flourished. Moreover, the tripartite interplay between the pathogen, the mosquito vector and its microbiota may answer the most thirsting queries about controlling various mosquito-transmitted diseases. Thus this complex relationship still needs more detailed investigation. The *Anopheles* microbiota can decrease malarial infection. They can also manipulate the host's physiological processes, including its lifespan. The microbiota-mediated effects remain compatible irrespective of the species of *Anopheles* and *Plasmodium,* which suggests that the interaction between mosquito, microbiota and *Plasmodium* is a stable system and each plays a significant role in shaping the tripartite interaction. Although our understanding of the bacterial part of mosquito microbiota continuously expands, numerous elements surrounding the microbiota-mediated prevention of vector-borne diseases have not been entirely clarified yet. This represents the main challenge of this domain. The bacterial components of the mosquito microbial flora have mostly received more research attention than the non-bacterial components. It has been suggested that eukaryotes and viruses could inhibit pathogenic infection. Yet, it is also unclear if the microbes found in the salivary glands or reproductive tract impair disease transmission or host fitness. Also, the majority of the microbiome study conducted up to now used mosquitoes reared in laboratories. Yet, their microbial community differs from that of mosquitoes that were taken from the wild.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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