Technical Opinion on Examination Request presented at the 171st Plenary Meeting of the National Technical Commission on Biosafety (CTNBio), held on April 10th, 2014

Procedure: 01200.002919/2013-77
Applicant: Oxitec do Brasil Participações Ltd.

1. Presentation

The Oxitec do Brasil Participações Ltda. (CQB 357/13) requests authorization for the commercial release of the OX513A lineage of *Aedes aegypti*, genetically modified for control – by population reduction – of the wild mosquito, carrier of the dengue virus (DENV).

Filed on 03/07/2013; Protocol 28300/2013; Previous Statement 3676/2013 published on 15/07/2013. The process received favorable opinions of the drafters Mário Hiroyuki Hirata, João Santana da Silva and Odir Antônio Dellagostin (in the Permanent Sector Subcomissions of Human and Animal Health) and Francisco José Lima Aragão and Fernando Hercos Valicente (in the Permanent Sector Subcomissions of the Plant and Environmental Areas).

The present report corresponds to an examination request of the commercial release process, solicited at the 170th Ordinary Meeting of CTNBio on March 13th, 2014, under the responsibility of Leonardo Melgarejo and Antônio Inácio Andrioli. Allan Edver (Permanent Sector Subcomissions of Human and Animal Health) and Orlando Cardoso (Permanent Sector Subcomissions of the Plant and Environmental Areas) who serve as advisors for CTNBio;

2. Initial Comments

The importance of the theme is unmistakable. The dengue fever advancing in the country, the emerging resistance – among vectors – to insecticides used, the harm to the health of the population , social and environmental economic costs and the need for innovative methods to combat the disease, which are more than well known, provide pressure for quick acceptance of alternative proposals.

The project is well informed and the three studies referred in Cayman, Malaysia and Brazil (Juazeiro, State of Bahia, during 2012 and 2013) present interesting preliminary results, showing it to be a promising alternative in the fight against dengue.

However, data is insufficient to assert a steady position, as is demonstrated below. In this perspective, the present report recommends the process should be put into DILIGENCE until the gaps referred to here are solved in a consistent manner.

Among the highlighted points, consider that:

2.1. The treatment provided by CTNBio deserves revision, for it differentiates itself from others in ways that are exceptional

The process regarding the Planned Release into the Environment (LPMA) that precede the request for commercial release are not yet concluded. It is possible to affirm this situation is unprecedented and the precedents already revealed threaten CTNBio's credibility. The LPMAs are instruments that provide inputs to commercial release processes and should be conducted in all ecosystems relevant to risk assessment and in all Brazilian biomes, in order to meet the demands of the current legislation.

What motives would justify the premature acceptance of preliminary data by CTNBio that, in this case, configures an anticipated assessment of the final reports, opposing the practices used so far, that are recommended by this commission? Furthermore, what circumstances would justify the fact representatives for the applicant of the technology have been invited to attend a meeting where the technology would be evaluated and, perform an exposure of merit that could be confused with institutional marketing and creating possibility of inducing CTNBio members to the approval of its demand?

If these conditions weren't enough to suspend the present assessment on their own, the impact of these concessions should be considered, regarding equality of treatment, considering all processes being currently evaluated and the ones to be evaluated in the future, forwarded by applicants of innovative technogies in the field of genetic engineering. From now on, are the requests for commercial release exempt from including completion reports of LPMA requests that sustain them?

What arguments justify the contempt for the Biosafety Law that demands LPMA studies in all Brazilian biomes? Would it be acceptable that allegedly "preliminary" information collected in Bahia, should attend to peculiarities from Pampa, the Amazon or Pantanal, where the environmental conditions that affect the dynamics of mosquito populations are clearly distinct? In addition, in this case, would it be prudent that

CTNBio continued breaching this requirement when a Brazilian court decision recently suspended the release of transgenic T-25 corn, based on the argument that no studies had been conducted in the North and Northeastern biomes, prohibiting its cultivation in those regions?

2.2. There is a glaring inadequacy of CTNBio protocols to assess winged insects risks

The implications of this matter are evident: when adequate guidelines to assess winged insects are not available, CTNBio is likely to decide on the unprecedented possibility to authorize the release of a living transgenic being that do not have effective restrictions in regards to spread, based on guidelines created for the purpose of assessing risks associated to cultivated plants. The fact that the vector to be controlled by transgenic mosquitoes that were to be eradicated from Brazil in the 1970s, is present throughout the country, does not make it a less severe issue, despite the mosquitoes' autonomous flight capability not exceeding 200 meters. Additionally, the fact that the basic control systems (release of males and sterility) possess recognized failures is anything but irrelevant. Even the mortality rate of larvae in the absence of tetracycline presents failure levels of 5%, in ideal lab conditions for research.

Therefore, the consideration that the valid guidelines have been met, does not seem sufficiently safe. They just do not apply to the problems in question. The applicant itself recognizes the serious fact that Normative Resolution No. 5 of CTNBio does not contemplate the peculiarities in the case, and does not offer an annex to specifically assess topics on health and environmental risks related to transgenic insects. It is worth noting that only cases related to "organisms consumed as food" and "microorganisms used as vaccines" are planned, concerning risk assessment efforts for human and animal health.

In this sense, since there are no normative instructions to assess the transgenic organism submitted by Oxitec, it is surprising that one of the opinions approved by the Permanent Sector Subcomissions of Human and Animal Health related to risks to animals that would eventually consume that mosquito affirmed that "the evaluation of these parameters was a result of complying with requirements on human and animal health, as present in CTNBio's Normative Resolution No. 5". In respect to the Precautionary Principle , the establishment of robust guidelines in advance would be wise, capable of guiding the evaluation process of transgenic insects, with effective conditions to decide their own implications for human health and the environment.

It has to be stressed that all opinions that support the request for commercial release (including the consolidated one) consider the OX513A mosquito **Risk Class I**, when the applicant company understands the issue as distinct and deserving of greater caution. On page 67 of the dossier presented by the applicant it can be read that "the risk classification of the *Aedes aegypti* OX513A was evaluated and in accordance with Normative Resolution No. 2 of November 27th, 2006, it was established as **Risk Class II**: moderate individual risk and low collectivity risk".

This topic should be clarified before any decision. In its statement, the company affirmed that it works with Risk Class II events, and it benefited from a Quality Certificate in Biosecurity Class NB-1 (in accordance with the evaluator Mário Hiroyuki Hirata) and developed the planned release into the environment based on CTNBio Normative Resolution No. 7, which is restricted to genetically modified, Risk Class I organisms. If the transgenic mosquito is classified as Risk Class II, the LPMA then followed, at least, the guidance of an "inadequate" Normative Resolution.

3. Risk assessment associated with the introduction of massive quantities of OX513A into the environment

The dossier presented by the applicant company presents a vast set of scientific data, complemented by a rich bibliographic review, covering aspects pertaining to the biology of the *A. aegypti*, associated risks on environementincluding the OX513A in trophic chains and potential consequences of releasing genetically modified females undesirably. However, the process lacks certain biosafety aspects:

3.1. The occupation of the ecological niche of A. aegypti by A. albopictus has not received sufficient attention from the dossier and the other evaluators

The large-scale release of OX513A, altering the reproductive performance of the *Aedes aegypti*, can trigger a population explosion of other vectors, with implications for adaptive dengue virus mechanisms in epidemiological terms and consequences for public health. Therefore, it is important to check the possibility of alterations in hosts, vectors, or even infectious profiles.

The data pointed to as preliminary were collected in three locations evaluated on a planetary scale, and suggested high effectiveness of the technology. The reduction of 95% of the local population of *A. aegypti* in

Brazil is impressive, after treating the area for six months (adult population estimated by marking-release-recapture statistics, according to page 36 of the dossier submitted by the company). These field results, in spite of the adversities of studies of this type would have surpassed even those obtained under controlled laboratory conditions. This successful endeavor should also be perceived as an additional reason for repeating tests.

The alterations made by releasing hundreds of thousand transgenic mosquitoes with the characteristic of letality passed down to *Aedes aegypti* descendants will benefit other insects. As local populations of *A. aegypti* compete with local populations of *A. albopictus* (species that have invasive ecologic characteristics) wouldn't the suppresion of the first favor a population explosion of the second?

Available references suggest *A. albopictus* is adapted to the peridomestic environment just as *A. aegypti*, where it feeds from human and animal blood, laying eggs in many natural and artificial water-accumulating containers (Hawley, 1988, quoted in Lambrechts et al., 2010). Scientific reports support the fact that up to the XVIII and XIX centuries, *A. albopictus* was the most frequent daytime biting species in the majority of the cities in Asia (Gilotra et al., 1967 quoted in Lambrechts et al., 2010), having since lost space due to conditions that benefited its main competitor. As the naval industry expanded (commerce, then tourism), *A. aegypti* started to dominate ecological niches occupied by *A. albopictus*, becoming progressively the main daytime biting species in some Asian cities. Urbanization conditions and *Aedes aegypti*'s greater adaptation to the urban environment (Macdonald, 1956 quoted in Lambrechts et al., 2010) were decisive for such changes, and tend to be eroded following massive releases of OX513A.

The inclusion of *A. albopictus* in the list of the world's 100 most invasive species leaves no doubts as to its agressiveness and potential to occupy that ecological niche. In other words: the almost complete suppresion of local populations of *A. aegypti* by the OX513A will possibly cause migration flows in local populations of *A. albopictus,* compromising the disease-reduction goal, for the simple fact that a new vector of the disease will occupy the ecological niche that was abruptly abandoned by the main competitor.

3.2. The ecological imbalance caused by mass introduction of the OX513A into the environment can cause implications for the epidemiological profile of the dengue virus, aside from transmitting other viral human and zoonotic diseases

In the dossier and opinions favorable to Oxitec's demand, a thesis on a smaller capacity/efficiency of the *A. albopitus* to transmit the dengue virus in an epidemic manner (compared to the *A. aegypti*) was found. Thus, this conclusion omits scientific literature which describes viruses' adaptation/mutation cases to other hosts and vectors. A more careful interpretation considers that evolutionary forces are at stake, highlighting mutation-selection pressures, which tend to stimulate responses to the dengue virus in the absence of its main vector (*A. aegypti*).

Some cases studied demonstrate that arboviruses could rapidly alter associations with hosts/vectors. For example, epidemics caused by the Venezuelan Equine Encephalitis virus (VEE) in several countries in Central and South Americas in the mid-1990s. According to Brault and collaborators, the Mexican epidemic in the 1993-1996 period was unleashed due to the virus adaptating to an alternative vector (with increased epizootic capacity), based on the substitution of a single aminoacid from a glycoprotein envelope (Brault et al., 2002 and 2004). According to Anishchenko et al. (2006), however, the epidemic/epizootic characteristic of the VEE would have been acquired/unleashed by a single mutation in viral strains only present (so far) in its enzootic form. It is possible to perceive in any of the hypotheses above that those studies point to high probability of alterations in the infectious profile of said viruses (starting from a single mutation), reaching high disease transmission capabilities in an epizootic/epidemic form.

Additionally, in the chikungunya epidemic in the island of La Réunion in the 2005-2006 period *A. albopictus* was the main vector, while that role is normally played by *A. aegypti*. Tsetsarkin et al. (2007 and 2009) concluded that a mutation in the CHIK virus was directly responsible for a significant increase of the patogen's inefectiveness, through a vector that was much involved in the transmission of the disease, *A. albopictus*. This mutation would have allowed the virus a greater dissemination efficency of the viral load in the mosquito's secondary organs and, consequently, greater efficacy in transmitting the disease to hosts.

Therefore, considering the hypothesis that mass releasing of the OX513 mosquito will cause mass occupation of the *A. aegypti*'s ecological niche by *A. albopictus*, this could cause changes in the dengue virus' epidemiologic profile, as well as in other viral diseases (human, animal and zoonotic). These are some of issues that were not examined in the dossier.

A reduction in the detected dengue cases can be expected at first. They would then occur sporadically and non-epidemically, due to the slow occupation efforts of ecological niches and the *A. albopictus*' lesser competence (compared to the *A. aegypti*) when transmitting the disease. Next, the suppresion of the virus' main epidemic vector will exert selective pressure potentially favorable to genetic mutations of local strains of the dengue virus, causing implications in the epidemiologic profile of the disease. In these conditions, considering the available scientific literature, we can elaborate at least two hypotheses:

a) Hypothesis based on the experience adquired with the Venezuelan Equine Encephalitis virus

Mutations in the dengue virus strains - which are present today in association with *A. albopictus* but without the capacity to unleash epidemics - could occur. These mutations could infect other vectors which are more prone to causing epidemics. Theoretically, any of the several species of mosquitoes that are vectors for arboviral pathologies present in Brazil (whether from the *Aedes* gender or a genetically close configuration) could take on this role. That species would then become a new epidemic vector for the dengue virus, cohexisting with the *A. albopictus* despite its competitiveness in urban zones.

b) Hypothesis based on the experience acquired with the recent epidemic caused by the chikungunya virus

Mutations in dengue strains that would allow *A. albopictus* to become a highly efficient transmission vector could occur, getting around the immunological properties provided by the symbiote bacteria Wolbachia (as it was with the CHIKV). In that case, *A. albopictus* would become the dengue virus' main epidemic vector.

In both cases, a new epidemic vector for the dengue virus would replace *A. aegypti*, followed by new risks. In said conditions, the change in vector would mean alterations in the infectivity mechanisms of the dengue virus itself, making its control by health agencies more complex.

Additionally, mass releases of the OX513A into urban zones could favor the entry of other human, animal and zoonotic viral diseases, which do not occur today thanks to the occupation of the ecological niche by *A. aegypti*, that is not a vector for these diseases. Considering that *A. albopictus* on its own, facing the current conditions, it is possible to speculate on risks involving the whole set of viral diseases, whether human, animal or zoonotic which that species hosts.

Considering the predictable hypothesis that some CTNBio members shall take the occurrence of mutation-selection processes as highly speculative, we draw attention to the fact that the greatest part of RNA-based viruses have a mutation frequency so elevated that it could reach 10E-4 (0.0001) mutants per nucleotide, according to Weaver et al. (1993). In the case of the EEV epidemic, Anishchenko et al. (2006) estimated that the mutant capable of creating an epidemic amplification (having suffered only one mutation – as in the chikungunya epidemic case already referred to) could be produced from the moment the total population of VEEV reached 10E4 (10,000) individuals (which represents a relatively small population for arboviruses).

These risks have been approached superficially in the dossier, and the favorable opinions on the commercial release of OX513A mosquitoes do not comment on them very much. The applicant and CTNBio's evaluators who are favorable to the applicant focused on the *A. aegypti*'s biology (adaptation capacity to the DENV and other viral diseases, especially), and did not assess the risks associated with the colonization of urban areas treated with the OX513A by the *A. albopictus* and other vector species.

It is a known fact that the *A. albopictus* is susceptible to infection and is capable of transmitting most viruses that have been tested on it. The list includes 8 alphaviruses, 8 flaviviruses and 4 bunyaviruses, representing the three main types of arbovirus that include human pathogens (revised in Paupy et al., 2009). In this sense, besides transmitting dengue, *A. albopictus* also transmits yellow fever and the chikungunya virus (Hochedez et al., 2006), as well as other viral diseases. It is worth noting the recent chikungunya epidemics in the Indian Ocean islands (especially La Réunion), in Central Africa (Gabon, among other countries) and in Italy, derived from the *A. albopictus* vector (Lambrechts et al., 2010).

Furthermore, *A. albopictus* feeds on a vast variety of animal species, and is recognized as a vector with high potential for transmitting zoonotic pathogens (from animals to humans). This is exactly why the La Cross and Eastern Equine Encephalitis (EEE) viruses are major causes for concern for public health care in the USA. The quoted authors also warn that *A. albopictus* deserves special attention in the South and Central Americas, for it is a vector of yellow fever and Venezuelan Equine Encephalitis viruses. At this point, it is worth noticing that the EEE, VEE and WEE (Western Equine Encephalitis) viruses are present in Brazil (Kotait et al., 2006;

Figueiredo, 2007). It is also worth noticing the West Nile virus (already detected in Brazil, as it is informed on page 350 of the dossier), although it has never caused an epidemic in Brazil. The virus is responsible for a zoonosis that's also transmitted to humans by *A. albopictus*.

Therefore, given the evidence presented in scientific studies, it is necessary to examine the possibility that the abrupt emptying of the ecological niche occupied by *A. aegypti* will tend to strengthen the invasive capacity of local populations of *A. albopictus*. Its implications aren't restricted to the dengue fever, for they extend to other arboviral diseases and several zoonoses that could be brought from peri-urban zones into urban zones. In this sense, considering the Precautionary Principle , this issue needs to be addressed more carefully.

3.3. The dossier presented by the applicant and the favorable opinions tend to minimize the consequences of ecologic disturbances for public health care

The applicant requests that the "target species of biological control" is the *A. aegypti* and in this perspective, elaborates answers for item E 1 in Annex IV of RN5 (p.560). However, the revelance of the matter is in the fact that the dengue fever is a viral disease of dramatic connotations. Thus, the target species only acquires practical sense regarding controlling the dengue virus, so the *Flavivirus sp. (DENV)* would be the target species for Biological Control.

Therefore, the company provided answers that approach the real problem indirectly, and that were wrong for a great deal of the subjects presented in item E. In these conditions the process is weak, omitting health risks associated with the occupation of the *A. aegypti* ecological niche by the *A. albopictus*, as well as possible consequences stemming from this fact, in terms of eventual viral adaptations (of the DENV and other human and animal viruses) and its implicatios, like new epidemics/epizootics and the increased complexity of treatment systems.

On the other hand, the applicant approached this question in a partial manner in item 2.5 of the dossier, where it refers to the "evaluation of the substitution potential for other pathogenic vectors" (p. 338). At that time, the applicant distorts the issue, minimizing its probability tof occurence as well as potential consquences. It literally affirms that: "however, there's still a slight risk that the *A. albopictus* takes over the ecological niche abandoned by the *A. aegypti*.", p.340. But, as we have explained earlier, the probability for this to occur in context, seems to range from "high" to “moderate”. It is worth noting that the group of specialists created within the scope of the *Capacity Building for Implementation of Malaysia’s Biosafety Act 2007* project, has pointed out that the risk associated with the *A. albopictus* occupying the ecological niche is moderate (Beech et al., 2009).

The company further states that “the *Aedes aegypti* is an invasive species in Brazil; it was eradicated and returned in the 1970s. As consequence, since the insect does not have a vast history in the country, its suppression or local elimination might be considered a reversion to the pre-introductory stage of the species” (p. 338). This assertion is obviously a mistake. It does not only disregard the set of socio-environmental changes that took place over the last 40 years, with its implications relating to changes in the species’ habitat, but it distances itself from the geographic expansion of *A. albopictus*. In addition, it ignores the revolution in urbanization, in means of transportation, in animal breeding systems, in the agroindustries around urban centers, in the standardization of rations and in tetracycline usage, among other factors related to this case of viral epidemiology. It would be naive to assume the specific and abrupt exclusion of *A. aegypti* locals populations today would simply reconstruct the same conditions observed in the 1970s, in terms of epidemiologic risk of viral diseases, including dengue fever.

The company also states that “the possible adverse effects for removing *A. aegypti* aren’t specific to the use of OX513A mosquitos, and would apply to any effective methods of mosquito control. Therefore, it is not a new issue”. Once again we are facing a piece of information that is clearly mistaken.

We have in our hands an unprecedented situation where, in terms of history of epidemiology, a technology seems capable of eliminating 95% of the local individuals of a specific species (*A. aegypti*) in the short period of 6 months. The control methods were, so far, unspecific, and systemically hit all mosquito populations of the majority of species (if not all) present in the treated area.

Concerning the possible consequences of ecological niche occupation by *A. albopictus* at the sites where the OX513A is to be mass released, the company affirms that “an important recent revision concluded that *A. albopictus* is a lot less effective as a vector for the dengue virus than *A. Aegypti”* and that “Lambrechts et al. (2010) clarified several aspects by observing lineages of *A. albopictus* becoming more susceptible to the dengue virus after various generations created in a laboratory and that, furthermore, lab studies have the tendency to overestimate the role of this species as a vector for the dengue virus”. In this aspect the available scientific bibliography suggests the transmission capacity of the DENV to humans (from *A. albopictus*) might derive from the presence of a symbiotic bacteria – of the Wolbachia genus – that hosts itself in *A. albopictus* individuals. That condition, representing a barrier for the infection of these mosquitoes by the DENV and other arboviruses, reduces its potential to transmit diseases to humans. The recent chikungunya epidemics have shown the arboviruses to be capable of avoiding immunological barriers of *A. albopictus*, - which has become the main disease vector in these specific cases, replacing *A. aegypti*.

On the same topic, the applicant hurries to conclude that “both *A. albopictus* and *A. aegypti* are capable of transmitting viruses and pathogens, but there is no reason to think the replacement of *A. aegypti* by *A. albopictus* might have any negative effect upon human health or the environment (Gratz, 2004; Lambrechts et al., 2010; Moore and Mitchell, 1997)”. At this stage, one can notice contempt in regards to the knowledge provided by the chikungunya epidemics – and the alterations in epidemiologic transmission profiles – contradicting references quoted in the dossier to support this conclusion. Lambrechts et al. (2010) indeed conclude – on the natural increase of the *A. albopictus* distribution zone – that this species could present lesser risks in relation to DENV transmission in its epidemic forms, in comparison to *A. aegypti*. But they also concluded that “however, we can not dismiss the fact that at some future date, the occupation of territories by *A. albopictus* will be followed by the virus adapting to this species of vector mosquitoes [*A. albopictus*], invasive and in constant effective increase, followed by a global reemergence of chikungunya among other arboviral diseases”. It is worth noticing that the expression “at some future date” should be interpreted in the context hereby described, where the occupation of territories by the *A. albopictus* in “natural” conditions is analyzed, where there is an intense competition between the two species, and not in a context where 95% of the *A. aegypti* pertaining to local populations would be supressed in 6 months.

Therefore, once again: the mass release of OX513A mosquitos shall prevail, unprecedented in the establishment of large and perennial populations of *A. albopictus* in the urban zones, which are normally competition areas against the *A. aegypti*. Alterations on the main competitive species’ fitness that are not very deep shall, doubtless, modify the dynamics of the populations of *A. albopictus*. In parallel, altering the fitness of the main vector for specific diseases will also change the dynamics of viral populations which will be unable to complete their reproductive cycles, favoring any mutation capable of rebalancing their infestation levels in those areas. The VEEV and CHIKV examples picture the high capacity (or in evolutionary terms, “probability”) of the arboviruses to change hosts and/or alter the vectoral competence of specific species, including *A. albopictus*.

Finally, it is worth mentioning that, at no time does the dossier evaluate the potential for transmitting zoonoses and epizootics for local human and animal populations, respectively, through *A. albopictus*. This species forms an efficient bridge to conect viral diseases from peri-urban zones to the urban zones to be occupied by it.

The risks for public health in mass releasing OX513A into urban areas due to the occupation of *A. aegypti*’s ecological niche also seem not to be appropriately considered in the favorable opinions submitted to analysis by CTNBio. Doctor Fernando Hercos Valicente, for example, dismisses these risks, affirming that “occupying empty niches left by a different species, in the case of the *Aedes albopictus* which can also be a vector species, is difficult to occur”. That is because “ *A. albopictus* is essentially wild and only appears at cities close to woods or large gardens with a great number of trees. It never invades the extense areas of the city, far from important plant coverage”. These affirmatives could be easily rejected based on the current knowledge on the ecology of *A. albopictus*. According to Lambrechts et al., 2010 and references quoted, *A. albopictus* can occupy large urban areas, especially in the absence of *A. aegypti*. The statements also neglect the ecologic consequences, in terms of population dynamics, of quick and abrupt supression of the *A. albopictus*’ main competitor.

On the other hand, the applicant company emphasizes the risks associated with the occupation of the ecological niche by *A. albopictus*, and recommends the monitoring of these populations. However, it is suggested that this surveillance effort takes place only after the commercial release of OX513A has been approved. What is the justification for analyses in of such great importance to take place only after the commercial release has been approved?

In these evaluators’ perspectives it is unnacceptable to delay the data collection to after the approval, for it should result from field studies requested by the Biosafety Law in all relevant biomes. This data should be provided to CTNBio in the dossier that requests the comercial approval of the event. Among the omissions which are necessary for a solid decision, we highlight that the rates and recolonization profiles of areas where the

OX513A was/will be released are not informed/known, both to *A. aegypti* and *A. Albopictus* populations.

It is surprising that in this request for the commercial release of a transgenic insect, the qualitative and quantitative presence of the second species to be impacted the most – *A. albopictus* – is no longer analyzed, and no bibliographic references nor field studies approaching this issue exist. These omissions reveal a structural failure in this commercial release process: the absence of CTNBio guidelines that are coherent with the risks involved in this kind of release.

Lastly, and still relevant, these evaluators consider that the dossier fails by not presenting information relative to the potential of epidemiologic adaptation of the main human, animal and zoonotic viral diseases in the *A. albopictus*, also considering the context at play, when the main vector tends to disappear almost completely from the treated areas, in an extremely reduced time interval.

4. Conclusion

At first, we should reflect on the potential consequences of the administrative mistakes that occurred during this process of commercial release, highlighting:

a) the absence of the Conclusion Report for Planned Release into the Environment (LPMA);

b) contradiction with the RN2 when considering the OX513A as Risk Class I in the LPMA processes and Quality Certificate in Biosecurity;

c) contradiction with the Biosafety Law, having submitted only two LPMAs in Brazil, while the referred law demands the establishment of at least one LPMA in each biome.

Second, it is worth noting the set of unprecedented difficulties CTNBio had to face when assessing this first transgenic insect. The evaluator does not have specific guidelines to assess health-related risks. Besides, the company has made a mistake when considering the target species for biological control was in fact the target insect for the transgenic project (or the commercial release), which also harmed the environmental assessment. Furthermore, the mass introduction of the OX513A mosquito illustrates the difficulty of socialization between areas of expertise considered to be separate at CTNBio. The position of the evaluators from the Permanent

Sector Subcommissions on Human and Animal Health seems to grant them greater “legitimacy” or “competence” when assessing alterations in the epidemiologic profiles of viral transmissions, after the disturbances in the dynamics of local populations of the main vector and its competitor took place. On the other hand, the Permanent Sector Subcommissions on Plant and Environmental Areas seem to be endowed with greater legitimacy or competence to assess questions pertaining to the population dynamics of insects. Also, the technical decisions will be transformed into conclusions that do not depend on knowledge and arguments involved, for they will be based on the number of votes.

We highlight that this type of decision becomes more fragile as it gets influenced by the procedures, by the non presentation of previous studies, by the admission of the interested party on arguments conducted before some (and not other) members and in the absence of the contradictory views. Evaluators from the Permanent Sector Subcommissions on Human and Animal Health state that they have not addressed environmental factors for there are two other Subcommissions charged with that task. The evaluators from the Permanent Sector Subcommissions on Plant and Environmental Areas, on the other hand, state that they have not addressed human and animal health aspects because there are two other Subcommissions charged with that task. Thus, the existence of a decision facilitator agreement is clear, distorting analytical procedures and running away from the scope of responsibilities attributed to CTNBio.

Finally, contrary to the evaluators who favor the commercial release of the OX513A, we examined a possible route for harm, not treated properly in the process. The damage could be caused through reemergence of human and/or animal viral epidemics of zoonotic origin (or not), pre-existing (or not) to the mass release of the OX513A, with a significant degradation of public health in these areas, as well as potencial negative social and economic consequences for the municipalities affected. The route will be carried out by *A. albopictus* occupying the ecological niche – resulting from mass releasing the OX513A mosquito – with associated changes in the epidemiologic profile of animal, human and zoonotic viruses, providing these with greater infectivity, through exchange of vectors and/or circumvention of immunological barriers of secondary vectors.

In this context, aggravated by the non-fulfillment of the current legislation; the non-existence of evaluation protrocols adequate to the assessment of risks involving flying insects; the insufficiency of studies presented; and the non-inclusion of final results from the field studies approved by CTNBio, we consider that the commercial release of OX513A in these conditions, presents relevant and irreversible risks for both health and environment, whose probability of occurrence ranges from high to moderate. We recommend the process should be put into DILIGENCE so it can be complemented, and that it should return for analysis in accordance with the guidelines to be established by CTNBio.

5. Forwarding Procedures

Once the diligence is approved, the applicant company shall:

a) Annex the Conclusion Reports on LPMAs carried out in Brazil;

b) Fulfil the Biosafety Law by performing LPMAs in all Brazilian biomes;

c) Provide extensive argumentation based on the published scientific literature and on the information obtained from the LPMAs, on the recolonization rates of the ecological niche left empty by the *A. aegypti*, monitoring the *A. aegypti* and *A. albopictus* species, as well as other vector species for human, animal and zoonotic arboviruses common to the region;

d) Provide extensive argumentation, both quantitative and qualitative on the capacity of epidemiologic adaptation of arboviruses – especially the ones with epidemic and epizootic profiles – to the main secondary vectors present in urban and peri-urban zones in Brazil.

In parallel, we request the Presidency of CTNBio to forward an evaluation request on the social and economic risks related to the OX513A technology to the National Biosafety Council (CNBS), taking into account the fact that information contained in the process suggests a negative/moderate cost-benefit ratio for the municipalities and general public health care services. We point out the human behavior is highlighted among the factors that unleash diseases. Recent studies associate epidemics to cases of assymptomatic infections, involving non-epidemic serotypes, where the role of human dengue reservoirs is not well understood in the dynamic of the disease. In this sense, several authors considerer human populations can disseminate the dengue virus more effectively than mosquitoes (Morrison et al., 1998; Harrington et al., 2005; Morrison et al., 2010; Honório et al., 2009), which raises scientific questions on the real advantages of controlling only the main vector in specific areas. In this sense, it is important to notice that the head of The Neglected, Tropical and Vector Borne Diseases Unit from the Pan American Health Organization (OPS), Luis Gerardo Castellano, said that there is not enough scientific evidence to clarify the benefits and advantages the genetically modified mosquito could bring to countries (Castellano, 2014).

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