Guidelines for Mosquito Control in Built-Up Areas in Europe

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European Mosquito Control Association (EMCA)

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Sections

1. Abbreviations 4
2. Glossary 5
3. Executive summary 9
4. Introduction 10
 4.1 Public Health context of mosquito-borne diseases (MBDs) 10
 4.2 Objectives of the document and expected outcomes 11
 4.3 Built-up areas 11
 4.4 Points of entry 12
 4.5 Public areas 12
 4.6 Private properties 12
 4.7 Governance – stakeholders 13
 4.8 Policies impacting built-up areas and mosquitoes 13
 4.9 Mosquito species relevant as pests and/or potential vectors of diseases in built-up areas 13
 4.10 References to Chapter 4 16
5. Methods 20
 5.1 General methodology 20
 5.2 Literature search by SRT teams 21
6. Surveillance/Monitoring 25
 6.1 Planning of a surveillance/monitoring project 25
 6.2 Surveillance/monitoring of adults 26
 6.3 Surveillance/monitoring of larvae 28
 6.4 Surveillance/monitoring of eggs 31
 6.5 References to Chapter 6 31
7. Control interventions 33
 7.1 Larval control 33
 7.2 Adult control 35
 7.3 Integrated vector management (IVM) 37
 7.4 Summary of non-evidence publications 38
8. Conclusions 42
9. Contributors and interests 43
References 44
Annex: All evidence profiles, sorted by sections 46

1. Abbreviations

Abbreviations

Bs	*Bacillus sphaericus*
Bti	*Bacillus thuringiensis* serotype *israelensis*
ECDC	European Centre for Disease Prevention and Control
EMCA	European Mosquito Control Association
ERG	External Review Group
GDG	Guidelines Development Group
GRADE	Grading of Recommendations Assessment, Development and Evaluation
IGRs	Insect growth regulators
IHR	International Health Regulations
IIT	Incompatible Insect Technique
IRS	Indoor residual spraying
IVM	Integrated vector management
JEV	Japanese encephalitis virus
LSM	Larval source management
MBD	Mosquito-borne disease
ORS	Outdoor residual spraying
PI	Principal Investigator
PICO	Population, Intervention, Comparator, Outcome(s)
PoE	Point of entry
PRISMA	Preferred Reporting Items for Systematic Reviews and Meta-Analyses
RVFV	Rift Valley fever virus
SINV	Sindbis virus
SIT	Sterile insect technique
SRT	Systematic review team
TDR	The Special Programme for Research and Training in Tropical Diseases
ULV	Ultra-low volume space treatment
USUV	Usutu virus
WHO	World Health Organization
WNV	West Nile virus

2. Glossary

(Adapted from WHO 2017 and ECDC 2014)

Adult mosquito control

Any intervention with or without chemical agents designed to kill adult mosquitoes, such as pyrethroid insecticides or trapping (e.g. mass trapping, lethal ovitraps) etc.

Bias

Bias is an overestimate or underestimate of the effect of an intervention.

Cluster-randomized controlled trial (cRCT)

This is a randomized controlled trial in which groups of structures, for example, rain water catch basins, households or villages are randomly allocated to receive either intervention or constitute the control group.

Confounding bias

This type of bias arises whenever a cause is common to both the outcome and the exposure. For example, in an observational study of the association between using house screening and the incidence of malaria, the relationship is likely to be confounded by socioeconomic status because people in houses that use screening are likely to have higher socioeconomic status, and they may have greater access to other protective measures against malaria, such as long-lasting insecticidal nets. This is a common type of bias in observational studies and non-randomized trials, but it can also occur in poorly randomized studies.

A variable that is on the causal pathway between the exposure and the outcome is not a confounder. For example, if indoor residual spraying reduces mosquito density, which results in lower malaria incidence, then mosquito density does not confound the relationship between exposure and outcome, even though it is associated with both.

Control group

This is the group of participants that receives no intervention, and this group thereby serves as a comparison group when the intervention results are evaluated.

Cross-sectional study

In an analytical cross-sectional study, information is collected at one point in time on the prevalence of the outcome of interest (for example, a disease or infection) and exposure (for example, the use of a protective intervention).

Cluster randomization

In this type of study, clusters are randomly assigned to either control or intervention groups. Clusters can be geographical areas (for example, sectors of a large city), communities (for example, villages), administrative units (for example, districts or regions), institutions (for example, schools), health facilities or households.

Confounding variables

The variables that are associated with both the exposure and the outcome but that do not lie on the causal pathway between exposure and outcome.

Consistency

Consistency refers to the level of heterogeneity in a study's results that remains after exploration of a priori hypotheses that might explain heterogeneity. The GRADE system (see below) suggests rating downgrading the quality of the evidence if a high level of inconsistency (heterogeneity) is present.

Controlled before-and-after

A study in which observations are made before and after the study implementation of an intervention in both the intervention group and a control group that does not receive the intervention; this is also known as a pre–post study.

Direct effect

In a cluster-randomized controlled trial, the direct effect of the intervention can be estimated within the intervention clusters by comparing the incidence of the outcome in those receiving the intervention with that in those not receiving it.

Directness

This refers to the generalizability of the population, intervention, comparator and outcomes from each study to the population of interest. For example, evidence from a study conducted in soldiers may be downgraded when applying this evidence to the general population. The GRADE approach suggests downgrading the quality of evidence if the study population, intervention, comparator or outcomes differ from those in the population of interest.

Dose response

This term refers to the relationship between the exposure and the outcome. There is a dose response when there is a decreasing or increasing trend in the outcome in relation to increasing levels of exposure.

Effectiveness study

These studies estimate the effect of an intervention under pragmatic (or real-life) conditions (for example, interventions delivered under routine conditions) so that the relevance of the findings for policy and practice is maximized.

Effect size

This refers to the magnitude of difference between the treatment and control groups, expressed, for example, as the risk, rate ratio or percentage reduction in prevalence.

Exotic mosquito species

A mosquito species or subspecies that is not native to an ecosystem, and if present, has been introduced. (syn: alien, foreign, non-indigenous, non-native)

Experimental study

In this type of study design, the interventions are allocated to study participants and the outcomes are observed.

Experimental unit

This refers to a participant or group of participants exposed to a particular treatment.

GRADE

The Grading of Recommendations Assessment, Development and Evaluation method is a systematic and explicit approach to making judgements about the quality of a body of evidence and the strength of recommendations made from that evidence.

Invasive mosquito species
is an exotic mosquitoe species that establishes and proliferates within an ecosystem and whose introduction causes, or is likely to cause, economic or environmental harm, or harm to human health.

Loss to follow-up

This refers to participants who are not able to be followed up during the study, generally because they are cannot be reached. High levels of loss to follow-up can introduce bias if the loss differs between study arms.

Mass effect

These are the additional effects of an intervention that are achieved when a substantial proportion of the population receives the intervention. For example, in some circumstances, mass killing of mosquitoes coming into contact with long-lasting insecticidal nets can reduce the transmission of a disease so that indirect protection is provided to those individuals not using the nets.

Matching

In this technique, clusters are formed into groups such that only one cluster in each group is assigned to each study arm. Matching is typically done using the baseline values of the end point of interest or a surrogate variable that is expected to be correlated with the end point.

Monitoring

Monitoring consists of procedures implemented for temporary or continuous observation (e.g. of species dynamics) and is not followed by any additional activities.

Observational study

A type of study in which the effect of the exposure on the participants is observed, but the investigator has no role in assigning participants to the exposure.

Observational unit

The unit in which a particular outcome is measured, for example, dengue infection may be measured in an individual, or mosquito density may be measured in a community.

Odds ratio

This is the ratio of the odds of a disease occurring in the exposed (intervention) group compared with the unexposed (control) group.

Outcome

This refers to a parameter that the study sets out to measure; it should be defined in advance of the study being conducted and reflect the question the study sets out to answer. The primary outcome is the outcome of greatest importance, and a secondary outcome is typically an additional effect of an intervention that is of lesser importance or is less optimal for assessing the question asked by the study. There can be multiple primary and secondary outcomes. Primary outcomes can be epidemiological or both epidemiological and entomological.

Overall effect

The overall effect of the intervention is obtained by comparing the overall incidence in the intervention and control arms.

Performance bias

This refers to “systematic differences in the care provided to members of the different study groups other than the intervention under investigation”. For example, if participants know they are in the control group of a trial of insect repellent, they may be more likely to use other forms of vector control, such as protective clothing. Alternatively, healthcare providers may care for patients differently if they are aware of the study group to which the participant is assigned. Performance bias can be reduced by using blinding to ensure that participants, healthcare providers and researchers are not aware of which intervention participants have received, although this is not always possible.

Precision

This refers to the level of confidence in an effect estimate. The confidence intervals around the effect estimate indicate the precision of the estimate. Random error can lead to large confidence intervals if the sample size is small or the number of events (or cases) is small. GRADE recommends downgrading for imprecision if

(i) a recommendation would differ if the upper or lower boundary of a 95% confidence interval represented the truth or

(ii) the effects are large and both the sample size and number of events are modest even if the 95% confidence intervals appear satisfactorily narrow.

Publication bias

This refers to bias introduced into a systematic review because positive study results are more likely to be published than negative ones.

Public health value

An intervention has public health value if it has proven protective efficacy to reduce or prevent infection or disease, or both, in humans.

Randomization

Individuals or clusters are allocated to intervention and control groups at random. Randomization consists of two interrelated steps: sequence generation and allocation concealment (not to be confused with blinding).

Randomized controlled

In this study design, individuals are randomly allocated to either the trial (RCT) intervention or control group. The intervention and control groups are then followed up for the outcome of interest.

Risk difference

This is the risk of disease (or infection) in the intervention group minus the risk of disease (or infection) in the control group.

Selection bias

This refers to “bias in the estimated association or effect of an exposure on an outcome that arises from the procedures used to select individuals into the study or the analysis”. Often the term is used to refer to systematic differences among the characteristics of the study population and those of other populations (that is, highlighting a lack of generalizability). In randomized controlled trials and cohort studies, selection bias can occur when there is loss to follow-up. In case–control studies, selection bias is introduced if cases are selected who are not representative of all cases within the population, or if controls are selected who are not representative of the population that produced the cases.

Stratification or stratified

This technique is used to ensure that study arms are balanced with randomization regard to a characteristic thought to affect response to the vector control intervention (for example, baseline incidence). Individuals or clusters are grouped to form strata based on a characteristic (for example, clusters with a low incidence versus high incidence of the disease) and are randomly allocated to the intervention or control group within the stratum such that equal numbers are assigned to each group in each stratum.

Superiority study

This type of study aims to show that one vector control intervention is more efficacious than another. This requires a one-sided test of statistical significance.

Surveillance

Surveillance consists of procedures developed in response to a risk and carried out to support subsequent actions.

Systematic review

A systematic review uses rigorous methods to identify studies and synthesize the results of these studies to answer a specific question. The Cochrane Collaboration produces gold standard systematic reviews that are conducted in a highly rigorous fashion.

3. Executive summary

There is a growing risk of mosquito-borne diseases (MBDs) worldwide, including in the European region. In Europe specifically, the continuously expanding range of invasive mosquito species, such as *Aedes albopictus* across all of Europe, and the introduction and establishment of *Aedes aegypti* populations in Cyprus and on Madeira, increase the risk of viral MBDs such as dengue and chikungunya, while native species such as *Culex pipiens* continue to spread West Nile virus. The movement of goods and people, increased urbanisation as well as favourable conditions generated by anthropogenic climate change contribute to an increasing frequency of locally acquired cases of MBDs, combined with the geographical expansion of cases recorded.

To address this problem, this report set out to establish clear guidelines and recommendations for effective surveillance and control measures against mosquitoes of public health concern. In the absence of systematic clinical studies assessing the impact of vector control interventions on the epidemiology of human MBDs, it was agreed to use entomological endpoints as proxy for the biting activity of mosquitoes (and thus the risk of disease transmission) instead.

A scientific review revealed a very heterogeneous landscape of various entomological endpoints supported by generally poor evidence, which reduced the strength of our recommendations for or against certain interventions. Some interventions might be recommended in specific settings, such as larviciding, or chemical control of adult mosquitoes under emergency conditions (outbreak of disease, high infestation with adult mosquitoes).

There is an urgent need to further evaluate the efficacy of mosquito control interventions in a systematic fashion that would also include epidemiological endpoints. However, given the comparably low numbers of MBD cases in Europe at this point, this would be unrealistic, and a future study should incorporate further studies from across the globe. We further recommend to standardise the type of data collected in each study by providing technical guidance to ensure a high standard in future studies.

4. Introduction

4.1 Public Health context of mosquito-borne diseases (MBDs)

Globally, mosquito-borne diseases (MBDs) have a huge impact on the health and well-being of humans. All vector-borne diseases combined account for more than 17% of all infectious diseases, causing more than 700,000 deaths annually (WHO 2023a). The most important MBDs are malaria (caused by *Plasmodium* parasites) and viral diseases, such as dengue, chikungunya, yellow fever or West Nile virus. While the disease burden from MBDs in Europe is comparatively low when viewed against other regions, the spread of invasive species that act as competent arboviral vectors (mainly *Aedes albopictus*), as well as the endemo-epidemic status of West Nile virus across Europe, are posing severe risks to public health (ECDC 2023a).

Cases of **malaria** reached 247 million in 2021, resulting in an estimated 619,000 malaria deaths worldwide (WHO 2022). In Europe, 4,856 malaria cases were reported in the EU/EEA in 2021, of which 4,855 were confirmed. Among 4,257 cases with known importation status, 99.7% were travel-related, while 13 confirmed cases were reported as acquired in the EU (four in Greece and nine in France) (ECDC 2023b). Malaria is still endemic in neighbouring regions, and the emergence of the urban-dwelling invasive vector species *Anopheles stephensi* across Africa is a major cause of concern (WHO 2023b).

Dengue is the globally most prevalent viral infection transmitted by *Aedes* mosquitoes. More than 3.9 billion people in over 129 countries are at risk of contracting dengue, with an estimated 96 million symptomatic cases and an estimated 40,000 deaths every year (WHO 2023a). In 2023, and as of 27 July, over 3 million cases and over 1,500 dengue-related deaths have been reported globally. In Europe, the dengue virus is transmitted via the mosquito vector *Aedes albopictus*, which is established in a large part of Europe: whereas in 2013, it was established in 8 EU/EEA countries, with 114 regions being affected, the mosquito is now established in 13 countries and 337 regions (see Fig. 1). *Aedes aegypti* is the main vector of dengue virus worldwide, but its distribution in the EU/EEA is currently limited to Madeira and Cyprus.

There have been autochthonous dengue cases reported in Europe since 2010, and in 2022, France reported nine outbreaks with a total of 65 locally acquired cases of dengue, which was the highest number of autochthonous cases and outbreaks in the EU/EEA so far. In August 2023, the first four autochthonous cases for the 2023 transmission season were reported in France, followed by 19 locally acquired cases in Italy as of 11 September 2023 (ECDC 2023d).

West Nile virus (WNV) infections are currently occurring in southern, eastern and western Europe, and expanding in range, with repeated cases occurring in Austria, Germany, and the Netherlands. Since the beginning of the 2023 transmission season and as of 13 September 2023, EU/EEA countries have reported 417 human cases of WNV infection, and five deaths (3 in Greece and 2 in Italy) (ECDC 2023c). The virus is transmitted among birds via the bite of infected mosquitoes (mainly *Culex* sp.), and incidentally humans and other mammals may become infected.

Chikungunya is not endemic in mainland EU/EEA and the majority of the cases are travellers infected outside of mainland EU/EEA. When the environmental conditions are favourable, in areas where *Ae. albopictus* is established, viraemic travel-related cases may generate a local transmission of the virus, as demonstrated by the sporadic events of chikungunya virus transmission since 2007, and outbreaks in Italy in 2007 (Angelini et al. 2007) and 2017 (Venturi et al. 2017).

In 2023 and as of 26 July, approximately 300,000 cases and over 300 deaths (in Brazil and Paraguay) have been reported worldwide as a result of Chikungunya virus disease. No autochthonous cases have so far been reported in Europe in 2023 (ECDC 2023c).

The underlying causes for the increase in MBDs include globalised trade, mobile human populations (eg. through travel or migration), unplanned urbanisation, and climate change.

The global trade in goods facilitates the movement of mosquitoes by passive transport, thus allowing exotic species to invade new areas (Tatem et al. 2006). This is especially pertinent for species whose eggs can survive dry periods and that breed in small human-made bodies of water (container breeding).

The increased mobility of human populations may result in higher frequencies of infected persons moving from disease-endemic regions, thus acting as potential reservoirs for local disease transmission in presence of suitable vectors. This is currently the main cause of dengue and chikungunya transmission in Europe.

Urbanised areas can provide a wide range of breeding grounds for mosquitoes, thus bringing these vectors into closer contact with large groups of susceptible human hosts (Kolimenakis et al. 2021).

As a result of climate change Europe is experiencing a shift in temperature and precipitation patterns, with Europe being the fastest-warming continent (WMO, 2023). This creates more favourable conditions for invasive mosquito species such as *Aedes albopictus* and *Ae. aegypti*, as the vectors benefit from longer phenological seasons, and conditions for vector competence improve with rising temperature. For example, a longer vector season will result in more generations (in 2022, 6 generations of *Ae. albopictus* were found in the Upper Rhine Valley, compared with 4-5 generations in previous years [A. Joest, pers. comm.]), higher population densities and more eggs, which in turn increase the risk of passive and active spreading. Invasive species like *Ae. albopictus* or *Ae. aegypti* are often more tolerable to higher water temperatures than native species, thus resulting in their displacement. Moreover, higher temperatures result in increased biting activity of *Ae. albopictus* (Heitmann et al. 2017; 2018).

4.2 Objectives of the document and expected outcomes

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This document builds on the knowledge generated during the 10th EMCA workshop on "Best practices for mosquito control in Europe", held in November 2022 in Mendrisio/Switzerland.

The aim of this document is to provide evidence-based best practices to policymakers and to mosquito control and pest control professionals for the built environment, taking the specific structural and socio-economic situation into account. It expands on the recommendations made in previous publications focusing on the surveillance (ECDC 2012a, 2014; AIM COST 2020) and the control of invasive and native mosquito species in the European region (EMCA 2013; Takken & van den Berg 2019; ECDC 2020a). The key differences of this document to these earlier publications are:

- its focus on the built environment
- the critical assessment of available practices for surveillance and control to make specific, evidence-based recommendations to policymakers and vector control professionals
- incorporating input by experts from the neighbouring region of the Middle East and North Africa (MENA)
- including *Anopheles stephensi* as a recently emerged invasive urban vector species.

The main objectives of this document are:

- the dissemination of available methods and tools for mosquito control in built-up areas
- harmonising strategies adapted to specific requirements, while complying with international recommendations/regulations, and in line with the Global Vector Control Response (2017–2030) (WHO 2017).

We thereby hope to make a major contribution to achieving the following outcomes:

- promoting the use of integrated vector management (IVM)
- reducing the resistance of vectors to biocides
- controlling high population densities of mosquitoes in built environments, thereby reducing nuisance and risk for MBDs to human populations
- monitoring to prevent and/or slowing the spread of invasive mosquito species
- helping to reduce the risk of MBD outbreaks, and responding adequately to the occurrence of MBDs
- reducing the overall burden of MBDs
- increasing citizen engagement in mosquito monitoring and control. Examples might include: citizen-aimed campaigns to prevent invasive mosquito spread via private vehicles, or campaigns to reduce stagnant water at private properties.

4.3 Built-up areas

"Built-up areas" are defined as the presence of buildings (roofed structures). This definition largely excludes other parts of urban environments or human footprint such as paved surfaces (roads, parking lots), commercial and industrial sites (ports, landfills, quarries, runways) and urban green spaces (parks, gardens) (OECD, 2023). Built up areas are of relevance in this document, as they provide a wide range of potential breeding sites for disease vectors (specified below), and through the increased chance of interacting with human, the highest risk of VBD outbreaks (Gubler 2011; Weaver 2013).

Built-up areas include all inhabited areas otherwise classified as urban, suburban, peri-urban, and rural. Europe's level of urbanisation is expected to increase to approximately 83.7% in 2050. Trends in the total population of EU27 and UK from 1961 to 2018 show a decline in the share of population living in rural areas over the total population, while towns and cities experienced a smooth and constant increase. Built-up areas are likely to expand by more than 3% between 2015 and 2030, reaching 7% of the EU territory by 2030 (Perpiña Castillo et al. 2019) The influx of vector species from outside the urban system (periurban, rural, wetlands) through active and passive movement into built-up areas needs to be taken into consideration when planning vector control activities.

4.4 Points of entry

As defined by WHO (2005), a point of entry (PoE) means a passage for international entry or exit of travellers, baggage, cargo, containers, conveyances, goods and postal parcels as well as agencies and areas providing services to them on entry or exit. Points of entry (PoE) include those designated under the International Health Regulations (IHR), such as airports, harbours and ground crossings. At and around these locations, a country is obliged to put in place methods and procedures for surveillance and control of vectors and vector breeding reservoirs (WHO 2005; 2016). Other potential PoE for introduction of invasive mosquitoes are locations in built-up areas that receive products associated to the importation of invasive mosquito species such as imported used tyres or lucky bamboo (*Dracaena sanderiana*) plants, international train and bus stations, car parks and petrol stations along main travel axes that originate in or pass through countries or areas which are colonised by invasive mosquitoes.

4.5 Public areas

Public area is any area that is open to the public for public use, whether owned or operated by the government or a private party. There is a wide range of public places in built-up areas that need to be monitored for vector surveillance and control efforts. These include:

- Cemeteries
- Zoos
- Watering- / pumping stations
- Municipalities' depots
- Parks and recreational areas
- Sensitive places such as hospitals, nursing homes etc.
- Rainwater catchment basins / gutters
- Ornamental fountains
- Underground water (inundated constructions and basements, cable manholes, etc.)
- Septic tanks
- Cisterns
- Rainwater/drain/grey water catchment tanks
- Barrels
- Small containers (e.g., flowerpot dishes, watering cans, hollow fence poles, clogged or sagging gutters, parasol stands, tree watering bags)
- Vegetation and protected sites

See WHO (2016) for an extensive list of potential mosquito breeding sites.

4.6 Private properties

Private property refers to the ownership of property by private parties (anyone or anything other than the government). As in the public places described above, containers located on private properties are especially relevant for the surveillance and control of container-breeding mosquito species.

The specific legal aspects of entering private properties need to be established before conducting these activities. Likewise, communication with landowners should be done in an official way, communicating the objectives and the importance of collaboration. Permission of backyard owners is crucial for monitoring and control, thus ensuring successful stakeholder engagement.

4.7 Governance – stakeholders

Different stakeholders need to be involved depending on the purpose of the intervention. These include the following categories:

- Administrative bodies
  - National/regional/local governments, municipalities
  - National/regional/local health departments
  - National/regional/local departments for the environment
  - National/regional/local departments for public orders
  - Biocide regulations authority
  - Urban planning/development agencies (rain, drain, wastewater management)
  - Local parks and gardens authorities (including allotment gardens and cemeteries)
  - Housing organisations (eg. responsible for gated communities, social housing, or cooperative housing complexes)
  - Education sector (primary schools)
  - National/regional/local press departments for public relations work
  - Mosquito control operators (public/private)
- Commercial pest & vector control operators
- Research institutions
- Communities: Local communities play a major role in and are key to the success and sustainability of vector control. While coordination between many stakeholders is required, vector control is critically dependent on harnessing local knowledge and skills within communities. Community engagement and mobilization requires working with local residents to improve vector control and build resilience against future disease outbreaks (WHO 2017).

Some types of local communities include:

- Social associations (help for elderly people, tenant associations, scouts, etc.)
- Private sector (sports clubs etc.)
- Touristic agencies / associations
- National and local media (TV, radio) and social media for information dissemination: media campaigns can provide local communities with the tools and information to improve their understanding of mosquitoes and MBDs and of ongoing operations (Lwin et al. 2016). Moreover, community participation through social media can contribute to tracking and even forecasting MBD outbreaks (Marques-Toledo et al. 2017).

4.8 Policies impacting built-up areas and mosquitoes

Through their focus on urban environments, some policies might play a significant impact on mosquitoes and MBDs. In the European context, some recent legislations and policies include:

The Urban Agenda for the EU acts as a multi-level working method for urban policy and practice, promoting cooperation between Member States, cities, the European Commission, and other stakeholders. Its scope focuses on better regulation, better funding, and better knowledge.

The EU Strategy on Adaptation to Climate Change sets out how the European Union can adapt to the unavoidable impacts of climate change and become climate resilient by 2050. The Strategy has four principle objectives: to make adaptation smarter, swifter and more systemic, and to step up international action on adaptation to climate change.

4.9 Mosquito species relevant as pests and/or potential vectors of diseases in built-up areas

4.9.1 *Culex pipiens*

Culex pipiens is a common species native to Europe that is known as a pest in urban environments. *Cx. pipiens* is found in all European countries, except Iceland and Faroe Islands, and in all Middle Eastern and North African countries (ECDC 2020b).

Cx. pipiens is an ubiquitous species which can develop in a multitude of breeding sites in the urban, periurban, rural and natural environment from containers and underground waters up to large surface water collections as in wetlands.

Eggs are laid as rafts on the surface of existing water bodies, where larvae can hatch very quickly. The larvae often stay on the water surface where they filter the water for food; in case of disturbances (movement of water surface, shadowing) they flee very quickly towards the bottom of the water but also quickly reappear. Thus, checks for larvae should be carried out with a close-meshed net. In Europe, *Cx. pipiens* consists of two forms (or biotypes) and their hybrids: the *pipiens* form and the *molestus* form. The *molestus* larvae prefer eutrophic water (e. g. cess pools), located underground, thus offering limited access to the breeding site for control purposes. The *pipiens* larvae on the other hand breed in less eutrophic water aboveground, in more open breeding sites, thus allowing for easier access (A. Jöst, pers. comm.).

Whereas females of the *pipiens* form mainly feed on birds, the *molestus* form feed on a variety of vertebrate hosts (including humans) and may therefore contribute to the amplification cycle of West Nile Virus (WNV) among birds, and also the occasional spill-over of viruses to human and other mammal populations (Haba & McBride 2022). Thus, *Cx. pipiens* mosquitoes are major vectors of both West Nile and Usutu Viruses (USUV) in Europe. They are also able to transmit several other arboviruses, such as Rift Valley fever virus (RVFV), Japanese encephalitis virus (JEV), and Sindbis virus (SINV) (ECDC 2020b).

4.9.2 *Aedes albopictus*

Native to Asia, the tiger mosquito *Aedes albopictus* has undergone a dramatic global expansion facilitated by human activities, in particular the transfer of used tyres and exotic plants (Paupy et al. 2009). Together with passive movement via commercial and private transport, this has resulted in a widespread global distribution of *Ae. albopictus*, and it is considered one of the most invasive species (Benedict et al. 2007) (see Fig. 1 for its current distribution in Europe). *Ae. albopictus* is expected to expand its range further across Europe into suitable habitats, aided by to climate change (Kraemer et al. 2019).

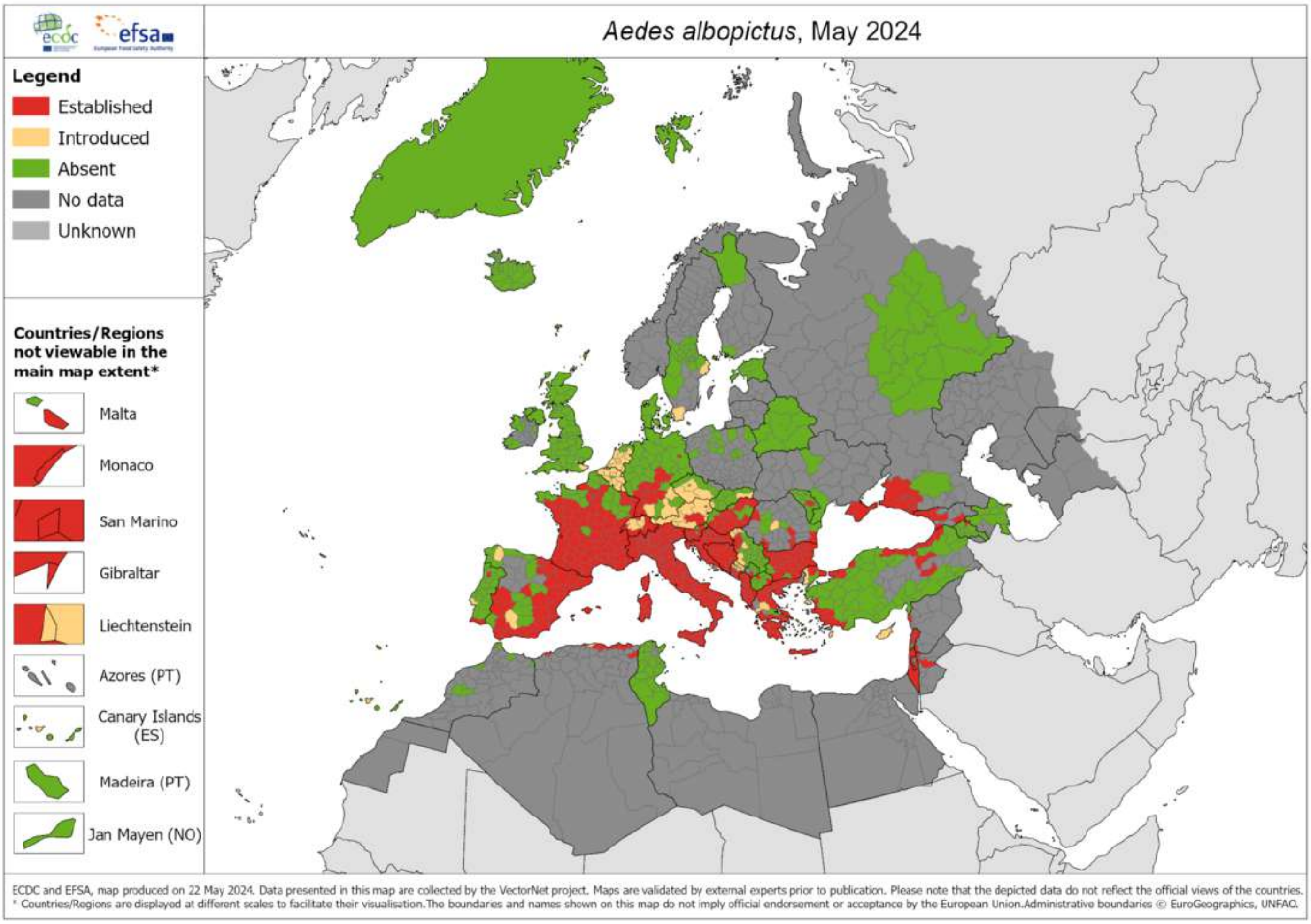

Figure 1: Presence of *Aedes albopictus* in Europe and surrounding areas, as of June 2024. Source: European Centre for Disease Prevention and Control and European Food Safety Authority. Mosquito maps [internet]. Stockholm: ECDC; 2024. Available from: https://ecdc.europa.eu/en/disease-vectors/surveillance-and-disease-data/mosquito-maps

Ae. albopictus feeds on a wide range of hosts and is an opportunistic feeder. Blood hosts include humans, domestic and wild animals, reptiles, birds and amphibians. It is also known to be a significant biting nuisance, with the potential to become a serious health threat as a bridge vector of zoonotic pathogens to humans. *Ae. albopictus* has the ability to breed in natural and artificial habitats, some of which include tyres, barrels, rainwater gulley catch basins and drinking troughs, and has a preference for urban and suburban habitats (Becker et al. 2020).

Eggs are placed individually on the inner edge of water-filled containers, with the female distributing the eggs to different breeding sites. Larvae can only hatch when the eggs have been flooded, even though the eggs are drought tolerant and can survive several months without flooding. Larvae are often found on the bottom of the water, where they graze food particles (detritus) from the ground. Like almost all other mosquito species, they absorb the oxygen via their breathing tube at the water surface. In case of disturbances (movement of water surface, shadowing) they flee towards the bottom of the water and can stay there for a considerable time. Checks for larvae should be carried out with a close-meshed net (A. Joest, pers. comm.).

Ae. albopictus is a known vector of dengue virus, chikungunya virus, and dirofilariasis, and it has been shown to be capable of transmitting 26 viruses (reviewed by Paupy et al. 2009).

4.9.3 *Aedes aegypti*

Of all mosquito species, *Ae. aegypti* is the most anthropophilic, feeding primarily on humans and closely associated with human habitation (Takken & van den Berg 2019). In subtropical climates, the species is found almost always in the close vicinity of human settlements. The larvae occur in a wide variety of small artificial containers and water recipients of all kinds, both inside and outside of human habitations in gardens and within a circle of 500 m around dwellings, e.g. in earthenware pots and water tanks for storing water, uncovered cisterns, rain-filled empty cans or flowerpots, broken bottles or discarded motor vehicle tyres (Becker et al. 2020).

Ae. aegypti is distributed in the tropical, subtropical and warm temperate regions of both hemispheres. The ability of *Ae. aegypti* to establish itself in more temperate regions is currently restricted, due to its intolerance of temperate winters and, in particular, the high mortality rate of eggs when exposed to frost (Gould & Higgs 2009). While its distribution in the EU/ EEA is currently limited to Madeira and Cyprus, before 1945, all Mediterranean countries and most major port cities had reported at least occasional introductions of *Ae. aegypti* (Mitchell 1995; cited by Becker et al. 2020). *Ae. aegypti* has been locally detected in the Netherlands at tyre yards, imported via shipments of tyres originating from Florida, USA (Scholte et al. 2010; Brown et al. 2011), and the species is yearly found introduced by aircrafts at Schiphol airport (Ibáñez-Justicia et al. 2017; 2020). It also has the potential to become re-established widely across the Mediterranean. Coastal regions of the Mediterranean, the Black Sea, and the Caspian Sea, and areas along large lowland rivers have been identified as suitable habitats for *Ae. aegypti* (ECDC 2012b). Moreover, this could change in the future, with global climate change resulting in the species' ability to expand further to the north and south (Weaver & Reisen 2010; Kraemer et al. 2019).

Aedes aegypti is a known vector of several viruses, including yellow fever virus, dengue virus, chikungunya virus, and Zika virus (ECDC 2023e).

4.9.4 Other *Aedes* species

Aedes atropalpus is an invasive North American species that has recently been reported in Europe. Its movement across America and into Europe has been a result of the commercial transport of used tyres which will continue to facilitate its further spread. Laboratory competency studies have shown the ability of *Ae. atropalpus* to transmit a number of viruses, including WNV (Turrell et al. 2001), but its importance as a vector of infectious diseases is still unknown (Scholte et al. 2009).

Aedes japonicus is an invasive Asian species, and while it is not considered an important disease vector in its normal native range of Japan and Korea, there is a concern that it might become a pest problem or be involved in the transmission of arboviruses such as WNV. *Ae. japonicus* colonises urbanised environments (Schaffner et al. 2009), increasing the potential contact this species could have with humans which in turn may result in disease transmission. The species is suspected to act as a bridge vector of WNV, and has shown vector competence (laboratorium only) for the transmission of dengue and chikungunya (Schaffner et al. 2011).

Aedes koreicus has been reported from small foci in Belgium and from the Veneto region in Italy in 2011 (Capelli et al. 2011), as well as from the Netherlands in 2022 (Teekema et al. 2022). It is not considered a major vector of disease (ECDC 2014b).

Aedes caspius is a Palaearctic species common in southern and dry regions, especially in Atlantic and Mediterranean coastal marches (Becker et al. 2020). While WNV has been detected in natural populations (Detinova and Smelova 1973; cited in: Becker et al. 2020), *Ae. caspius* is not considered a major disease vector.

Aedes vexans is a floodwater mosquito widely distributed throughout the Holarctic region. This species inhabits a variety of habitats, especially within rural areas (Becker et al. 2020). In Africa, *Ae. vexans* is considered one of the primary vectors of Rift Valley fever virus (RVFV) (Sang et al. 2017).

4.9.5 *Anopheles stephensi*

Anopheles stephensi is a highly competent vector of *Plasmodium falciparum* and *P. vivax*, and is considered an efficient vector of urban malaria. Until 2011, the reported distribution of *An. stephensi* was confined to certain countries of South Asia and parts of the Arabian Peninsula. Since then, the vector has been found in Djibouti (2012), Ethiopia (2016), Sudan (2016), Sri Lanka (2017), Somalia (2019), Nigeria (2020), Yemen (2021), and, most recently, Ghana and Kenya (2022) (WHO 2023b, 2023c).

An. stephensi larvae are found in domestic and other artificial water containers, and may also exploit a wide variety of larval habitats in the local environment (including cryptic habitats such as deep wells). *An. stephensi* has been shown to be resistant to multiple insecticide classes in many locations, including Africa, posing challenges to its control (WHO 2023b).

4.9.6 Other *Anopheles* species

Anopheles atroparvus: Native to Europe, and historically a major malaria vector in Spain and Portugal, as well as involved in winter transmission of malaria at the start of the 20th century in Britain, coastal areas in the Netherlands and Germany (Becker et al. 2020).While malaria has been eradicated in the European Union and European Economic Area countries since 1975, there is a risk that imported cases may act as a source of further infections in regions inhabited by *An. atroparvus*, although the risk of this occurring is considered low (Bueno-Marí & Jiménez-Peydró 2012).

Anopheles plumbeus: Native to Europe, distributed throughout Europe, the Caucasus, the Middle East, Iran and Iraq and in North Africa (Becker et al. 2020). Continued growth of this species in urban habitats could potentially increase contact with humans (Bueno-Marí et al. 2011). This species is a known persistent biter of humans. It is suspected of having contributed to malaria transmission in the past and has been shown to be a competent vector of both tropical and Eurasian strains of malaria. In recent decades, it has proliferated in huge numbers as a result of its adaptation from natural (tree holes) to artificial breeding sites (water catch basins and septic tanks contaminated with organic waste) (Becker et al. 2020).

Anopheles sacharovi: Its distribution is limited to south-eastern Europe mainly in coastal areas. Historically, it was one of the main vectors for malaria in the eastern Mediterranean (Hertig 2019), and has been involved in recent outbreaks of vivax malaria in Greece (ECDC 2014c). It is a thermophilic species, with a long hibernation period. It is anthropophilic and endophilic biting also cattle. Adults are frequently found in stables and human dwellings during the day. The larvae are tolerant to salinity and usually occur in coastal swamps and marshes, lagoons and nearby streams, irrigation, drainage and roadside ditches, rice fields, and grassy ponds, pools or seepages (Becker et al. 2020).

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5. Methods

5.1 General methodology

WHO/TDR had issued the contract for developing this guideline document to EMCA, with Sandra Gewehr (President of EMCA) as principal investigator (PI). Monthly meetings were held between EMCA and WHO/TDR to coordinate the efforts and clarify any questions.

The EMCA secretariat (Dr. Francis Schaffner, Dr. Frederik Seelig) developed the search terms for the Scientific Review Teams (SRTs), as well as inclusion/exclusion criteria.

The two SRTs conducted separate literature searches on mosquito surveillance and on mosquito control interventions, respectively, that have been used in built-up areas in Europe. Following the literature search and screening process, as described in 5.2 below, the final results were shared with the Secretariat.

The Secretariat screened the publications provided by the SRTs and subsequently developed the relevant PICO questions for each intervention.

The Guideline Development Group (GDG), consisting of 10 EMCA members, experts in mosquito control, assessed the quality of the evidence available from the published literature that informed their recommendations for or against any specific type of intervention according to GRADE principles. The final assessment of the evidence resulted in the recommendations on each intervention type made by the GDG.

As Van Hulk et al. (2021) emphasised, studies for the assessment of the efficacy of vector control interventions should include both entomological and epidemiological endpoints. However, as none of the studies collected in the frame of the scientific literature reviews included epidemiological endpoints, it was agreed between the EMCA secretariat, the Principal Investigator and TDR/WHO to use entomological endpoints as proxy for the biting activity of mosquitoes and thus the potential transmission of a disease.

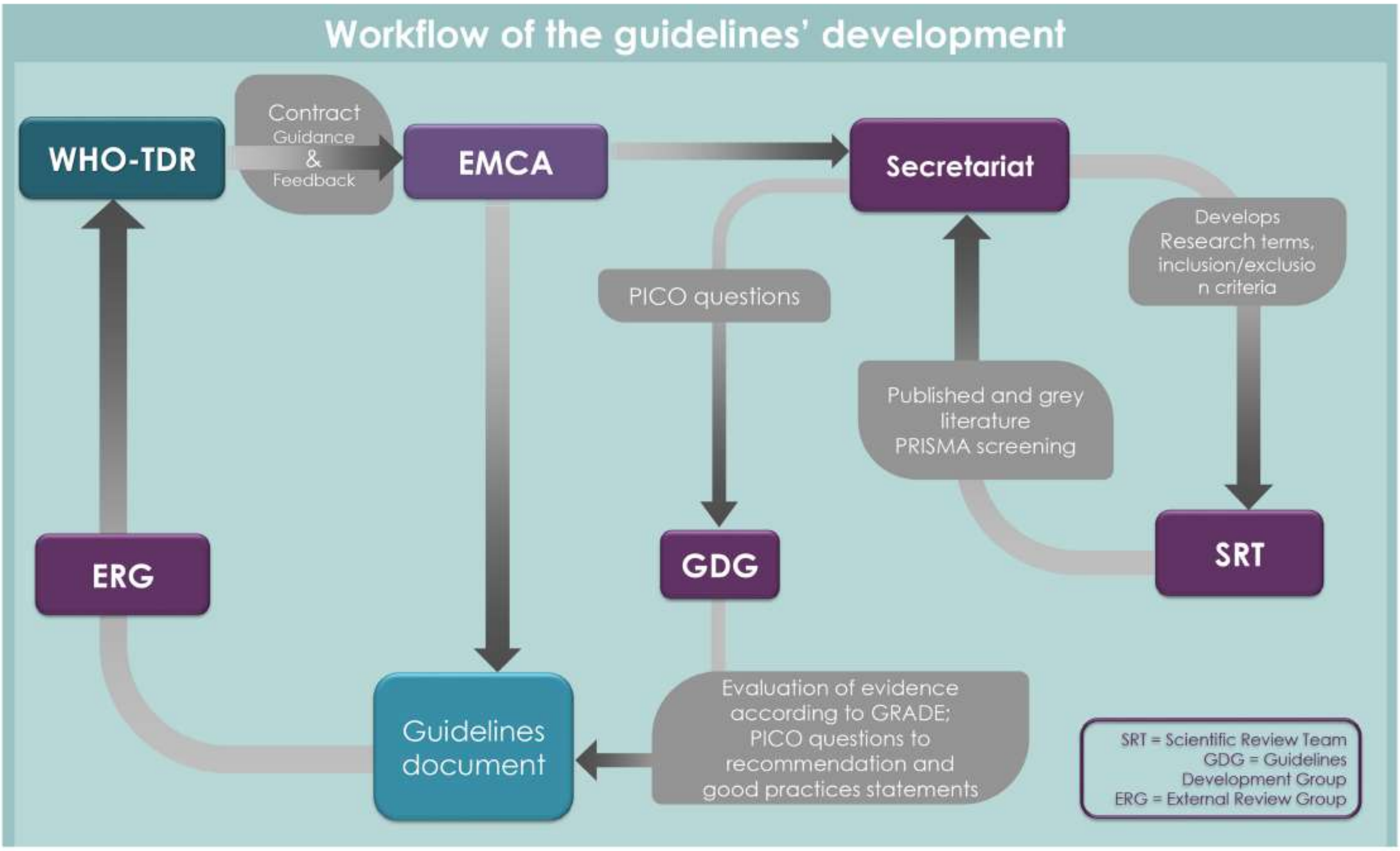


Fig. 2: Workflow of guideline development.

5.2 Literature search by SRT teams

5.2.1 Mosquito surveillance

Methods: A literature search was conducted on 26 January 2024 in three databases: Web of Science, Google Scholar and Pubmed, with similar search terms (Tab. 1). A total of 782 records were retrieved from all three databases (Fig. 3). A table of all titles in one column and all abstracts in a second column was collated and duplicates were removed. If an abstract was not provided by the databases, it was searched and added manually if available. This resulted in a total of 553 records, which were independently screened by two researchers (Renée Zakhia and Renke Lühken). Title and abstract of each record were screened and two exclusion criteria were used. Records were excluded if they (1) involved laboratory studies only and (2) did not use mosquito control methods, including studies only focusing on screening of insecticide resistance.

Results: From the 553 records, 10 studies did not match the two exclusion criteria (Fig. 1). In addition, there were another 16 records where no decision was possible, as the title and abstract did not give enough information. 10 of these 16 records were artefacts from the Google Scholar search and were not real articles (e.g. only citations). For the 6 remaining records, we conducted a full text screening and only 2 did not match our exclusion criteria. Thus, resulting in a total of 12 studies. From these, 8 presented evidence, while 3 were without evidence and one study was excluded as it was not in English. In conclusion, only 8 studies presented evidence of surveillance in combination with mosquito control techniques in the field.

Table 1: Search terms per literature database

Literature database	Search term	Hits
Web of Science	TI=(mosquito) AND TI=(surveillance OR monitoring) AND ALL=("Aedes aegypti" OR "Stegomyia aegypti" OR "Aedes albopictus" OR "Stegomyia albopicta" OR "Culex pipiens" OR "Anopheles stephensi")	218
Google Scholar	(intitle:mosquito) AND (intitle:surveillance OR intitle:monitoring) AND ("Aedes aegypti" OR "Stegomyia aegypti" OR "Aedes albopictus" OR "Stegomyia albopicta" OR "Culex pipiens" OR "Anopheles stephensi")	471
PubMed	(mosquito[Title]) AND (surveillance[Title] OR monitoring[Title]) AND ("Aedes aegypti" OR "Stegomyia aegypti" OR "Aedes albopictus" OR "Stegomyia albopicta" OR "Culex pipiens" OR "Anopheles stephensi")	93

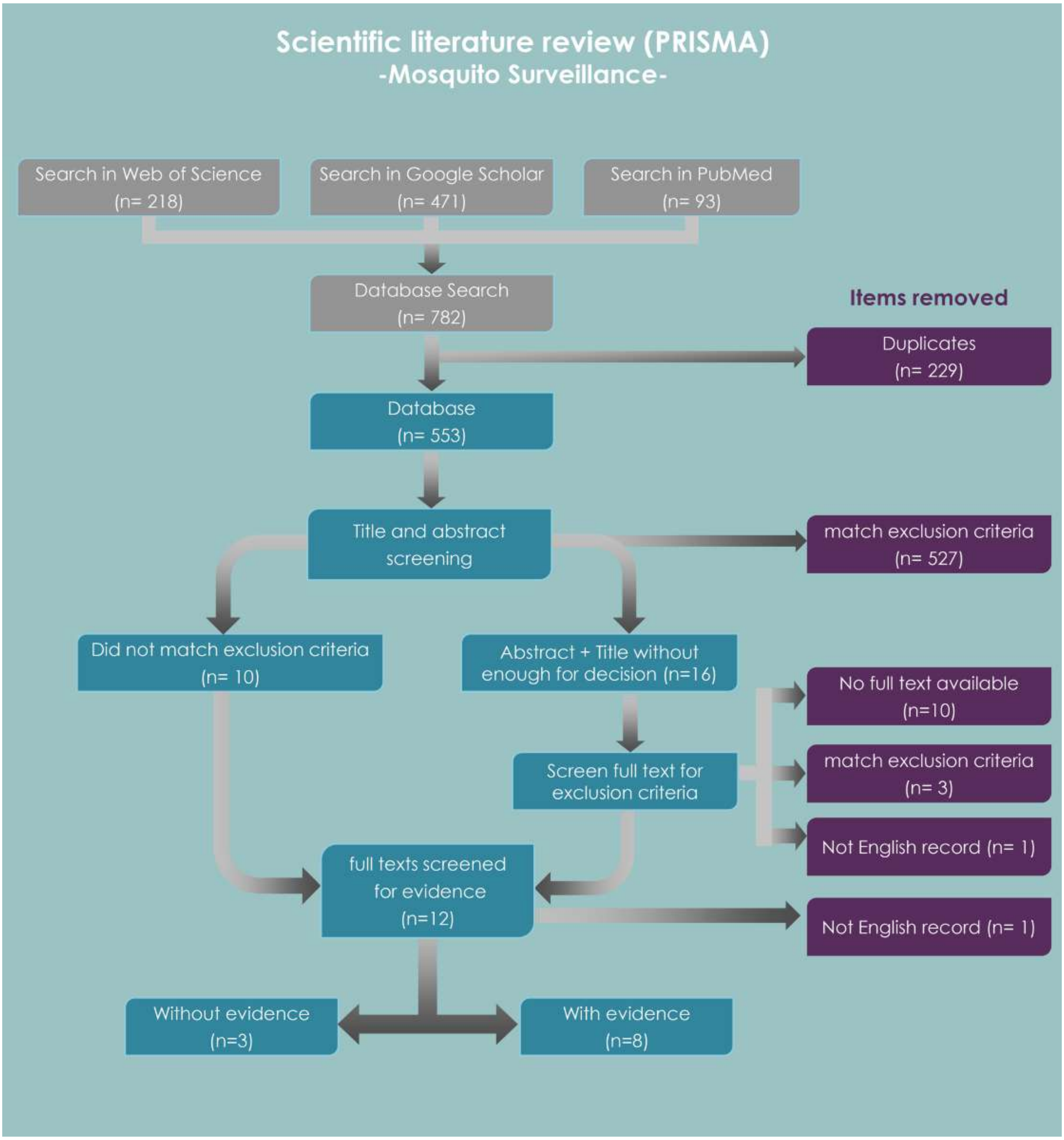


Fig. 3: Workflow of scientific review on mosquito surveillance studies.

5.2.2 Mosquito control

The aim of the team for mosquito control was to conduct a search of the literature on mosquito control methods and assess them for the presence of evidence on the efficacy of these methods. To this end, a literature search was conducted in Google Scholar to target both the grey literature and the scientific literature, as control activities can be managed by government dependencies or other health institutions that may not necessarily publish their assessments as scientific papers.

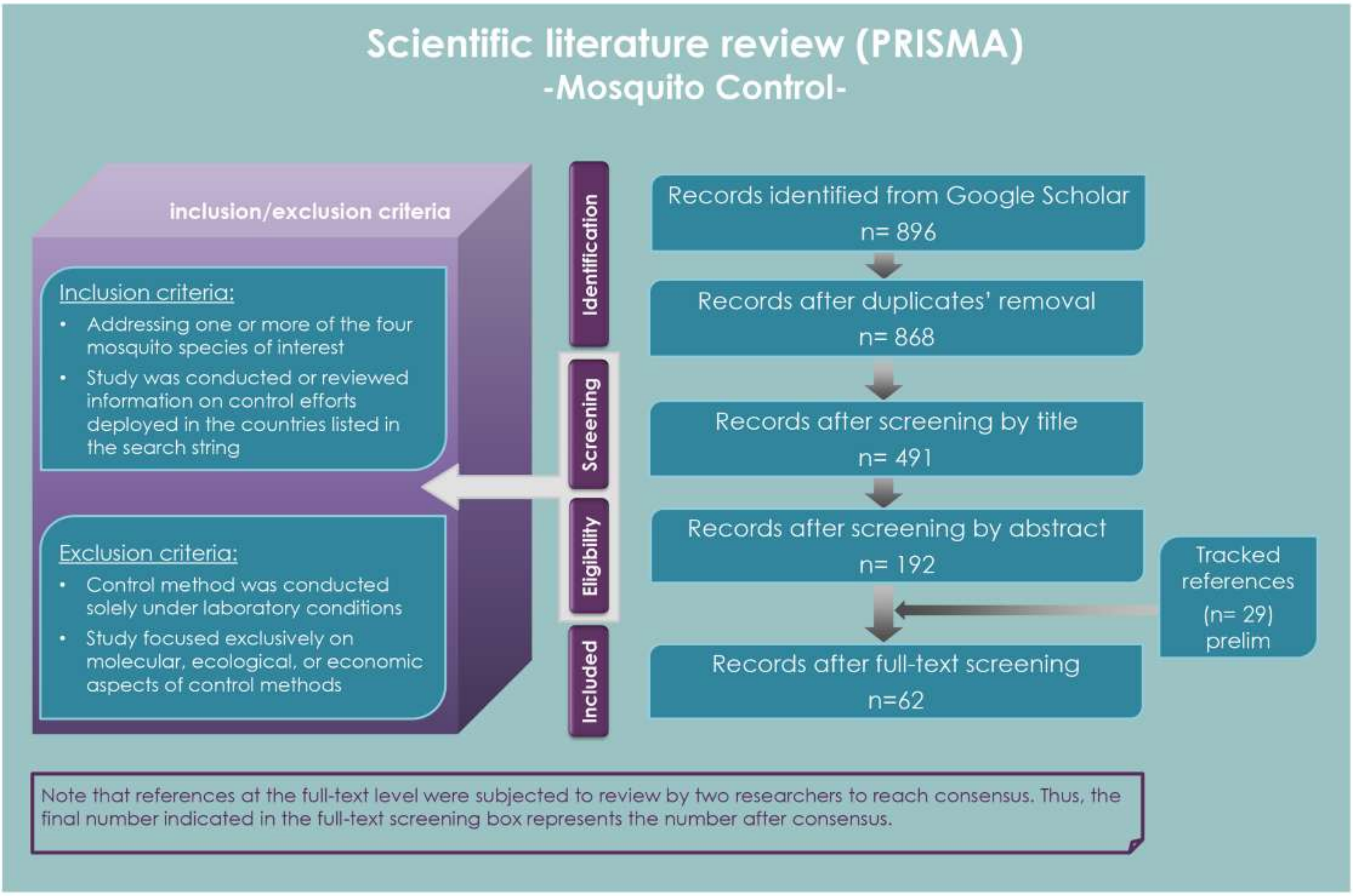


Fig. 4: Workflow of scientific review on mosquito control studies.

The search was limited to countries in the WHO European region and included the following countries of the Eastern Mediterranean and African WHO regions: Jordan, Lebanon, Syria, Morocco, Algeria, Tunisia, Libya and Egypt.

The search targeted four mosquito species:
Aedes aegypti, *Ae. albopictus*, *Culex pipiens* and *Anopheles stephensi*.
After going through several iterations of search string combinations and discussing with the EMCA secretariat and the GDG, the following search string was used in Google scholar:

(intitle:mosquito OR intitle:vector) AND -intitle:laboratory AND -intitle:journal AND (intitle:control OR intitle:management OR intitle:suppression OR intitle:elimination) AND ("Aedes aegypti" OR "Stegomyia aegypti" OR "Aedes albopictus" OR "Stegomyia albopicta" OR "Culex pipiens" OR "Anopheles stephensi") AND (Europe OR Albania OR Andorra OR Austria OR Belgium OR Bosnia OR Herzegovina OR Srpska OR Bulgaria OR Croatia OR Cyprus OR Czech-Republic OR Denmark OR Germany OR Spain OR Estonia OR Finland OR France OR Greece OR Hungary OR Ireland OR Italy OR Kosovo OR Latvia OR Liechtenstein OR Lithuania OR Luxembourg OR Macedonia OR FYROM OR Malta OR Montenegro OR Netherlands OR Norway OR Poland OR Portugal OR Slovenia OR Romania OR Serbia OR Slovakia OR Switzerland OR Sweden OR United-Kingdom OR Wales OR England OR Scotland OR Turkey OR Israel OR Palestine OR Jordan OR Lebanon OR Syria OR Morocco OR Algeria OR Tunisia OR Libya OR Egypt OR Armenia OR Azerba OR Belarus OR Bielorussia OR Georgia OR Moldova OR Russia OR Ukra).

For each reference returned by the search string, the title, authors, year published, source (name of journal or website where it was published), and URL were retrieved, using a web scraping script of our own design (Python v.3.12.1) and exported it as an xls file. The exported search results were checked for duplicates (references that appeared more than once in the search, which can happen when the same reference is published in different languages, for example) and each reference was screened using Preferred Reporting Items for Systematic Reviews and Meta-Analysis (PRISMA) standards. Using this method, references were systematically screened at three separate levels, title, abstract and full text to determine at each step if a reference was kept or dropped, based on the following inclusion and exclusion criteria:

- Included if one or more of the four mosquito species of interest were addressed
  • Included if the study was conducted or reviewed information on control efforts deployed in the countries listed in the search string
  • Excluded if the control method was conducted solely under laboratory conditions
- Excluded if the reference did not contain information on control methods or mosquito management strategies and instead focused exclusively on molecular, ecological, or economic of control methods (e.g., willingness-to-pay studies that focus on quantifying the economic cost of mosquito nuisance).

References were double-checked at the full-text stage by two members of the mosquito control SRT to ensure consensus and consistency with the inclusion/exclusion criteria. For each full-text reference retained, the pdf was retrieved and screened for evidence of efficacy assessment. References were classified as containing evidence if it listed quantifiable outcomes of the intervention (i.e., compared the effects of intervention versus non-intervention treatment), or as not containing evidence if no quantifiable outcomes were available (e.g., review articles on control methods).

At this stage, the cited literature in review documents (no-evidence) was compared with the full-text list, to account for the possibility that the search string employed might have left out some references of interest. Documents that were identified through this assessment were also retrieved and processed to a full-text stage. This additional search was only performed for no-evidence documents rather than evidence ones, as the formers represent compilations of literature to review specific subjects rather than an assessment of a specific method.

For each entry in the final list of references, which included the results of the systematic search and the additional tracked refences, the following information was extracted and compiled: a reference identifier to match the entry to the pdf document, the reference title, the abstract (when relevant), whether the reference contained evidence or not, the URL to the document, the authors, the year of publication, additional notes detailing the control methods, species and countries (when relevant), and additional notes on whether the intervention considered is currently under review for assessment of public health value.

Summary of search results

The search string reported a total of 896 references. Of these, a total of 868 references were retained after removing 28 duplicates. At the title screening stage, 491 references remained, of which 192 were retained after abstract screening. After full-text screening and consensus, 36 references were kept as a result of the systematic search on Google scholar.

An additional 29 references were identified through tracking review literature, of which 26 were included. The final list of references was made up of 62 studies, 36 of which contained evidence and 26 did not.

6. Surveillance/Monitoring

As described in the methodology section (5.2.1), the scientific review of available publications on mosquito surveillance yielded only eight studies with evidence and three without evidence, out of which only four studies (two each with evidence and without) referred to the European context. This restricted collection of references did not allow for a systematic approach to the surveillance of mosquitoes at their different stages (eggs, larvae, adults), as topics and methodologies were very restricted and specific. The predominant surveillance method was larval surveillance in natural and/or anthropogenic habitats (wetlands, refillable water containers, rainwater catch basins, sewage gutters [Khan 2014; Vakalis et al. 2007]) or in autocidal ovitraps for mapping the distribution of mosquitoes in an area (Gualberto et al. 2015). Another important objective was the assessment of the efficacy of different larviciding treatments (Copepods, IGRs, Bti, larvivorous fishes) by means of larval monitoring (Russel et al. 1996; Lamaninagao et al. 2020; Caputo et al. 2015; Lonc et al. 2004; Manna et al. 2016; McKie and Goedkoop 2010). Only two studies focused on adult surveillance for the evaluation of resistance to biocides and the behavior of adult mosquitoes in treated eave tubes (Khan 2014; Sperling et al. 2017).

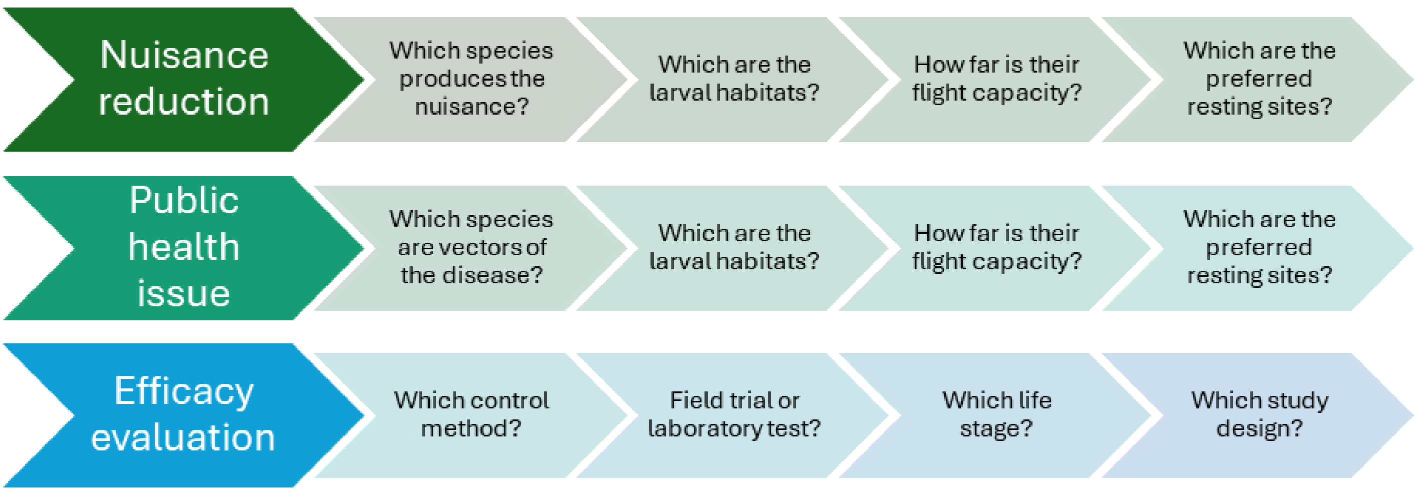
Moreover, the references yielded from the scientific review did not allow for the creation of PICO questions, or an assessment of the evidence provided following GRADE principles. Readers looking for guidance concerning specific surveillance and monitoring methods and procedures for different scopes and in different settings are referred to the guidelines for the surveillance of invasive and of native mosquitoes in Europe published by the ECDC in 2012 and 2014, respectively (ECDC 2012, 2014).

In order to provide good practices for the surveillance and monitoring of mosquitoes, we summarize below key results from two sessions of the 10th EMCA Workshop about "Best Practices in Mosquito Control", held 28-29 November 2022 in Mendrisio, Switzerland, entitled "Surveying adult mosquitoes to define the target" and "Larval monitoring for decision-making on larviciding applications".

6.1 Planning of a surveillance/monitoring project

The planning phase of mosquito surveillance is critical, and the method or strategy used must align with the specific objectives of the project. We differentiate between surveillance for actionable responses and monitoring for routine, broad-scale assessments. **Surveillance** typically aims at guiding immediate control interventions, whereas **monitoring** is a continuous effort to assess mosquito presence and population dynamics across larger areas over time.

Objective Definition: Clearly defining a project objective is paramount, as it will determine the surveillance strategy, including target species, areas to be surveyed, and the timing of interventions. When planning, one should consider whether the focus is on nuisance reduction, public health concerns, or evaluating the effectiveness of mosquito control efforts.



Nuisance Reduction: For this objective, systematic and repetitive sampling of larvae and adults should be carried out throughout the entire mosquito reproductive season. This approach enables the observation of population dynamics, facilitating timely decisions on larviciding for sustainable control effects and adulticiding (where appropriate and permitted) for short-term adult population reduction. Both qualitative (genus/species identification) and quantitative (population abundance classes) data should be collected for larvae and adults, with control efforts focusing on larval habitats that consistently produce large numbers of larvae and/or resting sites which present high abundances of adults of the nuisant species.

Public Health: If the objective is reducing health risks, surveillance should focus on detecting adult vector species and the pathogens they may carry. Sampling should occur throughout the vector activity period, and mosquitoes collected should be systematically analyzed for the presence of pathogens. In this context, the identification at species level is crucial, and even rare or patchy populations of potential vector species should be prioritized for control interventions. Additionally, all potential larval habitats of these species should be monitored systematically for the presence of larvae, with control efforts focusing on all sites in which the target species is detected, even in low abundances.

Efficacy Evaluation: In some cases, the objective for surveillance may be to evaluate the effectiveness of mosquito control interventions using entomological endpoints. This requires a tailored surveillance approach, often involving pre- and post-intervention comparisons in intervention and control areas using specific techniques that match the target life stages (eggs, larvae or adults) and the type of intervention (e.g., larviciding or adulticiding, IVM).

In order to perform surveillance activities, it is important to develop a comprehensive protocol that specifies:

- Locations, time, density, and frequency of sampling
- Mosquito identification procedures
- Pathogen detection methods
- Data storage, management and analysis, and
- Dissemination strategies

6.2 Surveillance/monitoring of adults

Mosquito adult surveillance plays a crucial role in managing mosquito populations and providing evidence for control efforts aimed at mitigating mosquito-borne diseases. Effective adult surveillance involves the systematic collection of data, including species identification, analysis of population dynamics, and evaluation of public health risks.

6.2.1 Identification of the source of the problem

The first step in mosquito surveillance is identifying the source of the problem. This includes determining which species are present and the extent to which they are connected to public complaints or disease outbreaks.

- **Species Identification:**
  - Conduct adult mosquito sampling using methods such as traps (CDC traps, BG traps etc.) or human bait (in areas without a risk for the presence of an MBD) to capture specimens.
  - Compare with historical data and consult literature to identify the species. In built-up areas, a focus on specific species such as *Aedes albopictus* or *Culex pipiens* is essential due to their roles in transmitting diseases like dengue, chikungunya and Zika, or West Nile Virus, respectively.
- **Assessing Public Complaints:**
  - Gather reports from the community to pinpoint hotspots of mosquito activity.
  - Geolocate the areas of reliable complaints to create a map of the intensity and spread of the mosquito problem.
- **Public Health Importance:**
  - Investigate whether there are ongoing health concerns related to mosquito-borne diseases.
  - Determine whether mosquitoes are vectors for pathogens affecting humans, pets, livestock, or wildlife in the area.
- **Transmission Evidence:**
  - Review reports of local transmission or circulation of mosquito-borne pathogens, such as arboviruses, in human and animal populations.
  - If possible, collect mosquitoes for pathogen testing to confirm the presence of disease.
- **Risk Assessment for Livestock and Pets:**
  - Assess the risk of mosquito-borne pathogens to animals, particularly livestock and pets. This may involve consulting veterinarians or local farms for any signs of mosquito-borne illnesses.
- **Economic Impact:**
  - Determine whether the mosquito problem is having a negative economic impact on agriculture, tourism, or property values, particularly in areas affected by high mosquito density or MBD transmission.
- **Species Adaptation:**

  - Investigate whether specific species, such as *Ae. albopictus*, are adapting to the local environment. This can be observed through shifts in habitat use, such as moving into urban areas, or changes in behavior, such as feeding at different times of the day.

6.2.2 Identification of the range of the problem

Understanding the spatial and temporal dynamics of mosquito populations is crucial for effective control measures. Therefore the existence of and access to timeseries of entomological data is essential.

- **Spatial Range:**
  - Determine the geographic extent of the mosquito infestation. This can be done by analyzing the areas of complaints, as well as performing adult mosquito surveys across various ecological zones (wetlands, urban areas, peri-urban, rural).
  - Flight capacity studies and/or empirical evidence are helpful in understanding how far mosquitoes can move (passively or actively), thus expanding the monitoring zone.

- **Seasonal Range:**
  - Mosquito populations are often seasonal. Define the start and end of the mosquito season, which will also help determine the timing of interventions.
  - Monitoring weather patterns, especially temperature and rainfall, is critical in predicting mosquito emergence and peak activity periods.

- **Species Behavior:**
  - Study the behavior of target species, including their feeding times (e.g., day-biting vs. night-biting), resting places, and larval habitat preferences. This information can influence when and where to apply control actions.

- **Abundance:**
  - Quantify mosquito abundance using traps and surveys to estimate population density. A higher density may require more aggressive intervention strategies, such as adulticiding.
  - Continual monitoring is necessary to track fluctuations in mosquito numbers throughout the season.

6.2.3 Involvement of partners and community engagement

Successful mosquito control programs often involve partnerships with local stakeholders and the community.

- **Partner Collaboration:**
  - Engage local governments, public health authorities, and environmental organizations to create a coordinated response.
  - Partners can help with data collection, provide additional resources for surveillance, and offer expertise in areas such as disease control or environmental management.

- **Community Involvement:**
  - Educate the public about the importance of mosquito control and how they can help, for example by eliminating standing water and reporting mosquito activity.
  - Organize community workshops or campaigns to raise awareness about vector-borne diseases and promote preventive actions.

- **Community Sensibility:**
  - Be mindful of community perceptions regarding control actions, especially insecticide spraying. Some communities may be sensitive to chemical interventions or have concerns about environmental impact.
  - Engaging in transparent communication about the safety and necessity of control measures can improve public acceptance.

6.2.4 Surveillance/Monitoring methods

Consistent monitoring is essential to detect changes in mosquito populations, identify new larval habitats, and evaluate the effectiveness of control interventions. Surveillance also aids in understanding the species composition, distribution, and behavior of mosquitoes in each area. Below is an overview of adult mosquito surveillance/monitoring methods and tools.

Trapping methods

Using a variety of traps is fundamental to gaining insights into the presence, density, and species of mosquitoes in different environments. Electronic real-time counters are not included in this section since they are not yet used at a sgnificant scale.

- **CDC Light Traps & BG-Sentinel traps:**
  - **Purpose:** Effective for capturing a wide range of mosquito species, including those active during the night. BG-Sentinel traps are particularly useful for attracting *Aedes* mosquitoes that are known vectors of diseases like dengue, Zika, and chikungunya.
  - **Considerations:** Traps should be placed in diverse ecological zones (e.g., wetlands, rural, urban areas) to get a comprehensive view of mosquito distribution and population dynamics (intrusion of flood water mosquitoes from adjacent areas into the urban

environment).

- **CO_2-Baited traps:**
  - **Purpose:** These traps mimic host respiration, attracting a variety of mosquito species. They are ideal for assessing *Culex pipiens* and most other relative species densities.

- **Human Landing Counts (HLCs):**
  - **Purpose:** Used to quantify the biting rate of human-biting mosquitoes in areas where there is no risk of MBDs. HLCs help identify species behavior and assess the level of human-vector interaction.
  - **Considerations:** This method involves exposure to mosquitoes, making it suitable only in regions where the risk of disease transmission is minimal.

- **Chemical lure-baited traps:**
  - **Purpose:** Uses chemical lures to mimic human odors to attract anthropophilic species such as *Aedes aegypti* and *Ae. albopictus*, known for their role in transmitting diseases. This method can target multiple species within the same ecological niche.
  - **Considerations:** Particularly efficient for identifying species with high public health importance, reducing the need for human exposure. Can be combined with other attractants such as CO_2 and light.

- **Gravid traps (GTs):**
  - **Purpose:** Gravid traps are used to monitor container-inhabiting mosquitoes by capturing females that have already taken a blood meal, helping assess pathogen infection rates. They are specifically attractive for *Aedes* species (e.g., *Aedes albopictus, Ae. aegypti*).
  - **Considerations:** These traps are effective for targeting all container-inhabiting mosquitoes and are used in both urban and rural settings.

- **Resting catches and sugar-baited sticky traps:**
  - **Purpose:** These methods aim to capture mosquitoes during their resting phase or those attracted to sugar sources, providing additional information about mosquito species and feeding behavior.
  - **Considerations:** Resting catches may require more effort, while sticky traps are relatively inexpensive and can be deployed in various habitats to complement other surveillance methods.

Involving citizen science and local participation

Engaging local communities in mosquito surveillance can enhance the efficiency of monitoring efforts while reducing costs. Methods such as:

- **Reporting nuisance:** Encouraging local citizens to report mosquito nuisance or bites can provide valuable real-time data on mosquito activity but also bears the risk of overestimates for political reasons.
- **Detecting invasive species:** Community-based monitoring efforts can help detect the presence of invasive mosquito species, such as *Ae. albopictus*, in new areas, e.g. through the Mosquito Alert App.

Monitoring ecological factors

To complement trapping and species identification, it is vital to monitor environmental conditions that influence mosquito resting and survival:

- **Vegetation cover:** Monitor vegetation density, which can provide shelter for resting mosquitoes.
- **Temperature variations:** Seasonal changes in temperature affect mosquito development and activity, so understanding these patterns can help optimize the timing of control interventions.

Systematic adult mosquito surveillance and monitoring enable the development of effective, targeted control strategies. By understanding the source of the problem, identifying the range of mosquito populations, and employing consistent monitoring, the goal is to minimize mosquito larvae and adult populations and, ultimately, to mitigate public health risks from mosquito-borne diseases.

6.3 Surveillance/monitoring of larvae

Effective mosquito larval surveillance/monitoring requires a structured approach, starting with defining the target species through both passive and active adult sampling, using historical data, literature, and a combination of sampling methods such as traps and human bait as presented in the previous chapter. The next step involves determining the area to be protected through larviciding-based mosquito control, whether on a small or large scale, considering the involvement of relevant stakeholders. The aim of mosquito larvae monitoring is to make only the necessary interventions at larval habitats (breeding sites) with proven presence of larvae and optimizing monitoring efforts in identifying the most productive larval habitats which need systematic follow up and control interventions. An inventory of geolocated, delineated and codified potential larval habitats across different environments—wetlands, rural/peri-urban systems, and urban systems—must be maintained. This will aid in rational monitoring and allow for the development of effective, site-specific intervention strategies.

6.3.1 Prerequisites for larval monitoring

Before initiating a larval monitoring program, it is essential to consider the following prerequisites to ensure accurate and effective surveillance:

Target species, bionomics and pathogens

- **Target species:** Determine which mosquito species is causing nuisance or represents a threat to public health related to the presence of a pathogen (see above on adult monitoring).
- **Flight capacity**: Determine the boundaries of the monitoring zone for each target mosquito species based on the known flight ranges to protect the area effectively.
- **Wetland habitats (in vicinity to urban areas)**: Use ecological indicators, such as vegetation types and hydrological regimes (inflows/outflows, ditches, channels), to classify wetlands. Use earth observation (EO) data to map inaccessible areas and identify accessible representative larval monitoring sites. Additionally, monitor access routes and protection status for potential larval habitats.
- **Rural/peri-urban systems**: Classify larval habitats by habitat types, such as rice fields, drainage channels, wastewater deposits, or periodically inundated areas. It is crucial to determine operationally manageable monitoring sites, whether they be area-, length- or point- based.
- **Population dynamics**: Identify potential larval habitats and understand how ecological factors such as water regime, vegetation, and temperature fluctuations influence mosquito larvae populations. Investigate the overwintering habits of the species, whether in egg, larval, or adult stages, to plan the start and end dates for larval monitoring.

Inventory of geolocated and codified potential larval habitats

- **Urban systems (public and private)**: For monitoring larval habitats in public areas (e.g., rainwater catch basins, fountains, cemeteries), and private premises (e.g., septic tanks, flowerpots, etc.), ensure compliance with data privacy regulations (GDPR). Establish reference scales such as “house” or “building block” to structure monitoring efforts.

6.3.2 Habitat typology and breeding sites

When identifying mosquito larval habitats, consider the following habitat typologies:

- **Stagnant temporary water bodies**: Include ditches, ponds, flooded meadows, or temporary forest pools.
- **Running waters**: Rivers, streams, and ditches, which may have temporary mosquito activity.
- **Bodies without vegetation**: Puddles, road tracks, or newly created ditches, often more temporary in nature.
- **Natural containers**: Tree holes, rock pools, or other naturally formed breeding containers.
- **Man-made (artificial) containers**: Pots, catch basins, and other human-made structures where water may accumulate.

Utilize GIS tools for mapping and tracking these breeding sites, ensuring that geolocated data is integrated with monitoring results.

6.3.3 Larval monitoring tools and techniques

Dipper

- **Application**: The dipper is the most commonly used tool for monitoring larvae in various habitats; it typically can hold about 250 ml and has a diameter of 10-15 cm.

- **Techniques**: Different dipping techniques – skimming, dumping, or inflow – are used, depending on the target species and habitat. The dipper is highly effective for shallow waters, and the most common measure is larvae per liter of water.
- **Advantages**:
  - Simple, requires no additional equipment.
  - Effective in shallow waters.
- **Disadvantages**:
  - Time-consuming for large areas.
  - Less effective for first-stage larvae.

Dipping Net

- **Application**: Suitable for larger areas with higher larval concentration, particularly useful in deeper waters, also suitable for rainwater catch basins
- **Techniques**: Involves different netting techniques, such as dipping, skimming, or dumping. A standard mesh width of 1.2 mm allows the capture of 1st-instar larvae, and transects of 1, 3, or 10 meters are commonly used.
- **Advantages**:
  - Highly effective for deep waters and particularly useful for *Anopheles* species.
  - Can be used for both dipping and netting, covering a large area.
- **Disadvantages**:
  - Difficult to use in shallow waters.
- **Common scale** (according to EID Méditerranée and Ecodevelopment technical guidelines, S. Gewehr, pers. comm.): Based on the number of larvae caught:
  - 0: No larvae.
  - 1: 1-8 larvae.
  - 2: 9-16 larvae.
  - 3: 17-24 larvae.
  - 4: >=25 larvae.

Container Water Aspiration Tools

- **Application**: These tools are used for cryptic breeding sites, such as small containers or sites with limited access.
- **Techniques**: Pipettes are used for easily accessible containers, while aspiration tools are useful for small openings. This method is effective for small cryptic habitats and allows complete drainage of containers.
- **Common Indices**:
  - **House Index (HI)**: Percentage of houses positive for mosquito larvae.
  - **Container Index (CI)**: Percentage of water-containing containers positive for larvae.
  - **Breteau Index (BI)**: Number of positive containers per 100 houses.

6.3.4 Data recording and thresholds

Effective larval monitoring also depends on accurate data recording and the establishment of action thresholds for interventions:

- **Descriptive characteristics**: Collect data on the breeding site's natural or anthropogenic origins, maximum water surface area, whether the water is permanent or temporary, organic matter content, and the presence of predators or competitors. Also, note the protection status of the habitat.
- **Seasonal larvae abundance**: Define and maintain a standardized abundance scale, ensuring accurate seasonal comparisons. For efficient data management, digitization is critical. Paper protocols should be replaced with electronic spreadsheets (e.g., MS Excel) or, preferably, relational databases (e.g. Access). Customized interoperable data recording systems, such as a field data recording app integrated with a geodatabase, are optimal for long-term analyses.
- **Ecological factors:** To complement larvae sampling and genus/species identification, it is vital to monitor environmental conditions that influence mosquito larval development and survival, such as the water levels, through regular assessment of wetlands, urban drainage systems, and other potential larval habitats for standing water, and the temperature variations since seasonal changes in temperature affect the development of mosquito larvae, so understanding these patterns can help optimize the timing of larvae and adult control interventions.

6.4 Surveillance/monitoring of eggs

While the surveillance and monitoring of mosquito eggs was not discussed during the 10th EMCA Workshop, it represents a common tool for the surveillance/monitoring of container-breeding mosquito species such as *Ae. albopictus*. Additionally, it is often used for the evaluation of the efficacy of specific mosquito control interventions against this species in Europe such as mass trapping, Sterile Insect Technique (SIT), adulticiding (ULV space treatments) and Integrated Vector Management approaches, and this often in combination with the surveillance of adult mosquito populations. Nevertheless, there is a lack of information in scientific literature about the correlation between the number of eggs collected by means of ovitraps and the adult population in a defined area and time, a fact which makes the use of this specific measurement as entomological endpoint for the efficacy evaluation of control interventions questionable.

For a qualitative approach the use of ovitraps is a reliable and easy-to-use tool, especially in areas where no other container-breeding *Aedes* species coexist. The visual differentiation of the eggs of different invasive *Aedes* species (such as *Ae. albopictus*, *Ae. japonicus* and *Ae. koreicus*) in areas where they coexist is difficult to achieve with standard stereoscope observations and need either a high-resolution stereomicroscope or molecular methods (Anicic et al. 2023). For a quantitative, wide-area and seasonal approach to *Ae. albopictus* population dynamics, due to the restricted active movement of this species and the unconfirmed correlation between the egg abundance and adult populations, the use of other monitoring tools should be preferred (such as specific adult traps and door to door inspections for the infestation of containers).

However, for the standardization of results obtained by the use of ovitraps, a harmonized protocol has been developed in the frame of the AIM Cost Action (CA17108), which is recommended to be used when scheduling a surveillance/monitoring program based on egg collections.

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7. Control interventions

7.1 Larval control

Larval Source Management (LSM) is the management of water bodies that are potential larval habitats for mosquitoes. Such management of water bodies is conducted to prevent the development of the immature stages (eggs, larvae and pupae) and hence the production of adult mosquitoes, with the overall aim of preventing or controlling nuisance and disease risk for human populations. At this point, all intervention types aimed at controlling mosquito larvae are part of integrated vector management (IVM) programs.

There are several types of LSM:

1. habitat modification: a permanent alteration to the environment, e.g. land reclamation, filling of water bodies
2. habitat manipulation: a recurrent activity, e.g. flushing of streams, drain clearance, physical control by using silicone-based products
3. biological control: the introduction of natural predators (fish or copepods) into water bodies, or by using microbiological agents (*Bti* or *L. sphaericus*)
4. chemical control: the regular application of chemical insecticides to water bodies
5. community involvement: includes the elimination of breeding sites and larval control in private areas and can be part of the environmental management and source reduction approaches.

Good practice statement , Low certainty evidence

Physical control by catch basin modification, silicone-based products and UNFO-PLS devices

- **Catch basin modification**, where applicable, should be used to constantly avoid the proliferation of *Ae. albopictus* and *Cx. pipiens* larvae. However, it should be complemented by other control measures in sewers where it cannot be applied and by control measures in other types of breeding sites that coexist in an area.
- **Silicone-based products** can be employed in catch basins and sewers to prevent the proliferation of *Ae. albopictus* and *Cx. pipiens* larvae. However, it should be complemented by alternative control methods and in various other breeding sites present in the target area.
- **UNFO-PLS devices** can be used in certain types of manholes and are effective against *Ae. albopictus* larvae. Other control measures should additionally be applied in other manhole types, sewers and in other existing breeding sites.

Evidence to decision

Certainty of the evidence Low

Catch basin modification: Very low certainty of evidence for reduction of immature stages for both *Ae. albopictus* and *Cx. pipiens*. The prevention of water accumulations in sewers will only prevent breeding in these sewers. It is unclear whether this treatment method can be transferred to other localities, due to differences in the construction of sewers and sewage systems.

Silicone-based products: Low to very low certainty of evidence on reduced immature stages and emerging adults of *Ae. albopictus* and *Cx. pipiens*. Silicone-based products may reduce number of mosquito larvae and pupae slightly. Their efficacy greatly relies on rain frequency and the construction of the water drains in which they are applied, as rain water flowing through the drains washes out the product. Retreatment after rain events is imperative.

UNFO-PLS devices: Low certainty of evidence on reduced immature stages and emerging adults of *Ae. albopictus* and *Cx. pipiens*.

Biological control by copepods

Biological control using copepods is not considered effective for reducing the risk of *Culex*- and *Aedes*-borne diseases and nuisance.

Evidence to decision

Certainty of the evidence

Very low

EP 7.1.4: biological control by using predators (copepods). Very low certainty; copepods may have little or no difference on larval reduction.

Biological control by Bti

Relying on biological control with Bti **alone** is not considered effective for mitigating the risk of *Culex*- and *Aedes*-borne diseases and nuisance.

Evidence to decision

Certainty of the evidence

Very low

EP 7.1.5: biological control by using microbiological agents (Bti). Very low certainty of evidence on decrease in immature stages.

Good practice statement , Low certainty evidence

Larviciding by insect growth regulators (IGRs)

The use of the IGR diflubenzuron in rain water catch basins is considered effective for the reduction of adult emergence but should be combined with other interventions.

Evidence to decision

Certainty of the evidence

Low

- **EP 7.1.6**: chemical larviciding using IGRs (pyriproxyfen). Low certainty of evidence. The intervention with pyriproxyfen does not reach 100% adult emergence inhibition as does diflubenzuron and has clearly less residual effect. By using the double of indicated dosage, results ameliorate and reach >95% Adult Emergence Inhibition (AEI) for three weeks in rain water catch basins.
- **EP 7.1.7:** chemical control by using insect growth regulators (IGRs) (diflubenzuron). Moderate evidence certainty. Three formulations of diflubenzuron showed significant efficacy against larval development for at least up to three weeks in rain water catch basins.
- **EP 7.1.8**: Autodissemination of IGRs. Very low certainty of evidence that IGR autodissemination reduces number of *Ae. albopictus* immature stages in containers. Further research in more locations and more repetitions is needed for supporting this evidence in other field conditions. Environmental factors (such as wind or shading) can affect the results in other habitats. Further studies are needed to prove and improve the auto-dissemination strategy under field conditions. Specifically, methods of application, concentrations and frequencies of treatment need to be clarified.

New

The autodissemination approach for IGRs **cannot be recommended** due to the lack of information about concentrations, frequencies and density of applications.

Evidence to decision

Certainty of the evidence

Very low

EP 7.1.8: Results seem promising but the study has been done in one city only, with 2 repetitions and at 2 sites only. Further research could have an important impact, which may change the estimates of the effect.

7.2 Adult control

Adult mosquito management focuses on the reduction and killing of adult mosquitoes, with the overall aim of preventing or controlling nuisance and disease risk for human populations. The main types of interventions are:

1. Spatial treatment - chemical control by using adulticiding
2. Residual spraying - chemical control by using adulticiding
3. Mass trapping
4. Irradiation-induced sterile insect technique (SIT) and *Wolbachia*-induced incompatible insect technique (IIT).

Good practice statement , Low certainty evidence

Mass trapping

Mass trapping is considered effective for small-scale focal interventions to control human disease and nuisance risk associated with *Ae. albopictus*.

Evidence to decision

Certainty of the evidence

Low

EP 7.2.1: Very mixed certainty (very low - moderate) of whether mass trapping reduced human landing rates of *Ae. albopictus* females, and thereby directly impacts disease risk of human populations in concerned areas.

Good practice statement , Moderate certainty evidence

Chemical control by adulticiding (outdoor residual spraying, ORS)

Under emergency conditions (outbreak of disease, high infestation with adult mosquitoes), the use of adulticides can be effective in quickly and massively reducing the population density and thus reduce the likelihood of disease transmission. However, the effect of adulticides is only short-term. For longer term impact, this method should be combined with more sustainable control strategies (e.g. reduction of larvae using Bti).

Evidence to decision

Certainty of the evidence

Moderate

EP 7.2.5: Use of pyrethroids. **Low to moderate certainty** whether ORS reduces number of *Ae. albopictus* adults. When measured by human landing catches, directly relevant for human health impacts.

Chemical control with adulticides (ultra-low volume space treatments, ULV)

Chemical control with ULV adulticides **alone** is not considered effective for reducing the risk of human disease and nuisance caused by *Ae. albopictus*.

Evidence to decision

Certainty of the evidence Very low

EP 7.2.3: chemical control by using adulticides (ultra-low volume space treatments, cold-fogging). **Very low certainty** whether adulticiding increases or decreases *Ae. albopictus* adult numbers.

EP 7.2.4: chemical control with adulticides (ultra-low volume space treatments, thermal fogging). **Very low certainty** whether thermal fogging decreases *Ae. albopictus* adult populations.

Irradiation-induced sterile insect technique (SIT)

Adult mosquito management with irradiation-induced SIT **alone** is not considered effectivefor controlling human disease and nuisance risk posed by *Ae. albopictus*.

Evidence to decision

Certainty of the evidence Very low

EP 7.2.6: use of irradiation-induced SIT only. **Very low certainty** whether SIT significantly reduced the number of *Ae. albopictus* fertile eggs.

EP 7.3.2: Use of irradiation-induced SIT plus source reduction. **Very low certainty** whether SIT applied for 10 weeks significantly reduced the mean number of *Ae. albopictus* eggs in a treated area compared to two control areas.

***Wolbachia*-induced incompatible insect technique (IIT)**

Adult mosquito management with IIT **alone** is not considered effective for controlling human disease and nuisance risk posed by *Ae. albopictus*.

Evidence to decision

Certainty of the evidence Very low

EP 7.2.7: While *Wolbachia* IIT decreases the percentage of viability in the eggs collected from the whole population by ovitraps, the percentage of decrease in egg viability is too small to determine whether *Wolbachia* IIT can make a difference in the *Ae. albopictus* population size

7.3 Integrated vector management (IVM)

IVM is defined by the WHO as "a rational decision-making process to optimize the use of resources for vector control" (WHO 2012). IVM is intended to utilize the best cost-benefit combination of all available control methods in a sustainable way respecting the environment in order to reduce the vector density and/or the vector-human contacts to levels not posing a public health concern.

Good practice statement , Low certainty evidence

Combination of interventions targeting both immature and adult mosquito stages

Integrated vector management (IVM) should include a diverse variety of complementary methods. In order to be more effective, IVM should involve an increasing number of control methods depending on the nuisance or human health risk level.

Evidence to decision

Certainty of the evidence

Low

1. **EP 7.3.1:** chemical control by using IGRs (pyriproxyfen) **and** adulticiding using permethrin, *B. bassiana* or Spinosad. **Low certainty** of evidence. Pyriproxyfen + permethrin may increase adult emergence inhibition slightly. The combination of adulticidal and larvicidal products can increase effectiveness by simultaneously controlling adults and larvae, and by expanding persistence.
2. **EP 7.3.2**: Use of irradiation-induced SIT plus source reduction. **Very low certainty** whether SIT applied for 10 weeks significantly reduced the mean number of *Ae. albopictus* eggs in a treated area compared to two control areas.
3. **EP 7.3.3**: source reduction **and** larviciding **and** adulticiding in sensitive areas. **Moderate certainty of evidence** that the combination of these three interventions reduces number of *Ae. albopictus* eggs.
4. **EP 7.2.2:** larviciding (Bti) **and** Sterile Insect Technique (SIT). **Very low certainty** whether combination of larviciding and SIT reduced number of *Ae. albopictus* eggs and adults.
5. **EP 7.3.4:** Larval source reduction (LSR) **and** biological larviciding (Bti) **and** SIT. **Very low certainty** whether combination of LSR, Bti and SIT reduced number of *Ae. albopictus* eggs. This intervention is favourable, but the measured effect is very low in the study area. It could be improved repeating the study at other locations and during several seasons. SIT in combination with source reduction and community participation could aid to reduce populations of *Ae. albopictus* in isolated areas. Introductions of specimens from other areas into the treated areas should be measured.
6. **EP 7.3.5:** Larval source reduction (LSR) **and** larvicide treatment **and** rubbish removal **and** adulticiding in recreation parks. **Very low certainty** whether combination of LSR, larviciding, solid waste management and adulticiding reduced number of *Ae. albopictus* eggs. Any difference in numbers of *Ae. albopictus* eggs in intervention and control areas might be attributed to additional non-documented interventions.
7. **EP 7.3.6:** chemical control by using IGRs (diflubenzuron) **and** adulticides (pyrethroids). **Very low certainty** whether combination of IGRs and adulticiding reduced number of *Ae. albopictus* adults.
8. **EP 7.3.7:** Larviciding in public road drains **and** door-to-door strategy (DtoD): monthly inspections of private properties **and** larval treatment of permanent breeding sites **and** removal/ inactivation of occasional breeding sites **and** predatory copepods in large permanent containers **and** direct information to residents **and** communication campaigns. **Low certainty** of evidence whether combined DtoD strategy reduced number of *Ae. albopictus* eggs and adults when compared with public larviciding only. However, as significantly reduced adult numbers were measured by HLC, this could have direct beneficial impact on human health risks.
9. **EP 7.3.8**: Source reduction, biological larviciding (Bti) and public education campaign. **Very low certainty** of evidence whether combined strategy reduced number of *Ae. albopictus* eggs.
10. **EP 7.3.9**: IGRs (Diflubenzuron) **and** Bti **and** extensive information campaign. **Low certainty** of evidence whether combined larviciding and information campaign strategy reduced number of *Ae. albopictus* eggs and adults.

7.4 Summary of non-evidence publications

The reviews conducted as described above yielded a broad range of publication types, including original research articles and review articles published in peer-reviewed journals, book chapters, and technical reports, mainly from WHO. Studies that provided real-world evidence on the efficacy of vector control interventions were included in the GRADE assessment process to inform the recommendations stated below. Studies without empirical evidence were assessed on whether their findings might contribute to the good practice statements. Owing to the mixed quality and relevance of the publications identified in the scientific review, not all of them were included in this section.

7.4.1 Research articles

A rapid communication article by La Ruche et al. (2010) describes the first autochthonous dengue cases detected in France in 2010. Local transmission of dengue calls for further enhanced surveillance, active case finding and vector control measures to reduce the spread of the virus and the risk of an epidemic.

Flacio et al. (2015) describe a planned surveillance and control program against the anticipated invasion of *Ae. albopictus* in the Swiss canton Ticino. As control interventions, larviciding was conducted using biocides (*Bti*) and IGRs (diflubenzuron), and adulticiding by permethrin and cypermethrin. Although the program was unsuccessful in preventing the invasion of *Ae. albopictus*, it allowed a management of the vector population and, compared to other regions in northern Italy, seemed to slow the spread. Owing to their effectiveness and safety, the use of *Bti* and diflubenzuron was recommended to control mosquito larvae. Pyrethroids are suggested as the best option for adulticiding.

7.4.2 Review articles

A review by Bellini et al. (2014) summarises the key control interventions directed against WNV vectors, especially *Culex* spp. While most of these interventions are routinely used in Europe, most studies evaluating their impact on WNV incidence are from the US. There, the preferred intervention is aerial ULV spraying of adulticides, which is currently banned in EU unless exempt under specific circumstances.

Key interventions recommended for use against WNV are environmental management and personal protection, followed by larviciding (biological, chemical and microbial), whereas community participation is considered potentially useful. The authors did not recommend ground adulticiding, and mass trapping lacks evidence of impact.

Baldacchino et al. (2015) provide a comprehensive overview of different types of control interventions for invasive *Aedes* mosquitoes, including environmental methods, mechanical control, biological and chemical control methods. The authors describe the efficacy and advantages/disadvantages of each method and draw on evidence from outside Europe, without giving specific data.

Of the control methods discussed in this review, several have been successfully used against *Ae. albopictus*, mainly outside of Europe. These include source reduction, predation by copepods, larvicide application, adulticide spraying and SIT. Mechanical methods have been evaluated in large areas, but only against *Ae. aegypti*; lethal ovitraps or gravid traps might also be effective against *Ae. albopictus*. New approaches such as pyriproxyfen autodissemination, ATSB or IIT based on Wolbachia infection have shown promising results in laboratory conditions or semi-field experiments, supporting their potential for future implementation at a larger scale.

Wilson et al. (2015) do not assess any specific interventions but make general and very useful recommendations on the correct design of trials to accurately assess efficacy of an intervention, including epidemiological outcomes. This is particularly useful, as the review mentions the adoption of GRADE by WHO in 2008, thus linking to the underlying rationale of our guidelines.

The authors recommended to ensure sufficient scale and follow-up time of intervention trials, with RCTs considered gold standard for evidence. Moreover, sufficient sample/group size must be ensured for statistical significance. Epidemiological outcomes should be used as primary endpoints, with entomological outcomes as possible secondary endpoints. Standardised data formats should be followed.

Chaskopoulou et al. (2016) give a summary review of *Culex* surveillance and control strategies used in the mid-2010s in Italy, France, Serbia and Greece, aimed at curbing WNV outbreaks. While no primary evidence was generated for this review, it is noteworthy that timing of vector control applications seems to be an important cornerstone for the implementation of effective WNV control and emergency measures can be largely ineffective if delayed until the index case appears. There is a need to refine the understanding of the most effective vector control tools in order to optimize the resources and design proactive, evidence based WNV control strategies.

Achee et al. (2019) reviewed alternative strategies for mosquito-borne arbovirus control, mainly from outside Europe. Owing to the fact that classical vector control has failed to prevent epidemics and arrest expanding geographic distribution of key arboviruses, such as dengue, there is a need for new methods. These alternative strategies include the use of novel larvicides, and new applications, such as entomopathogenic fungi, pyriproxyfen and autodissemination, but also spatial repellents, traps, attractive targeted sugar baits (ATSBs), insecticide-treated materials, SIT, release of insects with dominant lethality, *Wolbachia* and gene drives.

Encouraging results of irradiation-based SIT have been obtained for *Ae. albopictus* in Italy. Many of the other methods or control agents listed still need to be tested for epidemiological effectiveness or are not applicable in Europe.

Bellini et al. (2020) outlined a practical management plan for invasive mosquito species in Europe, focusing on Ae. albopictus. While no empirical evidence was generated for this article, the authors provide comprehensive control recommendations against Ae. albopictus. In particular, this management plan to control Ae. albopictus in areas where the species is well established includes several activities which may be modulated according to local resource availability and cost-benefit evaluation. The management plan primary component activities are as follows:

- Public health risk assessment
- Monitoring by ovitraps
- Standard control measures in public and private areas
- Community participation
- Door-to-door control measures in private areas
- Emergency control measures in response to the detection of imported cases of dengue, chikungunya or Zika
- Quality control of treatment efficacy
- Prevention of resistance to insecticide

Each component is explained in detail to provide a comprehensive practical and technical guidance to local authorities in organizing the vector control activities in the best possible way. Together with the routine actions aimed at reducing the population density of Ae. albopictus, the management plan includes an emergency vector control plan to reduce the risk of an epidemic in case of detection of infected persons.

Allen et al. (2021) provide an overview of community participation for vector control of *Aedes* species, with most studies identified from USA or Europe, but very few from low-income countries that are much more affected by VBDs. Citizen Science platforms such as MosquitoAlert are being cited a positive example to ensure community engagement and support.

The citizen science studies in this review showed promising results in terms of the feasibility and cost-effectiveness for incorporating these strategies into routine mosquito management. Long-term effectiveness of collaborating with the community on sustainable mosquito management outcomes requires further research.

Giunti et al. (2023) give an overview of invasive mosquito species in Europe, with a special focus on *Aedes albopictus*. The authors also summarise the currently employed methods for vector surveillance and control. Moreover, they call for increased funding to manage and control invasive species, and to monitor insecticide resistance as a growing threat.

IVM should be adopted according to the local conditions as well as to the national/regional regulations and should comprise all appropriate available surveillance and control tools. In areas where the invasive species is/are established, control should focus on the containment of the species below the noxious and epidemiological thresholds.

7.4.3 Book chapters

Becker & Zgomba (Chapter 21 in Takken & Knols 2007) give an historical overview of mosquito control in Europe, including the founding and role of the EMCA. Programs in various European countries are being described, with a strong focus on the use of *Bacillus thuringiensis israelensis* (*Bti*) and *Lysinibacillus sphaericus* (previously known as *Bacillus sphaericus*). Furthermore, the authors recommend that the best strategies and techniques for mosquito control should be integrated, well suited to achieving the objectives of the programmes, and at the same time they should preserve the ecological balance. This approach has gained much support, not only among scientists, but also among the general public in the last 20 years.

On a more localised level, Becker (2010) reviews the communal control programme implemented in the upper Rhine Valley in Germany. The article describes the prerequisites for the development of a microbial mosquito control strategy, the planning of the intervention, and the importance of monitoring the control program, including with the support provided by GIS. The application of microbial control agents (MCAs) against mosquitoes is thoroughly discussed, highlighting their effectiveness, safety and possible advantages over adulticiding treatments.

While Becker & Lüthy (2017) do not provide primary sources of evidence for the efficacy of *Bti* and *L. sphaericus*, they describe analytically their use in different European countries for wide-area mosquito control against nuisance mosquitoes, emphasizing the environmental safety of both products. The authors describe that *Bti* and *L. sphaericus* introduction can effectively reduce the number of nuisances by *Culex* mosquitoes to a tolerable level in comparison to non-treated areas.

7.4.4 Technical & meeting reports

In 2015, the WHO Vector Control Advisory committee (VCAG) assessed the genetically modified OX513A *Aedes aegypti* prototype developed by the company Oxitec and concluded that entomological efficacy should be quantified using measures directly related to mosquito population abundance (adult density, pupa/person measures) in addition to the ovitrap indices that had been incorporated into the monitoring procedures. Moreover, the effects of releasing homozygous OX513A *Ae. aegypti* females and of the less than 100% penetrance of lethality in heterozygous offspring on vectorial capacity and dengue virus transmission dynamics should be evaluated. Lastly, the VCAG concluded that this prototype was supported by sufficient information to warrant moving into Stage 3 development, i.e. cluster randomized trials to evaluate epidemiological effectiveness.

In 2017, the VCAG assessed the combined use of SIT and IIT to control *Aedes* populations based on presented evidence that the combined approach confers high levels of sterility to *Aedes* mosquitoes (as high as 100%). The VCAG considered that this combined technology has important potential for long-term control of *Aedes*-borne diseases. However, the VCAG strongly recommended detailed review of results from entomological field trials prior to the validation of this intervention. More specifically, the VCAG requested to perform additional laboratory studies to assess the transmission blocking effects of different *Wolbachia* strains in the mosquito for different dengue virus strains and serotypes and other alpha- and/or flaviviruses.

Taken together, these articles, reviews, book chapters and technical reports broadly recommend a wide spectrum of interventions on surveillance and control of MBDs in Europe as part of successful IVM campaigns tailored to specific local settings and conditions. Entomological surveillance is considered essential for the assessment of the effectiveness of IVM. For larviciding, *Bti* has most uniformly been recommended for inclusion in IVM programs against MBDs, especially when combined with environmental management, personal protection, and community engagement (without, however, providing evidence in form of raw data from field experiments). Pyrethroids are suggested as a suitable option for adulticiding, owing to their knockdown effect and persistence. Novel interventions (such as *Wolbachia*-based IIT) deserve further testing in larger-scale trials to evaluate their effectiveness.

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8. Conclusions

The risk of MBDs (such as WNV or dengue) is increasing in Europe, expressed through the increasing frequency of locally acquired MBD cases in combination with the geographical expansion of recorded cases. This increased risk is linked to climate change, environmental changes, global trade and travel, and the continously expanding range of invasive vector species, such as *Aedes albopictus* across Europe, and the recent introduction and establishment of *Ae. aegypti* in Cyprus. Mosquito control measures are crucial for managing these risks. Given the absence of drugs or vaccines against the medically most important MBDs in Europe, mosquito control interventions remain the only available countermeasure to reduce or prevent human infections.

There is a need for continuous entomological monitoring in areas at risk in order to define the targets for any planned mosquito control intervention, and would allow for setting entomological endpoints for any control interventions in the absence of epidemiological endpoints (such as reduced human disease incidence). Owing to this lack of epidemiological endpoints in all studies used for this guideline document, it was agreed to use entomological endpoints as a proxy for the biting activity of mosquitoes and thus the potential transmission of a disease.

Overall, we found poor evidence for the efficacy of all analysed mosquito control interventions, coupled with very heterogenous reporting of a wide variety of entomological endpoints, which made comparison of the outcomes from different studies very difficult. Taken together, this did not allow for more stringent recommendations for or against specific control interventions.

Analysis of grey literature showed that several mosquito control interventions (either active ingredients or methodologies), while being used in practice, seem not to have been evaluated yet for their efficacy in Europe and for the target species (eg, s-methoprene, spinosad, larvivorous fish, etc), according to the systematic literature review performed for these guidelines.

There is a need for further, systematic research about the efficacy of mosquito control interventions. To this aim, the types of appropriate studies should be defined and examples should be provided (eg, before-after studies, clustered Randomized Control Trials, case-control studies, etc).

Moreover, there is a need for technical guidance to plan and conduct high-quality studies in order to assess the efficacy of an intervention. It is therefore necessary to define a minimum standard set of data that should be recorded for each type of environment investigated, as well as setting out standardised methodologies for data analysis in order to ensure comparability of results.

There is also a lack of research on the environmental side effects of the systematic use of mosquito control interventions under real-world conditions. This would be important for making informed environmental cost-benefit decisions about adequate interventions in each ecological setting. Likewise, insecticide resistance in both immature and adult mosquitoes should be systematically monitored.

In a next step, it should be considered to expand the literature search to other continents to include any studies containing human epidemiological endpoints, thus improving the certainty of evidence for different mosquito control interventions.

These guidelines represent the first attempt to implement the GRADE system, which had originally been developed for clinical trials, to evaluate the efficacy of mosquito control method in urban settings in Europe in a systematic and rigorous fashion. We hope this approach will inspire the development of similarly systematic guidelines elsewhere.

9. Contributors and interests

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Declaration of interests:

Members of the GDG and SRT reported relevant financial or intellectual interests. Where relevant, these members were recused from assessing interventions based on the use of their products or studies in which they had participated.

The relevant declared interests are summarized as follows:

- Dr. Eleonora Flacio is the leader of a surveillance and control group based in Southern Switzerland, which is co-author of the following articles chosen: Parrondo Monton et al., Evaluation of the Manhole Physical Pest Management System UNFO-PLS for Mosquito Control in Southern Switzerland. 2023, https://doi.org/10.52004/JEMCA2022.0007; Suter et al., Surveillance and control of Aedes albopictus in the Swiss-Italian border region: Differences in egg densities between intervention and non-intervention areas. 2016, https://doi.org/10.1371/journal.pntd.0004315; Ravasi et al., Effectiveness of integrated Aedes albopictus management in southern Switzerland. 2021, https://doi.org/10.1186/s13071-021-04903-2; Flacio et al., Strategies of a thirteen year surveillance programme on Aedes albopictus (Stegomyia albopicta) in southern Switzerland. 2015, https://doi.org/10.1186/s13071-015-0793-6.
- Dr. Andreas Rose (https://orcid.org/0000-0003-1899-7710) is founder and co-proprietor of the company, Biogents AG (www.biogents.com), which develops, produces, and distributes mosquito traps such as the BG-Sentinel, the BG-Mosquitaire, the BG-Pro, the BG-Suna, the BG-GAT, or the BG-Counter. These devices are used in the surveillance and control of mosquitoes, with one focus being Aedes aegypti and Ae. albopictus. He and other Biogents employees (co-)authored scientific publications on the use of Biogents traps as surveillance or mass trapping tools (e.g. Englbrecht et al. 2015, DOI: 10.2987/14-6444.1; Degener et al. 2015, DOI: 10.1590/0074-02760140374; Degener et al. 2014, DOI: 10.1590/0074-0276140234; Degener et al. 2014, DOI: 10.1603/ME13107; Hiscox et al. 2014, DOI: 10.1186/1475-2875-13-257; Becker et al. 2013, DOI: 10.1007/s00436-012-3230-1; Kröckel et al. 2006, DOI: 10.2987/8756-971X(2006)22[229:NTFSOA]2.0.CO;2).

- Dr. Francis Schaffner is co-author of several articles on mosquito surveillance methodology: Bellini et al. Practical Management Plan for invasive mosquito species in Europe: I. Asian tiger mosquito (Aedes albopictus). 2020, doi: 10.1016/j.tmaid.2020.101691. Martinou et al. A call to arms: Setting the framework for a code of practice for mosquito management in European wetlands. 2020, doi: 10.1111/1365-2664.13631. Schaffner et al., Development of guidelines for the surveillance of invasive mosquitoes in Europe. 2013, doi: 10.1186/1756-3305-6-209

All other members declared no potential conflicts of interest.

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albopictus and Culex pipiens in Italy. Journal of the American Mosquito Control Association 2015;31(1):32-43 Pubmed Journal

29. Veronesi R, Carrieri M, Albieri A, di Cesare S, Panziera M, Bazzocchi G, et al. Field evaluation of AQUATAIN AMF TM as a mosquito larval and pupal control agent in different breeding sites in Northern Italy. Poster E-SOVE Conference 2016 2016. Website

Annex: All evidence profiles, sorted by sections

1. Abbreviations

2. Glossary

3. Executive summary

4. Introduction

4.1. Public Health context of mosquito-borne diseases (MBDs)

4.2. Objectives of the document and expected outcomes

4.3. Built-up areas

4.4. Points of entry

4.5. Public areas

4.6. Private properties

4.7. Governance – stakeholders

4.8. Policies impacting built-up areas and mosquitoes

4.9. Mosquito species relevant as pests and/or potential vectors of diseases in built-up areas

4.10. References to Chapter 4

5. Methods

5.1. General methodology

5.2. Literature search by SRT teams

6. Surveillance/Monitoring

6.1. Planning of a surveillance/monitoring project

6.2. Surveillance/monitoring of adults

6.3. Surveillance/monitoring of larvae

6.4. Surveillance/monitoring of eggs

6.5. References to Chapter 6

7. Control interventions

7.1. Larval control

Clinical question/ PICO

Population: human populations in built-up European areas
Intervention: larval source reduction through environmental management (catch basin modification)
Comparator: no control action

Outcome Timeframe	**Study results and measurements**	**Comparator** no control	**Intervention** environmental management	**Certainty of the evidence** (Quality of evidence)	**Summary**
Reduction of Ae. albopictus larvae & pupae numbers [1]	Measured by: plastic dipper & hand collector High better [2]	68.11 % reduction in mean Ae. albopictus larvae + pupae per visit and sewer between 2018 and 2020. Difference:	100 % reduction in mean Ae. albopictus larvae + pupae per visit and sewer between 2018 and 2020. **MD 31.89 lower**	Very low Due to serious indirectness. Low comparability to other settings and locations. Outcome measured on population of immature mosquitoes, not	The prevention of water accumulations in sewers will only prevent breeding in these sewers. There needs to be combined with reduction measures of other mosquito sources = breeding sites. Unclear if treatment method can be transferred to other

Outcome Timeframe	**Study results and measurements**	**Comparator** no control	**Intervention** environmental management	**Certainty of the evidence** (Quality of evidence)	**Summary**
				human target population. [3]	localities, due to differences in the construction of sewers and sewage systems.
Reduction of Cx pipiens larvae & pupae numbers [4]	Measured by: plastic dipper & hand collector High better [5]	75.28 % reduction in mean Cx pipiens larvae + pupae per visit and sewer between 2018 and 2020. Difference:	100 % reduction in mean Cx pipiens larvae + pupae per visit and sewer between 2018 and 2020. **MD 24.71 lower**	Very low Due to serious indirectness. Low comparability to other settings and locations. Outcome measured on population of immature mosquitoes, not human target population. [6]	The prevention of water accumulations in sewers will only prevent breeding in these sewers. There needs to be combined with reduction measures of other mosquito sources = breeding sites. Unclear if treatment method can be transferred to other localities, due to differences in the construction of sewers and sewage systems.

2, 5. [22]. **Comparator:** [22].
3, 6. **Risk of Bias: no serious.** Unclear how sewers were selected for control and treatment arm.. **Inconsistency: no serious.** Visit intervals vary between years and sewers.. **Indirectness: serious.** Differences between the population of interest and those studied. **Imprecision: no serious.** Only data from one study..

References

22. Montalvo T, Higueros A, Valsecchi A, Realp E, Vila C, Ortiz A, et al. Effectiveness of the Modification of Sewers to Reduce the Reproduction of Culex pipiens and Aedes albopictus in Barcelona, Spain. Pathogens (Basel, Switzerland) 2022;11(4) Pubmed Journal

Clinical question/ PICO

Population: human populations in built-up European areas
Intervention: physical control by UNFO-PLS
Comparator: no such control action

Outcome Timeframe	**Study results and measurements**	**Comparator** no such control action	**Intervention** physical control by UNFO-PLS	**Certainty of the evidence** (Quality of evidence)	**Summary**
Mosquito larvae reduction	Measured by: 3 dips + aquarium net High better Based on data from 24 participants in 1 studies. [1] (Randomized controlled) Follow up: 4 months / year,	0 % of larvae reduction per manhole (Mean) Difference:	95.26 % of larvae reduction per manhole (Mean) **MD 95.26 higher** (CI 95% 92.6 higher	Low Data from only one study Little information is given about the surroundings of the manholes. The	Physical control by using unfo-pls may improve slightly mosquito larvae reduction

Outcome Timeframe	**Study results and measurements**	**Comparator** no such control action	**Intervention** physical control by UNFO-PLS	**Certainty of the evidence** (Quality of evidence)	**Summary**
	2 years.		— 97.2 higher)	authors write about urban areas and industrial areas. However, it is not known whether there are many trees or bushes around, for example. After heavy rain showers or storms, the leaves could clog the closure of the UNFO-Pest device and this would have a massive impact on the result. It would therefore be desirable if the sample size were larger. It is certainly good for an initial study, but it would be desirable if there were a larger study with more gullies in different municipalities with very different settlement structures. This method does not kill mosquitoes, but merely prevents females from using appropriately equipped gullies as breeding sites. The females can fly elsewhere and lay their eggs there if they are able to find another breeding site. Therefore, it cannot be said that the reduction of larvae and pupae in the gullies reduces the nuisance caused by tiger mosquitoes and thus the risk of virus transmission. [2]	

1. [23]. **Comparator:** Primary study [23]. **Supporting references:** [23],
2. **Inconsistency: no serious. Indirectness: no serious. Imprecision: serious.** Data from only one study Little information is given about the surroundings of the manholes. The authors write about urban areas and industrial areas. However, it is not known whether there are many trees or bushes around, for example. After heavy rain showers or storms, the leaves could clog the closure of the UNFO-Pest device and this would have a massive impact on the result. It would therefore be desirable if the sample size were larger. It

is certainly good for an initial study, but it would be desirable if there were a larger study with more gullies in different municipalities with very different settlement structures.. **Publication bias: no serious.** The authors stated that they cooperated with the inventor of the product and the manufacturer of the device and they received compensation from them for the field evaluation. They would not benefit from the sale or non-sale of the product. I am very sure that this is not the case either. .

References

23. Parrondo Monton D., Würsch G., Tanadini L.G., Wyman M.T., Haverkamp P.J., Flacio E.. Evaluation of the manhole physical pest management system UNFO-PLS for mosquito control in southern Switzerland. Journal of the European Mosquito Control Association 2023. Journal Website

Clinical question/ PICO

Population: human populations in built-up European areas
Intervention: physical control by silicone-based products
Comparator: no such control action

Outcome Timeframe	**Study results and measurements**	**Comparator** no control	**Intervention** silicone-based products	**Certainty of the evidence** (Quality of evidence)	**Summary**
Number of emerging Cx pipiens adults [1] 1-3 weeks 1 Not Important	Measured by: Emergence traps Lower better Based on data from 50 participants in 1 studies. [2] (Observational (non-randomized)) Follow up: 3 weeks.	16.9 mean number of emergent adults (Mean) Difference:	1.2 mean number of emergent adults (Mean) **MD 15.7 lower** (3.2 lower — 34.3 higher)	Low Outcome measured on population of adult mosquitoes, not human target population.	Silicone-based products may improve number of emerging Cx pipiens adults. Their efficacy greatly relies on rain frequency, as rain water flowing through the drains washes out the product. Retreatment after rain events is imperative.
Number of emerging Ae. albopictus adults [3] 1-3 weeks 1 Not Important	Measured by: Emergence traps Lower better Based on data from 50 participants in 1 studies. [4] (Observational (non-randomized)) Follow up: 3 weeks.	31.9 mean number of emergent adults (Mean) Difference:	2.3 mean number of emergent adults (Mean) **MD 29.6 lower** (6.6 lower — 40.6 higher)	Low Outcome measured on population of adult mosquitoes, not human target population.	Silicone-based products may improve number of emerging Cx pipiens adults. Their efficacy greatly relies on rain frequency, as rain water flowing through the drains washes out the product. Retreatment after rain events is imperative.
Reduction of Cx pipiens larvae & pupae [5] 3 weeks	Measured by: Dipping net Lower better [6] (Randomized controlled)	25.2 Mean density of Cx pipiens larvae + pupae per catch basins 28 d post-treatment (Mean) Difference:	9.9 Mean density of Cx pipiens larvae + pupae per catch basins 28 d post-treatment (Mean) **MD 15.3 lower**	Very low Due to serious imprecision. Low comparability to other settings and locations. Limited rain may have skewed results to the positive. Outcome measured on population of immature	Silicone-based products (in this assessment, 5 ml Aquatain AMF per catch basin) may improve reduction of cx pipiens larvae & pupae slightly. Their efficacy greatly relies on rain frequency and the construction of the water drains in which they are applied, as rain water flowing through the drains

Outcome Timeframe	**Study results and measurements**	**Comparator** no control	**Intervention** silicone-based products	**Certainty of the evidence** (Quality of evidence)	**Summary**
				mosquitoes, not human target population. [7]	washes out the product. Retreatment after rain events is imperative.
Reduction of Ae. albopictus larvae & pupae [8] 3 weeks	Measured by: Dipping net Lower better [9] (Randomized controlled)	54.5 Mean density of Ae albopictus larvae + pupae per catch basins 28 d post-treatment (Mean) Difference:	9.4 Mean density of Ae albopictus larvae + pupae per catch basins 28 d post-treatment (Mean) **MD 45.1 lower**	Very low Due to serious imprecision. Low comparability to other settings and locations. Limited rain may have skewed results to the positive. Outcome measured on population of immature mosquitoes, not human target population. [10]	Silicone-based products (in this assessment, 5 ml Aquatain AMF per catch basin) may improve reduction of cx pipiens larvae & pupae slightly. Their efficacy greatly relies on rain frequency and the construction of the water drains in which they are applied, as rain water flowing through the drains washes out the product. Retreatment after rain events is imperative.
Number of dead or alive mosquito larvae and pupae [11] 1-42 days 1 Not Important	[12] (Observational (non-randomized)) Follow up: 42 days.	Water drains before and 1 to 42 days after the treatment with Aquatain AMF were inspected for mosquito larvae using a dipper. Immediately after the application of the product, dead mosquitoes of all stages were found in the drains. Frequent rainfalls however washed the product out of the drains, leading to a recolonization of the drains with larvae within a week, especially in drains with larger drain pipes. An efficacy of the product was observed for up to two weeks after application.		Very low Due to very serious indirectness, Due to extremely serious imprecision. Outcome measured on population of immature mosquitoes, not human target population. [13]	Silicone-based products may improve number of dead or alive mosquito larvae and pupae slightly. Their efficacy greatly relies on rain frequency and the construction of the water drains in which they are applied, as rain water flowing through the drains washes out the product. Retreatment after rain events is imperative.

2, 4. [16]. **Comparator:** [16]. **Supporting references:** [16], We calculated mean values for the three first weeks after each application in order to be comparable for mean numbers..
6, 9. [29]. **Comparator:** [29]. **Supporting references:** [29], Mean densities taken from measuring height of bars for trial 3 post 28 d in figures, and relating them to values given on the y axis..
7, 10. **Imprecision: serious.** Only data from one study, Low number of catch basins.
12. **Supporting references:** [18],
13. **Indirectness: very serious.** Direct comparisons not available.. **Imprecision: extremely serious.** Wide confidence intervals due to inconsistent data collection times and unclear variation in teh construction of treatment sites..

References

16. Drago A, Simonato G, Vettore S, Martini S, Marcer F, di Regalbono AF, et al. Efficacy of Aquatain® Against Culex pipiens Complex and Aedes albopictus in Catch Basins in Italy. Journal of the American Mosquito Control Association 2020;36(1):51-54
Pubmed Journal

18. Kalan K, Šušnjar J, Jugovic J, Ivović V. Pilot Study of Invasive Mosquito Species Control in Selected Slovenian Municipalities.

Acta Veterinaria 2022;72(2):256-267 Journal Website

29. Veronesi R, Carrieri M, Albieri A, di Cesare S, Panziera M, Bazzocchi G, et al. Field evaluation of AQUATAIN AMF TM as a mosquito larval and pupal control agent in different breeding sites in Northern Italy. Poster E-SOVE Conference 2016 2016. Website

Clinical question/ PICO

Population: human populations in built-up European areas
Intervention: biological control by predators (copepods)
Comparator: no such control action

Outcome Timeframe	**Study results and measurements**	**Comparator** no control	**Intervention** copepods	**Certainty of the evidence** (Quality of evidence)	**Summary**
Larvae reduction [1] 103 days in 2007 1 Not Important	Measured by: Dipper Lower better Based on data from 20 participants in 1 studies. [2] (Observational (non-randomized)) Follow up: 103 days.	23.2 (Larvae +pupae) per dip (Mean) Difference:	12.02 (Larvae +pupae) per dip (Mean) **MD 11.18 lower**	Very low Due to serious risk of bias: Only results from catch basins where Copepods survived were reported. The suggested treatment cannot be applied to all breeding sites in the urban environment. [3]	Copepods may have little or no difference on larvae reduction. The study showed no statistical evidence of larvae reduction in treated populations. Besides, suggested treatment sites are not representative of the diverse breeding sites in Urban environment.

2. [28]. Only the field experiment in catch basins is used for evidence. Semi-field and laboratory experiments are not regarded.. **Comparator:** [28]. Only the 2007 experiment can be used due to additional treatment with Bti in the 2008 experiment, data extracted from graph 4 b,c. **Supporting references:** [28], Only the 2007 experiment in catch basins is used. Data are extracted from graph 4b,c. Data from Culex pipiens and Ae. albopictus are added up..
3. **Risk of Bias: serious.** Selective outcome reporting: larvae count was performed in catch basins where the copepodes survived. . **Imprecision: no serious.** Only data from one study.

References

28. Veronesi R, Carrieri M, Maccagnani B, Maini S, Bellini R. Macrocyclops albidus (Copepoda: cyclopidae) for the Biocontrol of Aedes albopictus and Culex pipiens in Italy. Journal of the American Mosquito Control Association 2015;31(1):32-43 Pubmed Journal

Clinical question/ PICO

Population: human populations in built-up European areas
Intervention: biological control by microbiological agents (Bti)
Comparator: no such control action

Outcome Timeframe	**Study results and measurements**	**Comparator** no control	**Intervention** microbiological agents	**Certainty of the evidence** (Quality of evidence)	**Summary**
Decrease mosquito larvae 24 hours 1 Not Important	Measured by: Ovitraps High better Based on data from 8 participants in 1 studies. [1] (Observational (non-randomized))	4 % larvae mortality (Mean) Difference:	99.9 % larvae mortality (Mean) **MD 95.9 higher** (94.7 higher — 100 higher)	Very low Due to serious imprecision, Due to serious indirectness [2]	We are uncertain whether microbiological agents will finally decrease mosquito larvae under field conditions. The semi-field trial setup of the reference is very restrictive.

1. [26]. semifield assay. **Comparator:** [26]. **Supporting references:** [26],
2. **Risk of Bias: no serious.** Small sample, clear methodology but not conclusive. **Indirectness: serious.** The study design does not meet the research question as set up by the title. The outcomes of the research are not extendible, generalizable and transferable. . **Imprecision: serious.** due to the use of 4 abundance classes through estimation instead of counting . **Publication bias: no serious.** The search in the review was not exhaustive. .

References

26. Toma L, Severini F, Bella A, Romi R. A semifield evaluation of Vectobac DT (ABG-6499), a new formulation of Bacillus thuringiensis israelensis for control of Aedes albopictus. Journal of the American Mosquito Control Association 2003;19(4):424-9 Pubmed

Clinical question/ PICO

Population: human populations in built-up European areas
Intervention: chemical control by insect growth regulators (pyriproxyfen)
Comparator: no such control action

Outcome Timeframe	**Study results and measurements**	**Comparator** no control	**Intervention** insect growth regulator (pyriproxyfen)	**Certainty of the evidence** (Quality of evidence)	**Summary**
Reduction of adult emergence 2 months 2006,2007 1 Not Important	Measured by: Emergence traps High better Based on data from 131 participants in 3 studies. [1] (Randomized controlled) Follow up: Weekly for 3 weeks.	20.6 Per cent inhibited adult emergence (Mean) Difference:	83.37 Per cent inhibited adult emergence (Mean) **MD 62.77 higher** (SD 7.16 higher — 17.28 higher)	Low Due to serious imprecision [2]	The intervention with pyriproxyfen does not reach 100% adult emergence inhibition as does diflubenzuron and has clearly less residual effect. By using the double of indicated dosage, results ameliorate and reach >95% AEI for three weeks.
Adult emergence inhibition 12 days - 9 months	Based on data from participants in 13 studies. [3] (Randomized controlled) Follow up: 12 days - 9	The most widely used outcome measure was the per cent adult emergence inhibition (AEI) and outcomes were analyzed per category of treatments. Here		Low Due to serious risk of bias, Due to serious	IGR (pyriproxyfen) may improve adult emergence inhibition slightly. Further studies are needed to prove and improve the

Outcome Timeframe	**Study results and measurements**	**Comparator** no control	**Intervention** insect growth regulator (pyriproxyfen)	**Certainty of the evidence** (Quality of evidence)	**Summary**
3 Not Important	months.	only the exclusive pyriproxyfen interventions are mentioned: Container treatment: Six of the studies showed a significant (82-100% AEI) and long lasting (up to 8 months) effect. The two RCTs reported less positive results with significant effects of 4 weeks only. No good results were reported regarding area-wide ULV application for larviciding and auto-dissemination only. The best persistence in container treatment was achieved using slow-release formulations reporting a mortality rate of >80% - 100% for 5 up to 8 months. Auto-dissemination: 5 Intervention - control studies demonstrated that this approach is efficacious and can be applied easily and at low costs. For *Ae. aegypti* an AEI of 49-84% as opposed to 7-8% in controls has been demonstrated and for *Ae. albopictus* an AEI of 20.8% vs 2.4 in controls. The persistence obtained was between 12 days and 6 weeks.		inconsistency, Due to serious indirectness [4]	auto-dissemination strategy under field conditions. Specifically, methods of application, concentrations and frequencies of treatment need to be clarified.

1. [6]. We used the all data for pyriproxyfen of the three experiments for the first three weeks (theoretical residual effect). **Comparator:** [6]. **Supporting references:** [6], We choosed AIE for the first three weeks (theoretical residual effect) and used an average of 2g/catch basin pyriproxyfen interventions. Data extracted from table 3..
2. **Imprecision: serious.** Only data from one study, dosage doubled (in relation to label indications) when low efficacy was observed. **Publication bias: no serious.** The search in the Systematic review was not comprehensive. **Upgrade: clear dose-response gradient.** when dosage was doubled clearly better effects.
3. Systematic review [20]. Results abalyzed per categories of treatments: Container treatment, fumigation, auto-dissemination and combination of pyriproxyfen with adulticides. **Supporting references:** [20],
4. **Inconsistency: serious.** Point estimates vary widely, The magnitude of statistical heterogeneity was high. Follow up times varied significantly between studies.. **Indirectness: serious.** Direct comparisons not available, applicability, generalizability, transferability, translatability, external validity not given.. **Imprecision: no serious.** Few observed events in included studies for auto-dissemination..

References

6. Bellini R. Efficacy and lasting activity of four IGRs formulations against mosquitoes in catch basins of northern Italy. Journal of the European Mosquito Control Association 2009. Website

20. Maoz D, Ward T, Samuel M, Müller P, Runge-Ranzinger S, Toledo J, et al. Community effectiveness of pyriproxyfen as a dengue vector control method: A systematic review. PLoS neglected tropical diseases 2017;11(7):e0005651 Pubmed Journal

Clinical question/ PICO

Population: human populations in built-up European areas
Intervention: chemical control by insect growth regulators (diflubenzuron)
Comparator: no such control action

Outcome Timeframe	**Study results and measurements**	**Comparator** no control	**Intervention** IGRs (diflubenzuron)	**Certainty of the evidence** (Quality of evidence)	**Summary**
Reduction of adult emergence [1] 2 months: 2006, 2007 1 Not Important	Measured by: Mosquito breeder High better Based on data from 211 participants in 3 studies. [2] (Randomized controlled) Follow up: Weekly for 3 weeks.	20.75 Per cent inhibited adult emergence (Mean) Difference:	99.27 Per cent inhibited adult emergence (Mean) **MD 78.6 higher** (61 higher — 96 higher)	Moderate [3]	Three formulations of Diflubenzuron showed significant efficacy against larval development for at least up to three weeks.

1. During the summers of 2006 and 2007: August 30 to September 28, 2006 and 17 May 2007 to 25 September 2007
2. [6]. **Comparator:** [6]. **Supporting references:** [6], We choosed AIE for the first three weeks (declared residual effect) and used an average of all diflubenzuron formulations in the experiments.
3. **Publication bias: no serious.** The search in the review was not comprehensive..

References

6. Bellini R. Efficacy and lasting activity of four IGRs formulations against mosquitoes in catch basins of northern Italy. Journal of the European Mosquito Control Association 2009. Website

Clinical question/ PICO

Population: human populations in built-up European areas
Intervention: Autodissemination of insect growth regulators
Comparator: no such control

Outcome Timeframe	**Study results and measurements**	**Comparator** no control	**Intervention** Autodissemination of insect growth regulators	**Certainty of the evidence** (Quality of evidence)	**Summary**
Reduction of Ae. albopictus larvae and pupae 7-20 days in 2010 and 2011 1 Not Important	Measured by: Ovitraps (with pyrex beaker) with L3 larvae High better Based on data from 50 participants in 2 studies. [1] (Observational (non-randomized)) Follow up: 7-20 days.	1.6 % of larvae/pupae mortality (Mean) Difference:	48.6 % of larvae/pupae mortality (Mean) **MD 47 higher** (CI 95% 18.4 higher — 70 higher)	Very low Results seem promising but the study has been done in one city only, with 2 repetitions and at 2 sites only. Further research could have an important impact, which may change the estimates of the effect. [2]	Study shows that autodissemination of igrs (such as PPE at concentration 5%) have an effect in the the mortality of Ae. albopictus pupae. This has been demonstrated at two sites in Rome (Italy) at field conditions. However, further research in more locations and more repetitions is needed for supporting this evidence in other field condicions. Environmental factors (wind, shade, ..) can affect the results in other habitats

1. [10]. **Comparator:** [10]. **Supporting references:** [10],
2. **Risk of Bias: serious.** due to [reason] only 2 sites investigated in a city. Results cannot be generalized for other areas.. **Inconsistency: no serious. Indirectness: no serious.** not applicable. Only comparison is one product and control. **Imprecision: serious.** due to [reason]not wiede comparison in other habitats/locations, Only data from one study, Low number of patients. **Publication bias: no serious. Upgrade: large magnitude of effect.** results evidence an significant effect on pupae mortality using PPE through autodisemination. .

References

10. Caputo B, Ienco A, Cianci D, Pombi M, Petrarca V, Baseggio A, et al. The "auto-dissemination" approach: a novel concept to fight Aedes albopictus in urban areas. PLoS neglected tropical diseases 2012;6(8):e1793 Pubmed Journal

7.2. Adult control

Clinical question/ PICO

Population: human populations in built-up European areas
Intervention: mass trapping
Comparator: no such control action

Outcome Timeframe	**Study results and measurements**	**Comparator** no control	**Intervention** mass trapping	**Certainty of the evidence** (Quality of evidence)	**Summary**
Reduction of	Measured by: Human landing rate	11	1	Low	Mass trapping may decrease the Aedes

Outcome Timeframe	**Study results and measurements**	**Comparator** no control	**Intervention** mass trapping	**Certainty of the evidence** (Quality of evidence)	**Summary**
Aedes albopictus human landing rate [1] 16 weeks, June-October 2018 1 Not Important	Lower better Based on data from 123 participants in 1 studies. [2] (Observational (non-randomized)) Follow up: weekly for 4 months.	number of female Ae. albopictus / 1.5 hour (Median) Difference:	number of female Ae. albopictus / 1.5 hour (Median) **MD 10 fewer** (0 fewer — 97 fewer)	In the concrete setting, with the density of traps positioned, we are quite confident for the estimates [3]	albopictus human landing rate
Human landing rates/biting rates of Aedes albopictus adults [4] 13 weeks 4 Important	Measured by: Human landing catch Lower better [5] (Randomized controlled)	4 Human landing rate (30 minutes) (Median) Difference:	1 Human landing rate (30 minutes) (Median) **MD 2.25 lower** (-4 lower — 7 lower)	Moderate Due to serious indirectness related to very small area of intervention [6]	Mass trapping probably decreases human landing rates/biting rates of Aedes albopictus adults
Reduction of Aedes albopictus egg numbers [7] 16 weeks, June-October 2018 1 Not Important	Measured by: Number of eggs in ovitraps Scale: 0 — 522 Lower better Based on data from 173 participants in 1 studies. [8] (Observational (non-randomized)) Follow up: weekly for 4 months.	28 no.eggs/ovitrap/ week (Median) Difference:	12.5 no.eggs/ovitrap/ week (Median) **MD 15.5 fewer** (0 fewer — 522 fewer)	Very low Due to serious indirectness related to very small scale intervention, not easily transferable, generalizable. [9]	Mass trapping may decrease Aedes albopictus egg numbers

1. Assessment of the impact of control of Aedes albopictus by mass trapping (BG-Sentinel trap + BG-Lure) on adult mosquito population by counting mosquito adults in human landing catches
2, 8. [17]. **Comparator:** [17]. **Supporting references:** [17],
3. **Inconsistency: no serious. Indirectness: no serious. Imprecision: no serious.** Low number of patients. **Publication bias: no serious.** The study may be commercially funded .
4. Evaluation of the effectiveness of a field trap barrier system, i.e. a “removal trapping” outdoor control strategy for Ae. albopictus in southern France.
5. [2]. **Comparator:** [2]. **Supporting references:** [2], Data extracted from supplementary file 3.
6. **Inconsistency: no serious. Indirectness: serious.** due to very small scale of intervention. **Imprecision: no serious.** Low number of patients. **Publication bias: no serious.**
7. Assessment of the impact of control of Aedes albopictus by mass trapping (BG-Sentinel trap + BG-Lure) on adult mosquito population by counting mosquito eggs in ovitraps
9. **Inconsistency: no serious. Indirectness: serious.** due to very restricted intervention area with very dense trap network, restricted feasibility. **Imprecision: no serious. Publication bias: no serious.** The study may be commercially funded .

Practical issues	**no such control action**	**mass trapping**	**Both**
Procedure and device Feasibility of methodology in wide-area setting		Need for electricity for traps allows only for very small scale interventions	

Costs and access Costs and accessibility to premises		has high cost (traps) and depends on access to private premises and provision of electricity	

References

2. Akhoundi M, Jourdain F, Chandre F, Delaunay P, Roiz D. Effectiveness of a field trap barrier system for controlling Aedes albopictus: a "removal trapping" strategy. Parasites & vectors 2018;11(1):101 Pubmed Journal

17. Englbrecht C, Gordon S, Venturelli C, Rose A, Geier M. Evaluation of BG-Sentinel Trap as a management tool to reduce Aedes albopictus nuisance in an urban environment in Italy. Journal of the American Mosquito Control Association 2015;31(1):16-25 Journal Website

Clinical question/ PICO

Population: human populations in built-up European areas
Intervention: chemical control by adulticiding (pyrethroids by outdoor residual spraying)
Comparator: no such control action

Outcome Timeframe	**Study results and measurements**	**Comparator** no such control action	**Intervention** ORS (pyrethroids)	**Certainty of the evidence** (Quality of evidence)	**Summary**
Decrease Aedes albopictus adult numbers 20-40 days 1 Not Important	Measured by: Sticky trap Lower better Based on data from 6 participants in 2 studies. [1] (Observational (non-randomized)) Follow up: 20-40 days.	**6.77** adult mosquitoes /day (Mean) Difference:	**1.19** adult mosquitoes /day (Mean) **MD 5.58 fewer** (SE 0.11 fewer — 0.45 fewer)	Low Due to severe risk of bias, Due to very severe insufficient precision, Due to severe indirectness Counting and comparing adults in a trap is not a direct indication of a possible reduction in nuisance to the population, especially as the absolute values and the differences are very small. It is not entirely clear how the two values 6.77 and 1.19 were determined [2]	Ors may improve decrease aedes albopictus adult numbers
Decrease Aedes albopictus adult numbers [3] 20 days 1 Not Important	Measured by: BG Sentinel trap Lower better Based on data from 3 participants in 1 studies. [4] (Randomized controlled) Follow up: 20 days.	**21.1** adult mosquitoes /day (Mean) Difference:	**3.4** adult mosquitoes /day (Mean) **MD 17.7 fewer** (SE 0.89 fewer —	Low Due to severe risk of bias, Due to very severe insufficient precision, Due to severe indirectness Counting and	Ors may improve decrease aedes albopictus adult numbers

Outcome Timeframe	**Study results and measurements**	**Comparator** no such control action	**Intervention** ORS (pyrethroids)	**Certainty of the evidence** (Quality of evidence)	**Summary**
			0.9 fewer)	comparing adults in a trap is not a direct indication of a possible reduction in nuisance to the population. A comparison of the values 21.1 and 3.4 (outcome) is actually not entirely correct, as the value 21.1 refers to a different site, where completely different conditions may exist. [5]	
Reduction of Aedes albopictus population [6] 24 hours 1 Not Important	Measured by: Human Landing Catches High better Based on data from 18 participants in 1 studies. [7] (Randomized controlled) Follow up: 24 hours.	0 % population reduction (Mean) Difference:	95 % population reduction (Mean) **MD 95 lower** (SD 8 lower — 8 higher)	Moderate The manufacturer did not provide any data on the persistence of Etox. Therefore, it cannot be completely ruled out that a certain effect of Etox was still present during the subsequent treatment with Microsin. However, the similar human landing rates on untreated and previously treated sites tend to speak against this. Unfortunately, there is no meteorological data on the day after application (e.g. wind, rain, temperature), which could have an influence on the attack rates. The results of the intervention were documented using HLR, which made it possible to record the effects of the measures directly in terms of public nuisance. Recommendation: In an emergency (outbreak of disease, high	Ors (pyrethroids) probably improves reduction of aedes albopictus population

Outcome Timeframe	**Study results and measurements**	**Comparator** no such control action	**Intervention** ORS (pyrethroids)	**Certainty of the evidence** (Quality of evidence)	**Summary**
				infestation in the mosquito population), the use of adulticides could be useful to quickly and massively reduce the population density and thus reduce the likelihood of disease transmission. However, the effect is only short-term. They should therefore be used carefully in combination with more sustainable measures (e.g. reduction of larvae using Bti), taking into account the weather conditions. 8	

1. [19]. multiple field experiments, use of singlie treatment and maximum follow up. **Comparator:** [19]. **Supporting references:** [19], long-term single treatment and short-term single and double treatment used.
2. **Risk of Bias: serious.** No inclusion of meteorological data (e.g.heavy rainfall, sunshine, temperature...), which is essential for the effectiveness of the products used and the catch rates.. **Inconsistency: no serious.** No explanation for the decrease in captured females at site G, while at site F the number of captured females increased.. **Indirectness: serious.** Monitoring for only 5 days before the intervention seems a little short to capture the population density. The weather could have a significant influence on the result., . **Imprecision: very serious.** Sample size far too small (1 site per method investigated). The inclusion of several traps on only one site in the evaluation could be a pseudo-replication, which is highly questionable. In experiment 2, the trapping results of a different number of STs are compared with each other. STs catch numbers are generally very low, a meaningful evaluation based on these numbers is very questionable (differences could also be due to different positioning of the traps). Coincidence could play a major role. . **Publication bias: no serious.** The article was published in a journal of the "Society of Chemical Industry". There may be a conflict of interest here.
3. measured with BG Sentinel traps
4. [19]. **Comparator:** [19]. **Supporting references:** [19], only short term first and second treatment analyzed.
5. **Risk of Bias: serious.** No inclusion of meteorological data (e.g.heavy rainfall, sunshine, temperature...), which is essential for the effectiveness of the products used and the catch rates.. **Inconsistency: no serious. Indirectness: no serious. Imprecision: very serious.** Sample size far too small (1 site per method investigated). 1 BG-S per site. BG-S trap competes with 4 STs traps at location D and only with 3 STs traps at location C. BG-S catch numbers are partly low, a meaningful evaluation based on these numbers is questionable (differences could also be due to different positioning of the traps). Coincidence could play a major role. No pre-monitoring was carried out at the treated sites. A comparison of the values 21.1 and 3.4 (outcome) is actually not entirely correct, as the value 21.1 refers to a different site, where completely different conditions may exist.. **Publication bias: no serious.** The article was published in a journal of the "Society of Chemical Industry". There may be a conflict of interest here..
6. Only 24 hrs efficacy was used as max. efficacy measure. Residual effect not regarded.
7. [21]. **Comparator:** [21]. **Supporting references:** [21], Only mean population reduction is provided. Data extraction from graph 1..
8. **Inconsistency: no serious. Indirectness: no serious. Imprecision: no serious.** The manufacturer did not provide any data on the persistence of Etox. Therefore, it cannot be completely ruled out that a certain effect of Etox was still present during the subsequent treatment with Microsin. However, the similar human landing rates on untreated and previously treated sites tend to speak against this. Unfortunately, there is no meteorological data on the day after application (e.g. wind, rain, temperature), which could have an influence on the attack rates.. **Publication bias: no serious.** Some of the authors have close ties to the chemical industry. In my opinion, however, this has no influence on the quality of the study. .

References

19. Manica M, Cobre P, Rosà R, Caputo B. Not in my backyard: effectiveness of outdoor residual spraying from hand-held sprayers against the mosquito Aedes albopictus in Rome, Italy. Pest management science 2017;73(1):138-145 Pubmed Journal

21. Marini L, Baseggio A, Drago A, Martini S, Manella P, Romi R, et al. Efficacy of Two Common Methods of Application of Residual Insecticide for Controlling the Asian Tiger Mosquito, Aedes albopictus (Skuse), in Urban Areas. PloS one 2015;10(8):e0134831 Pubmed Journal

Clinical question/ PICO

Population: human populations in built-up European areas
Intervention: chemical control by adulticides (ultra-low volume space treatments, cold-fogging)
Comparator: no such control action

Outcome Timeframe	**Study results and measurements**	**Comparator** no control	**Intervention** adulticiding	**Certainty of the evidence** (Quality of evidence)	**Summary**
Decrease Aedes albopictus adult number [1] 1 hour 1 Not Important	Measured by: Cages Scale: 20.1 — 68.6 High better Based on data from 7 participants in 1 studies. [2] (Observational (non-randomized)) Follow up: 1 hour.	3.63 % mosquito adult reduction (Mean) Difference:	44.1 % mosquito adult reduction (Mean) **MD 40.47 higher** (CI 95% 20.1 lower — 68.6 lower)	Very low Due to serious inconsistency, Due to serious indirectness, Due to serious imprecision, Due to serious publication bias [3]	We are uncertain whether adulticiding increases or decreases Aedes albopictus adult numbers with this experimental setting (cages, night time of treatments)
Decrease Aedes albopictus adult number 4 months 1 Not Important	Measured by: Sticky traps Scale: 0 — 100 High better [4] (Observational (non-randomized)) Follow up: 72 hours.	-3.9 % mosquito adult reduction (Mean) Difference:	37.05 % mosquito adult reduction (Mean) **MD 40.95 higher** (CI 95% 0 lower — 100 lower)	Very low Due to serious inconsistency, Due to serious indirectness, Due to serious imprecision [5]	We are uncertain whether adulticiding increases or decreases the Aedes albopictus adult numbers. Sticky traps are not a reliable measure for the effectiveness of ULV applications on wild Ae. albopictus populations.
Decrease Aedes albopictus adult number [6] 24h 1 Not Important	Measured by: Cages High better [7] (Observational (non-randomized)) Follow up: 24 hours.	15.83 % mosquito adult reduction (Mean) Difference:	68.3 % mosquito adult reduction (Mean) **MD 52.47 higher** (40.27 higher — 71.67 higher)	Very low Due to serious inconsistency, Due to serious imprecision [8]	There is a very low evidence that ULV cold fogging might reduce significantly the adult population of Ae. albopictus. However due to the multitude of factors tested in the study a clear evaluation of the ULV cold fogging applications cannot be concluded.
Aedes albopictus adult female	Measured by: BG Sentinel adult trap	-3.02 % reduction of	10.9 % reduction of	Very low Evidence against	We are uncertain whether adulticiding with cold

Outcome Timeframe	**Study results and measurements**	**Comparator** no control	**Intervention** adulticiding	**Certainty of the evidence** (Quality of evidence)	**Summary**
numbers [9] 5 days 1 Not Important	High better [10] (Observational (non-randomized)) Follow up: 5 days.	adult female Aedes albopictus (Mean) Difference:	adult female Aedes albopictus (Mean) **MD 14.1 higher** (0 higher — 42.4 higher)	the cold fogging methodology, however very low evidence due to serious inconsistency bias due to very few repititions with ambiguous results and in general low numbers of adults in the traps even before the intervention. [11]	fogging ULV interventions increases or decreases Aedes albopictus adult female numbers
Aedes albopictus egg number 5 days 1 Not Important	Measured by: Ovitraps Lower better [12] (Observational (non-randomized)) Follow up: 5 days.	-8.68 % reduction of Ae. albopictus eggs (Mean) Difference:	-21.12 % reduction of Ae. albopictus eggs (Mean) **MD -12.44 lower** (-97.6 lower — 9 lower)	Very low Evidence against the cold fogging methodology; very low evidence due to serious inconsistency bias due to very few repititions with ambiguous results [13]	We are uncertain whether adulticiding through cold fogging ULV interventions increases or decreases Aedes albopictus egg number

2. [12]. **Comparator:** [12]. **Supporting references:** [12], Mortality from cages placed at 10m, 30m, 50m was used as the average mortality for the total buffer zone of 50 m around each ULV applications. The total average was calculated from the 7 treatments..
3. **Inconsistency: serious.** Point estimates vary widely, The magnitude of statistical heterogeneity was high.. **Indirectness: serious.** For the assessment of the effectiveness of ULV treatments, the use of cages is not appropriate because Aedes albopictus is resting on the vegetation. The time of treatments was inappropriate (during inactive time). **Imprecision: serious.** Only data from one study. Need for more empirical evidence.. **Publication bias: serious.** The search in the Systematic review was not comprehensive/ did apply language restriction.
4. [12]. **Comparator:** [12]. **Supporting references:** [12],
5. **Inconsistency: serious.** The direction of the effect is not consistent (only 50% of treatments showed expected result).. **Indirectness: serious.** Differences between the outcomes of interest and those referred to in the title. . **Imprecision: serious. Publication bias: no serious.** The search in the Systematic review was not comprehensive/ did apply language restriction.
6. Only 24 hrs evaluation taken into account
7. [8]. **Comparator:** [8]. **Supporting references:** [8], Only adult reduction in 24hrs for all distances (up to 25m) used.
8. **Inconsistency: serious.** Point estimates vary widely, The magnitude of statistical heterogeneity was high between study site D and the others. Explanation lacking.. **Imprecision: serious.** due to lack of repititions of the treatment and multifactor experimental setup (6 treatments with two different active ingredients, different meteorological settings (up-wind, down-wind, no wind), 1 indoor vs 5 outdoor study sites out of which one yields completely different results). **Publication bias: no serious.** The search in the review was not comprehensive.. **Upgrade: large magnitude of effect.** The additional test for residual effect on Ae. albopictus on vegetation in various distances from treatment pathway is very important as it proofs generally a complementary impact of the application.
9. Susceptibility to the insecticide was high but there was no discernable change in the oviposition rate or the catch of adult female mosquitoes, nor was there any change in the parous rate. In contrast, hand-held thermal foggers were highly effective, with more than 90% reduction of both laid eggs and females.
10. [9]. **Comparator:** [9]. Percentage of adult reduction before/after treatment extracted from Fig.5 for intervention and control.. **Supporting references:** [9], Percentage of adult reduction before/after treatment extracted from Fig.5 for intervention and control..
11. **Risk of Bias: no serious.** due to small sample size, very low numbers of Ae. albopictus adults in traps. **Inconsistency: serious.** The direction of the effect is not consistent between the included studies, Point estimates vary widely. **Imprecision: no serious.** Results due to chance, only few trials/measurements. **Publication bias: no serious.** The search in the review was not extensive.

12. [9]. **Comparator:** [9]. % of eggs reduction extracted form Fig. 5.. **Supporting references:** [9],
13. **Risk of Bias: no serious.** due to small sample size. **Inconsistency: serious.** The direction of the effect is not consistent between the included trials, Point estimates vary widely. **Imprecision: no serious.** Results due to chance, only few trials/measurements. **Publication bias: no serious.** The search in the review was not extensive.

References

8. Bengoa M, Eritja R, Lucientes J. Ground ultra-low volume adulticiding field trials using pyrethroids against Aedes albopictus in the Baix Llobregat region, Spain. Journal of the American Mosquito Control Association 2014;30(1):42-50 Pubmed

9. Boubidi SC, Roiz D, Rossignol M, Chandre F, Benoit R, Raselli M, et al. Efficacy of ULV and thermal aerosols of deltamethrin for control of Aedes albopictus in nice, France. Parasites & vectors 2016;9(1):597 Pubmed

12. Caputo B, Manica M, D'Alessandro A, Bottà G, Filipponi F, Protano C, et al. Assessment of the Effectiveness of a Seasonal-Long Insecticide-Based Control Strategy against Aedes albopictus Nuisance in an Urban Area. PLoS neglected tropical diseases 2016;10(3):e0004463 Pubmed Journal

Clinical question/ PICO

Population: human populations in built-up European areas
Intervention: chemical control by adulticides (ultra-low volume space treatments, thermal fogging)
Comparator: no such control action

Outcome Timeframe	**Study results and measurements**	**Comparator** no control	**Intervention** ULV adulticiding (thermal fogging)	**Certainty of the evidence** (Quality of evidence)	**Summary**
Ae. albopictus adult reduction	Measured by: BG Sentinel trap High better [1] (Observational (non-randomized)) Follow up: 5 days.	-18.5 % reduction of adult female Aedes albopictus (Mean) Difference:	71.6 % reduction of adult female Aedes albopictus (Mean) **MD 88.3 higher** (CI 95% 84.4 higher — 92.2 higher)	Very low Due to serious risk of bias, due to serious imprecision related to small intervention area, extremely small sample and one repitition only, due to serious publication bias [2]	There is very low evidence that ULV adulticiding (thermal fogging) decreases Ae. albopictus adult populations and we are uncertain if this is valid for all real-world circumstances and applicable in wide-area.
Ae. albopictus egg reduction	Measured by: ovitraps High better [3] (Observational (non-randomized)) Follow up: 5 days.	-83.6 % reduction of Ae. albopictus eggs (Mean) Difference:	64.15 % reduction of Ae. albopictus eggs (Mean) **MD 147.75 higher** (CI 95% 69.6 lower — 225.9 lower)	Very low Due to serious risk of bias, Due to serious imprecision, Due to serious publication bias [4]	There is very low evidence that ULV adulticiding (thermal fogging) decreases Ae. albopictus egg populations and we are uncertain if this is valid for all real-world circumstances and applicable in wide-areas.

1. [9]. **Comparator:** [9]. **Supporting references:** [9], Percentage of reduction extracted from Fig. 5..
2. **Risk of Bias: serious.** due to very small intervention area . **Imprecision: serious.** due to very few observed results by only one repitition. . **Publication bias: serious.** The search in the review was not extensive.
3. [9]. **Comparator:** [9]. Percentage of eggs reduction extracted from Fig. 5.. **Supporting references:** [9], Percentage of egg reduction

extracted from Fig. 5..
4. **Risk of Bias: serious.** due to very small treatment area . **Imprecision: serious.** due to only one repitition and two samples. **Publication bias: serious.** The search in the review was not extensive.

References

9. Boubidi SC, Roiz D, Rossignol M, Chandre F, Benoit R, Raselli M, et al. Efficacy of ULV and thermal aerosols of deltamethrin for control of Aedes albopictus in nice, France. Parasites & vectors 2016;9(1):597 Pubmed

Clinical question/ PICO

Population: human populations in built-up European areas
Intervention: Sterile insect technique (SIT) (irradiation-induced)
Comparator: No intervention

Outcome Timeframe	**Study results and measurements**	**Comparator** no intervention	**Intervention** SIT	**Certainty of the evidence** (Quality of evidence)	**Summary**
Number of Ae. albopictus eggs [1] 4 months 1 Not Important	Measured by: Ovitraps Lower better [2] (Observational (non-randomized)) Follow up: 4 months.	77.9 number of eggs / trap / week (Mean) Difference:	56.47 number of eggs / trap / week (Mean) **MD 21.43 lower** (CI 95% 0 lower — 62.44 lower)	Very low Due to serious imprecision: only 1/ 3 intervention areas and only 2/5 trials with expected results [3]	We are uncertain whether SIT increases or decreases the number of Ae. albopictus eggs

2. [7]. **Comparator:** [7]. **Supporting references:** [7],
3. **Imprecision: serious.** due to only 1/3 intervention areas and only 2/5 trials with expected results .

References

7. Bellini R, Medici A, Puggioli A, Balestrino F, Carrieri M. Pilot field trials with Aedes albopictus irradiated sterile males in Italian urban areas. Journal of medical entomology 2013;50(2):317-25 Pubmed

Clinical question/ PICO

Population: human populations in built-up European areas
Intervention: Sterile insect technique (SIT) (irradiation-induced) and source reduction
Comparator: Source Reduction

Outcome Timeframe	Study results and measurements	Comparator SR	Intervention SIT + SR	Certainty of the evidence (Quality of evidence)	Summary
Reduction in egg hatching [1] 10 weeks 4 Important	Measured by: mean reduction in egg hatching of eggs collected by ovitraps High better [2] (Observational (non-randomized))	12.4 Egg hatched (Mean) Difference:	67.6 Egg hatched (Mean) **MD 55.2 higher** (CI 95% 17 higher — 92 higher)	Very low SIT applied for 10 weeks was capable to strongly reduce the mean egg hatching in the treated area compared to two control areas. To increase the certainty level, the study should be repeated for several years and in different locations. Addiontally, the ratio of the sterile males compared to the wild males should be provided to allow to better estimate the effectiveness and sustainability of the control method., Due to serious indirectness, Due to serious imprecision. [3]	SIT applied for 10 weeks significantly reduced the mean Ae. albopictus egg hatching in a treated area compared to two control areas.
Mean number of Ae. albopictus eggs [4] 10 weeks 1 Not Important	Measured by: ovitraps Lower better [5] (Observational (non-randomized))	12.5 Weekly average number of eggs per ovitrap (Mean) Difference:	12.91 Weekly average number of eggs per ovitrap (Mean) **MD 0.41 higher** (CI 95% 29 lower — 18.5 higher)	Very low SIT was not capable to reduce the mean number of eggs per trap in the treated area compared to two control areas. It is possible that a larger time frame could have higlhighted a better result. Additionally, the migration of wild individuals from the surrounding areas could have confounded the results., Due to serious indirectness, Due to serious imprecision [6]	SIT applied for 10 weeks have had no effect on the mean number of Ae. albopictus eggs collected in the treated area compared to two control areas

2. [3]. **Comparator:** Primary study [3]. **Supporting references:** [3],
3. **Indirectness: serious.** Diapause could have a confounding effect on the hatching of the eggs which is not evaluated or discussed on ths study. **Imprecision: serious.** Only data from one study. The study did not provide the ratio between released sterile males and wild males that is necessary to estimate effectiveness and sustainability of the method. **Upgrade: all plausible confounding would**

have reduced the effect.
5. [3]. **Comparator:** [3]. **Supporting references:** [3],
6. **Indirectness: serious.** the effect of SIT in terms of reduction of eggs or adult mosquitoes can require several months to be appreciated and migration of individuals from surrounding areas can confound the result..

References

3. Balatsos G, Puggioli A, Karras V, Lytra I, Mastronikolos G, Carrieri M, et al. Reduction in Egg Fertility of Aedes albopictus Mosquitoes in Greece Following Releases of Imported Sterile Males. Insects 2021;12(2) Pubmed Journal

Clinical question/ PICO

Population: human populations in built-up European areas
Intervention: Wolbachia Incompatible Insect Technique (IIT)
Comparator: No intervention

Outcome Timeframe	**Study results and measurements**	**Comparator** No intervention	**Intervention** Wolbachia IIT	**Certainty of the evidence** (Quality of evidence)	**Summary**
Egg viability [1] 8 weeks 1 Not Important	Measured by: Ovitrap Scale: 80 — 99 Lower better Based on data from 44,542 participants in 2 studies. [2] (Observational (non-randomized)) Follow up: 8 weeks.	97.95 % viability (Mean) Difference:	78.35 % viability (Mean) **MD 19.6 fewer**	Very low Due to serious indirectness, Due to serious imprecision [3]	While Wolbachia IIT decreases the percentage of viability in the eggs collected from the whole population by ovitraps, the percentage of decrease in egg viability is too small to determine whether Wolbachia IIT can make a difference in the Ae. albopictus population size
Fertility rates of Aedes albopictus females [4] 8 weeks 1 Not Important	Measured by: HLC, oviposition Scale: 6.2 — 85.5 Lower better Based on data from 17,298 participants in 2 studies. [5] (Observational (non-randomized))	86.63 % of viable eggs laid (Mean) Difference:	7.84 % of viable eggs laid (Mean) **MD 78.79 fewer** (CI 95% 78.97 fewer — 79.06 fewer)	Very low Due to serious risk of bias, Due to serious indirectness, Due to serious inconsistency [6]	Wolbachia IIT may decrease fertility rates of Aedes albopictus females

1. Viability rates of Aedes albopictus eggs collected by ovitraps
2. [13], [14]. As participants the collected and checked for viability eggs are regarded. **Comparator:** [13], [14]. **Supporting references:** [14], Repitition of experiment from 2018 in 2019. [13],
3. **Inconsistency: no serious. Indirectness: serious.** egg viability reduction not enough to influence significantly mosquito population. **Imprecision: serious.** Wide confidence intervals, only small reduction of egg viability.
4. Fertility rate of single ovipoiting females
5. with included studies: [14], [13]. Ae. albopictus eggs from collected females are considered as participants. **Comparator:** [13], [14]. **Supporting references:** [14], [13],
6. **Risk of Bias: serious.** Incomplete data because data related to unmated females were discarded and only an average of 45.7% probability of mating with ARwP males was estimated, Selective outcome reporting. **Inconsistency: serious.** due to comparison of

viability of eggs from mated females of treated areas vs viability of eggs from unmated females of treated and control areas. **Indirectness: serious.** due to collection of wild Ae. albopictus females in proximity of release spots of infected males. The outcome time frame in studies were insufficient..

References

13. Caputo B, Moretti R, Manica M, Serini P, Lampazzi E, Bonanni M, et al. A bacterium against the tiger: preliminary evidence of fertility reduction after release of Aedes albopictus males with manipulated Wolbachia infection in an Italian urban area. Pest management science 2020;76(4):1324-1332 Pubmed Journal

14. Caputo B, Moretti R, Virgillito C, Manica M, Lampazzi E, Lombardi G, et al. A bacterium against the tiger: further evidence of the potential of noninundative releases of males with manipulated Wolbachia infection in reducing fertility of Aedes albopictus field populations in Italy. Pest management science 2023;79(9):3167-3176 Pubmed Journal

7.3. Integrated vector management (IVM)

Clinical question/ PICO

Population: human populations in built-up European areas
Intervention: Sterile Insect Technique (SIT) and larviciding (Bti)
Comparator: larviciding (Bti)

Outcome Timeframe	**Study results and measurements**	**Comparator** larviciding	**Intervention** larviciding and SIT	**Certainty of the evidence** (Quality of evidence)	**Summary**
Decrease Aedes albopictus egg numbers 10 month for 3 years 1 Not Important	Measured by: Ovitrap Lower better [1] (Observational (non-randomized))	3.6 Eggs per trap per day (Mean) Difference:	1.78 Eggs per trap per day (Mean) **MD 1.85 lower** (CI 99% 0.5 lower — 4.7 lower)	Very low Due to very serious risk of bias [2]	Larviciding + SIT may slighlty improve the decrease Aedes albopictus egg numbers.
Decrease Aedes albopictus adult numbers 8 months for 2 years 1 Not Important	Measured by: BG Sentinel trap Lower better [3] (Observational (non-randomized)) Follow up: April-November.	1.7 Adult Aedes albopictus per day per trap (Mean) Difference:	0.45 Adult Aedes albopictus per day per trap (Mean) **MD 1.55 lower** (CI 99% 0.1 lower — 2.9 lower)	Very low Due to very serious risk of bias [4]	Larviciding + sit may slightly improve the decrease of Aedes albopictus adult numbers
Egg hatching [5] 10 month 3 years 1 Not Important	Measured by: Incubator Lower better [6] (Observational (non-randomized))	59.77 percent eggs hatched (Mean)	43.6 percent eggs hatched (Mean)	Very low Due to serious indirectness [7]	Larviciding + SIT may decrease egg hatching slightly. Only a very slight difference between eggs hatched in the intervention

Outcome Timeframe	**Study results and measurements**	**Comparator** larviciding	**Intervention** larviciding and SIT	**Certainty of the evidence** (Quality of evidence)	**Summary**
		Difference:	**MD 16.17 lower** (CI 99% 4.7 lower — 30.3 lower)		and the control can be stated.

1, 3. [27]. **Comparator:** [27]. **Supporting references:** [27],
2. **Risk of Bias: very serious.** Use of unvalidated and/or subjective outcome measures because of the use of an extensive network of lethal ovitraps, and because of the arbitrary use of adulticiding treatments between both intervention and control areas.
4. **Risk of Bias: very serious.** Use of unvalidated and/or subjective outcome measures because of the use of an extensive network of lethal ovitraps, and because of the arbitrary use of adulticiding treatments between both intervention and control areas, Use of unvalidated and/or subjective outcome measures.
6. with included studies: [27]. **Comparator:** [27]. **Supporting references:** [27],
7. **Indirectness: serious.** Only a very slight difference between intervention and control can be stated.

References

1. Abramides GC, Roiz D, Guitart R, Quintana S, Guerrero I, Giménez N. Effectiveness of a multiple intervention strategy for the control of the tiger mosquito (Aedes albopictus) in Spain. Transactions of the Royal Society of Tropical Medicine and Hygiene 2011;105(5):281-8 Pubmed Journal

27. Tur C, Almenar D, Zacarés M, Benlloch-Navarro S, Pla I, Dalmau V. Suppression Trial through an Integrated Vector Management of Aedes albopictus (Skuse) Based on the Sterile Insect Technique in a Non-Isolated Area in Spain. Insects 2023;14(8) Pubmed Journal

Clinical question/ PICO

Population: human populations in built-up European areas
Intervention: chemical control by insect growth regulators (pyriproxyfen) and adulticiding by permethrin, B. bassiana or Spinosad
Comparator: no such control action

Outcome Timeframe	**Study results and measurements**	**Comparator** no control	**Intervention** pyriproxyfen + permethrin	**Certainty of the evidence** (Quality of evidence)	**Summary**
Adult emergence inhibition [1] 3 weeks to 5 months 4 Important	Based on data from participants in 9 studies. [2] (Randomized controlled) Follow up: 9 weeks to 5 months.	Fumigation: In two studies a combination of Pyriproxyfen and permethrin reported a significant AEI and BI for *Ae. aegypti*. The residual effect was < 9 weeks. Adding outdoor ULV application of permethrin increases the effectiveness of the intervention. Combination of pyriproxyfen and adulticides: Combined results are used instead of stratified effects by product. In two RCTs significant reduction fo mosquito populations were measured by means of Breteau Index (BI), Container Index (CI) and seroprevalence studies for the combination of pyriproxyfen and		Low Due to serious inconsistency, Due to serious indirectness [3]	Pyriproxyfen + permethrin may increase adult emergence inhibition slightly. The combination of adulticidal and larvicidal products can increase effectiveness by simultaneously controlling adults and larvae, and by expanding persistence.

Outcome Timeframe	Study results and measurements	Comparator no control	Intervention pyriproxyfen + permethrin	Certainty of the evidence (Quality of evidence)	Summary
		permethrin. Residual effect dpending on application methodology reached 9 weeks and 5 months respectively.			

1. Seven studies with combined intervention pyriproxyfen + adulticide
2. Systematic review [20]. RCTs results respected. Similar results in non-RCTs.. **Supporting references:** [20],
3. **Inconsistency: serious.** Not all trials used the same adulticide, nor the same application methodology or efficacy evaluation methodology (BI, CI, seroprevalence, RD etc.) Point estimates vary widely.. **Indirectness: serious.** Direct comparisons not available, no generizable, transferable results..

References

20. Maoz D, Ward T, Samuel M, Müller P, Runge-Ranzinger S, Toledo J, et al. Community effectiveness of pyriproxyfen as a dengue vector control method: A systematic review. PLoS neglected tropical diseases 2017;11(7):e0005651 Pubmed Journal

Clinical question/ PICO

Population: human populations in built-up European areas
Intervention: Sterile insect technique (SIT) (irradiation-induced) and source reduction
Comparator: Source Reduction

Outcome Timeframe	Study results and measurements	Comparator SR	Intervention SIT + SR	Certainty of the evidence (Quality of evidence)	Summary
Reduction in egg hatching [1] 10 weeks 4 Important	Measured by: mean reduction in egg hatching of eggs collected by ovitraps High better [2] (Observational (non-randomized))	12.4 Egg hatched (Mean) Difference:	67.6 Egg hatched (Mean) **MD 55.2 higher** (CI 95% 17 higher — 92 higher)	Very low SIT applied for 10 weeks was capable to strongly reduce the mean egg hatching in the treated area compared to two control areas. To increase the certainty level, the study should be repeated for several years and in different locations. Addiontally, the ratio of the sterile males compared to the wild males should be provided to allow to better estimate the effectiveness and sustainability of the	SIT applied for 10 weeks significantly reduced the mean Ae. albopictus egg hatching in a treated area compared to two control areas.

Outcome Timeframe	Study results and measurements	Comparator SR	Intervention SIT + SR	Certainty of the evidence (Quality of evidence)	Summary
				control method., Due to serious indirectness, Due to serious imprecision. [3]	
Mean number of Ae. albopictus eggs [4] 10 weeks 1 Not Important	Measured by: ovitraps Lower better [5] (Observational (non-randomized))	12.5 Weekly average number of eggs per ovitrap (Mean) Difference:	12.91 Weekly average number of eggs per ovitrap (Mean) **MD 0.41 higher** (CI 95% 29 lower — 18.5 higher)	Very low SIT was not capable to reduce the mean number of eggs per trap in the treated area compared to two control areas. It is possible that a larger time frame could have higlhighted a better result. Additionally, the migration of wild individuals from the surrounding areas could have confounded the results., Due to serious indirectness, Due to serious imprecision [6]	SIT applied for 10 weeks have had no effect on the mean number of Ae. albopictus eggs collected in the treated area compared to two control areas

2. [3]. **Comparator:** Primary study [3]. **Supporting references:** [3],
3. **Indirectness: serious.** Diapause could have a confounding effect on the hatching of the eggs which is not evaluated or discussed on ths study. **Imprecision: serious.** Only data from one study. The study did not provide the ratio between released sterile males and wild males that is necessary to estimate effectiveness and sustainability of the method. **Upgrade: all plausible confounding would have reduced the effect.**
5. [3]. **Comparator:** [3]. **Supporting references:** [3],
6. **Indirectness: serious.** the effect of SIT in terms of reduction of eggs or adult mosquitoes can require several months to be appreciated and migration of individuals from surrounding areas can confound the result..

References

3. Balatsos G, Puggioli A, Karras V, Lytra I, Mastronikolos G, Carrieri M, et al. Reduction in Egg Fertility of Aedes albopictus Mosquitoes in Greece Following Releases of Imported Sterile Males. Insects 2021;12(2) Pubmed Journal

Clinical question/ PICO

Population: human populations in built-up European areas
Intervention: Source reduction, larviciding and adulticiding in sensitive areas
Comparator: no control action

Outcome Timeframe	**Study results and measurements**	**Comparator** no control	**Intervention** SR + Larviciding + adulticiding (sens)	**Certainty of the evidence** (Quality of evidence)	**Summary**
Reduction Aedes albopictus egg numbers [1] July - November 2012 and 2013 1 Not Important	Measured by: ovitraps Scale: 0 — 1537 Lower better Based on data from 6,440 participants in 1 studies. [2] (Randomized controlled) Follow up: 5-7 months.	52.2 number of eggs per trap per 14 days (Mean) Difference:	21.95 number of eggs per trap per 14 days (Mean) **MD 30.25 fewer** (CI 95% 1.4 fewer — 3.65 fewer)	Moderate The proof of the connection between intervention or non-intervention and the number of eggs is somewhat indirect. As the authors themselves write, an approach in which the effects can be measured more directly in response to the measures implemented would be somewhat more meaningful. However, this is difficult to implement. A further inaccuracy is that the extent and type of breeding sites in the study area could not be recorded, nor was the quantity of insecticides used documented. The authors also drew attention to this. However, due to the extensive study, this should not have had a decisive influence on the basic results. Counting eggs is not direct evidence of a significant reduction in the nuisance to the population and is therefore not a clear indication of a lower probability of pathogen transmission. Furthermore, the stated reduction in egg counts is not necessarily sufficient to achieve a significant reduction in public nuisance. [3]	Sr + larviciding + adulticiding (sens) probably improves reduction aedes albopictus egg numbers

1. Assesment of the efficacy of source reduction+larviciding on populations of Aedes albopictus
2. [25]. **Comparator:** [25]. comparison intervention (source reduction + Bti larviciding) and non intervention areas. **Supporting references:** [25],
3. **Inconsistency: no serious. Indirectness: no serious.** The proof of the connection between intervention or non-intervention and the number of eggs is somewhat indirect. As the authors themselves write, an approach in which the effects can be measured more directly in response to the measures implemented would be somewhat more meaningful. However, this is difficult to implement.. **Imprecision: no serious.** A further inaccuracy is that the extent and type of breeding sites in the study area could not be recorded, nor was the quantity of insecticides used documented. The authors also drew attention to this. However, due to the extensive study, this should not have had a decisive influence on the basic results.. **Publication bias: no serious.**

References

25. Suter TT, Flacio E, Feijoó Fariña B, Engeler L, Tonolla M, Regis LN, et al. Surveillance and control of Aedes albopictus in the Swiss-Italian border region: Differences in egg densities between intervention and non-intervention areas. PLOS Neglected Tropical Diseases 2016;10(1):e0004315 Journal Website

Clinical question/ PICO

Population: Aedes albopictus mosquitoes
Intervention: Source reduction, biological larviciding (Bti), and SIT
Comparator: Source reduction, Biological larviciding (Bti)

Outcome Timeframe	**Study results and measurements**	**Comparator** SR, Bti	**Intervention** SIT + SR + Bti	**Certainty of the evidence** (Quality of evidence)	**Summary**
Number of Ae. albopictus eggs 10 weeks 1 Not Important	Measured by: Ovitraps Lower better [1] (Observational (non-randomized))	5.38 mean number of eggs per trap per week (Mean) Difference:	4 mean number of eggs per trap per week (Mean) **MD 1.38 lower**	Very low Due to serious risk of bias, Due to serious inconsistency, Due to serious indirectness, Due to serious imprecision, Due to serious publication bias [2]	Intervention is favourable but measured effect is very low in the study area. It could be improved repeating the study at other locations and during several seasons. SIT in combination with source reduction and comunity participation could aid to reduce populations of Ae. albopictus in isolated areas. Introductions of specimens from other areas into the treated areas has not been measured.
Reduction in Ae. albopictus egg fertility [3] 10 weeks 1 Not Important	Measured by: egg hatching High better [4] (Observational (non-randomized))	14.55 % egg sterility (Mean) Difference:	77.4 % egg sterility (Mean) **MD 62.85 higher** (49.3 higher — 89.19 higher)	Low The effect determined by the intervention has a large magnitude but open field studies should be repeated for several years and in several areas to reach a	The application of the Sterile Insect Technique following interventions to reduce the larval breeding sites and the application of Bti significantly increased the efficacy of IVM compared to areas where only the first two measures were applied. This positive

Outcome Timeframe	**Study results and measurements**	**Comparator** SR, Bti	**Intervention** SIT + SR + Bti	**Certainty of the evidence** (Quality of evidence)	**Summary**
				high level of certainty. Also, the number of eggs per trap is quite small. At last, the sterile/ wild male ratio is a parameter that is strictly necessary to estimate the effectiveness of the control measure and its sustainability. However, this study reflects the effectivennes for reducing populations of Aedes albopictus using community participation and source reduction and SIT. Due to serious risk of bias, Due to serious inconsistency, Due to serious indirectness, Due to serious imprecision, Due to serious risk of bias [5]	result is revealed by the significantly reduced mean egg fertility in the treated areas. A repetition of the same experiment in different locations and years could reinforce the value of the findings. An estimation of the ratio of the released sterile males compared to the wild males could help in measuring the specific effectiveness of the method and its sustainability on the long term.

1, 4. [5]. **Comparator:** [5]. **Supporting references:** [5],
2. **Risk of Bias: serious.** There is no comparision with other areas that could be also representative in other locations. Biased towards residential areas. Study SIT released different amount of males in the 2 locations measured. . **Inconsistency: serious.** Study SIT released different amount of males in the 2 locations measured. **Indirectness: serious.** Differences between the population of interest and those studied, The outcome time frame in studies were insufficient, Direct comparisons not available. **Imprecision: serious.** There is a significant difference in amout of traps deployed at the locations. **Publication bias: no serious.** Not other studies published using this approach..
5. **Risk of Bias: serious.** Few data and selective for CI measurements in 2019. Control location (no treatments) absent in dataset. **Inconsistency: serious.** Too observational. In open field with many factors that can alter the results. No replicas in different seasons.. **Indirectness: serious.** The outcome time frame in studies were insufficient. **Imprecision: serious.** Only data from one study. **Publication bias: no serious.** There are no other similar publications for comparison. **Upgrade: large magnitude of effect.** The large magnitide of the effect in the publication could rate up the certainty..

References

5. Becker N, Langentepe-Kong SM, Tokatlian Rodriguez A, Oo TT, Reichle D, Lühken R, et al. Integrated control of Aedes albopictus in Southwest Germany supported by the Sterile Insect Technique. Parasites & vectors 2022;15(1):9 Pubmed Journal

Clinical question/ PICO

Population: human populations in built-up European areas

Intervention: Source reduction, larvicide treatment, rubbish removal, and adulticiding in recreation parks
Comparator: No intervention

Summary

This study was undertaken to evaluate the effectiveness of four complementary and combined strategies to minimize the presence of the invasive mosquito Aedes albopictus, firmly established in Sant Cugat del Vallès, Catalonia, Spain. A quasi-experimental design including six neighbourhoods was performed in 2008–2009. The abundance of mosquitoes was monitored through ovitraps. The multiple intervention strategy consisted of four actions: source reduction; larvicide treatments (Bacillus thuringiensis israelensis and diflubenzuron); adulticide treatments (alfacipermetrin); and cleaning up uncontrolled landfills. The results showed the number of eggs significantly reduced in the areas with intervention. In 2008, the accumulate median of eggs was 175 and 272 in the intervention and control areas, respectively. In 2009, these medians were 884 and 1668 eggs.

Outcome Timeframe	**Study results and measurements**	**Comparator** No intervention	**Intervention** IVM	**Certainty of the evidence** (Quality of evidence)	**Summary**
Number of Ae. albopictus eggs [1] 3-7 months 1 Not Important	Measured by: Ovitraps Lower better [2] (Observational (non-randomized)) Follow up: 3-7 months.	83.05 Number of eggs/ trap/2 weeks (Mean) Difference:	48.77 Number of eggs/ trap/2 weeks (Mean) **MD 34.28 lower**	Very low Due to very serious risk of bias, due to serious indirectness. [3]	Difference in numbers of Ae. albopictus eggs in intervention and control areas might be attributed to additional non-documented interventions.

1. The multiple intervention strategy (IVM) consisted of four actions: source reduction; larvicide treatments (Bacillus thuringiensis israelensis and diflubenzuron); adulticide treatments (alfacipermetrin); and cleaning up uncontrolled landfills.
2. [1]. The results showed the number of eggs significantly reduced in the areas with intervention. In 2008, the accumulate median of eggs was 175 and 272 in the intervention and control areas, respectively. In 2009, these medians were 884 and 1668 eggs.. **Comparator:** [1]. Data for control (average of 2008 and 2009) extracted from Fig.2 A+B. **Supporting references:** [1], Data for intervention (average of 2008 and 2009) extracted from Fig. 2A+B.
3. **Risk of Bias: very serious.** Adulticiding interventions in the study areas, the effect of which were not taken into consideration.. **Indirectness: serious.** Non-conclusive setup of experimentation. Slight differences between intervention and control. Time frames, number of samples not equal in both experimental years. Results of standard area not displayed..

References

1. Abramides GC, Roiz D, Guitart R, Quintana S, Guerrero I, Giménez N. Effectiveness of a multiple intervention strategy for the control of the tiger mosquito (Aedes albopictus) in Spain. Transactions of the Royal Society of Tropical Medicine and Hygiene 2011;105(5):281-8 Pubmed Journal

Clinical question/ PICO

Population: human populations in built-up European areas
Intervention: Chemical control by insect growth regulators (diflubenzuron) and adulticides (pyrethroids)
Comparator: No control action

Outcome Timeframe	**Study results and measurements**	**Comparator** No control action	**Intervention** diflubenzuron + pyrethroids	**Certainty of the evidence** (Quality of evidence)	**Summary**
Reduction of adult emergence 15 weeks 1 Not Important	Measured by: Mosquito Emerging Trap (MET) Lower better Based on data from 10 participants in 1 studies. [1] (Observational (non-randomized)) Follow up: 8 weeks.	5 Adult/trap/48h (Mean) Difference:	0.75 Adult/trap/48h (Mean) **MD 4.25 lower** (CI 95% 0.3 lower — 0.3 lower)	Very low Due to serious imprecision, Due to very serious risk of bias [2]	There seems to be a reduction of emerging adults but this reduction is not clear to which of the two interventions it can be attributed. Repititions are very few.

1. [11]. **Comparator:** [11]. **Supporting references:** [11],
2. **Risk of Bias: very serious.** due to combination of two different intervention types: chemical larviciding and adulticiding. **Imprecision: serious.** due to very few intervention and control catch basins.

References

11. Caputo B, Ienco A, Manica M, Petrarca V, Rosà R, della Torre A. New adhesive traps to monitor urban mosquitoes with a case study to assess the efficacy of insecticide control strategies in temperate areas. Parasites & vectors 2015;8:134 Pubmed Journal

Clinical question/ PICO

Population: Human population in built-up areas in Europe

Intervention: Larviciding in public road drains + door-to-door strategy (DtoD): monthly inspections of private properties + larval treatment of permanent breeding sites + removal/inactivation of occasional breeding sites + predatory copepods in large permanent containers + direct information to residents + communication campaigns

Comparator: Larviciding in public road drains

Outcome Timeframe	**Study results and measurements**	**Comparator** Larviciding in public road drains	**Intervention** Larviciding in public road drains + DtoD strategy	**Certainty of the evidence** (Quality of evidence)	**Summary**
Number of Aedes albopictus eggs [1] 3 years 1 Not Important	Measured by: Ovitraps Lower better [2] (Observational (non-randomized))	454.4 Number of eggs/ ovitrap/14 days (Mean) Difference:	274.9 Number of eggs/ ovitrap/14 days (Mean) **MD 179.5 lower** CI 95%	Low 39% reduction compared to the control population. The control population is not as well described as the treated population. [3]	Combined DtoD strategy may decrease number of Aedes albopictus eggs compared to control areas where only breeding sites present in public areas were treated with larvicide.
Number of Aedes albopictus adults [4] 3 years 4 Important	Measured by: Human Landing Catch (HLC) Lower better [5] (Observational (non-randomized))	11.7 Number of females/ session (Mean) Difference:	3.4 Number of females/ session (Mean) **MD 8.3 lower**	Low Significant reduction in adult mosquito population as measured with HLC	Combined dtod strategy may decrease number of Aedes albopictus adults compared to control areas where only breeding sites present in public areas

Outcome Timeframe	**Study results and measurements**	**Comparator** Larviciding in public road drains	**Intervention** Larviciding in public road drains + DtoD strategy	**Certainty of the evidence** (Quality of evidence)	**Summary**
			(SD 4.5 lower — 12.8 lower)	(71% reduction compared to mean control values). The control population is not as well described as the treated population. [6]	were treated with larvicides.

1, 4. The applied DtoD strategy consists of the following actions: • six larval control treatments in public road drains in the period April-September, using manual pump spraying Diflubenzuron (Arysta LifeScience France) or Vectomax®FG (and , Sumitomo Chemical Italia) or Aquatain AMF ™ (polydimethylsiloxane, Bleu Line Italia); • six DtoD interventions in the period April-September, including source removal, larval treatment of permanent breeding sites using the same products as in public areas and direct information to the residents; the introduction of predatory copepods in the large permanent containers[10]; • the activation of a green phone line to support citizen contact; • organization of a database to store the contact phone number of each resident, the most appropriate timing for the visits and the number and exact locations of permanent breeding sites in each property; • communication campaigns including on-site meetings, web news, notes on the major local aggregation points, a personal letter sent by the Municipality to each resident, the notice of the next date of the visit by the Alert System (this is a system some Municipalities have put in place to directly communicate with citizens through SMS if need be).
2, 5. [15]. **Comparator:** [15]. **Supporting references:** [15],
3. **Indirectness: serious.** Differences may exist between the intervention/comparator of interest and those studied. The control population was not well described. No information on the number of breeding sites, residences, surface area etc..
6. **Inconsistency: no serious.** Point estimates vary widely. The SD values are relatively high when compared to the Mean values in the control and treated population. . **Indirectness: no serious.** The control population was not well described. No information on the number of breeding sites, residences, surface area, ...

References

15. Donati L, Carrieri M, Bellini R. A Door-to-Door Strategy for Aedes albopictus Control in Northern Italy: Efficacy, Cost-Analysis and Public Perception. Vector Biology Journal 2020. Website

Clinical question/ PICO

Population: human populations in built-up European areas
Intervention: Source reduction, biological larviciding (Bti) and public education campaign
Comparator: No interventions

Outcome Timeframe	**Study results and measurements**	**Comparator** No interventions	**Intervention** SR + Bti + education	**Certainty of the evidence** (Quality of evidence)	**Summary**
Number of Aedes albopictus eggs [1] 7 months 1 Not Important	Measured by: Ovitraps Lower better Based on data from 70 participants in 1 studies. [2] (Observational (non-randomized))	95.04 Eggs/trap/14 days (Mean) Difference:	43.27 Eggs/trap/14 days (Mean) **MD 51.77 fewer**	Very low Due to serious imprecision, Due to serious indirectness [3]	Source reduction plus biological larviciding in public and private areas in combination with public education campaigns may decrease the number of

Outcome Timeframe	Study results and measurements	Comparator No interventions	Intervention SR + Bti + education	Certainty of the evidence (Quality of evidence)	Summary
	Follow up: Fortnightly.		(SE 2.55 fewer — 5.52 fewer)		Aedes albopictus eggs slightly.

1. 7 months: April to October 2015, only full intervention is evaluated here
2. [4]. **Comparator:** [4]. **Supporting references:** [4],
3. **Indirectness: serious.** Direct comparisons not available due to combination of three interventions. **Imprecision: serious.** Only one intervention and one control area. Few participants.

References

4. Baldacchino F, Bussola F, Arnoldi D, Marcantonio M, Montarsi F, Capelli G, et al. An integrated pest control strategy against the Asian tiger mosquito in northern Italy: a case study. Pest management science 2017;73(1):87-93 Pubmed Journal

Clinical question/ PICO

Population: human populations in built-up European areas
Intervention: IVM by larviciding (IGRs and Bti) and information campaign
Comparator: no IVM

Outcome Timeframe	Study results and measurements	Comparator no IVM	Intervention IVM through the combination of IGRs and Bti	Certainty of the evidence (Quality of evidence)	Summary
Reduction of Ae. albopictus eggs 5 months 1 Not Important	Measured by: Ovitraps Lower better [1] (Randomized controlled) Follow up: 5 months.	234.53 mean number of eggs per trap / 2weeks (Mean) Difference:	65.4 mean number of eggs per trap / 2weeks (Mean) **MD 169.13 lower** (CI 95% 138.5 lower — 204.4 lower)	Low Specification of type and frequency of larviciding treatments insufficient. No report concerning use of freely available larvicides by citizens, nor of percentage of efficient communication effort. [2]	The proposed IVM intervention (monthly treatment with Diflubenzuron or weekly treatment with Bti + extensive information campaign) reduced by about 3.8 times the number of Ae. albopictus eggs collected by ovitraps
Reduction of Ae. albopictus adults [3] 5 months 1 Not Important	Measured by: Sticky traps (GAT) Lower better [4] (Observational (non-randomized)) Follow up: 5 months.	13.3 number of adult Ae. albopictus per trap / 2weeks (Mean) Difference:	4.53 number of adult Ae. albopictus per trap / 2weeks (Mean) **MD 8.77 lower**	Low Specification of type and frequency of larviciding treatments insufficient. No report concerning use of freely available larvicides	The proposed IVM intervention (monthly treatment with Diflubenzuron or weekly treatment with Bti + extensive communication campaign) reduced by 3.5 times the mean number of Ae. albopictus adults

Outcome Timeframe	**Study results and measurements**	**Comparator** no IVM	**Intervention** IVM through the combination of IGRs and Bti	**Certainty of the evidence** (Quality of evidence)	**Summary**
				by citizens, nor of percentage of efficient communication effort. [5]	collected by sticky traps (GAT adhesive plastic sheets).

1. [24]. **Comparator:** [24]. **Supporting references:** [24],
2. **Risk of Bias: serious.** Specification of type and frequency of larviciding treatments insufficient. No report concerning use of freely available larvicides by citizens, nor of percentage of efficient communication effort..
3. IVM using IGRs and Bti caused a reduction in the number of adult Ae. albopictus peaking two months after the beginning of the experimentation
4. [24]. **Comparator:** Primary study [24]. **Supporting references:** [24],
5. **Risk of Bias: serious.** Specification of type and frequency of larviciding treatments insufficient. No report concerning use of freely available larvicides by citizens, nor of percentage of efficient communication effort.. **Upgrade: all plausible confounding would have reduced the effect.** Other factors that could have affeted the value of the study (location, altitude) have been analyzed and they were found to not affect differentially the areas of intervention and the areas not subjected to IVMlt.

References

24. Ravasi D, Parrondo Monton D, Tanadini M, Flacio E. Effectiveness of integrated Aedes albopictus management in southern Switzerland. Parasites & vectors 2021;14(1):405 Pubmed Journal

7.4. Summary of non-evidence publications

8. Conclusions

9. Contributors and interests