Handbook

# Vector Surveillance and Control at Ports, Airports, and Ground Crossings





International Health Regulations (2016)

Vector Surveillance and Control at Ports, Airports, and Ground Crossings

# Vector Surveillance and Control at Ports, Airports, and Ground Crossings

Control of Neglected Tropical Diseases Vector Ecology and Management and Department of Global Capacity, Alert and Response Support for Interational Health Regualtions Capacity Devleopment Ports, airports and ground crossings



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## CONTENTS

Preface	5
Acknowledgements	6
ACRONYMS AND TERMS	8
Executive summary	9

1.	Overview of the IHR legal framework	11
	1.1 Introduction	11
	1.2 International Health Regulations: from conception to the present	11
~		40
<u>Z.</u>	Purpose and scope	13
3.	Roles and responsibilities	14
	3.1 Competent authorities (Article1 of 2005 IHR)	14
	3.2 Surveillance	14
	3.3 Service providers involving travellers, conveyances, containers, cargo and,	
	postal parcels	9
	3.4 Multi-sector cooperation	14
	3.5 Threshold levels of native and invasive species	15
	3.5.1 Mosquito species-specific threshold levels	16
4.	IMPORTANT VECTOR-BORNE DISEASES AND TARGET VECTOR SPECIES	17
	4.1 Medically-important mosquitoes	18
	4.2 Rodents	21
	4.3 Fleas	21
	4.4 Sandflies	22
	4.5 Cockroaches	24
	4.6 Triatomine bugs	24
	4.7 Houseflies	25
	4.7.1 Stable flies	26
5.	Surveillance at points of entry	26
	5.1. Identification of main issues at PoE	26
	5.1.1 Description of the environment	26
	5.1.2 The local entomological situation	28
	5.1.3 The epidemiological context	28
	5.1.4 Summary and conclusions about the receptivity and vulnerability of PoE	28

5.2 Establishment of a surveillance plan	29
5.3 Vector-risk free zone	30
5.4 Essential elements of vector surveillance	31
5.5 Mosquitoes: Surveys of immature stages and adults	32
5.5.1 Eggs	32
5.5.2 Larvae	32
5.6 Adult surveillance	35
5.6.1 Material required for adult catches of mosquitoes	36
5.6.2 Epidemiological considerations to initiate control measures for adul	t
mosquitoes	37
5.7 Rodents: surveillance of rats and mice	37
5.7.1 Runways and rub marks	37
5.7.2 Tracks	38
5.7.3 Gnawing	38
5.7.4 Droppings	38
5.7.5 Urine	38
5.7.6 Rodent hair	38
5.7.7 Techniques for surveillance of rodents	39
5.7.8 Traps	39
5.7.9 Tracking tunnels	42
5.8 Flea surveillance	43
5.8.1 Flea Index	43
5.9 Sandflies surveillance	44
6. VECTOR CONTROL AT THE POINT OF ENTRY	45
6.1 Principle and purpose	45
6.1.1 Mosquitoes	45
6.1.2 Making mosquito control choices	46
6.1.3 Preparedness for mosquito control	46
6.2 Larval control	46
6.2.1 Reduction of vectors	46
6.2.2 Environmental management	47
6.2.3 Biological control	51
6.2.4 Chemical insecticides	52
6.3 Adult control	52
6.3.1 Problem of insecticide resistance and management	52

6.4 Disinsection of aircraft	53
6.4.1 Pre-flight	53
6.4.2 Blocks away	53
6.4.3 Top-of-descent	53
6.4.4 Residual spraying	54
6.5 Rodent prevention	54
6.5.1 Sanitation	54
6.5.2 Rodent-guards	54
6.5.3 Illumination for rodent movement restrictions	55
6.5.4 Pier-side inspections/surveillance on-board ships	55
6.6 Rodent control	55
6.6.1 Conventional rat cage traps	55
6.6.2 Spring board traps	55
6.6.3 Glue traps	56
6.6.4 Electronic rat traps	56
6.6.5 Sherman trap	56
6.7 Sandfly: personal protection and control	56
6.8 Flea: personal protection and control	57
6.9 Cockroach control	58
6.9.1 Dust	58
6.9.2 Aerosol	58
6.9.3 Baits	58
6.10 Housefly control	58
7.Emergency measures	60
8. MONITORING AND EVALUATION.	61
9.Regulations on wildlife and animalsat PoE	63
10. References	64
Annex 1. Personal protective equipment	66
Annex 2. Laboratory Requirements for the Vector Work	68
Annex 3. Surveillance methods of mosquitoes and rodents at PoE	70
Annex 4. Identification, preservation, and transportation of specimen	
to designated laboratory	71
Annex 5. Laboratory requirements for pathogen detection	72
Annex 6. Potential mosquito breeding sites at PoE and interventions to address them	73

Annex 7. Onsite record form for mosquito surveillance	75
A 7.1. Surveillance of immature mosquitoes	75
A 7.2. Record format for surveillance of adult mosquitoes	76
A 7.3. Rodent vector ectoparasites surveillance form	77
Annex 8. Statistical indices for mosquito vector surveillance	78
Annex 9. Chemical/biological insecticides*for vector control	79
Annex 10. Disinsection of aircraft	82
Annex 11. Model ship sanitation control exemption certificate/ship sanitation control	
certificate	83
Annex 8. Statistical indices for mosquito vector surveillance	84

## PREFACE

In May 2005, the Fifty-eighth World Health Assembly adopted new International Health Regulations (IHR), which came into force in July 2007. One of the essential elements of the IHR is capacity building for vector surveillance and control at points of entry (PoE). To that end, State Parties had five years in which to designate the airports and ports (Article 21) that shall develop core capacity requirements for vector surveillance and control (Article 13).

The purpose of this handbook is to provide guidance to Member States on the practical aspects of maintaining sanitary standards at international borders and points of entry (i.e. ports, airports, and ground crossings) as prescribed under International Health Regulations (Articles 3 & 9).

This technical guidance is aimed to assist State Parties' compliance with those obligations by providing technical advice for developing a comprehensive programme for systematic monitoring of disease vectors and integrated vector control at PoE. This includes standardizing the procedures at PoE, ensuring a sufficient monitoring and response capacity, which includes the necessary infrastructure for surveillance and control of vectors at PoE and up to a 400-metre perimeter around them.

In addition, this handbook is intended to be used as reference material by port health officers, regulators, port operators, and other competent authorities in charge of implementing the 2005 IHR at PoE and on conveyances. Its further role is to assist in the development of a management plan for preparing and performing vector surveillance and applying public health measures within the framework of the IHR (2005).

Suggestions and comments are welcome to strengthen future editions of this document.

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## ACRONYMS AND TERMS

Bti:	Bacillus thuringiensis israelensis
DDT:	Dichloro-diphenyl-trichloroethane
DHF:	Dengue Haemorrhagic Fever
DNA:	Deoxyribonucleic acid
dNTPs:	deoxyribonucleotide triphosphates
DPX:	Distrene-Plasticiser-Xylene (mountant)
EC:	Emulsifiable Concentrate
HRP-2:	Histidine-Rich Protein-2
IHR:	International Health Regulations
Instar:	A developmental stage of mosquito larva
IRS:	Indoor Residual Spray
IVM:	Integrated Vector Management
JSB:	Jaswant-Singh-Bhattacharya (stain)
LDH:	Lactodehydrogenase (enzyme)
LLIN:	Long Lasting Insecticide Net
MSDS:	Material Safety Data Sheet
Ootheca:	A small brown egg mass (of cockroaches)
OP:	Organophosphate
PCR:	Polymerase Chain Reaction
PoE:	Points of Entry
PPE:	Personal Protection Equipment
ppm:	parts per million
TBE:	Tris/Borate/EDTA (Buffer)
UV:	Ultraviolet
WHO:	World Health Organization

### EXECUTIVE SUMMARY

In May 2005, the Fifty-eighth World Health Assembly adopted the new International Health Regulations (IHR), which came into force in July 2007. The updated regulations provide that, within five years, all the State Parties must develop core capacities and infrastructure to designate airports and ports that shall develop core capacities requirements (Articles 13, 17 and 20 and Annex 1), and may also designate ground crossings where justified for public health reasons (Article 21). One of the areas stressed in the IHR is capacity building for vector surveillance and control at points of entry and up to (at least) a 400-metre perimeter around them. This is due to the continuous threat of the spread of vectors and vector-borne diseases by ships, aircraft, and conveyances from one country to another. There have been innumerable examples of vector entry and disease transmission in and around airports and ports available in the literature since the 1950s. «Airport malaria» in Europe through infected *Anopheles* vectors from malaria-endemic areas and widespread distribution of *Aedes albopictus* in the US and Europe are two such examples.

Of all the vectors that require attention the most important ones are mosquitoes, rodents, fleas, sandflies, houseflies, and cockroaches; these vectors are specifically dealt with in this handbook, as are the different methods of vector surveillance. The important facets of the latter discussed include the roles and responsibilities of the concerned authorities at points of entry, recognizing the threat of vector-borne diseases, instituting vector surveillance at points of entry, organizing evidence-based vector control strategies, and if necessary invoking emergency measures to prevent the spread of vector-borne disease. Various physical, chemical, biological, mechanical, and environmental methods of vector control are known and many of these have been highlighted with the help of photographs in the text and annexes. Material required in surveillance programmes has been listed, as have different techniques recommended for disinsection of aircraft.

Finally annexes have been included to serve as a guide on situation-specific integrated vector management, including examples of potential mosquito breeding sites observed at points of entry and physical/minor engineering methods of intervention, surveillance methods for conveyances, containers, cargo, postal parcels and baggage, requirements of a site laboratory, preservation and transportation of specimens to a referral laboratory, methods of pathogen detection, on-site record forms for vector surveillance and control, statistical tables for vector surveillance, and physical methods of vector control used by entities stationed in ports.

#### 1. OVERVIEW OF THE IHR LEGAL FRAMEWORK

#### **1.1 Introduction**

Vector-borne diseases (such as malaria, dengue, chikungunya, Zika virus, yellow fever) are reported in over 100 countries, and put up to 60% of the world's population at risk of infection; more than 500 million cases are reported each year (WHO 2014). International travel and transport play an important role in the rapid spread of vector-borne diseases all over the world – as borders become more porous and the speed and extent of travel and shipping increase, so too does the potential of the spread of reservoirs and pathogens related to vector-borne diseases.

The global development of the shipping industry and expansion of port cities during the past two centuries has led to the global spread of Aedes aegypti and Aedes albopictus in countries within the Americas and European Regions. This has resulted in the spread of several diseases: yellow fever in the Americas, dengue and severe dengue in tropical countries (especially in South-East Asia and the Pacific) and, more recently, Zika virus in the Americas and Western Pacific Regions. A member of the Anopheles gambiae species complex<sup>1</sup> believed to originate in Dakar, Senegal, invaded Natal, Brazil in the 1930s. Originally arriving via a fast ship, the species spread further via car, train, and boat. It is a classic example of an invasion of a highly potent disease vector from one continent to another. It took the Brazilian Government assisted by the Rockefeller Foundation – nearly 18 months of a highly-disciplined campaign to stamp out this vector from north-eastern Brazil with larvicide Paris green (Parmakelis et al. 2008). Other documented examples of invasive disease vector species of mosquitoes include the Culex pipiens complex in Europe and the United States of America in the 1990s, Culex quinquefasciatus in tropical Asia, India, Haiti and Brazil in the 19th century, Anopheles darlingi in Amazonian Peru in the 1990s, Aedes aegypti linked with yellow fever in the Region of the Americas from the 16th to the 20th century, and Aedes albopictus linked with chikungunya in Italy from 2005 to 2008 (Lounibos-Philip 2010).

#### 1.2 International Health Regulations: from conception to the present

Globalization and industrialization have opened and expanded trade and commerce, which in turn have provided impetus to increased air traffic in the post world war era. The rapid global growth of connectivity has been responsible for the spread of vectors and phenomenon such as «airport malaria». To address the threat of global spread of vectors and vector-borne diseases, through points of entry (PoE), i.e. ports, airports, and ground crossings between nations, WHO brought Member States under the umbrella of the International Health Regulations (IHR) in 1969 to which all the Member States were signatory. Member States were required to notify to WHO any specific disease outbreaks/public health emergencies of international concern within 24 hours through focal point (Article 5 and 6), maintain sanitary standards at international borders and PoE as prescribed under IHR. In May 2005, the Fifty-eighth World Health Assembly adopted the new International Health Regulations (IHR, 2005), which came into force in July 2007. In the IHR, State Parties are requested to designate airports, and ports

<sup>&</sup>lt;sup>1</sup> Identified through DNA as Anopheles arabiensis in the 1960s.

that shall develop core capacities requirements (Art.19, Art. 20, and Annex 1 of the IHR), and where justified for public health reasons, may also designate ground crossing (Art. 21). One of the underlined areas is capacity building for vector surveillance and control at PoE.

As stated in Annex 5 of the IHR (2005), State Parties shall establish programmes to control vectors that may transport infectious agents constituting a public health risk. Such programmes must ensure that vectors are controlled to a minimum distance of 400 metres from those areas of point-of-entry facilities that are used for operations involving travellers, conveyances, containers, cargo, and postal parcels, with extension of the minimum distance if vectors with a greater range are present. The IHR (2005) also stipulates, in light of Articles 22, 24, 27 and Annex 4, that competent authorities are required to ensure that facilities used at PoE are maintained in a sanitary condition and are kept free of sources of infection and contamination, including vectors and reservoirs; conveyance operators are so tasked for conveyances. Furthermore, Annexes 3 and 9 of the IHR (2005) encompass technical requirements respectively on vector surveillance and control with regard to ship inspection and those of disinsecting or sanitary treatment measures in aircraft. Vector surveillance and control at PoE has become an essential and pressing issue for the implementation of the IHR. Figure 1 shows an inspection being conducted at a laboratory at an entry point in China.



Figure 1. A bird being examined for West Nile virus infection in a PoE laboratory in China, in compliance with IHR © Chunxiao Liu, Shenzhen Entry-Exit inspection and quarantine bureau, China

### 2. PURPOSE AND SCOPE

Due to the new challenges/requirements brought about by the 2005 IHR, necessary efforts are being made by State Parties to ensure effective vector surveillance and control at PoE, and on conveyances. This handbook is aimed at assisting State Parties to comply with the obligations of the IHR by providing technical advice for developing a comprehensive programme for systematic monitoring of vectors and an integrated vector-control plan at points of entry. It is also intended for use by port health officers, airport health officers, border health officers, regulators, port operators, and other competent authorities in charge of implementing the 2005 IHR at points of entry and on conveyances.

The handbook is based on provisions in the IHR, regarding vector surveillance and control and should be used as a basis for the development of a management plan for preparing and performing vector surveillance and applying public health measures within the framework of the 2005 IHR.

The primary aim of developing this handbook is to assist public health authorities at PoE in strengthening core capacities and managing vector surveillance and control programmes by providing technical guidance on the optimal use of resources, planning, monitoring, and decision-making.

This handbook is also intended to provide examples of accepted best practices. However, it is acknowledged that there may be equally effective alternative solutions that could be deployed to achieve the desired objectives. Such solutions, should they be evidence-based, are welcomed for future editions of this handbook. Figure 2 highlights an overview of several steps recommended by the IHR.

Figure 2. An overview of various steps recommended for implementing the 2005 IHR provisions for vector control at points of entry



#### 3. ROLES AND RESPONSIBILITIES

#### 3.1 Competent authorities (Article1 of 2005 IHR)

The competent authorities should, subject to International Health Regulations, establish and revise national or regional relevant laws and regulations and ensure their effective implementation [Article 3 (4)].

The competent authorities should establish a national or regional intersectoral cooperation mechanism especially for the implementation of integrated vector management (WHO, 2012b).

The competent authorities are responsible for the supervision of vector surveillance and control within the scope set out in this guidance, as well as for transferring technical know-how and operational vector-control skills to port owners or operators, conveyance operators on an international voyage and relevant port service providers involving travellers, conveyances, containers, cargo, and postal parcels to carry out vectors control (IHR: Annex 5).

#### 3.2 Surveillance

The PoE authorities or operators, conveyance operators, competent service providers should conduct surveillance and apply public health measures with an aim to keeping vectors density below the threshold level set by national policies and practices (IHR:Annex 5).

## 3.3 Service providers involving travellers, conveyances, containers, cargo and, postal parcels

Service providers/port authorities for travellers, conveyances, containers, cargo and postal parcels shall be responsible for vector control within the area they are responsible for, keeping vectors density below the threshold level set by national policies and practices.

Service providers for travellers, conveyances, containers, cargo, and postal parcels shall assist competent authorities, port authorities or operators, and operators of conveyances in promoting technical know-how and skills of vector surveillance and control to their staffs.

#### 3.4 Multi-sector cooperation

Technical cooperation for vector surveillance and control should be strengthened between Member States and other relevant international organizations or other public organizations, between Member States, or between public sectors and private sectors. This includes establishing and updating relevant technical regulations, information communication, personnel training, and technical support. PoE should identify partner institutions/laboratories which will assist in identification of vectors, reservoirs of infections and pathogens. A network of such institutions will be of great value for early detection and accurate identification of invasive species, a prerequisite to initiating an appropriate response. Figure 3 shows identification of a mosquito vector species in a designated laboratory in China based on digital image transfer.



Figure 3. Digital image of an invasive mosquito being readied to be sent by email to an expert for species identification © Chunxiao Liu, Shenzhen Entry-Exit inspection and guarantine bureau, China

#### 3.5 Threshold levels of native and invasive species

The threshold for initiating control action against any invasive species at PoE should be zero. Mosquitoes are a case in point: even if the number of larvae/dip or adults captured during landing or trap collection are few, full-scale surveillance would be warranted to know the extent of the invasion at PoE – with the aim of eliminating the invasive species with all available resources. The phase of the intense elimination campaign should also serve as vector monitoring to confirm whether the goal of elimination has been achieved or not. In the example of the mosquito, monitoring of larvae and adults must continue until such time that the zero status of the imported vector has been confirmed beyond any reasonable doubt.

The threat level of an invasive mosquito species would depend upon multiple factors: its capacity to create a foothold in an alien land, how abundant the adult populations of these species could become in a short span of time, as well as their flight ranges, host preferences, biting aggressiveness, and disease transmission potential.

Continuous surveillance and monitoring of native mosquito species (to prevent their exit) and/or any newly invaded species will form the basis of when, where and what methods of control will be initiated at PoE. In addition, monitoring of virus activity in sentinel birds, wild birds and animals has enabled authorities to adjust the threshold of when the vector-control action would begin.

#### 3.5.1 Mosquito species-specific threshold levels

Mosquito species differ in their choices of breeding habitats, flight ranges, feeding behaviour, and activity rhythms. All these factors are critical in defining a threshold of tolerance above which larvicidal or adulticidal action must be undertaken. Determining the threshold values of either nuisance causing-or disease-vector species should be a priority.

Threshold values can be based on:

- larval surveillance in potential breeding sites at PoE e.g. salt marshes, wetlands, construction sites, container habitats, tyres, etc.;
- adult surveillance e.g. landing catches or trap catches;
- pathogen monitoring e.g. sentinel chicken stations, wild birds (for West Nile virus) or horses (for Eastern equine encephalitis) or in the vector population itself;
- public complaints related to mosquitoes.

#### 3.5.1.1 Larval counts

Mosquito larvae go through four developmental stages called instars, usually referred to as 1<sup>st</sup> to 4<sup>th</sup> instar larvae, which eventually turn into pupae. In the case of *Aedes* breeding, if the combined larval and pupal count exceed on average >5/dip (also taking zeros into account) and if >25% of all samples are positive for immature mosquitoes, it should mark the threshold for control action (Strickman and Kittayapong, 2003). Studies have found varying risk of dengue transmission due to fluctuations in weather parameters and food stress during larval period reflected in adult sizes (Strickman and Kittayapong, 2003).

Larval sampling should be systematic. It should take into account temporal and spatial coverage. Larval sampling sites should include either fixed or random sampling (or both) depending upon whether the habitats are permanent or temporary in nature. Experience has shown that lower densities of salt marsh mosquitoes that invade human habitations with a long flight range of 3-15 miles (~5-24 km) would warrant larviciding at distant breeding 'hot spots' as compared to fresh water or container breeder species with a limited flight range of a few hundred metres. However when located near human inhabitation, larval counts of >2 larvae/dip in manmade containers or natural habitats might warrant control.

For Aedes species, a House Index of >5% and Breteau Index of >20% should sound an alarm and would warrant immediate control action at PoE. The detection of invasive Aedes species should sound the alarm and would warrant immediate control action at PoE.

#### 3.5.1.2 Adult mosquito threshold

Various threshold levels have been suggested for native mosquitoes based on different levels of incidence and types of traps. Ovitraps are particularly useful in monitoring Aedes activity in an area and can help determine population levels in space and time (Musah et al (2008); http://www.ncbi. nlm.nih.gov/pubmed/18344071).

A few examples are given below.

- 1. Human landing rates: >2-3 mosquitoes/minute in a populated area; 5-10 mosquitoes/minute near wet woodlands (50-100 mosquitoes/minute might also occur in exceptional circumstances).
- 2. New Jersey-style adult light trap: >25 adults of a nuisance species caught/trapped per night.
- 3. CDC trap is more attractive: >50 mosquito females caught/trapped per night (Smallegange et al. 2010).

#### 4. IMPORTANT VECTOR-BORNE DISEASES AND TARGET VECTOR SPECIES

Diseases such as malaria, dengue, chikungunya, yellow fever, Zika virus, plague, leptospirosis, etc. are vector-borne diseases that pose major public health risks via PoE to a non-endemic area (Table 1).

Organisms of high priority are those that are disease vectors, nuisance causing, or reservoirs of infection, which when successfully transported have the potential to invade, establish and cause harm to public health or to stored products in non-endemic areas. Every country needs to do its own risk assessment. The level of risk will vary depending upon local environmental factors and disease transmission potential of the local area.

Table 1. Selected vector-borne diseases that could be potentially transported by conveyances	
through sea, air, and ground entry points	

Disease	Vector	Causal organism	Reservoir	Risk at PoE
I. Mosquito-borne d	diseases			
Filariasis Culex, Anopheles Aedes niveus group		Plasmodium species W. bancrofti (nocturnal, periodic) W. bancrofti (diurnal s	Humans Humans Humans Humans	High/Moderate Low High High
Chikungunya Dengue fever & Zika virus Yellow fever Japanese encephalitis and West Nile virus	Aedes species Aedes species Aedes species Aedes & many Culex spp. Culex spp	ub-periodic) Brugia malayi Alphavirus Flavivirus Flavivirus Flavivirus Flavivirus Flavivirus	Humans Humans Humans/monkeys Mammals/ birds Birds	High Moderate Moderate to high
II. Sandfly-borne di	seases			
Visceral Leishmaniasis Cutaneous Leishma- niasis Espundia Sandfly fever	Phlebotomus spp. P. papatasi, P. sergenti, etc. Lutzomyia spp. P. papatasi	Leishmania donovani L. tropica L. major, L. braziliensis Sandfly fever virus	Humans/ Mammals Humans/ Mammals Humans	Moderate Low Low Moderate
III. Fly- and cockroa	ch-borne diseases (can	act as passive vector	s)	
Examples (selected) Bacillary dysentery Amoebic dysentery Gastroenteritis Typhoid Paratyphoid Cholera Poliomyelitis Viral hepatitis (Type A) Trachoma Yaws Asthma	M. domestica & Cockroach M. domestica & Cockroach M. domestica & Cockroach M. domestica & Cockroach M. domestica M. domestica M. domestica M. domestica M. domestica Cockroach	<i>E. histolytica</i> Specific/Non specific	Humans Humans Humans/Animals Humans Humans Humans Humans Humans Humans	Low High High High High High High High Low
IV. Flea-borne disea	ases			
Plague (Bubonic) Ende- mic/Murine Typhus Dipylidium caninum- Hymenolepis diminuta H. nana	Xenopsylla species Xenopsylla species Ctenocephalides felis/ canis X. cheopis / N. fasciatus X. cheopis / C. canis/ Pulexirritans	Yersinia pestis R. typhi Dipylidium caninum Hymenolepis diminuta H. nana	Rodents Rodents, domestic Animal Dogs/Cats,wild Carnivores Rats, Mice	High Moderate Low Low Low
V. Louse-borne dise	ases			
Epidemic typhus Epidemic relapsing fever Trench fever Dermatitis	PediculushumanusPedicu- lushumanusPediculushu- manusPediculus humanus/ capitis	R. prowazeki Borreliare currentis Bartonella quintana Bacterial super infection	Humans Humans Humans/Animals Humans	Moderate Moderate Moderate Moderate

Disease	Vector	Causal organism	Reservoir	Risk at PoE
VI. Tick-borne dise	ases			
Kyasanur Forest Disease Tick typhus Tularaemia Relapsing fever Crimean Congo Hae- morrhagic Fever	Hard ticks species Hard ticks species Hard ticks species Soft Tick Soft Tick Soft Tick species	Arbovirus group B R. conorii P. tularensis B. duttoni B. unyaviridae	Monkeys/Birds Dogs Rabbits/Rodents/ Cattle/Rats Mammals, birds	Low Low Low Low Low
VII. Mite-borne dis	eases		,	
Scrub typhus Rickettsial pox	Leptotrombidium deliense Allodermanyssus san- guineus	Orientiatsutsugamushi R. akari	Rodents Rodents Humans	Moderate High
VIII. Reduviid bugs	(Triatoma)			
Chagas disease	Triatoma infestatation and Rhodnius prolixus	Trypanosoma cruzi	Domestic animals/ humans	Low
IX. Tsetse flies	1		1	1
Trypanosomiasis	Glossina species	T. gambiense and T. Rhodesiense	Wild animals/ Cattle/ Humans	Low

#### 4.1. Medically-important mosquitoes

There are 490 species in the Anopheles genus (Figure 4) and more than 3 100 in the Culex genus distributed all over the world. However, only a handful of them have the potential to be disease vectors and of medical significance. Between the vectors, some play a primary role and have either limited or vast geographical distribution, while others are secondary vectors of regional importance and might play a limited role in disease transmission. Mosquito vectors of different regions are listed in Tables 2–7.



Figure 4. Anopheles mosquito © Shanghai Entry-Exit inspection and quarantine bureau, China.

Anopheles	Aedes	Culex	Mansonia	Armigeres	Others
Anopheles (Ano.) bancrofti barbirostris belenrae campestris claviger donaldi kleini labran- chia elesteriletifer messeae nigerrimus pullussa charovisinensis sineroides whartoni <b>Anopheles (Celia)</b> annulais annulipes s.l. culicifacies s.l. farauti s.l. flavirostris fluviatilis gambiae complex hancocki jeyporiensis kanvari koliensis leucosphyrus group baimai balabacensis dirus sulawesi ludlowae maculates s.l. minimus partoni philippinensis suphilippinensis suphilippinensis pucherrimus punctulatus superpictustessellatus vagus	Aedes (Adm.) vexans Aedes (Fin.) fijiensis harinasutai japonicus kochi niveus complex. oceanicus poicilius samoanus togoi tutuilae Aedes (Och.) dorsalis normanensis vigilax Aedes (Stg.) aegypti albopictus cooki hensili Polynesiensis scutellaris polens	Culex (Cux.) annulirostris gelidus pipiens quinquefasciatus sitiens tritaeniorhynchus vishnui complex Culex (Ocu.) bitaeniorhynchus	Mansonia (Mnd.) annulata bonneae dives indiana uniformis		Coquillettidia (Coq. ) crassipes

Table 2. Mosquito vectors of Australia, South-East Asia, and the Pacific

Source: Walter Reed Biosystematics Unit (http://www.wrbu.org/index.html).

Anopheles	Aedes	Culex	Mansonia	Armigeres
Anopheles (Ano.) Atroparvus Claviger Messeae sacharovi Anopheles (Celia) arabiensis culicifacies s.l. gambiae complex moucheti multicolor pharoensis s.l. pulcherrimus sergentii stephensi superpictus	Aedes (Adm.) vexans Aedes (Och.) dorsalis Aedes (Stg.) aegypti albopictus	Culex (Cux.) antennatus gelidus pipiens quinquefasciatus sitiens tritaeniorhynchus uni vittatus Culex (Ocu.) bitaeniorhynchus	Mansonia (Mnd.) annulata bonneae dives indiana uniformis	Armigeres (Arm.) subalbatus

Table 3. Mosquito vectors of central Asia

Source: Walter Reed Biosystematics Unit (http://www.wrbu.org/index.html).

#### Table 4. Mosquito vectors of Africa

Anopheles	Aedes	Culex	Mansonia	Others
Anopheles (Celia) arabiensis funestus gambiae hancocki melasmerus moucheti multicolor nili s.l. pharoensis s.l. wellcomei multicolor sergentii	Aedes (Adm.) vexans Aedes (Alb.) stocksi Aedes (Dic.) furcifertsylori Aedes (Neo.) mcintoshi Aedes (Stg.) aegypti africanus albopictus bromeliaeluteocep halus	Culex (Cux.) antennatus pipien quinquefasciatus sitiens theileri tritaeniorhynchus uni vittatus Culex (Ocu.) bitaeniorhynchus	Mansonia (Mnd.) uniformis	Coquillettidia (Coq.) fuscopennata

Source: Walter Reed Biosystematics Unit (http://www.wrbu.org/index.html).

#### Table 5. Main mosquito vectors of Europe

Anopheles	Anopheles	Aedes	Culex
Anopheles (Ano.) atroparvus claviger labranchiae maculipennis s.s. (secon- dary vector of malaria) messeae (secondary vector of malaria) subalpinus (secondary vector of malaria	Anopheles (Celia) cinereus (secondary vector of malaria) multicolor (secondary vector of malaria) sergentii (secondary vector of malaria) superpictus	Aedes (Stg.) aegypti albopictus	Culex (Bar.) modestus Culex (Cux.) Perexiguus/vittatus pipiens

Source: Schaffner et al.(2001).

#### Table 6. Mosquito vectors of North America

Anopheles	Aedes	Culex	Mansonia	Others
Anopheles (Ano.) aztecus crucians freeborn ipseudo punctipennis punctimacula punctipennis quadrimacu- latus walkeri Anopheles (Ker.)	Aedes (Fin.) japonicus Aedes (Och.) angusti vittatus canadensis dorsalis infirmatus melanimon scapularisti vittatus	Culex (Culex) antennatus gelidus pipiens quinquefasciatus sitiens tritaeniorhynchus	<b>Mansonia (Man.)</b> titillans	Coquillettidia (Rhy.) venezuelensis Coquillettidia (Coq.) perturbans Culiseta (cli.) melanura Culiseta (Cus.) inornata
Anopheles (Nys.) Anopheles (Nys.) albimanus argyritarsis darling	Aedes (Pro.) triseriatus Aedes (Stg.) aegypti albopictus	uni vittatus <b>Culex (Ocu.)</b> bitaeniorhynchus		<b>Psorophora (Jan.)</b> ferox

Source: Walter Reed Biosystematics Unit (http://www.wrbu.org/index.html).

#### Table 7. Mosquito vectors of South America

Anopheles	Aedes	Culex	Mansonia	Others
Anopheles (Ano.) calderoni pseudo punctipennis punctimacula Anopheles (Ker.) bellatorcruzi lepidotus neivai Anopheles (Nys.) albimanus albitarsis aquasalis argyritarsis benarrochi braziliensis darling marajoara nuneztovaris.l. oswaldi triannulatus	Aedes (Och.) albifasciatus angusti vittatus scapularistae niorhynchus Aedes (Stg.) aegypti albopictus	Culex (Cux.) nigripalpuspipiens- quinquefasciatus Culex (Mel.) ocossa Portesi spissipestaeniopus vomerifer	Mansonia (Man.) titillans	Coquillettidia (Rhy.) venezuelensis Haemogogus (Hag.) janthinomys Psorophora (Jan.) ferox Trichoprosopon digitatum

Source: Walter Reed Biosystematics Unit (http://www.wrbu.org/index.html).

#### 4.2 Rodents

Rodents are a part of human history. Rodents infest human dwellings, and their urine and faeces contaminate foodstuffs in enormous quantities. Rodents can also cause destruction aboard ships, crafts, rail cabins and motor lorries by gnawing through fittings, wiring, and circuitry, which can even cause short circuits resulting in fire hazard. Rats in particular occur in almost all human communities in both rural and urban areas. They have great propensity for dispersal. Rats can invade and establish in new lands easily. Many species of medical importance have been found and may pose serious danger to humans. Withinto the family Muridae, the prominent species are *Rattus norvegicus* (Figure 5), *Rattus rattus* (Figure 6), *Tetera indica, Bandicoota bengalensis, Mus musculus* (Figure 7) and *Funumbulus palmarum*. Of these, features of three species are mentioned in Table 8.

Species	Rattus norvegicus (Figure 5)	Rattus rattus (Figure 6)	Mus musculus (Figure 7)	
	© Shanghai Entry-Exit inspection and quarantine bureau, China.	© Shanghai Entry-Exit inspection and quarantine bureau, China.	© Shanghai Entry-Exit inspection and quarantine bureau, China.	
Common name	Brown rat or Norway rat	Roof rat or black rat	House mouse	
Weight	500 g	250 g	20 g	
Length	45 cm	40 cm	18 cm	
Habitat	Principally lives in sewers and holes and feeds on garbage	Under the roof of any type of building	Around supplies of grain, cereals, and flour	

Table 8. Common rodent species of medical importance and their	features
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Rodents can carry serious communicable diseases such as plague and murine typhus transmitted by fleas, leptospirosis through urine and food-borne illnesses by contamination of food with faeces (e.g. Salmonellosis).

#### 4.3 Fleas



Figure 8. A flea © Mathieu Bangert Fleas are holometabolous insects with 2 380 described species placed in 15 families and 238 genera. Fleas are laterally compressed, wingless insects; the head is shield or helmet shaped, compound eyes are absent, and mouthparts are specialized for piercing and sucking (Figure 8). Fleas are entirely ectoparasites of mammals and birds. There are 137 flea species representing 22 genera and 6 families that have birds as hosts, 5 species that are found on both bird and mammal hosts, and the remainder of the species are mammal specific. Fleas are of tremendous medical and economic

importance as vectors of several diseases to human health including bubonic plague, murine typhus, and (seldom) tularemia.

#### 4.4 Sandflies

Phlebotomine sandflies are important disease vectors. They transmit viral, bacterial, and protozoan diseases to humans. The most important and widespread disease that sandflies transmit is leishmaniasis (visceral and cutaneous). About 500 000 cases of this disease are reported annually. Five countries, namely India, Sudan, Nepal, Bangladesh and Brazil account for nearly 90% of the global burden. Within the endemic countries, Leishmaniasis occurs among the socially marginalized and poorest communities. Although there are several genera of Phlebotomine sandflies, *Phlebotomus* is the most important and widespread (Table 9).

Sandflies live for about a month of which about 20 days are spent as larvae. They also breed in soil. Adults are poor fliers; they usually hop for short distances at a time. Sandflies breed in the presence of organic debris, making homes with mud walls plastered with cow dung an ideal breeding ground. They can be found on rocks, in tree hollows and other cracks and crevices. They take blood meals usually in the evening and at night and can travel in a radius of a few hundred metres around their habitat.

Leishmaniasis is among the most recognized diseases transmitted by sandflies. *Phlebotomus* species are also vectors of *bartonellosis, verrugaperuana, pappataci fever* (sandfly fever), caused by Naples and Sicilian strains of *Phleboviruses* (family Bunyaviridae), which also includes closely related Toscana virus. Sandflies are also known to transmit Chagres virus and Punta Toro virus.

Adults are small sized about 1.5–3 mm, brownish in colour with conspicuous black eyes, hairy body, wings and legs. The oval lanceolate wings are carried in a semi erect posture. Males possess long prominent genital terminalia. The life-cycle of a sandfly is similar to that of other insects, going through four stages i.e. egg, larvae, pupae and the adult stage. Females lay 30 to 70 eggs in the soil, in cracks and crevices which are dark, humid and rich in organic matter and retain water by capillarity. The eggs are small, elliptical and brownish in colour. The female sandfly spreads them around in several different sites. Within one to two weeks, the eggs develop and larvae hatch. If conditions become very cold, the eggs will enter in diapause or hibernation. During this state, development is arrested and the egg does not develop further. It will hatch once temperatures begin to warm. Thus the emergence of the 1st generation of sandflies generally coincides with the onset of spring/summer.

Larvae are small and whitish in colour with a black head capsule. They feed on dead organic matter and are often found in the crevices of walls or rocks, animal burrows, caves, or in decaying leaves. Following four larval stages, the fully-grown larva metamorphoses into the pupal stage. The pupa is brownish in colour and shortly before the fly emerges, the wings and eyes turn black. Pupal development takes five to 10 days with the adult often emerging just before dawn. The male flies generally emerge about 24 hours prior to the females. Only the female sandfly will suck blood, which is necessary for egg production. Both males and females feed on plants. The adults are about 2.5 mm in length. The sandflies often mate near the host. Some Indian sandfly species have been observed mating and swarming on the bodies of buffaloes, which occurs most often near saltmarshes. Some species do, however, exist in fresh water areas and tree holes.

Australia, Pacific and Asia	Central Asia and Middle East	Africa	Europe	North America	South America
	Middle East Phlebotomus (Adl.) balcanicus brevis halepensis longiductus simici tuarnicus Phlebotomus (Eub.) argentipes Phlebotomus (Lar.) guggisbergi kandelakii keshishiani	Africa Phiebotomus (Lar.) ariasi guggisbergi orientalis pedifer perfiliewi perniciosus Phiebotomus (Pab.) sergenti Phiebotomus (Phb.) duboscqipapatasi Phiebotomus (Syb.) martini	Phiebotomus (Adl.) balcanicus halepensis kyreniae longiductus simici Phiebotomus (Lar.) ariasi guggisbergi kandelakii langeroni longicuspis neglectus orientalis perniciosus tobbi Phiebotomus (Pab.) alexandri andrejevi caucasicuschabaudi mongolensis saevus Phiebotomus (Phb.) bergeroti duboscqipapatasi Phiebotomus (Syb.) martini rossi Sergentomyia (Ser.) dubia	North America Lutzomyia (Lut.) diabolica longipalpis Lutzomyia (Nys.) olmeca olmeca ylephiletor Lutzomyia (Psy.) panamensis	South America Bichromomyia flaviscutellata olmeca olmeca olmecareducta Dampfomyia (Dam.) anthophora Evandromyia (Can.) pinottii Lutzomyia (Hel.) ayacuchensis hartmanni peruensis Lutzomyia (Lut.) longipalpis Lutzomyia (Lut.) longipalpis Lutzomyia (Lut.) longipalpis Lutzomyia (Lut.) longipalpis Lutzomyia (Lut.) longipalpis Lutzomyia (Lut.) longipalpis Lutzomyia (Lut.) longipalpis Lutzomyia (Pfi.) christophei torvida townsendi verrucarum youngi Pintomyia (Pfi.) christophei torvida townsendi verrucarum youngi Pintomyia (Pin.) pessoai Psychodopygus carrerai carreraichagasi squamiventris maripaensis squamiventris maripaensis squamiventris wellcomei yucumensis Sciopemyia fluviatilis

Table 9. Important sandfly species in different regions of the world

#### 4.5 Cockroaches



Figure 9. An adult cockroach © Shanghai Entry-Exit inspection and guarantine bureau, China.

Cockroaches are distributed worldwide and are one of the most common pests aboard ships, aircraft, and lorries especially in the areas where food is kept or stored (Figure 9). Three types of cockroaches are most common: the American, Oriental, and German.

Cockroaches prefer dark and warm areas, crevices and a variety of other human-made hideouts. They are usually nocturnal, agile, and live in colonies where often adults and nymphs share the habitat (Figure 10). They are characterized bytwo pairs of wings, a flattened appearance, and yellow-brown to dark brown coloura-

tion. Their length varies from 5 to 73 mm. In the tropics they may live and breed outdoors. Foul-smelling cockroach faeces are an indication of infestation. Adult female cockroaches produce a visible brown egg capsule (called an ootheca) located at the tip of the abdomen. The



Figure 10. Cockroach life-cycle: ootheca, nymphs and adults © Shanghai Entry-Exit inspection and quarantine bureau, China.

#### 4.6 Bugs



Figure 11. A Triatomine bug © Mathieu Bangert

egg capsule protects the developing 30-40 eggs inside until they are ready to hatch. Gravid females may carry the egg capsule up to three weeks, until the eggs are within 24 hours of hatching. Young cockroaches are called nymphs and can look very similar to the adults. The nymphs moult several times and finally mature to an adult. Cockroaches act as mechanical vectors and may cause asthma and transmit dysenteric or diarrhoeal diseases and typhoid fever. Some diarrhoeal diseases are viral ones like Norovirus disease.

Triatomine bugs are large blood-sucking insects that occur mainly in Latin America and the southern USA. A number of species have adapted to living in and around houses and are important in the transmission to humans of *Trypanosomacruzi*, a protozoan parasite that causes Chagas disease (also known as American trypanosomiasis). Chagas disease, which occurs in most South and Central American countries (particularly in rural areas), is incurable in its chronic form and may cause damage to the heart and intestines. In 1996

it was estimated that between 16 and 18 million people were infected, of whom over 6 million would develop clinically overt disease and 45 000 would die per year.

Although different *Triatoma* species occur in various countries, all are similar in appearance and life-cycle, and are easy to distinguish from other insects (Figure 11). In Latin American countries, the bugs are known in a variety of local names, including barbeiros, vinchucas, pitos, and chinches. The life-cycle of the triatomine bug varies from 4 to 24 months, depending on the species and environmental conditions. The most important vector species usually have one or two cycles per year. The adults differ from the immature stages (nymphs) by the presence of fully developed wings and genitalia. The adults and immature stages occupy similar habitats and have similar feeding habits.

The bugs occur in both forested and dry areas in the Americas. The adult and immature stages live in the burrows and nests of wild animals, including birds, bats, squirrels, opossums and armadillos, on which they feed during the night by sucking blood when the animals are asleep.

A number of species have adapted to living in and near houses, where they feed on humans and domestic animals, including chickens, cattle, goats, cats and dogs. Feeding may take 10–25 minutes. The triatomine bug species that transmit Chagas disease rest during the day in dark places close to their source of blood.

During daytime the triatomine bugs prefer to hide in dark crevices, which are abundant in unplastered cracked walls of mud or mud-brick. Other hiding places are behind pictures, among furniture, boxes, and clothes hanging from pegs in walls, and in beds. An important vector species, *Rhodnius prolixus*, which is found in Colombia, Venezuela, and Central America, often hides in palm-thatched roofs. *Triatoma infestans*, which is the most important vector species in South America, often hides in roofs of wood and soil. A vector species in Central America, *Triatoma dimidiata*, also hides in cracks in floors. Some of the triatomine bug species find suitable resting places in areas surrounding houses, from which they may re-enter houses to feed. Resting occurs in all sorts of stored objects, such as firewood, lumber, tiles, stones and bags of food. Resting bugs are also found in animal quarters, such as chicken coops and goat corrals.

Biting is usually relatively painless and most people are not woken up when it occurs. In some cases severe itching and other skin problems occur afterwards. The Trypanosomes are discharged with the faeces of the insects while they are feeding on the blood and enter the human body when scratching occurs or via the mucous membranes. Bites from numerous triatomine bugs can cause chronic anaemia through loss of blood. Transmission can be successfully interrupted by removing nests of triatomine bugs from around human homes.

#### 4.7 Houseflies



Figure 12. Housefly © Shanghai Entry-Exit inspection and quarantine bureau, China.

Houseflies, *Musca domestica*, are found worldwide and constitute over 90% of all flies. It is one of the most successful of insect species. A housefly in its lifespan can lay around 500 eggs, mostly in dead or decaying matter such as garbage and faeces, cow dung, etc. The fly maggots are pale in colour and feed on organic matter and after the fourth instar metamorphose into reddish brown pupae within 36 hours. Eventually adults emerge from the pupae stage and live for 2 to 4 weeks. The female fly mates one time only, 36 hours after emerging from the pupae stage, and receives enough sperm to fertilize all the batches of eggs.

Houseflies tend to aggregate near food and decaying matter. Given their distribution, they can be found at PoE and also on vessels, particularly the kitchen and service areas. They feed by process of sponging: their saliva or vomit can dissolve solid food. Houseflies are

quite dangerous in that they can transmit over 100 pathogens causing cholera, dysentery,

salmonellosis, typhoid, tuberculosis, poliomyelitis, viral hepatitis A & E, etc. They also transmit anthrax, parasitic worms, pyogenic cocci, *E. coli*, enteroviruses, etc.

#### 4.7.1 Stable flies

These flies (Stomoxys calcitrans) closely resemble houseflies except that they are smaller



Figure 13. Stable Fly © Shanghai Entry-Exit inspection and quarantine bureau, China.

in size, lighter in colour, have a wider spotted abdomen and mouth meant for biting (Figure 13). Both male and female feed on animal blood and get engorged in 2-5 minutes, which in milking animals can lead to anaemia and lower milk yields. As the name suggests the flies are found in abundance in the cattle sheds and cause tremendous annoyance to animals. The flies bite humans but the bites are painless and go unnoticed.

The male dies after mating, and the female dies after laying eggs. The maggots can be seen in rotting organic matter. Stable flies are known to carry *Trypanosoma evansi*, *Trypanosoma brucei*, anthrax, fowlpox virus, brucellosis, equine infectious anaemiavirus and African horse sickness.

#### 5. SURVEILLANCE AT POINTS OF ENTRY

#### 5.1. Identification of main issues at PoE

Programmes of surveillance and vector control are expected to vary considerably and hence have to be proportionate and adapted to the local context of each PoE. For this purpose, it is crucial to design appropriate monitoring and control measures at each PoE, which identify the local characteristics and critical issues regarding the risk of import and/or export of vectors. Information on vector-borne diseases in circulation or vectors' activity should be collected over the course of at least one year, particularly in territories with marked seasonality. The data collected should focus on:

- the description of the environment (natural and urban) of PoE and surrounding 400-metre perimeter or wider;
- local entomological situation;
- epidemiological context (endemic or potential health risks associated with invasive vectors).

As much as possible, data should be mapped to improve clarity, and facilitate the sharing of information with stakeholders. This first cartographic analysis will establish priorities for action.

#### 5.1.1 Description of the environment

The analysis should include a description of the environment around PoE (be it urban or rural) and the 400-metre perimeter. Such a description should include the following points.

a) Description of the area and its environment in relation to vector risks

Existing cartographic documents on which the initial description can rely are:

- site plan (delimitation of the closed zone, buildings, accesses, loading/unloading areas, cargo, warehouses, handling and maintenance, etc.);
- maps of sanitation and drainage systems, etc.
- maps of hydraulic structures (retention basins, pollution basins, storm-basin);
- map of area with vegetation (mowing plan, network of hedges, bushes, etc.);
- plans of buildings and hangars with specification of their assignment (e.g. transit of animals).

#### b) Activities and organization of PoE

The activities at PoE should identify the different flows from a qualitative and quantitative viewpoint in order to identify the risks at origin: the loading/unloading areas, storage areas, controlled areas requiring special access, etc.

It is recommended to collect:

- list of ground, air or maritime routes of origin (source: PoE authorities);
- passenger/wild life flows by origin (source: PoE authorities);
- the types and quantities of goods through the port of entry (source: customs);
- access to restricted area or information of contact person to get access (source: PoE authorities).

#### c) Description of the perimeter of 400 m around PoE

The following information has to be mapped, especially to determine the exact perimeter of the surrounding area, which can exceed 400 metres depending on the site typology and vector flight ranges. Each type of *land use includes different vulnerabilities with respect to disease vectors*.

- Urban area: Urban areas should be mapped, as they are particularly vulnerable mosquitoes breed in open containers found often in urban areas. Such areas could be divided into sectors in order to ascertain entomological indices (e.g. Breeding Index, Container Index, House Index, Breteau Index and Pupal Index) during monitoring activities. Defined areas could exceed the 400 m buffer zone to include a sufficient number of houses or to cover homogeneous blocks.
- **Commercial and industrial areas:** Inventorying the activities in these areas will facilitate the identification of which carry risks for disease vectors. It will also facilitate monitoring such areas and implementing recommendations (planning, waste management, etc.) that were issued. It is recommended to carry out an inventory of all businesses, companies, and stakeholders present within the perimeter of 400 m. The identification of contact persons can be helpful for activities that need to be monitored regularly.
- **Public domain:** Those areas in the public domain that could be reservoirs for disease vectors should be identified (rainwater drainage/harvesting systems, urban wasteland, cemeteries, parks and gardens, etc.).
- **Sensitive sites:** Sensitive sites (schools, kindergartens, retirement homes, nursing homes, etc.) should be identified for the implementation of vector control. A list of contact persons in these establishments could be helpful.

#### d) Identification of areas where insecticide treatments could be excluded

This will depend mainly on local regulations with respect to sensitive areas – e.g. aquatic harbourages, fountains, hives, catchment areas of drinking water, areas of ecological interest, etc. Here too, the peoplein charge of these sensitive areas and official contact persons should be identified.

#### 5.1.2 The local entomological situation

An inventory of species must be readied on the basis of existing bibliographic information as well as dedicated surveys (both larval and adult collection). The surveys will focus on PoE and the perimeter to 400 metres. However, the survey may be expanded to a radius of 1000 metres when justified, especially in case of *Anopheles* risk.

A finding of exotic mosquitoes already intercepted on site or nearby will indicate that control measures be given priority.

The different species listed should be described considering the following items:

- vector status (proven, potential)
- seasonal activity and dispersal ability
- preferred breeding sites (in order to facilitate the implementation of a control and monitoring programme).

#### 5.1.3 The epidemiological context

The main potential health risks linked to vectors relevant for the area where PoE is located should be identified and listed.

The health risks associated with the presence of vectors must be characterized. To do this, it is necessary to propose the epidemiological evidence for assessing the possibility of contact between a vector, a pathogen, and a host.

For this purpose, it is necessary to understand:

- the risk of introducing infected vectors regarding route of origins of ships and aircraft (risk of airport or port malaria for example);
- the risk of local transmission of imported pathogens by autochthonous vectors (competence and vectorial capacity of local vectors regarding these pathogens);
- the risk of exporting pathogens through dissemination of infected vectors (PoE is located in a malarious area or in an area where arboviruses are circulating according to an epidemic or endemic mode).

#### 5.1.4 Summary and conclusions about the receptivity and vulnerability of PoE

The analysis of the different elements of data assessed in previous sections must be made according to the conventional matrices for risk assessment and prevention in order to appreciate the context in terms of:

- Gravity/hazard: Is the area particularly vulnerable to major threats?
- Frequency/risk: Has the introduction of exotic vectors already been identified?
- How important are imported or autochthonous cases at the PoE or nearby?

- Risk management: Are such issues already included in general risk management documents available at PoE?
- Have preventive or curative actions already been implemented?
- Is the staff trained or sensitized to the problem?

Based on these elements, the objectives of the surveillance and monitoring programme will be identified, particularly in terms of the species of concern to focus on. Such identification will lead to the determination of the most appropriate methods to monitor (type of traps, breeding sites to sample, time of the day to perform surveillance, etc.) and control the species that pose a disease risk.

Programmes will also be adapted according to seasonal risks, if applicable. It should therefore be ascertained if the programme requires year-round or seasonal implementation.

The size of the area where the surveillance and control programme should be implemented needs to be determined. A perimeter of 400 metres will be sufficient for certain situations. However, for certain contexts (risk of malaria for example), this perimeter could be extended according to flight range of the vector and potential of the area to harbour breeding sites.

Note: The situation in and around PoE will continually evolve with respect to disease vectors, and seasonal variability may also influence disease transmission risk. Therefore, information about such risks should be continuously gathered and monitored. Situations that could lead to the emergence of a vector-borne disease have to be detected, monitored, and addressed promptly.

#### 5.2 Establishment of a surveillance plan

The surveillance plan will depend on the current epidemiological situation and risk of importation/exportation of vectors and their associated pathogens at PoE.

Under normal circumstances: Establish a routine surveillance plan.

In epidemics/outbreaks: Establish an emergency surveillance plan for rapid action.

In these circumstances, the following issues are essential:

- assessment of ecological conditions
- assessment of epidemiological risk
- adherence to local laws and regulations
- identification of surveillance target, areas and methods
- creation of surveillance plan.

Field operations: The following actions will assist in drawing up a surveillance plan:

- identification of target vector and extent of public health threat
- assessment of the extent of invasion, adaptation, breeding, and capacity to become established within local ecosystem
- geographical spread in the local environment
- collection of field samples
- preservation of samples
- transportation of samples to the laboratory

- identification of sample in local laboratory or transportation of sample to a designated referral laboratory within the country or overseas (or digital images sent for identification to a designated laboratory)
- follow-up plan for re-surveillance/recurrent surveillance and its frequency
- preparation of surveillance report
- discussion on surveillance report and initiation of routine control action in normal circumstances or rapid action/response in case of an emergency.

Entomological risk factors at PoE to consider include:

- 1. Number of invasive vector species of a particular disease at PoE
- 2. Breeding potential and population growth
- 3. Feeding preferences i.e. zoophilic versus anthropophilic
- 4. Conducive environmental parameters for vector growth
- 5. Inherent vector capacity
- 6. Dispersal: flight range, passive transportation of ectoparasites, etc.
- 7. Lack of vector surveillance and control.

Epidemiological risk factors at PoE to consider include:

- 1. Virulence of pathogens/parasite
- 2. Immune status of local population/reservoir (in case of zoonotic disease) and human hosts
- 3. Size of local reservoir of infection
- 4. Environmental factors conducive to disease transmission
- 5. Window of transmission
- 6. Amenability of vector(s) and pathogens to control measures/tools locally used.

#### 5.3 Vector-risk free zone

The International Health Regulations (2005) advocate achieving a vector-free zone at seaports, airports, and ground crossings and withina 400-metre perimeter around these entry points. The intent is to maintain vector free status through regular active surveillance and vector control so that transmission risk of pathogens imported with vectors/reservoirs could be nullified or minimized. This would also prevent local vectors from dispersal to distant lands via ships, aircraft, and rail/road conveyances and therefore prevent transmission of local vector-borne diseases from establishing in other countries. As discussed above, there is evidence that vectors have been introduced in distant lands via ships (e.g. *Aedes albopictus*), aircraft (e.g. Airport malaria in France), and conveyances (see Box 1). At every point of entry, effective vector monitoring can be achieved with the necessary infrastructure by a professionally organized surveillance and control programme.

Although the presence of a negligible number of invaded vectors may initially pose a limited real public health risk, experience has shown that in the long-term a few cases can spiral into an outbreak or epidemic/pandemic. Hence the aim of a routine surveillance and control programme is to achieve zero levels of exotic vector population by denying breeding opportunities as far as possible and taking timely and appropriate control action in order to stamp out exotic species. Keeping points of entry free of the breeding of native vector species is equally important to deny them access to conveyances and luggage/cargo in order to prevent their exit and spread to other countries.
#### Box 1. Review of Mosquito vector species intercepted at PoE in New Zealand since 1929

```
Number of interceptions: 171 (Since 1929)
Number of species identified: 27 (Including vectors Aedes aegypti, Aedes albopictus and
Culex annulirostris)
Disease-associated pathogens with invasive vectors (disease potential):
Arboviruses: Barmah Forest, chikungunya, dengue, Eastern equine encephalitis, Edge Hill,
GanGan, Japanese encephalitis, Kokobera, Kunjin, La Crosse encephalitis, Murray Valley
encephalitis, Rose River, Rift Valley Fever, Sindbis, Startford, Trubanaman, West Nile and
yellow fever
Parasites: Plasmodium spp., Dirofilaria immitis and Wuchereria bancrofti or Brugia malayi.
Number of countries of origin: 26
Place of origin of exotic species known: 152
South Pacific: 100 (66%) [Australia: 42 (28%) + 11 other countries: 58 (44%)]
Asia: 40 (26.3%) [Japan: 22 (14.5%) + 7 other countries: 18 (11.8%)]
Others: 12 (8%) [USA: 7 (4.6%)+ 5 other countries (3.4%)]
Mode of entry: Aircraft 94 (62%) and ship 57 (38%)
Total interceptions from 1989–2004: 62
Mode of entry: Ships 51 (82%) and Aircrafts: 11 (18%)
Main port of entry: Auckland (75)
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Source: Derraik JG (2004).

Country: New Zealand

## 5.4 Essential elements of vector surveillance

The following are essential elements of vector surveillance:

- professionally trained staff for laboratory and field services with required knowledge and skills;
- access to laboratory with infrastructure, equipment, and recurring requirements of chemicals, reagents, glassware, and plasticware;
- reorientation of field staff on methods of surveillance of mosquitoes (immature and adult), sandflies, rodents, etc. and also shipboard surveillance if necessary for mosquitoes, flies, sandflies, cockroaches, and rodents;
- standard operating procedures to be available for each type of surveillance methodology, piece of equipment used and time intervals, etc;
- effective personal protective equipment to be available at all times (Annex 1).

## 5.5 Mosquitoes: surveys of immature stages and adults

## 5.5.1 Eggs

It is difficult to institute surveillance against *Anopheles* eggs as they are laid singly over water surfaces where they remain buoyant due to lateral floats). Eggs of *Culex* species are easy to recognize from their rafts. *Aedes* eggs are laid singly in container habitats and appear shaped like a small black spindle and can be seen with a hand lens easily. Ovitraps in the form of 500 ml containers, which are painted black on the outside and filled with water (3/4capacity), can be placed randomly in an area where surveillance is intended. Within approximately one week, female *Aedes aegypti, Aedes albopictus* or any other container breeder *Aedes spp.* prevalent in the area will lay eggs in the placed ovitraps. Experience has shown that a flat 2x4 inch cardboard strip immersed in the ovitrap water and hay infusion will enhance attractiveness of the trap. These traps are useful for determination of vector populations and assessment of impact of interventions in a given area. It is suggested placing 10% more ovitraps than calculated as needed, to allow for loss of traps due to natural disturbance or unintentional removal.

Gravid traps can be deployed to collect gravid *Culex* mosquitoes and are powered by four D-cell batteries or 6V DC. The gravid trap contains a solution that is attractive to gravid female mosquitoes seeking an ovipositional site (Figure 17). This solution can be made by using a hay infusion or similar organic materials. The trap is portable and can be set wherever required.

## 5.5.2 Larvae

Surveys of immature mosquitoes are important aspects of an effective mosquito surveillance and control programme at PoE. They are used to determine the location, species, and population densities of native or/and introduced pest and vector mosquitoes. *Anopheles* larvae float parallel to the water's surface (Figure 14) while *Culex* and *Aedes* hang by piercing the water film with their siphon (Figures 15,16). With little experience and keen observation one can easily differentiate larvae of *Culex* from that of *Aedes* as the latter are longer and have whiplike movements. Larval surveillance is vital for predicting adult emergence and establishing optimal frequency of application of larval control measures. Larval surveillance is also helpful in forecasting the need for adult mosquito control, as well as to assess the effectiveness of control measures. Various indices are used to express results of larval monitoring. The most popular indices are the Breeding Index, House Index, Container Index, Breteau Index, and Pupal Index (Annex 8).



Figure 14. Anopheles Larva © Shanghai Entry-Exit inspection and quarantine bureau, China.



Figure 15. *Culex* Larva © Shanghai Entry-Exit inspection and quarantine bureau, China.



Figure 16. Aedes Larva © Shanghai Entry-Exit inspection and quarantine bureau, China.

#### Box 2. Purpose and scope of egg, larval and pupal sampling at PoE

- Egg and larval surveillance is vital in detecting any invasive vector species trying to establish in the local ecosystem.
- Surveillance provides first-hand information on habitat preferences, which allows prioritizing control in the habitats preferred by vector or pest species.
- Routine egg and larval surveillance provides a more complete and accurate picture of sources of mosquito breeding spatially and temporally, thereby elucidating the extent of mosquito production as a basis for treatment in space and time.
- Egg and larval surveillance allows for continuous evaluation of insecticide application and control measures.
- Egg and larval surveillance provides understanding of species distribution, density, seasonal occurrence, and abundance.
- It supplements the knowledge gained from adult mosquito surveillance (e.g., light traps, bite counts, landing rates, etc.).
- Insecticide resistance can also be monitored through a larval surveillance programme.

#### 5.5.2.1 Components of a survey kit for sampling of immature mosquitoes

- 1. A Canvas collection bag of sufficiently large size
- 2. Dipper in the form of a bowl-shaped ladle (300 ml capacity)
- 3. Sieve/strainer
- 4. Screw-cap plastic containers
- 5. Glass pipettes (long and short) fitted with rubber bulbs
- 6. Hand magnifier lens
- 7. Torch and batteries
- 8. Forceps/tweezers
- 9. Field notebook and pen
- 9. Soap
- 10. Small towel
- 11. Mosquito repellent lotion or cream
- 12. Sticker paper labels for labelling containers

One field kit may be provided to a team comprising a worker and a supervisor.

#### 5.5.2.2 Larval sampling technique

Different sampling techniques are used in different types of breeding habitats (<u>http://www.who.</u> <u>int/malaria/publications/atoz/9789241505819/en/</u>). Anopheles, Culex and Aedes larvae display unique behaviour. While Anopheline larvae lie parallel to the surface of water in order to breathe through spiracles, Culex larvae hang from the water surface with their respiratory siphon piercing the water film. Aedes larvae hang like Culex larvae but are unique as they prefer to stay in 'schools' with fellow larvae and show whipping sideway movements. These basic facts are known to most of the field entomologists and workers engaged in routine larval surveillance. Mosquito species show marked preferences for habitats for breeding and this knowledge can be systematically exploited for sampling of immatures and organizing species-specific control. For example if there is suspicion or confirmation of invasion of an *Anopheles* species at any point of entry, one should look for clean water habitats (either natural or man-made) for immatures. On the other hand, *Aedes* larvae can be found in stagnant water in container habitats, scrap, cement mixers or tyres and such similar habitats. *Culex* immatures will most likely be found in organically rich waters.

Approach larval habitats with care, slowly and facing the sun to prevent your shadow over the breeding site or else the larvae will dive to the bottom and may take some time before ascending to the surface again. Some species like *Anopheles stephensi* can remain submerged for a long time. If water surface is disturbed by heavy winds, the windward side should be preferred for sampling. In ponds, pools, and lakes mosquito immatures will be found close to edges, mostly near floating vegetation especially in the clear pockets created in floating vegetation or grasses and algal mats where small predators of larvae find it hard to reach. Visual assessment of larval presence can be made before dipping. Larvae will be most likely found adjacent to floating twigs or logs where sampling can be most rewarding.

There exists a variety of sampling methods including skimming for Anopheline larvae in shallow water with a bowl, or partial or complete submersion of sampling device or using dipper as background to spot larvae and/or pupae and then collecting them. The most commonly used technique is simple scooping to collect larvae in the dipper. Siphons can be used for sampling larvae from tree holes, leaf axils or containers and tyres. The other sampling devices used for sampling larvae are well nets and strainers.

Breeding sites	Resting places
Ponds	Human dwellings/restrooms
Puddles	Sheds
Ditches	Indoor hanging objects
Surface drains	Crevices
Grassy and marshy land	Bushes/vegetation – wild and in gardens
Pits and depressions	Underneath furniture
Scrap- and water-containing depressions in sheeting	Curtains
Containers of different varieties and shapes	Underneath or on the sides of tanks
Water chambers	Cargo boxes/holds
Hydrants	Work stations
Tyres including fenders	Walls of buildings and underneath roofs
Ground and overhead water tanks	Open luggage/cargo boxes or containers
Septic tanks	Cartons and containers
Terraces/roof tops/lintels	Scrapped crafts, vehicles, vessels, etc.
Curing waters in construction/development sites	Tyre dumps
Wells	Abandoned structures/buildings
Iron ore loader buckets	Tree holes

# Table 10. Potential breeding and resting places of mosquitoes at ports, airports,ground crossings and in the periphery

The following is the step-by-step approach of larval surveys and identification of mosquitoes at the harbour area of ports and at airport platforms, hangers, baggage, cargo areas, and at ground crossings.

Step 1: Detect and map all potential breeding sites of vectors at the point of entry and surrounding 400-metre perimeter where human habitation occurs or the port is established.

Step 2: Take photographs of the breeding sites.

Step 3: Make an inventory of potential breeding sites of vectors/mosquitoes and update it seasonally and temporally. Categorize them into temporary and permanent breeding sites.

Step 4: Organize larval surveys using appropriate sampling methods (Annex 4).

Step 5: Collect larvae (also pupae if found in the same habitat) in screw-cap plastic containers along with water from the larval habitat and bring samples to the insectary for rearing and emergence. Fill the container 3/4 with water and secure the cap tightly. Label each container, mentioning breeding site, its specific location, and date of collection of sample. Note that during transportation of sample in a vehicle, jerks must be avoided as these can result in mechanical injury and mortality in larvae.

Step 6: Gently pour out water along with immature stages of mosquitoes into a 300 ml bowl or basin or a plastic/enamel tray.

Step 7: Mix finely ground powdered yeast and ground dog biscuits in a proportion of 60:40 as larval feed daily. Baby formula food along with fish meal flakes ground to powder and mixed in equal proportion may also be used.

Step 8: Cover the bowl with muslin cloth secured outside on the brim of the bowl with a rubber band.

Step 9: Segregate pupae from larvae and keep them separately in similarly secured bowls and wait till the emergence of adults.

Step 10: Remove adults carefully from the bowl by inserting the plastic tube of a hand-held standard mouth aspirator through the hole in the netting or use a motorized aspirator. As far as possible use a mouth aspirator, which will avoid injury to adults that may occur when using a motorized aspirator. Transfer the adults gently into a test tube and close the tube using a cotton plug to prevent mosquitoes from escaping.

Step 11: Anaesthetize the adults using a few drops of ether and wait till the mosquitoes' are knocked down and their movements stop completely.

Step 12: Identify knocked-down adults under a dissecting binocular microscope using standard morphological keys, transfer them to a designated referral laboratory, or retain the specimen for PCR-based molecular identification if local capability exists.

# 5.6 Adult surveillance

Before planning collection of adults some important considerations include the following.

1. A list of native species at PoE and surrounding areas must be available.

2. It must be appreciated that mosquito populations build up in relation to seasonal weather conditions. Hence when monitored at regular intervals, natural variations in the numbers of the collected adults are bound to occur.

3. Different mosquito species may have different biting rhythms, host choices, and resting preferences. Hence for adult collections, one must take these factors into consideration during the planning stage.

4. When an invasive/exotic species has been collected, recommended procedures may be followed and the identity of the species should be confirmed by sending it to the referral laboratory identified for the purpose (Annex 4). More collections may be organized in the area if necessary. Prompt control action for risk reduction/elimination must be prioritized to deny opportunity to the vector for disease transmission and establishment in and around the point of entry. In case of the occurrence of adverse events such as outbreak, traps may be preferred over other methods of adult collection for the monitoring of impact assessment. Identity of pathogens/parasites may be established either by their identification in local laboratory (if facilities exist) or by sending the sample to a designated regional or global referral laboratory.

## 5.6.1 Material required for adult catches of mosquitoes

The following is needed to catch adult mosquitoes.

- 1. Field bag of a sufficiently large size
- 2. Hand held and/or motorized aspirator and adult traps (light traps with or without carbon dioxide, CDC, UV, or BG-Sentinel traps) [Figure 17]
- 3. Test tubes
- 4. Paper sticker labels for labelling test tubes containing adult collection
- 5. Cotton roll
- 6. Torch and batteries
- 7. Forceps (one blunt and one sharp)
- 8. White cloth sheets for space spray
- 9. Knockdown atomized insecticide can
- 10. Field notebook and pen
- 11. Soap
- 12. Small towel
- 13. Protective gear

The frequency of adult collection should be at least monthly in vulnerable areas or more frequently (weekly/fortnightly) in areas with weather conditions that allow for a greater probability of vector survival and establishment.



Figure 17. Gravid trap for adult *Culex* © Shanghai Entry-Exit inspection and quarantine bureau, China.

# 5.6.2 Epidemiological considerations to initiate control measures for adult mosquitoes

The following epidemiological points should be considered when initiating control measures of adult mosquito populations.

- Interpret seasonal and temporal data on abundance of key native species for risk assessment.
- Use mapping to study whether the vector/mosquito distribution is patchy or random and its epidemiological significance.
- Identify risk/vulnerable areas at PoE and surroundings to monitor vectors. Monitoring can be expanded beyond the recommended 400-metre perimeter, when invasion by *Anopheles* species is suspected and also depending upon its flight range.
- Characterize the health risks associated with the presence of vectors, pathogen/parasite, and host. Determine whether ships from endemic areas call on particular berths of the port. Similarly determine the timings of aircraft coming from disease endemic/ epidemic countries where viruses/malaria parasites are currently in circulation. This will enable selective monitoring for risk assessment in vulnerable areas.
- Delineate risk of local transmission from an imported vector.
- Assess risk of exporting pathogen via a local vector, based on local epidemiological situation, whether endemic or epidemic.
- Where insecticide use for mosquito control could harm predators, pollinators or other economic species such as silkworms, such areas/pockets may be excluded from vector control according to government policy.

Based on these factors surveillance, monitoring, and control of mosquitoes may be organized, in addition to the creation of a risk management strategy.

# 5.7 Rodents: surveillance of rats and mice

Rodents have been implicated in transmitting plague, Leptospirosis, murine typhus, and food-borne Salmonellosis. Rodent surveillance is essential to identify species, extent of their infestations, and location for initiating control measures. By their behaviour and activity pattern, rodents lend themselves to tracking, surveillance, and control.

#### 5.7.1 Runways and rub marks

Routes frequented by rodents are called runways. When rats repeatedly travel over the same passage, they leave behind marks often discoloured that identify runways (Figure 18). These runways are created when oil from rodent hair rubs against a surface and over time this mark turns dark in colour; it is called a scuffmark. Runways may not be readily visible as they may be made inside pipes or casings where they must be followed to trace the openings of the tunnels. The roof rat will leave these marks over overhead wires or pipes, casings, air ducts, iron rods, electric cables or the top of the sheathing, etc.



Figure 18. Rub marks of rats © Shanghai Entry-Exit inspection and quarantine bureau, China.

#### 5.7.2 Tracks

Hold a flashlight at an angle to the deck to observe tracks in the dust or talcum powder used for the rodent footprints. The tracks will cast a distinct shadow. Fresh tracks will appear sharp and distinct (Figure 19), whereas old tracks may be less distinct, because of dust accumulation. The five-toed tracks of the rear paws are more commonly observed than the four-toed front paws, yet both may be present. It is very useful to spread a thin band of talcum powder along runways to check for rodent direction and the amount of recent activity.



Figure 19. Fresh track of a rat © Shanghai Entry-Exit inspection and quarantine bureau, China.



#### 5.7.3 Gnawing

Rodents gnaw to keep their incisors sharp. They gnaw on wood, plastics pipes/containers, cement, or metal surfaces (Figure 20). This leaves behind gnaw markings on the surface and fine material scrapings, which is an indicator of rat infestation. As the rat rubs its body against the gnawed surface it gradually becomes smooth.

Figure 20. Gnawing

#### 5.7.4 Droppings

Fresh faecal droppings appear soft, shiny, and dark and vary in shape and size, depending on the species (Figure 21). After a few days, droppings may appear dry and hard. Old droppings appear dull and grey in colour and easily crumble when pressed with a stick. Droppings are usually abundant near the food source, but they may also be found along runways.



Figure 21. Rat droppings; Norway rat blunt ¾", roof rat pointed ½", house mouse pointed ¼" © Shanghai Entry-Exit inspection and quarantine bureau, China.

#### 5.7.5 Urine

Rodents cannot regulate or control their urine output, so they constantly urinate. To search for rodent signs along runways, hold a black (UV) light at an angle to the deck. Fresh rodent urine fluoresces a lime green colour. Old rodent urine appears bluish-white (Figures 22, 23.)

#### 5.7.6 Rodent hair

Rodent hair, particularly rat hair, would appear bluish-white in colour when seen under a black light.



Figure 22. Shipboard inspection for rodent urine with UV light © Tianxiao Chen, Shenzhen Entry-Exit inspection and quarantine bureau, China.



Figure 23. Urine spotted with UV light © Tianxiao Chen, Shenzhen Entry-Exit inspection and quarantine bureau, China

#### 5.7.7 Techniques for surveillance of rodents

The following surveillance techniques will be useful to check for the presence of rodents and to assess the impact of control measures.

#### 5.7.8 Traps

Traps specifically designed for rodents must be used for surveillance. A distinction must be made between a mouse and rat trap. As rats are usually bigger than mice, they can often escape from a mouse trap. Likewise, smaller mice may not be able to set off a rat trap. Traps should be durable and of standard quality. They should not rust easily and should be sensitive to the touch of the rodent it is designed for. Professional snap-traps, being portable and effective, are generally recommended.

#### 5.7.8.1 Trap usage and care

Below are considerations when using traps.

- The end of the trigger arm of new traps often needs filing to make it smooth for maximum sensitivity.
- Traps should be stored indoors when not in use to prevent rusting of spring.
- All metal parts should be treated with fish oil or molten wax to reduce rusting before and after use. Anti rust sprays (e.g. CRC) should not be used as their smell may deter rodents.
- Secure the trap firmly with a wire, vegetation, or other suitable object so it is not dragged away by injured rats, large mice, or scavenging predators attracted by the rodent.

## 5.7.8.2 Trap deployment

- Rodents frequent areas where there are food grains, leftover food logs, vegetation, or food scraps. If rat droppings, tracks, or rub marks are visible, deploy trap there.
- Traps should be deployed near the platform where ships anchor. Note that rats tend to move down the anchoring ropes unless rat guards are used.

- The number of traps required depends upon the area to be covered, the number of skilled personnel handling the traps, and trap availability.
- Rodents, particularly Norway rats, are very sensitive to human odour or perfume. Traps should be placed firmly on the ground and efforts made to prevent frightening the rats away.
- If the site is infested by more than one species, different traps may be deployed proportionately (Figure 24). The captured rats should be examined for fleas etc. (Figure 25).
- Deploy two traps together in one area by keeping them a little distance from each other.
- Since rodents prefer to travel alongside walls, traps should be placed there if present.



Figure 24. Rat trap being deployed to collect fleas for plague surveillance © Chunzhong Zhao, Shenzhen Entry-Exit inspection and quarantine bureau, China.



Figure 25. Trapped rat being combed for flea collection © Chunzhong Zhao, Shenzhen Entry-Exit inspection and guarantine bureau, China.

#### 5.7.8.3 Trap covers

- Trap covers ensure that the rodent approaches the trap from the best side and gets caught.
- Covers protect traps from disturbance by weather or other animals, and help reduce the possibility of catching non-target animals.
- Covers can be made of whatever material is available and portable (e.g. wire mesh, clear plastic sheet, or plastic drain pipe).
- Traps should always have a cover (tunnel).
- The black plastic tunnels for the Black Trakka™ tracking pads are also suitable for covering snap-traps.
- Use stones, sticks, or wire pegs to hold covers in place. Bent wire hoops, or similar, should be placed across the entrances to exclude non-target animals.
- If covers are used, both traps may be set back-to-back under the one cover. A wire hoop or forked stick placed in the space between them helps prevent one setting off the other.
- Ensure the cover does not impede the action of the traps. To test this, set the selected trap off when it is within the selected cover. If the arm of the trap hits the cover at any point, the cover needs to be larger.

#### 5.7.8.4 Baiting of traps

- Always use the same type of bait for consistency in surveillance.
- A stiff mixture of peanut butter and rolled oats is recommended as a reliable standard bait for index trapping; it lasts well. Other suitable baits include roasted coconut, cheese, nuts, chocolate, bacon, or leather soaked in fish oil.
- Renew the bait whenever its attractiveness has been reduced by rain, hot weather, mould, or partial consumption by ants or other insects.

#### 5.7.8.5 Index trapping

- The Abundance index provides useful information on changes in rodent abundance in the same location between seasons.
- The more trapping that is is done the more likely you are to trap more individuals. For example, take two locations (site 1 and site 2) with the same abundance of populations. If at site 1 you deploy 50 traps and at site 2 you deploy 100 traps, with all else constant you would expect to trap twice as many individuals at site 2 than at site 1. The Abundance Index is a measure of the number of individuals captured adjusted by the number of traps deployed.

An example to calculate the Rodent Abundance Index is given below.

## No. of traps deployed =100 No. of nights = 3 Total trap Nights = 100 × 3 or 300 Suppose 14 rats are trapped and 36 traps are sprung or empty Trap nights lost: = $\frac{1}{2}$ (captures + sprung, empty traps) = 14 (Rats trapped) +36 (traps sprung with no catches) = $\frac{1}{2} \times 50$ = 25 Therefore the corrected number of trap nights is: = Total no. of nights – trap nights lost = 300–25 = 275

- **Index of abundance** = captures x 100 / corrected trap nights
- = 14×100/ 275

#### = 5.09 captures per 100 trap nights

Source: Seniloli E and Rasalato S. Bird Life International Pacific Secretariat. Unpublished information: Post eradication monitoring report (Ringgold Islands), 2009.

#### 5.7.8.6 Method of index trapping

- Always use the same brand of trap for index trapping (otherwise differences in their effectiveness will bias your results making them incomparable).
- An index line should ideally have at least 25 sites spaced evenly apart. Each site should have two traps within the same cover.
- Index lines are usually run for three nights to capture night-to-night variability and rodent activity.
- Plan index trapping so as to give a minimum of 100 corrected trap-nights in each habitat.

For example 50 traps for three nights gives a maximum of 150 trap-nights.

- The spacing between sites should be as large as possible within the range of 25 and 50 metres. Use measuring tape if necessary. For recurrent rodent surveillance, permanent trapping index lines should be accurately measured and trap sites marked.
- To maintain consistency during index trapping, covers should either be present at every site or be absent from every site. Experience shows that trap covers may influence trapping success so they should be left in place between trapping sessions to accustom the animals to their presence.

#### 5.7.8.6.1 Census by trapping

A rodent is trapped alive by using live traps such as a Wonder trap or Sherman trap. The rat is marked by toe clipping and released. In about a week or so the rat is captured again. The Lincoln Index is calculated as below:

 $N = \frac{Mn}{m}$ 

Where:

N= Population size M= No. of rodents trapped for the first time N= No. of rodents trapped for the second time M= No. of rodents re-trapped (marked) in second trapping

#### 5.7.8.6.2 Live burrow counting method

This is one of the most reliable methods of rodent population estimation as it is based on direct observation of live burrows of the rodents in a given area. The method involves closing all the burrows on day 1, followed by re-closing all the active (open) burrows on day 2 with pre-bait mixture and finally counting the actual number of opened live burrows on day 3. This gives a rough idea of rodent population in an area. However the limitation of this method is that the number of rodents living in one burrow cannot be ascertained.

## 5.7.9 Tracking tunnels

An alternative to rodent traps is tracking tunnels – rectangular cardboard boxes with tasty baits such as roasted coconut scraping, chocolate, peanut butter, or fish – which lure small animals including rodents (Gillies and Williams 2013). These animals can be tracked from the footprints they leave behind. The footprints of an invasive rodent can be readily distinguished by an experienced surveyor. There are many advantages to using tracking tunnels:

- They are much more effective devices to track rodents than live burrow counting method.
- They can be placed over many nights if necessary.
- They do not kill the rodent and hence do not scare other animals from approaching the tunnel.
- They are easily deployed and maintained.
- They do not harm non-target species.

The biggest disadvantage is that if invasive rodents have been identified during surveillance then they have to be killed using aggressive control methods, which the tracking tunnel does not assist with.

#### 5.7.9.1 Placing the tracking tunnel

Tunnels can be placed at a distance of 50 metres apart for large rodents along a transect line of a grid (which makes it easier to locate and monitor tracking tunnels), or 10-25 metres in case of mice. Tracking tunnels can be made easily but ready-made Black Trakka™ tunnels are more effective and will allow comparable results from the tracking. Footprints can be identified with the help of information given at <u>http://www.doc.govt.nz/Documents/science-and-technical/inventory-monitoring/im-toolbox-animal-pests-using-tracking-tunnels-to-monitor-rodents-and-mustelids.pdf</u>

## 5.8 Flea surveillance

Fleas are the primary vectors of plague; knowledge of local flea species and their hosts is essential for estimating risks of human plague infection and designing specific control measures appropriate for local situations. The relative importance of local flea species as plaque vectors can usually be determined by analysing relevant surveillance data, including the numbers of fleas per host, host preferences, and Y. pestis infection rates for the species of fleas collected. Future surveillance efforts can then concentrate on important vectors and their hosts, thereby reducing costs while providing the most relevant information for control efforts. Host/flea data also provide indirect clues about which mammalian hosts are involved in local epizootics. For example, mortality among rock squirrels (Spermophilus variegatus) is high during plaque epizootics, and it is not unusual at these times to find their usual flea parasite Oropsylla montana (Diamanus montanus) on other hosts such as other sciurids, rabbits, mice, or wood rats. The number of fleas per host is also important. An increase in the average number of fleas per host may be of little concern when the flea species is a poor vector of plague. However, when the species is a disease vector (e.g. Xenopylla cheopis) and their numbers on Rattus species increase above a certain level, it may be necessary to initiate control measures to decrease the risk of human cases and plague epizootics (WHO 2009b).

## 5.8.1 Flea Index

Rat fleas are ectoparasites that are found on the neck, below the neck, and on the back of the rodent. Capture the rodents live by setting up Wonder or Sherman traps. Collect all the traps next morning. Bring the traps in bags to the laboratory and anaesthetize the rats with ether. Use gloves at all times while handling rats. Comb the live rodent and collect the fleas in a white enamel basin by suction tube. Look for free fleas in the rodent bag and collect them as well. There are over 1 500 species of fleas. It is expected that a key to the identification of local flea species and main global species will be available at PoE to facilitate identification of native and invasive flea species. Calculate the Flea Index by using following formula.

Absolute Flea Index =

Total number of fleas collected

Total number of rats collected

In other words absolute flea count is the average number of fleas collected per rodent captured.

Total number of fleas of a particular species collected
Specific Flea Index =

Total number of rats collected

In other words specific flea count is the average number of specific fleas collected per rodent captured.

## 5.9 Sandflies surveillance

For sampling populations of sandflies human landing collections (HLCs), sticky castor oil traps, CDC simple un-baited, incandescent or UV light traps (Figures 26 and 27), with or without  $Co_2$  have been used. Although baited HLCs are far more effective than other methods and have the obvious advantage of catching live sandflies, the main disadvantage is the exposure to risk of infection of leishmaniasis to the human bait and the collector, hence a serious ethical issue. The success of HLC also depends on the skill and alertness of the collector. The relative attractiveness of different human baits may also influence collection outcome. On the other hand, the sticky traps, unlighted and un-baited traps collect sandflies from their immediate vicinity, while the CDC incandescent and UV traps can attract flies from a distance, attraction being enhanced when  $Co_2$  is combined with UV light (Kline et al. 2011).



Figure 26. Sandfly CDC incandescent light trap © Shanghai Entry-Exit inspection and quarantine bureau, China.



Figure 27. Sandfly CDC UV light trap © Shanghai Entry-Exit inspection and guarantine bureau, China.

# 6.1 Principle and purpose

Vector control at PoE assumes great importance in order to prevent invasive species form establishing in local environs and also to prevent exportation of local vector species to other countries by land (through lorries and trains), by air (through aircraft) and water (via ships at ports). In the past there have been many instances where vectors have spread to other countries and not only survived alien weather conditions but also transmitted diseases. A case in point is airport malaria in France, Morocco, and Tunisia to name a few countries. Another example is the geographical spread of *Aedes albopicta* to Mediterranean countries in the past decade.

#### Integrated vector management

Integrated vector management (IVM) is defined as «a rational decision-making process for the optimal use of resources for vector control» and includes five key elements: 1) evidence-based decision-making; 2) integrated approaches 3); collaboration within the health sector and with other sectors; 4) advocacy, social mobilization, and legislation; and 5) capacity-building (Beier et al. 2008).

According to WHO, IVM is a dynamic and still-evolving field. IVM strategies are designed to achieve the greatest disease-control benefit in the most cost-effective manner, while minimizing negative impacts on ecosystems (e.g. depletion of biodiversity) and adverse side-effects on public health. Possible health risks range from acute exposures to pesticides and their residues to bio-accumulation of toxic chemicals, as well as the development of vector resistance to some widely-used pesticides and drugs. More about IVM can be found at: http://www.who.int/heli/risks/vectors/malariacontrol/en/.

A new WHO global strategic framework for integrated vector management defines IVM as a strategy to improve the efficacy, cost-effectiveness, ecological soundness and sustainability of disease vector control. IVM encourages a multi-disease control approach, integration with other disease control measures, and the considered and systematic application of a range of interventions, often in combination and synergistically. In 2004, WHO adopted IVM globally for the control of all vector-borne diseases.

## 6.1.1 Mosquitoes

IVM is a key to the control of vectors at points of entry. Various well-tested control measures in order of priority are the following.

- 1. Environmental management:
  - source reduction
  - habitat modification/manipulation.
- 2. Mechanical control:
  - window and door screening
  - drilling holes in fenders for drainage
  - removal and safe storage of scrap.

#### 3. Biological control:

- larvivorous fish
- biological larvicides.
- 4. Chemical control/insecticide treatment:
  - larvicidal application
  - adult control
    - fogging
    - indoor spraying
    - insecticide treated material/special circumstances
    - repellents.

## 6.1.2 Making mosquito control choices

The choice of which vector control to employ would depend upon a situation analysis at the point of entry, ground crossing, or conveyance. Biological control agents should be preferred over insecticidal methods for being safer and more cost-effective over time. In outbreak/emergency situations however, drastic and immediate action might be necessary to stamp out an invader species or stop an outbreak in conjunction with other disease control methods. For example, thermal/cold fogging might be preferred in an outbreak situation or where evidence of vector invasion exists and where quick knockdown is intended.

## 6.1.3 Preparedness for mosquito control

Basic preparedness and infrastructure for vector control includes the following:

- Sufficient numbers of trained laboratory and field human resources
- Resources for procurement of biolarvicides/chemical insecticide, spray, and personal protection equipment
- Support of the engineering department/contractors for implementing physical, mechanical, and environmental control measures
- Routine surveillance plan that provides evidence for action and timely decisions on the choice of control method
- Larval and adult bio-assay kits
- Linkage with referral laboratory
- Impact assessment plan.

Note: The above points are self-explanatory. Vector control is to be carried out by a professional agency or staff.

# 6.2 Larval control

## 6.2.1 Reduction of vectors

With the support of civil engineers at the point of entry, measures such as dredging ditches, filling up pits and clearing accumulated water should be undertaken to permanently eliminate habitats of vectors.

Environmental sanitation should be improved at PoE, including measures such as regularly removing rubbish and faeces, properly arranging rubbish bins with covers and their routine clearance. There should be flush toilets instead of dry pail latrines in ports. Septic tanks should be threechamber style and have sealed lids to prevent leakage and to eliminate fly breeding sites.

#### 6.2.2 Environmental management

Environmental measures against vectors have proven their usefulness over time; these methods are often long lasting, safe, and cost-effective in the long-term. These include source reduction, habitat modification, and manipulation using minor engineering methods so that breeding opportunities to vectors are denied. Being non-insecticidal, these methods are safe for fragile ecosystems near seaports. These methods can be implemented at PoE through engineering and maintenance departments/agencies as part of their routine maintenance services. Alternatively, a special drive for vector control may be undertaken from time to time.

#### 6.2.2.1 Scrap

Scrap generated at the port commercial area and residential colonies provide a range of breeding habitats to vectors especially during rainy seasons (Figure 28). Scrap disposal from PoE must be efficiently organized and done on a regular basis. In the intermittent period scrap items which can hold water such as safety helmets, sanitary flush tanks, troughs, fibreglass containers, buckets, barrels, drums, etc. must be stored upside down (Figure 29).



Figure 28. Scrap at PoE can provide a foothold to mosquitoes, especially *Aedes* mosquitoes © Ashwani Kumar



Figure 29. Scrap awaiting disposal should be kept facing the ground © Ashwani Kumar

#### 6.2.2.2 Tyres

Tyres are notorious for providing safe niches for *Aedes* to breed in. Tyres have been implicated in shipping *Aedes albopictus* from Japan to USA where it is completely entrenched. When kept disorderly or even in rows, tyres are difficult to treat individually with insecticides (Figure 30).



Figure 30. Tyres accumulate water in rains and are preferred sites of *Aedes* breeding (*Anopheles* species may occasionally breed there as well) © Ashwani Kumar



Figure 31. Tyres stacked vertically covered with tarpaulin and flat sheet to prevent water stagnation on the sheet during rains © Ashwani Kumar

Tyres should be either stored indoors in dry condition or stacked vertically and the top of each such stack should be covered with plastic sheeting above which a wooden/metal flat sheet should be kept to prevent water from entering (Figure 31). Tyres are also tied along jetties as fenders to prevent vessel damage (Figure 32). These tyres should have holes drilled at 3, 6, 9 and 12 o'clock to prevent water accumulation and breeding of mosquitos, especially of *Aedes* vectors (Figure 33). The holes have to be large enough to prevent water clogging.



Figure 32. Tyres abutting jetty when filled with rain water can be extremely dangerous in providing the first foothold to any mosquito arriving with a ship © Ashwani Kumar



Figure 33. Holes cut in the tyres should be at 3, 6, 9 and 12 o'clock to prevent waterstagnation © Ashwani Kumar

#### 6.2.2.3 Mineral loader buckets

Large mineral loading buckets which lie disused accumulate water and support vector breeding during rains (Figure 34). These buckets can be stored safely in an inverted manner (Figure 35).



Figure 34. Mineral ore loader buckets with water accumulation © Ashwani Kumar



Figure 35. Mineral ore loader buckets stored inverted © Ashwani Kumar

#### 6.2.2.4 Ground tanks

Water tanks kept open near construction sites can pose health risks by allowing mosquito breeding (Figure 36). These must be kept closed at all times and stored upside down when disused (Figure 37). Partially open overhead tanks should also be kept closed with air-tight lids, and overflow pipes should be covered with gauze at all times to prevent mosquito entry (Figure 43).



Figure 36. Open ground water tank near a construction site © Ashwani Kumar



Figure 37. Disconnected overhead tank should be kept inverted © Ashwani Kumar

#### 6.2.2.5 Surface drains

Surface drains should have smooth margins to ensure smooth flow of water as wavy margins can create pockets where culicine mosquitoes could breed (Figures 38 and 39).



Figure 38. The wavy margins of the drain and floating plastic bags have created pockets where *Culex* species breed @ Ashwani Kumar



Figure 39. Lined margins of a surface drain ensure free flow of water © Ashwani Kumar

#### 6.2.2.6 Mosquito-proof service chambers

Mosquito-proof lid assemblies should be installed on service chambers at all locations of ports and airports (Figure 40).



Figure 40. When closed this mosquito-proof lid assembly fits tightly on the margins of the service chambers © Ashwani Kumar



Figure 41. Sealed septic tanks prevent culicine breeding especially of *Culex quinquefasciatus* © Ashwani Kumar

**Septic tanks:** Septic tanks can support heavy breeding of nuisance mosquitoes and the filarial vector *Culex quinquefasciatus*. Sealing these tanks ensures that mosquitoes cannot access them to breed (Figure 41). Vent pipes of septic tanks should be covered with nylon mesh as well (Figure 42).



Figure 42. Distal end of vent pipe covered with nylon mesh prevents mosquito entry © Ashwani Kumar



Figure 43. Vent pipe of an overhead tank covered with nylon mesh © Ashwani Kumar

**Mosquito-proof lid of overhead tank:** Overflow pipe of tank should be covered with nylon mesh (Figure 43). A wrought iron mosquito-proof lid assembly of overhead tank prevents mosquito entry. The circular lid rests outside the rim when closed (Figure 44).



Figure 44. Mosquito-proof lid assembly of overhead tank © Ashwani Kumar



Figure 45. Wrought iron hose pipes may support vector breeding at PoE © Ashwani Kumar

#### Other situations at PoE conducive to mosquito breeding:

**Hose pipes:** Hose-pipe connectors tend to accumulate water following rain and support breeding of mosquitoes. These should be stored indoors or kept vertical to prevent water stagnation (Figure 45).

**Iron platforms:** At ports iron float platforms collect water in the rainy season (Figure 46). These require safe storage.



Figure 46. Iron platform with water © Ashwani Kumar



Figure 47. Blocked terrace with water © Ashwani Kumar

**Blocked terrace:** When terrace drain pipes are blocked and water stagnates, ideal breeding opportunities are offered to mosquitoes (Figure 47).

**New development activities at PoE:** Construction works around PoE should be carefully monitored and adequate measures taken to prevent mosquito breeding (Figures 48 and 49). Areas that could hold water and allow it to stagnate could provide ideal breeding opportunities to exotic vectors, allowing them a foothold.



Figure 48. Construction site at PoE with an enormous potential for mosquito breeding © Ashwani Kumar



Figure 49. Water stagnation at new development site at a port © Ashwani Kumar

## 6.2.3 Biological control

Biological control agents (Bti or fish) can be introduced to affect reproduction, growth, and activity of vectors or change the transmission dynamics of vector-borne diseases on condition that they do not harm the environment or the ecosystem.

#### 6.2.3.1 Biological larvicides

Biological larvicides formulated as wettable powders, granules and briquettes of *Bacillus thuringiensis israelensis* are commercially available. Wettable powders are water suspensions and can be applied using knapsack sprayers while granules can be dispensed manually.

#### 6.2.3.2 Larvivorous fish

Gambusia affinis and Lebistes reticulatus are two widely used species of larvivorous fish in vector control. They are small surface feeding fish that self propagate (breeding ever 2-3 months), giving rise to juvenile fish (being ovo-viviparous). They actively and preferentially devour mosquito larvae in their feeding zone. There are many native species of top minnows distributed all over the globe, which can be locally evaluated and used for vector control.

## 6.2.4 Chemical insecticides

Liquid formulation of organophosphates (OP) compound is usually effective against *Anopheles* immatures. It can also provide effective control of *Culex* and *Aedes* species when sprayed with knapsack or compression pump @1ppm at an interval of two weeks, or to be used as recommended on the product label.

## 6.3 Adult control

Residual chemical insecticides can be used depending upon the target species, susceptibility to insecticide used, and amenability to control and residual efficacy. It is important to plan spray operations carefully, informing the communities in advance. The public should be well informed about the goal of the insecticide spraying and necessary precautions during and after spraying operations.

Barrier residual sprays may be practical to prevent vector invasion at land borders. Depending upon the insecticide used and weather conditions, either two or three rounds of spray may be required to provide protection during the most sensitive seasons particularly in tropical countries. In addition, sheeting impregnated with residual insecticide can be affixed to interior walls to bring about effective control of mosquitoes, bed bugs, cockroaches, houseflies, and sandflies. Insecticide-treated bed nets can be advised especially in the break rooms of port workers and the residential areas within a 400-metre perimeter of the point of entry. Insecticide-treated curtains can be advised in buildings and offices at the port/airport and residential buildings in the periphery (within the 400-metre perimeter).

Thermal (hot) or cold fogging can be effective in achieving quick knockdown of both local and exotic vectors at PoE. Equipment and insecticides for thermal and cold fogging differ. The conditions in which fogging is most useful (e.g. timing of fogging, taking into consideration weather conditions especially wind speed and direction) are critical for successful operations. Thermal fog is visible and has greater penetration than cold fog, which is not visible but is good for focal application in buildings and on vegetation outdoors. Precautions are necessary when fogging to avoid exposure to asthma patients, infants, and the public.

For more information on recommended insecticides, dosage, methodology, equipment, and protective gear for thermal and cold fogs visit:

http://whqlibdoc.who.int/ publications/2010/9789241500791\_eng.pdf

#### 6.3.1 Problem of insecticide resistance and management

Repeated exposure to the same insecticide may lead to the development of resistance. Hence there is a need to have a resistance management plan in place.

- Organize surveillance to ascertain whether recently carried out insecticidal sprays have had the desired impact on the target populations.
- Conduct standard bioassays (both larval and adult) to ascertain efficacy of the insecticide used.
- Cross-resistance to the same class of insecticide occurs quickly in vector populations. Hence insecticide of different classes may be used in rotation or in a mosaic format to delay acquisition of resistance against a particular insecticide.

 Use of variable vector control techniques such as alternate use of chemical and biological larvicides or selective vector control involving multiple methods (e.g. use of insecticidal and non-insecticidal methods) may retard the development of resistance. For more information on management of insecticide resistance management visit: http://www.who.int/iris/bitstream/10665/44846/1/9789241564472 eng.pdf.

# 6.4 Disinsection of aircraft

Air traffic has increased enormously due to the globalization of economies and increase of leisure and medical tourism. There have been several incidents of malaria being transmitted by invasive and infected vector mosquitoes to individuals living in the vicinity of airports in malaria-free countries (such as France, Switzerland, Netherlands, Belgium, Italy, and Germany). The affected individuals had neither visited a malaria endemic area in the past nor had history of blood transfusion or intravenous drug use. The vectors are transported from endemic areas (e.g. in Africa) by aircraft and cause sporadic cases of malaria. This phenomenon of malaria infection in the vicinity of airports is popularly known as 'airport malaria'. The most recent example is four cases in Tunisia in 2013 (Siala et al. 2015). To prevent airport malaria and other vector borne diseases four methods are currently recommended by WHO for aircraft disinsection: pre-flight; blocks away; top-of-descent; and residual treatment.

## 6.4.1 Pre-flight

A pre-flight aerosol containing an insecticide with rapid action and limited residual action is applied by ground staff to the flight deck, passenger cabin including toilet areas, open overhead and side-wall lockers, coat lockers and crew rest areas. The spray is applied before the passengers have boarded the aircraft but not more than one hour before the doors are closed. A 2% permethrin cis: trans (25:75) formulation is currently recommended for this application, at a target dose of 0.7 g a.i./100 m<sup>3</sup>. This requires application at 35 g of formulation per 100 m<sup>3</sup> to various types of aircraft, with a droplet size of 10–15  $\mu$ m. Pre-flight spraying is followed by a further in-flight spray (i.e. top-of-descent) as the aircraft starts its descent to the arrival airport.

#### 6.4.2 Blocks away

Spraying is carried out by crew members when the passengers are on board, after closure of the cabin door and before the flight takes off. An aerosol containing an insecticide for rapid action is used. The air-conditioning system should be switched off during cabin spraying. (The flight deck is sprayed before the pilot and passengers board.) The doors of overhead luggage racks should be closed only after spraying has been completed. An aerosol containing 2% d-phenothrin is currently recommended by WHO and should be applied at a rate of 35 g offormulation per 100 m<sup>3</sup> (i.e. 0.7 g a.i./100 m<sup>3</sup>). Cargo holds should also be disinsected.

## 6.4.3 Top-of-descent

Top-of-descent spraying is carried out as the aircraft starts its descent to the arrival airport. An aerosol containing 2% d-phenothrin is currently recommended by WHO for this purpose and is applied with the air recirculation system set at normal flow The amounts applied are based on a standard spray rate of 1 g/s and 35 g of the formulation per 100 m<sup>3</sup> (i.e. 0.7 g a.i./100 m<sup>3</sup>). As stated in Annex 9 of the IHR, the details of each disinsecting (place, date, time, method) during the flight should be noted on the Health Part of the Aircraft General Declaration form.

### 6.4.4 Residual spraying

The internal surfaces of the passenger cabin and cargo hold, excluding food preparation areas, are sprayed with a compression sprayer that has a constant flow valve and flat fan nozzle according to WHO specifications (WHO, 2010)<sup>2</sup>. Permethrin 25:75 (cis:trans) emulsifiable concentrate is currently recommended by WHO at a target dose of 0.2 g/m<sup>2</sup> applied at intervals not exceeding two months. The emulsion is applied at 10 ml/m<sup>2</sup> to avoid runoff. Residual sprays are applied by professional pest control operators on aircraft interior surfaces and are intended for long-term residual activity. In electrically sensitive areas, it may be necessary to use an aerosol instead of a compression sprayer. After treatment is completed, air-conditioning packs should be run for at least one hour before the crew and passengers embark to clear the air of the volatile components of the spray. Areas that undergo substantial cleaning between treatments require supplementary 'touch-up' spraying. The pesticide formulations, including spray cans, should comply with national regulations and international standards as well as with WHO specifications for pesticides. Spray operations should follow international regulations and WHO recommended procedures and also comply with quarantine requirements of the country of arrival.

## 6.5 Rodent prevention

#### 6.5.1 Sanitation

Rodents are readily attracted to food and rubbish and will attempt to gain repeated access to it by hiding in nearby shelters. Aboard ships, proper handling and storage of dry food items and elimination of garbage will considerably reduce chances of rodent infestation. Highquality sanitation must be maintained at all times on passenger and cargo ships.

## 6.5.2 Rodent-guards



Figure 50. Anchored ship with rat guards © S. Senthil Nathan

Using rat guards on all tending lines can prevent rats from invading the ship or moving from the ship to port (Figure 50). A standard rat guard has a 36-inch (91.4 cm) minimum diameter, a cone angle of  $30^{\circ}$ , and should be made of 18 gauge steel or aluminium plate. Rat guards must be mounted with the point of the cone toward the ship. Rat guards should be at least 6 feet (1.8 m) from the pier and farther than 2 feet (0.6 m) from the ship.

Rags may be used to plug gaps in the rat guards. Rags should be secured tightly to prevent loosening or being pulled apart by the rat. Stray lines if any should be kept out of the water. If two lines are in close proximity to each other, either the lines should be grouped to pass through a single rat guard, or rat guards should be installed side-by-side or

touching each other. This will prevent rats from jumping from one line to another, skirting the

<sup>2</sup> https://extranet.who.int/iris/restricted/handle/10665/44836

rat guards and making them ineffective. It may be noted that International health regulations no longer require the use of rat guards by ships except when berthed in ports where plague is endemic. However, rat guards are still recommended to prevent rodent movement between ships and ports, especially in ports where large populations of rodents exist. Regardless of the use of rat guards, proper sanitary measures of food and food waste aboard each vessel should be maintained at all times to minimize attracting rodents.

#### 6.5.3 Illumination for rodent movement restrictions

Rodents are nocturnal and therefore shun light. If the ports, cargo holds, and luggage areas in airports, warehouses and ship gangways are well illuminated at night, rodents will be discouraged from moving about freely and moving between the port and vessel. When not deployed, cargo nets must be raised and removed to prevent movement of rodents across them.

#### 6.5.4 Pier-side inspections/surveillance on-board ships

All incoming items and substances must be thoroughly checked for any signs of rodent activity. As recommended elsewhere, rodent droppings, hair, urine, and gnawing marks strongly suggest possible rodent access into a ship. Hence surveillance and control of live rodents should be initiated as soon as any of these are discovered.

# 6.6 Rodent control

Rodents can be controlled using different types of traps. Rats are suspicious of new objects and if they see other rats already in a trap, they may avoid it. Traps which do not address these issues are likely to catch only young, inexperienced rats. There are different types of traps, which are discussed below.

#### 6.6.1 Conventional rat cage traps



Figure 51. Conventional rat cage with conical entrance (also known as Wonder trap) © S. Senthil Nathan

A rat cage trap is a metal box-shaped cage that is designed primarily to catch rats without killing them (e.g. the Wonder trap, Figure 51). Food bait consisting of bread with peanut butter, etc. (and not poisoned) is put in the cage trap. When an animal enters the cage and moves toward the bait, the mechanism triggers and closes a door over the entry point. The animal is caught alive and without injury. The animal can be subsequently killed.

## 6.6.2 Spring board traps

Traps designed for rats are powerful enough to break the rat's neck or spine; they may break human fingers as well (whereas an ordinary spring-based mousetrap is very unlikely to break a human finger). Note that spring traps designed for rats may not be sensitive enough to spring when a mouse takes the bait.

#### 6.6.3 Glue traps

Glue traps are non-poisonous with sticky glue spread over cardboard. When a rat tries to pass over it, it becomes stuck and eventually dies from dehydration and asphyxiation. Bait may also be placed on the cardboard to attract the rats. These traps are especially effective in reducing populations of mice. Glue traps should be deployed continuously for two to three weeks in order to trap a good number of rodents. Their use is forbidden in some countries as the trapped animal suffers for a long time before death; hence this method should be discouraged.

#### 6.6.4 Electronic rat-traps

Electronic rat-traps detect the presence of a rodent via metal plates on the floor of the trap, then delivers a lethal dose of high voltage electricity stepped up from batteries. Some brands offer remote indication to tell you when the trap has operated.

#### 6.6.5 Sherman trap

These traps are made of eight hinged plates, are fully collapsible, and designed to trap the rodent alive. The rodent is attracted to bait placed at the rear of the trap. When it passes over the floor plate, its weight springs the trap, which closes the door behind it.

## 6.7 Sandfly: personal protection and control

For prevention of sandfly bites and effective sandfly control the following measures, which have been found effective, can be considered.

#### 1. Personal protection:

- Use of proper clothing that covers arms and legs (no exposed skin to sandflies)
- Use of head netting gear to cover head and face during sandfly surveillance and control operations to prevent bites
- Application of DEET lotion on exposed skin as a repellent
- Uniformed personnel can treat their uniform with 40% permethrin concentrate as per label instructions
- Ready-to-use individual dynamic absorption (IDA) kits and permethrin aerosols have been used for treating individual uniforms (<u>http://www.afpmb.org/sites/default/files/</u><u>whatsnew/2012/Sandfly\_PocketGuide\_2012.pdf</u>).

#### 2. Sandfly control:

Spraying of synthetic pyrethroids such as permethrin, bifenthrin, and lambda-cyhalothrin can effectively control sandflies. These can be delivered via hand-pressurised compression pumps for small focal applications; engine-pressured backpack and truck-mounted hydraulic sprayers may be deployed for vegetation and larger areas. Thermal fogs and ULV applications have shown promise in sandfly control as well. It should be noted that before, during, and after insecticide applications all necessary precautions mentioned elsewhere in the handbook must be taken to maximize effectiveness of the sprays and to minimize exposure to humans and animals. Member States may assess the resistance profile of the transmitting species to various insecticides before choosing an insecticide if the local data near PoE exist, or may make efforts to generate data in coordination with national health services and sandfly control

make efforts to generate data in coordination with national health services and sandfly control programmes wherever necessary.

# 6.8 Flea: personal protection and control

Flea control measures should be undertaken during the following situations:

- When in any locality rats are observed or signs of their passage noted.
- When an increase in the population of fleas/flea nuisance (an increase of the Flea Index) is reported.
- Specific Flea Index found to be more than 1.0 through active surveillance at PoE especially in areas infested by rodents such as warehouses.

The following measures are recommended for plague vector control: personal prophylaxis, insufflation and residual insecticide spray.

Personal prophylactic measures include the following:

- Use of repellents like benzylbenzoate, diethyltolumide (DEET), dimethylphtholate (DMP) on skin or clothing to avoid flea bites.
- Use of high-necked shoes or socks up to the knees.
- Sleeping on cots at least 0.5 metre from ground level.

Insufflation measures involve treating rodent burrows and rat runs with 10% DDT or 5% malathion dust powder (wettable powder). Insecticide dusts should be blown with a rotary plunger-type duster or cyanogas pump into the mouth of the rodent burrow, and a patch of dusting powder about 0.5 cm to 1.0 cm thick and 20–25 cm wide should be left around its mouth.

Insecticides used for residual insecticide spray are malathion, deltamethrin, cyfluthrin, and lambda cyhalothrin. Formulations for each, including treatment areas, are discussed below.

#### Malathion 25% WP spray formulations

- Suspension is applied @ 2.0 g/m<sup>2</sup> active ingredient.
- To get 5% suspension, 2.0 kg of 25% WP is mixed in 10 litres of water.
- Spraying may be undertaken annually or as per local requirements.

#### Deltamethrin 2.5% WP

- Suspension is applied @ 20 mg/m<sup>2</sup> active ingredient.
- To get 0.125% suspension, 400 g of 2.5% deltamethrin is mixed in 10 litres of water.
- Spraying may be undertaken annually or as per local requirements.

#### Cyfluthrin 10% WP

- Suspension is applied @ 25 mg/m<sup>2</sup> active ingredient.
- To get 0.125% suspension, 125 g of 10% cyfluthrin is mixed in 10 litres of water.
- Spraying may be undertaken annually or as per local requirements.

#### Lambda-cyhalothrin 10% WP

• Suspension is applied @ 25.0 mg/m<sup>2</sup> active ingredient.

- To get 0.125% suspension, 125 g of 10% lambda-cyhalothrin is mixed in 10 litres of water.
- Spraying may be undertaken annually or as per local requirements.

#### Treatment areas

Residual spraying in indoor situations should be up to a height of one metre in the affected areas because fleas can hop only up to half a metre.

Areas where people sleep, animal bedding, under rugs, and cracks and crevices in floors should also be sprayed.

Exterior treatment should include all areas frequented by rodents, cats, dogs, etc. Handcompression pumps should be used for residual spraying, following the techniques used for the control of adult mosquitoes.<sup>3</sup>

## 6.9 Cockroach control

#### 6.9.1 Dust

Boric acid is a well-tested weapon against cockroach infestations. It may be formulated into bait or as a residual that cockroaches walk through. When a cockroach walks through a treated area, boric acid powder adheres to its legs and body. Upon grooming itself, the cockroach ingests the boric acid dust. Boric acid dust has to be applied carefully to be effective. Avoid contact with food and kitchen utensils and before treatment cover all surfaces that come into contact with food.

#### 6.9.2 Aerosol

A 2% aerosol formulation of d-phenothrin or recommended formulation of pyrethrins is suggested for aerosol applications. It should be applied in cracks and crevices using an extender tip.

#### 6.9.3 Baits

Place bait in areas frequented by cockroaches. Gel baits are available with a variety of active ingredients, several of which can work on cockroaches. The cockroaches consume the bait and are killed by the active ingredient. Since they also share the bait between them, knockdown and mortality of sizable populations can be achieved quickly.

Note: If the cockroach infestation is extensive or if oothecae are detected or remain even after the cockroaches have been eliminated, the population can build up rapidly from the nymphs that hatch from oothecae.

## 6.10 Housefly control

Houseflies are not only a cause of nuisance but also the source of many infections in humans as mentioned earlier. Flies are easily attracted to food, kitchens, service areas, and garbage. Their numbers can be legion when favourable conditions of feeding and breeding are present. Being ubiquitous and a close companion of humans, they are a major concern at any point of entry. For example, they can easily fly into an anchoring ship, aircraft or lorry or train , particularly if favourable breeding conditions are available at these premises.

Disease control basically relies on sanitation and hygiene: preventing flies from coming into contact with food, food areas, and humans and encouraging proper hygiene practices. In

<sup>&</sup>lt;sup>3</sup> - For further reading visit: (http://www.searo.who.int/entity/emerging\_diseases/documents/ISBN\_9789\_92\_9022\_376\_4/en/)

addition, fly populations can be controlled through the reduction of breeding places; sanitation and hygiene will also help minimize an area's attractiveness to flies.

Elimination of fly breeding sites involves repeated flushing of floors of kitchens, using sanitizers to clean cooking surfaces, utensils and other food preparation and dining areas (e.g. in galleys), and covering leftover food. Garbage, cow dung, faeces, and waste in drains should be cleared and kept covered until such time as it is disposed of. Surface drains at PoE particularly should be cleaned regularly, as any sludge or organic waste will serve as a breeding site for flies. Ideally all drains at PoE should have a concrete base.

The garbage generated at PoE can be transported several kilometres away to sanitary landfills (where the garbage is compacted and covered with 15-30 cm layer of fresh soil in order to prevent fly breeding). Window and door screens keep flies away. In areas where food is prepared or eaten air curtains fixed above doors can prevent fly entry as well. Devices that electrocute insects can also be used – they generally use a blue light to attract, and an electric grid to kill the insect. Another method is the use of sticky traps, hung vertically from ceilings, where fly activity is common. These can be effective until such time that the trap is covered with flies. Attractive sugars used as lure attract flies to the sticky traps.

Chemical methods are used for either quick knockdown with short residual effect or for longterm control with a residual insecticide. It is essential to ascertain the status of resistance in the fly population to an insecticide prior to its use. Dichlorvos vaporizer strips slowly release insecticide, which is effective for two to three months especially in places where ventilation is limited. Flies tend to congregate and rest at night on curtains, strings, hangings, wires etc. in large numbers. This behaviour can be exploited to control them by hanging toxic insecticide materials cut in to strips, strings, etc. and hung from the ceilings. The flies will be exposed to insecticides when resting leading to their control. Many organophosphorus and carbamate compounds are available for use in lethal traps. A variety of attractants add value to the trap including traditional syrups, malt, ammonium carbamates, whole egg to a more advanced Synthetic Fly Attractant (SFA), a mixture of 88% commercial fish-meal, 5% ammonium sulfate, 5% trimethylamine hydrochloride, 1% linoleic acid and 1% indole. A variety of baits such as dry baits, liquid sprinkle bait,liquid bait dispensers and viscous paints are available.

Residual insecticides are recommended in animal sheds and farms. A number of organophosphorus and pyrethroid formulations are available for use as residual insecticides against flies. These can be sprayed over larger areas using hand-held sprayers, and should be effective for a few weeks. It is important that spraying is done selectively in areas preferred by flies. Indoor space sprays can be used to achieve quick reduction in fly populations and complement all other methods. Outdoor space sprays can be used repeatedly for control of flies in larger areas but their main disadvantage is that they are ineffective against flies already indoors and those that are yet to emerge from pupae. Hence repeated control is necessary, which may be expensive. Where flies congregate, direct spraying can result in knockdown of existing flies and also those that will come into contact with treated surface subsequently. Breeding sites such as garbage dumps or cow dung dumps can be directly treated with insecticides. Since the maggots will be dwelling deep in the breeding site much greater quantities of the insecticide solution will be necessary to penetrate and wet the breeding site surface to be effective. Note that at an active dump site more garbage will be continuously dumped necessitating repeated treatment. For more details about houseflies (WHO 1997). Visit http://www.who.int/water sanitation health/resources/vector302to323.pdf.

# 7. EMERGENCY MEASURES

If an outbreak of vector-borne disease occurs or an exotic vector species is detected at PoE during routine surveillance that merits emergency measures, action must be initiated immediately on a scale commensurate with the risk posed (Table 11). As indicated in the monitoring and evaluation section (Chapter 8), an emergency coordination meeting of the task force should be called to discuss the outbreak/infestation. Full-scale containment measures must be conducted followed by impact assessment of the measures used. The containment operations must continue until the exotic vector/rodent species is eradicated.

Vector	Emergent situation	Surveillance measure	Control measure
Mosquito	Detection of exotic vector species immatures and/ or adults during routine surveillance or following the report of disease outbreak (e.g. malaria, dengue, chikungunya or yellow fever)	Conduct extensive surveillance in and around/the site where the species was detected or outbreak reported	Undertake antilarval sprays in breeding sites and anti-adult measures, particularly fumi- gation, with recommended insecticides (thermal/cold fog) for quick knockdown of the invading vector populations. IRS to be done if necessary
Rodent	Detection of a large indi- genous rodent population infestation, exotic rodents, or evidence of plague occurrence	Deploy sentinel traps for rodent surveillance	Deploy rodent baits (anticoa- gulants) in the path frequented by rats; in case of plague, both rodents and fleas should be targets for control measures (insecticides)
Housefly	Fly infestation or a gastrointestinal disease outbreak detected	Conduct surveillance for possible breeding grounds (e.g. garbage dumps)	Remove garbage to deny breeding grounds to flies. Use anti-adult sprays, fly baits, fly traps and maggot control measures
Sandfly	Sandfly invasion/ infestation detected or indigenous case/s of leishmaniasis reported	Conduct emergency surveillance for sand- flies in the affected area	Spraying of recommended insecticides (IRS) in infested locality particularly. The use of larvicides in sandfly control pro- grammes is generally regarded as impractical
Cockroach	Reports of cockroach infestation particularly of an exotic species or population	Carryout surveillance for adults, nymphs and ootheca of cockroaches	On confirmation, use aerosol especially in crevices, deploy baiting traps. Undertake focal IRS with recommended insec- ticide for quick cockroach flush out or fumigation particularly thermal fog for deep penetra- tion and flushing effect

#### Table 11. Actions to take in the event of vector species or disease detection at PoE

# 8. MONITORING AND EVALUATION

Monitoring and evaluation are essential elements of vector surveillance and control at PoE. They ensure sound management practices and success of the programme. Table 12 lists the indicators associated with the steps towards implementing the applicable IHR, that is, establishing vector surveillance and control capacity at PoE including the capacity to respond to emergencies.

Category	Process indicator	Outcome indicator
Policy related to 2005 IHR	Notification issued by State Party on IHR implementation in the key ports of entry and ground cros- sings as national policy.	National level implementation of 2005 IHR made mandatory by an act of legislation or notification.
Advocacy for 2005 IHR implementation	Advocacy for national-level imple- mentation of 2005 IHR initiated by stakeholders.	Practical steps taken and road map for implementation of 2005 IHR prepared.
PoE designation	Major PoE designated and shared with WHO.	Designated PoE start developing specific proposals for implementation of IHR provisions.
IHR implementation at PoE	Requirements of infrastructure and human resources development inter alia for vector surveillance and control assessed.	Necessary infrastructure is in place and human resources of different categories with required skills recruited; outsourcing as mandated to specialized companies.
Vector surveillance and control core capacity	Need for extensive training of human resources for capacity buil- ding/skill development assessed and training schedule planned, resource persons and appropriate materials identified for class-room and practical training on vector surveillance and control including PPE; necessary laboratory and field data formats discussed; control measures of mosquitoes (immatures and adults) identified as needed; control measures for other vectors (e.g. cockroaches, sandflies, houseflies, and rodents) identified and discussed.	Vector surveillance and control infrastructure fully developed, human resources trained, laboratory and field formats developed, field-tested, and refined after pilot-scale testing; field operations are initiated; adequate PPE procured and supply chain established.

Category	Process indicator	Outcome indicator
Impact assessment	Vector monitoring programme designed, and impact assessment indicators listed; vector data formats for impact assessment developed; frequency of impact assessment decided.	Performance indicators, vector data formats tested, impact assessment exercise completed; vector popula- tions assessed from surveillance data; further action planned if abnormal events observed; if exotic vector(s) detected during surveillance emer- gency control measures invoked until the vector is eradicated.
Preparedness for emergencies	Emergency control measures for each category of vector identified and tested in mock drills. The exercises repeated till expertise achieved. Emergency plan shared with entire vector control team.	Emergency task force and coor- dination mechanism established, command identified and made known to all members. Plan revised from time to time if necessary especially to accommodate new knowledge, skills and vector control developments.

## 9. REGULATIONS ON WILDLIFE AND ANIMALS AT POE

Birds and mammals – whether pets, livestock or game – brought through PoE may pose serious threats to human health and local animal populations. There are stringent licensing and health requirements for people travelling across borders with their pets and livestock, such as vaccinations, which if not met can result in extended quarantine or refusal of entry (e.g. travellers with dogs must possess a valid certificate of vaccination against rabies). Veterinary sanitary regulations of State Parties might require destruction of animals that are denied entry at ports and ground crossings. Pets that return to their native country with travellers are also subject to health checks and quarantine similar to pets entering for the first time. Exotic species can be subject to restrictions as well (e.g. many countries do not allow importation of monkeys as pets), as can species that belong to an endangered category.

There are international agreements according to which several nations have created regulatory systems that "screen out" introduction of potentially harmful animals that may become invasive or carry human or animal pathogens. The global imports of animals and their products are subject to agreements signed by nations as per the Convention on Biological Diversity (CBD), International Plant Protection Convention (IPPC), World Organisation for Animal Health (OIE), and World Trade Organization (WTO). The applicability of these regulations is subject to their agreement by State Parties.

In addition, there is a need to maintain vigilance on borders in the light of recent global pandemics such as the highly-infectious avian influenza H5N1, severe acute respiratory syndrome (SARS), and other zoonotic diseases.

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# Annex 1. Personal protective equipment

Personal protective equipment (PPE) includes protective clothing, helmets, goggles, shoes, and other garments or equipment designed to protect those spraying insecticides from exposure and injury.

PPE	Size	Function
Coveralls, disposable (weight and thickness determined by climate)	Variable	Single use; protects entire body especially the skin from exposure to droplets of insecticides
Coveralls, re-usable (weight and thickness determined by climate)	Variable	Multiple use; protects skin from exposure to droplets; to be washed separately before reuse
Broad-rimmed hat or helmet	Variable	Protects head and face from droplets
Face shield or goggles	Standard	Protects eyes from droplets
Respirator, with pre-filter	Standard	Protects nose and mouth from airborne particles and prevents inhalation
Gas mask	Standard	Protects against fine droplet sand fume inhalation
Protective gloves	Variable	Protects hands; sleeves to be inside of gloves
Boots	Variable	Protects feet; overalls to be worn outside of boots
Rain coat	Standard, for men and women	Protects body from rain during spraying

Table A1-1. Description and functions of personal protective equipment (Figure A1-1 and A1-2)



Figure A1-1. Workers wearing PPE while spraying containers with insecticides



Figure A1-2. Illustration of itemized PPE
WHO health and safety regulations advise that all persons spraying insecticides must be adequately protected against potential harm due to exposure from pesticides during handling, transportation, storage, use, and cleaning of pesticides or pesticide-contaminated materials. All such persons must wear appropriate personal protective clothing in accordance with the safety instructions on the product label or material safety data sheet (MSDS).

The following guidelines should be followed:

- Overalls should be made of cotton and of a weight and thickness appropriate for the climate.
- Coveralls and boots must be available in sizes appropriate for the specified work force (women's and men's sizes).
- There must be extra gloves, boots, face shields, and helmets available to replace items broken or lost.
- Spraying staff must have a minimum of two uniforms to allow for frequent changes.
- Filters in masks must be replaced each day for spray operators. For the 8-hour filter mask, the mask itself should be changed once every two days.
- Gloves should be examined carefully before use for any signs of damage, particularly in the areas between the fingers. If there is any doubt about their protectiveness, they should be replaced. At the end of each day's use, gloves must be washed inside as well as outside before they are used again.
- Worn out gloves should be replaced with new ones immediately.
- Overalls must be changed daily for spray operators using carbamates, pyrethroids, or organophosphates; every two days for spray operators using DDT (to minimize effluent waste).
- Overalls must be changed immediately when a direct spill occurs onto the overalls.
- Overalls should not be tucked into boots.
- Personnel while washing the spraying equipment should wear their long gloves over sleeves.
- The Supervisor shall ensure that all workers wear protective clothing.

Source: Adapted from (WHO 2015) and (President's Malaria Initiative 2013).

# Annex 2. Laboratory requirements for the vector work

The list below includes suggested items to stock in the laboratory that is to conduct vector testing. However, laboratory requirements should be subject to national legal or policy requirements, as well as any national laboratory quality-assurance system. The list below is not exhaustive and may be expanded as necessary.

#### I. Furnishings

- 1. Adequate lighting
- 2. Air conditioning
- 3. Air curtains
- 4. Working desk
- 5. Paper/cork pin-up board
- 6. Comfortable revolving chair/stool
- 7. Iron racks for adult mosquito cages or desks
- 8. Storage cabinets
- 9. Insect cabinet

#### II. Equipment

1. Binocular dissecting microscope with spare parts (especially bulbs when equipped with internal light source)

2. Binocular compound microscope with spare parts (especially bulbs when equipped with internal light source)

- 3. Hand lens
- 4. Refrigerator
- 5. –20° C deep freezer
- 6. CDC light traps or similar traps for mosquito surveillance with adequate spares
- 7. Desktop computer, printer, uninterrupted power supply with Internet connection
- 8. Photocopier
- 9. Laboratory thermometer and hygrometer
- 10. Rat traps (spring board, conventional iron cage, glue trap)

# III. Chemicals and reagents

- 1. Immersion oil
- 2. Distrene-Plasticiser-Xylene (DPX), mountant
- 3. Para-dichlorobenzene
- 4. NaCl
- 5. Giemsa stain (ready to use)

#### IV. Glassware, plasticware, and minor instruments and materials

- 1. Petri plates
- 2. Glass slides
- 3. Cover slips
- 4. PCR tubes
- 5. Test tubes
- 6. Watch glasses
- 7. Tweezers
- 8. Dissecting needles

- 9. Coupling jars
- 10. Measuring cylinders (50, 100, 500ml capacities)
- 11. Cotton-wool
- 12. Tissues
- 13. Paper napkins
- 14. Gloves
- 15. Droppers with bulbs
- 16. Plastic basins
- 17. Plastic bowls (capacity 300ml)
- 18. Net cloth
- 19. Rubber bands
- 20. Mosquito larval food (baby formula of a trusted brand and fishmeal flakes in equal proportions; yeast and dog biscuit mixture in a ratio of 60:40)
- 21. Hand-held mouth and hand-held motorized aspirators for mosquitoes
- 22. Mosquito cage stands (1 ft<sup>3</sup> or 2 ft<sup>3</sup>)
- 23. Fine hair brushes (0 number)
- 24. Plastic containers for sample packaging and transportation to a sentinel lab
- 25. Bubble wrapping

# Annex 3. Surveillance methods of mosquitoes and rodents at PoE

Vector (growth stage)	Method	Tools
Mosquitoes (immature)	<ol> <li>Mapping breeding habitats</li> <li>Inventory of habitats and photos taken</li> <li>Focused sampling in 400 m radius of PoE</li> <li>Sending immature samples to laboratory</li> <li>Rearing in insectary and spe- cies identification of emerged adults</li> <li>Analysis of field data and breeding habitat preferences of vectors</li> <li>Writing evidence-based vector control strategy (IVM)</li> </ol>	<ol> <li>Immature surveillance field kit (Ref. 5.5.2.1)</li> <li>Camera for photography of breeding habitats</li> </ol>
Mosquitoes (adult)	<ol> <li>Listing native species</li> <li>Sampling plan to cover space and time</li> <li>Sampling method to be based on mosquito biting activity, host and resting site preferences</li> <li>Seeking identification of and expert advice on collected samples of invasive species</li> <li>Sample(s) may be sent to refer- ral laboratory for confirmation</li> <li>In the meantime more mosquito collections must be undertaken with CDC traps/ similar traps in and around area where sample of species was originally collected</li> <li>In case of disease outbreak, pathogen to be identified locally or in a designated referral facility</li> </ol>	<ol> <li>Adult collection field kit (Ref. 5.6.1)</li> <li>CDC light traps or similar traps</li> </ol>
Rodents	<ol> <li>Studying runway and rub marks</li> <li>Studying tracks of rodents</li> <li>Observing gnawing activity</li> <li>Observing droppings and rodent identification</li> <li>Observing urine with UV light</li> <li>Deploying traps for surveillance</li> </ol>	<ol> <li>Traps with cover and/or snap traps</li> <li>Bait for traps</li> <li>Rat tunnels</li> <li>Hand lens (to observe droppings)</li> <li>UV light torch</li> </ol>

# Annex 4. Identification, preservation, and transportation of specimen to designated laboratory

Vector (growth stage)	Specimen condition	Method	Packaging	Remarks
Mosquitoes (adult) Sandflies Cockroaches	Dry	Mount adult specimen or nymphs on an entomo- logical pin, piercing the thorax between the legs and placing the other end of the pin in the cork. With forceps gently slip a few crystals of para- dichlorobenzene into the bottom of the tube and heat on a candle flame until the crystals melt (it will solidify rapidly when cooled). This will prevent fungal growth on the specimen. Insert mounted specimen in a glass tube of sufficient size, close the mouth with a cork and label the tube appropriately	Secure the specimen tube in bubble wrap and pack it in a box. Fill the gaps with polystyrene beads or other packing material to provide sufficient cushion. Label the pack fragile. Dispatch the specimen(s) to the desired address by a courier which accepts biological materials	Send with a cover letter and contact reference laboratory by phone or email to explain the emer- gency, and ask for prompt identification
Mosquitoes (immature)	Wet	Preserve in 70% alcohol in a glass tube with a leak- proof lid. Label the tube appropriately	Seal well to pre- vent evaporation of alcohol	There might be restrictions on the shipping of speci- mens preserved in alcohol (due to flam- mability). If feasible the specimen may be carried perso- nally to reference laboratory
Mosquitoes (adult)	Digital image	Take a set of clear and sharp photographs of the specimen under a dissecting microscope highlighting important morphological features/characters for a convenient taxonomic classification of specimen to the species level	Send images to the designated laboratory via e-mail along with list of obser- ved characters recorded by a qualified and trained personnel. Mention tentative identification if possible	Request for an urgent identification/ confirmation of the specimen by the refe- rence laboratory for an appropriate and rapid action against exotic species. Many countries prohibit transfer of biological specimens overseas. Hence digital image transfer is not only quick, convenient and inexpensive but also hassle free

# Annex 5. Laboratory requirements for pathogen detection

# 1. Microscopy (detection of Plasmodium and Leishmania species):

- Phase contrast microscope with high power (100x objective)
- Sterile pricking lancets
- Cotton swab ready-to-use sachets (or cotton soaked with isopropyl alcohol)
- Glass slides
- Ready-made stains (Giemsa/Leishman/Field/JSB)
- Coupling jars
- Methanol (for fixation of thin smear)
- Beakers
- Measuring cylinders
- Drying rack
- Lead pencil for labelling thin smear

## 2. Rapid diagnostic tests (detection of malaria):

## • Rapid test ready-to-use kit of standard quality

- o Either monovalent (HRP-2 based) for *P. falciparum* or
- o Bivalent for P. falciparum (HRP-2 based) and P. vivax (LDH based)
- o Multivalent for all human *Plasmodium* species detection (HRP-2 and species-specific LDH and pan LDH combination)

# 3. Requirements for ELISA/PCR detection of Plasmodium or Leishmania infection\*

- ELISA readers
- PCR machine
- Agarose gel electrophoresis unit
- Micropipettes
- UV transilluminator
- Gel documentation system

#### Chemicals/reagents required for PCR

- ELISA reagents
- PCR buffer
- Primers
- Deoxyribonucleotide triphosphates (dNTPs)
- Taq polymerase
- DNA template
- Deionized water

#### Chemicals/reagents required for electrophoresis

- Agarose
- TBE buffer
- Ethidium bromide
- Tracking dye

<sup>\*</sup>Not viable for plague, dengue, chikungunya.

# Annex 6.Potential mosquito breeding sites at PoE and interventions to address them

Breeding site	Potential problem	Corrective measure
Surface drains	Unlined with wavy margins, obstruction due to dumping of garbage or solid waste, faulty gradient	Provide lined margins and proper gradient; conduct regular inspections; ensure waste remo- val and smooth flow of water
Potable water tanks	Open, missing or broken lids, overflow pipes open, without mesh, or torn mesh	Install mosquito-proof lid assembly; mouth of overflow pipes fitted at all times with perforated screw-cap or covered with sturdy plastic mesh with <2.5mm holes
Septic tanks	Open or partly open; vent open	Lid sealed with mortar ensuring no gaps for mosquito entry/escape; vents to be covered with sturdy plastic mesh on regular basis; inspect routinely
Inspection/service chambers	Open or partly open	Inspect routinely; they should have tight-fitting mosquito proof lids
Sluice valve chambers	Open with leakage of water from valve	Have to be kept covered; water leakage pre- vented with regular maintenance
Ore loader buckets	Stored upright	Always store facing down
Tyres as fenders, or disused or rejected	Accumulate fresh water during rains	Cut 2 sq. inch holes at 3, 6, 9 & 12 o'clock in tyres; organize regular disposal of used tyres; in the interim stack horizontally one above the other and cover the top with plastic sheets held in position by a flat heavy plank above the top sheeting (Figure 25)
Construction sites at harbour or residen- tial neighbourhoods near ports	Water stagnates in pits, on slabs for curing, in barrels and masonry tanks, etc.	Avoid standing water (beyond one week); introduce larvivorous fish (guppies/ <i>Gambusia</i> or a proven native species) in larger pits and monitor; treat all shallow stagnant waters with permitted biological or chemical larvicide; conti- nue monitoring until complete
Blocked terraces	Water tends to stagnate, creating a site suitable for breeding	Remove any obstruction to drainage pipes or correct gradient if faulty to ensure normal run-off of water
Dock workers/dri- vers breakrooms	Open windows or doors, eves	Install mosquito screens on doors and windows (preferably metallic) as rats can gnaw through fibreglass or nylon mesh easily
Scrap	A variety of rejected articles, tanks, etc. holding fresh water	Ensure continuous disposal of scrap, at least on a monthly basis
Discarded boats	Kept upright holding rain water	Should be stored facing down
Flat floats with depressions	Iron floats covered with shal- low waters	Store vertical and prevent water stagnation

Breeding site	Potential problem	Corrective measure
Ornamental fountains	When not cleaned on a regular basis, fountains can harbour plankton and mos- quito larvae	Ornamental fountains should be avoided at PoE. If present they should have larvivorous fish of either exotic or local species at 5 fish/m <sup>2</sup> surface area at all times; clear fountains of any debris or algal mats for free fish movement
Grassy and marshy land	Continued stagnation causing mosquito breeding	These may be filled with soil and levelled or be cleared of grasses and stocked with larvivorous fish
Puddles	Filled with fresh stagnant water serve as breeding source during rains	To be filled up or drained off
Wells	Perennial breeding potential	Should have larvivorous fish at 5 fish/m <sup>2</sup> surface area; ensure fish presence at all times
Water hydrant	Margins have floating vegeta- tion or algae	Surface should be cleared routinely; introduce larvivorous fish at 5 fish/m² surface area
Large hoses and pipes	Not stored correctly, pipes will hold water and support breeding of mosquitoes	Should be stored indoors or in a manner that prevents them holding water
Cargo holds/ containers	Water might stagnate when open	Ensure these are kept closed at all times preven- ting water entry

# Annex 7. Onsite record form for mosquito surveillance

# A7.1. Surveillance of immature mosquitoes

S.	Name of	Location/ If positive, no. of larvae and/or pupae in sample							Remarks		
s. No.	breeding	complete	Anop	heles	Cu	lex	Ae	des	$\sim N$	lix	Remarks
INO.		address	L	Р	L	Р	L	Р	L	Р	

# Name of the port/airport/ground crossing: Date:

\*Make inventory of breeding sites in your area; L,Larvae of all instars; P, Pupae.

Total no. of houses/buildings/areas surveyed: Total no. of houses/buildings/areas where larvae found: Total no. of containers checked: Total no. of containers found positive: Breeding Index: House Index: Container Index: Breteau Index: Pupal Index:

Actions taken: Source reduction/biological control/chemical control/insect growth regulator used

Give details:

Signatures of field worker:

Signatures of field supervisor:

Signatures of port health officer:

# A7.2. Record format for surveillance of adult mosquitoes

Name of the port/airport/ground crossing: Type of collection: hand catch/spray sheet collection/CDC traps Time of collection: Date:

c	Location	Random		Species with numbers						Remarks						
S. No.	where sample	or fixed	or fixed	or fixed			or fixed	Anop		Cu		Ae		Other:		List species
INO.	collected	station	М	F	М	F	М	F	М	F						

M,Male; F, Female.

Average density per working hour (hand catch): Per room density (spray sheet collection): Per trap per night density (CDC trap collection): Total no. of containers found positive:

Actions taken: IRS/thermal/cold fogging/insecticide-treated materials used

Give details:

Signatures of field worker:

Signatures of field supervisor:

Signatures of port health officer:

# A 7.3. Rodent vector ectoparasites surveillance form

# Name of the port:

#### Country

Date	No. of	Location	Ro specie	dent es found	Sex		Sex No. of traps		Collection from combing			nbing	Sample no.	Remarks
	uaps		Туре	No.	Μ	F	Pos.	Neg.	Mites	Ticks	Lice	Fleas	10.	

Total no. of traps placed: Total no. of traps sprung: Total no. of traps found with rats: Total no. of rat fleas/mites/ticks/lice collected: Actions taken: Rodent traps/baits (anticoagulants) deployed; insulfation done with insecticide malathion 25%WP/deltamethrin 2.5%WP/cyflithrin 10%/ lambda-cyhalothrin 10%WP

Give details:

Signatures of field worker:

Signatures of field supervisor:

Signatures of port health officer:

# Annex 8. Statistical indices for mosquito vector surveillance

	Index	Definition
А.	Immatures	
1	Breeding Index	The percentage of breeding sites (all kinds) positive for mos- quito immatures from those inspected
2	House Index (HI)	The percentage of houses infested with larvae or pupae. House index indicates spatial distribution of vector breeding in a locality
3	Container Index (CI)	The percentage of water-holding containers infested with larvae and pupae out of those inspected. This index indicates infesta- tion of vector breeding among containers present in a locality
4	Breteau Index (BI)	Number of positive containers per 100 houses inspected (Aedes immatures). BI indicates density of immature-harbouring containers in a locality
5	Pupal Index (PI)	Number of pupae per 100 houses inspected. Pupal index is a good measure of risk of transmission especially in case of dengue by <i>Aedes</i> mosquitoes
6	Per dip density	Average density of immatures (larvae and pupae) in a habitat calculated from a minimum representative sample. Usually a minimum of 5 dips/habitat drawn randomly from the entire body of water are considered
В.	Adults	
1	Hand catch: density per working hour	Number of adults caught in an hour. It can be expressed for all species or a particular species
2	Total catch	Number of adults caught after being knocked down by space spray using white cotton sheets placed on the floor (check after 15 minutes of spraying)
3	Per trap per night density (or per trap per day density in case of <i>Aedes</i> adults)	No. of adults caught in 12 hours (6PM-6AM in case of nocturnal Anopheles and Culex species or 6AM-6PM in case of Aedes species which are diurnal) in a continuously running mosquito trap. It can be expressed for total mosquitoes and for different species in the collections

Vector	Recommended insecticide/ formulation	Application method	Frequency of application	Precaution
Mosquitoes 1. Immature	OP in EC insect growth regula- tors Bti (Bacillus thuringiensis israelensis WP/ EC/GR)	Compression sprayer/ knapsack sprayer. Sprayers/hand dispersal in the case of BTI GR formulation	As per national guidelines and recommendations on product label	
2. Adult (Quick knock- down) aerosols	IRS: recom- mended insecticide Insecticide- treated materials Window and door screens	Compression spray pumps; spray canisters; cold fogger/ thermal fogger Long-lasting insecticidal nets/curtains Fibreglass/iron mesh	<2.5mm mesh size	All persons spraying or handling insecticides to wear approved PPE; dosage of insecti- cide as specified by national control programme
Rodents	Prevention 1. Sanitation 2. Rat guards	Keeping cargo, stores and godowns at PoE and shipboard as clean as pos- sible and garbage free Rat guards should be used on all tending lines when the ship is anchored both at pier side (at least 6 ft.) and 2 ft. from the ship. Rat guards must be mounted with the point of the cone toward the ship. Rat guards should have a 3 ft. minimum outside diameter, a cone angle of 30 degrees, and be made of 18 gauge steel or aluminium. <b>Illumination and</b> <b>Movement</b> <i>Restrictions</i> : Rodents are nocturnal, there should be proper lighting in the cargo area, cargo holds, godowns and on ship lighting up gangways and landing ramps at night can discou- rage rodents from coming on board		

# Annex 9. Chemical/biological insecticides\*for vector control

Vector	Recommended insecticide/ formulation	Application method	Frequency of application	Precaution
	Rodent control	Pier-side inspections Inspect all incoming supplies for signs of rodent infestation (e.g. droppings, urine, hair, gnawing, or live rodents) and reject such supplies that may be contaminated with rodent faeces and urine. Handle such items carefully, avoi- ding direct contact Appropriate rat-traps should be used, e.g.rat cage trap, springboard trap, electronic trap or glue trap Ingestible poison baits (anticoagulants) are avai- lable which are effective in killing rats		
Sandflies	IRS: recom- mended insecticide Aerosol/space sprays Insecticide- treated materials	Compression sprayers Atomized canisters/ cold or thermal foggers Long-lasting insecticidal nets/curtains; wall linings	-3 rounds or as per national guidelines As necessary, when evidence of infestation is evident Regular use. For personal protec- tion from sandfly bites and sandfly control	All persons spraying or handling insecticides to wear approved PPE; dosage of insecti- cide as specified by national control programme Read label for pesticide safety precautions and antidotes
Houseflies	Sanitation Window screens Electrocution	Keeping PoE clear of organic garbage is the best way to prevent housefly breeding. Prevents flies to get access indoors and to food area Mount electrocution gad- get on kitchen/dining area, UV attracts and the grid electrocutes the flies	At all times Ensure proper onetime installa- tion but maintain regular upkeep Regular basis When necessary	Avoid food/utensil contamination / co- ver all food contact surfaces/Keep the treated area out of bound for at least half an hour. Read label for pesticide safety precautions and antidotes
	Poison baits	There are poisonous sugar baits that attract and kill flies	When infestation is acute and source identified	Rodents gnaw fibreglass nets hence iron mesh may be preferred

Vector	Recommended insecticide/ formulation	Application method	Frequency of application	Precaution
	Focal insecticide sprays (should be avoided as much as possible)	Compression pumps		
Cockroaches	Dust	Boric acid is a well accepted weapon against cockroach infestations. It may be formulated in a bait or as a residual that cockroaches walk through. When a cockroach walks through a treated area, boric acid powder adheres to its legs and body. Upon grooming itself, the cockroach ingests the boric acid dust	Depending upon seriousness of infestation. If ootheca are detected or remain adhering even after the cockroaches have been eliminated, the population builds up rapi- dly from hatching	Boric acid dust has to be applied carefully in areas ge- nerally frequented by cockroaches to be effective.
	Aerosol	2% aerosol formulation of d-phenothrin or recom- mended formulation of pyrethrins apply in cracks and crevices using extender tip	nymphs from ootheca	contamination / co- ver all food contact surfaces before treatment. Keep the treated area free of people for at least
	Bait pesticide	Keep baits in area frequented by cockroaches. Gel baits are available with a variety of active ingre- dients, several of which can work on cockroaches. The cockroaches consume the bait and are killed by the active ingredient		30 minutes.

\*Note: Choice of insecticide is as per country registration/label specification/dosage recommended by national programme.

# Annex 10. Disinsection of aircraft

# Requirements and assessment indicators of WHO-recommended application of disinsection products.

Method of application	Requirements for application	Assessment methods and indicators
insecticide applied before		Cage bioassays: assess knockdown 60 min after application and mortality 24 hr after exposure to insecticide
Pre-flight spraying	action and limited residual action (at least within 1 h before the doors are closed after boarding)	Cone bioassays on treated surfaces 1 hr after application for 30 min: Assess knockdown 60 min after application and mortality 24 hr after exposure to insecticide
Blocks-away	Aerosol applied before take- off and after doors are closed, for rapid action	Cage bioassays: Assess knockdown 60 min after application and mortality 24 h rafter exposure to insecticide
Top-of-descent	Aerosol applied as aircraft starts its descent, for rapid action	Cage bioassays: Assess knockdown 60 min after application and mortality 24 h rafter exposure to insecticide
Insecticide applied by com- pression sprayer for long-term residual activity on aircraft		Cone bioassays on treated surfaces, each for 30 min: Assess knockdown 60 min after application and mortality 24 hr after exposure to insecticide
Residual application	interior surfaces	One day after application and then at regular intervals (e.g. weekly) until mortality within 24 hr is <80%

Source: (WHO, 2012a).

Annex 11. Model ship sanitation control exemption certificate/ship sanitation control certificate (Annex 3 of The International Health Regulations)

Port of ..... Date: ......

This Certificate records the inspection and 1) exemption from control or 2) control measures applied Registration/IMO No. ..... At the time of inspection the holds were unladen/laden with ..... tonnes of ...... cargo ......Flag ..... Name of ship or inland navigation vessel .....

**Comments regarding** 

conditions found

_													-	_				_		_	
<b>Re-inspection</b> date																					
Control measures applied																				: : : : :	
			_			_								_	_	_				-	
Documents reviewed	Medical log	Ship's log	Other																		
Sample results <sup>2</sup>																					
Evidence found <sup>1</sup>																					
Areas, [systems, and services] inspected	Galley	Pantry	Stores	Hold(s)/cargo	Quarters:	- crew	- officers	<ul> <li>passengers</li> </ul>	- deck	Potable water	Sewage	Ballast tanks	Solid and medical	waste	Standing water	Engine room	Medical facilities	Other areas specified - see attached	Note areas not applicable, by marking	N/A.	
	Evidence found <sup>1</sup> Sample         Documents reviewed         Control measures applied           results <sup>2</sup> results <sup>2</sup> results <sup>2</sup> results <sup>2</sup> results <sup>2</sup>	[systems, and evidence found <sup>1</sup> Sample Sample         Documents reviewed         Control measures applied           ces] inspected         results <sup>2</sup> Medical log         Evidence	[systems, and evidence found <sup>1</sup> Sample Sample     Documents reviewed     Control measures applied       ces] inspected     results <sup>2</sup> Medical log        Ship's log     Ship's log	Isystems, and     Evidence found <sup>1</sup> Sample     Documents reviewed       cesh inspected     results <sup>2</sup> Medical log       cesh inspected     sinp's log       cesh inspected     Other	Isystems, and cell inspected     Evidence found <sup>1</sup> Sample     Documents reviewed       cell inspected     results <sup>2</sup> Medical log       cell inspected     results 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Other        Notargo           Notargo          Notargo	

No evidence found. Ship/vessel is exempted from control measures. Name and designation of issuing officiar

Control measures indicated were applied on the date below.

Date ...

Signature and seal ..... Name and designation of issuing officer .....

<sup>1</sup> (a) Evidence of infection or contamination, including: vectors in all stages of growth; animal reservoirs for vectors; nodents or other species that could carry human disease, microbiological, chemical and other risks to human health; signs of inadequate sanitary measures. (b) Information concerning any human cases (to be included in the Maritime Declaration of Health).

<sup>2</sup> Results from samples taken on board. Analysis to be provided to ship's master by most expedient means and, if re-inspection is required, to the next appropriate port of call coinciding with the re-inspection date specified in this certificate.

Sanitation Control Exemption Certificates and Sanitation Control Certificates are valid for a maximum of six months, but the validity period may be extended by one month if inspection cannot be carried out at the port and there is no evidence of infection or contamination.

# Table A10-1 is a sample of an Evidence Report Form.

This form lists the evidence found, samples and documents reviewed, and control measures or corrective actions to be performed after a ship inspection, and supports the ship sanitation certificate (SSC).

When attached to the SSC, each page of this attachment needs to be signed, stamped and dated by the issuing authority. If this form is used as an attachment to a pre-existing SSC, this attachment must be noted in the SSC (e.g. by using a stamp).

Evide	nce Report Form										
This fo	orm supports the ship sani	tation certificate (SS	C), and provid	es a list	of evidence found and co	ontrol	measu	ures to be performed.			
When this de	attached to the SSC, eac ocument is used as an atta	ch page of this attac achment to a pre-ex	hment needs isting SSC, thi	to be s s attach	igned, stamped and date ment must be noted in t	ed by ne SSI	the co C (e.g.	ompetent authority. If by using a stamp).			
Ship's	name and IMO no. or re	gistration:			Name and signature of r	espor	nsible (	onboard ship officer:			
Name	e of issuing authority:	<u> </u>		Actual inspection date (dd/mm/yyyy):							
Date	of referred SSC (dd/mm/	уууу):			SSC issued in the port of:						
Indica	ate areas that <u>have not</u>	been inspected:									
Qua	rters	Galley, p	antry, service	area	Stores	nild-care facilities					
Med	lical care facilities	Swimmi	ng pools/spas	;	Solid and medical wa	ste	e Engine room				
Pota	ble water	Sewage			Ballast water	Ca	Cargo holds				
		Other (e	.g. laundry and	d washir	ig machine)						
Dete	cted heath events on bo	pard			Yes		N	о			
Evidence code	Evidence found (brief description accord checklist; draw a line und of evidence to ensure ite separated)	der each item	Measure to	be app	lied	Required	Recommended	Measure successfully performed (stamp and signature of re-inspecting authority)			
Name	e of issuing inspector:	Signature of issuin	g inspector:	Stamp	o of issuing authority: Page of						

#### Table A10-1. Sample Evidence Report Form

IMO, International Maritime Organization; SSC, ship sanitation certificate; WHO, World Health Organization. Source: (WHO, 2011). Reproduced with permission.



