FINAL REPORT
DIPA BIOTROP 2017

BIOLOGICAL CONTROL OF *ACACIA NILOTICA* SUBSP. *INDICA* BY USING BIO AGENT *CHIASMIA ASSIMILIS* (WARREN)

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(SEAMEO BIOTROP)
2017

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4. Research Schedule	: 10 Months
5. Research Budget	: Rp

Bogor, 1 December 2017

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ABSTRACT

The invasion of *Acacia nilotica* in Baluran National Park is threatening the integrity of the savanna ecosystems, the greater portion of the park. Recent works on the mechanical and chemical controls by cutting *A.nilotica* trees with a chainsaw and brusing the left stump with triclopyr (5-10% GARLON dissolved in diesel oil) killed the treated trees. When this is preceeded by spraying triclopyr (GARLON 2 lt/ha in 400 l of water mixed with 2% Agristick an adjuvant) the broadleaved weeds and sapplings, poles and smaller trees of *A.nilotica* and *Azadirachta indica*, will be controlled that helps to recover the domination of grasses in the savanna. However the seed bank of *A.nilotica* in the soil still has to be addressed acordingly. Application of herbicides to control seeds under the soil is difficult beside a possible environmental problems related to residual herbicides. Therefore, a set of research works on biological control of *A.nilotica* using *Chiasmia assimilis* to overcame the ever emerging seedling from the seed bank of *A.nilotica* were proposed. The biocontrol agent was imported from Australia, and reared at the laboratory of BIOTROP for further works on host specificity testing to estimate the host range of *C.assimilis*. According to a recent review genus Acacia being polyphyletic is split into new 5 genera, and *Acacia ni*lotica was not anymore an Acacia but *Vachellia nilotica*, taxonomically far from current genus Acacia, which was previously called Phyllodiniae. The Australian imported *C.assimilis*, the biocontrol agent, from Kenya collected *C.assimilis* from *A.nilotica* subsp. *leiocarpa* and *A.nilotica* subsp. *subalat*a. and from South Africa collected from *A.nilotica* subsp. *kraussiana* although *A.nilotica* subsp. subalata and *A.nilotica* subsp, *leiocarpa* were around and attacted the same C.assimilis. However colony of *C.assimilis* imported from Kenya did not establish in Australia, while that from South Africa established immediately and showed a significant impact by defoliating *A.nilotica* extensively in the northeat of Queensland state. This is the type of *C. as*simlis imported by BIOTROP to Indonesia. When tested on 60 plant species with host specificity testing with and without choices proved that host range of *C.assimilis* in Indonesia was very narow in fact it was able to complete its life cycle only on *A.nilotica*. It is expected that with this result *C.assimilis* is allowed to be released in the field by the the Minister of Agriculture of Indonsia.

Keywords : Biocontrol agens, *Acacia nilotica, Chiasmia assimilis* Warren Lepidoptera-Geometridae.

CONTROL OF *ACACIA NILOTICA* SUBSP. *INDICA* BIOLOGICALLY USING ITS NATURAL ENEMY *CHIASMIA ASSIMILIS* (WARREN)

I. INTRODUCTION

I.1. BACKGROUND

A. nilotica was planted as a fench along the savana Bekol bordering with the adjoining teak plantation in 1969 to prevent savana fire from encroaching into the teak plantation. The effort of preventing savanna fire from encroaching the teak forrest is successful, and *A. nilotica* grows prolifically to produce a substantial amount of pods. *A. nilotica* flowers during the wet season and the pods ripes during the dry season. The riped pods of *A. nilotica* falls on the ground during the dry season when grasses are dried out, and eaten by big herbivores banteng (*Bos javanicus*- indigenous cattle), buffaloes, and dears. The pod of *A.nilotica* is nutricious , containing high concentration of protein, carbohidrtae and lipids. However seeds of *A. nilotica* is not digested by the digestive tract of those big herbivores, the seeds remain viable and are excreted together with their feces. The excreted seeds enjoy a good growth media with a relatively moist environment, rich of nutrients from decomposed organic matter. Those herbivores during the dry seasson move to seek water and vegetation shade away from direct sunlight. In this way those herbivores spread *A.nilotica* seeds around the park. With this symbiotic mutualism, where herbivores feed on nutricious pods and *A.nilotica* is benefitted from spreading its seeds around the park rapidly. In 2013 *A. nilotica* invaded aproximately 6.222 ha (Setabudi, *et.al.* 2013), and *A. nilotica* canopy coverage varied from very dense (> 3000 tree/ha) to rare (< 50 batang/ha). *A. nilotica* canopy coverage inhibits the growth and reduces the biomass production of grasses , reduces the availability of herbage for herbivores, and facilitates the growth of broadleaved weeds . *A.nilotica* invasion not only reduces the growth of grasses but also alters savanna ecosystems into that of shrubs. Broadleaved weeds in the park are benefitted from the current climatic change. For example the elevation of CO_2 concentration in the atmosphare benefits them as they are C3 plants, where the photosynthetic enzime mainly Rubisco catalizes more fixation of CO_2 under higher atmospheric CO_2 concentration, therefore the photosunthetic rate may increase. They are able to survive and grow under the shade of *A.nilotica,* because with their relatively wide leaf surface they are still able to intercept a good amount of light. Savanna grasses such as *Dicanthium caricosum*, a C4 plant adapted to growing under the sun compates out those broadleaved weeds , and for so long the savanna has been dominated by grasses. However, under the shade of *A.nilotica* trees, this C4 grass does not obtain an optimum amount of sunlight and is not benefitted from

the elevation of CO_2 as inherently it has a menchanism to concentrate CO_2 concentation, any atmospheric increase of CO_2 concentration will not be responded as effective as that of C3 plant, and this grass is currently competed out by broadleaved weeds such as *Thespesia lampas*, *Bidens bitternata*, *Vernonia cymosa*, and others. The savanna condition is changing rapidly, perennial grasses that used to grow in the savanna are competed out by broadleaved weeds, and grass species are replaced by less palatable grasses, adapted to shade of plants such as *Brachiaria reptans*, *Oplesmenus burmanii.* It is indeed a bad omen for the fate of this only savana in Java island.

1.2. OBJECTIVES

A mechanical combined with a chemical cotrol successfully kills trees of of *A.nilotica*. A tree of *A.nilotica* was cut with a chainsaw and the stump was soon brushed or sprayed with triclopyr (10% of Garlon diluted in diesel oil) killed the treated tree. It is suggested that before killing the tree it should be preceeded with controlling shrubs or other broadleaved vegetation using triclopyr (as 1-2% Garlon) diluted in 400 lt/ha with 2% Agristick a non-kathionic adjuvant. The later treatment was effective not only broadleaved vegetation but also on young *A,nilotica* or *Azadirachta indica*. These combined treatments are actually a form of integrated control for *A.nilotica* in this Baluran National Park. If all activities embodying the integrated control of *A.nilotica* are carried out conrectly the standing *A.nilotica* vegetation will be well controlled and killed, leaving grasses which are not impacted by triclopyr. However seeds of *A.nilotica* and broadleaved weeds are still numerous under the ground. Some of seeds will die under the ground including that of that of *A.nilotica* will germinate and develop into sapling, pole and tree, and the cycle of *A.nilotica* invasion will be repeated. It is the objective of this research to complete the integrated control of *A.nilotica* using biological control component directed toward the growth of seedling from seeds under ground using its natural enemy *Chiasmia assimilis* . The detail of the objectives are:

1. To import *Chiasmia assimilis* a biological control agent of *A.nilotica* from Australia
2. To rear *C.assimilis* on *A.nilotica* and to carry out host specificity testing to find out the host range of the
3. To report the result to the Commision of Biological Agent to get the release permit in the field

1.3. EXPECTED OUTPUT

When the standing crop of *A.nilotica* is controlled leaving only a limited number of poles of *A.nilotica* the remaining seeds underground when germinate and emerge above the ground will soon be attacked by *C.assimlis*. Broadleaved weeds will still be in the field, but in open condition perennial grass will compete out those seedling of those broadleaved weeds. The management of savanna will be important to ensure the doimance of grasses and reduce the population of broadleaved weed. It is necessary to release this *C.assimilis* persistently until it is able to establish in the field and carry out its job to control *A.nilotica* as a biological control component under an integrated approach

II. THE BENEFIT AND IMPORTANCE OF RESEARCH WORKS

Baluran National Park consists of mostly savanna, and the only savanna ecosystems in Java island. This savanna is the home of banteng (*Bos javanicus*) an endemic herbivore of Jawa island recognised by IUCN and known as "World Heritage", therefore, the Indonesia government is responsible for guarding the sustainability of Baluran NP ecosystems. In term of the national interest, Baluran NP is very important. Bali cattle is known domesticated from banteng, and this cattles has been raised in Indonesia extensively as beef cattle, mainly in the eastern part of Indonesia region. Bali cattle has formed the main cattle husbandary in Indonesia providing an great number of beef cattle to Java island (mainly Jakarta) since the Indonesia independence up to 2000. The cattle systems had been developed since the colonial time, up to the end of 20th century where only a certain type of cattles was allowed to be exported out (mainly to Jakarta). Unfortunately the systems were not appropriately maintained. After the year of 2000 almost all type of cattles were allowed to be exported and the Bali cattle experinced genetic deterioration leading to the reduction of its productivity, therefore could not compete against imported Brahman from Australia. Recently experts in cattle husbandry suggest that Bali cattle can be improved genetically through crossbreeding with male banteng and female Bali cattle through artificial insemination, as the number of banteng bull is limited. Thus, nationally as well as internationally banteng and its ecosystems of Baluran National Park, are apt to be conserved. This research is dedicated to support the goverment effort to rehabilitate and restore the ecosystem of Baluran National Park as a home of banteng to function optimally.

III. LITERATURE REVIEW

3.1. *Acacia nilotica*

Acacia genus covers 1350 species distributed in tropical and subtropical regions found in all continents accept in Antartica. The recent research discoveries on molecular phylogenetics revealed new facts and understanding that Acacia is *polyphyletic*

Bentham (1840) stated that for the period of 80 years after the description by Miller (1754) *Acacia* was so diveres and the definition used was so inconsistent, therefore, producing a genus as a collection of plants with a great variations and they were polyphyletic. Bentham (1842) improved the definition by limiting *Acacia* only under Mimoseae with independent, indefinite stamen, this definition is still utilized up to now. A series of articles was published and culminated in 1875 as a *magnum opus* (Bentham,1875) not only redefining *Acacia*, but also improving internal classification . Following 60 years further , 15 genera were described as congeneric of *Acacia sensu lato*.

In 1986 Pedley published a reassessment of *Acacia* classification deviding the genus into 3, *Acacia sensu lato*, *Senegalia* and *Racosperma* (DC) Mart. These 3 genera were corresponded with subgenera of *Acacia*, *Aculeiferum* Vassal, and *Heterophyllum* Vassal, (=subgenera *Phyllodiniae*, (DC) Seringe). At that time classification of Pedley was less appreciated by experts considered as inconclusive and incomplete. However since 1986 an extensivw comparative molecular studies based on the analyses of chloroplast and nucleus DNA provided convincing evidence to decide a strong informative decission on the phylogenetic and classification of *Acacia*. The studies included not only *Acacia sensu lato* and *Faidherbia* A.Chev. (together including *Acacieae* tribe) a more important one, also included representation of *Ingeae* and *Mimoseae* . tribe . These studies demostrated that *Acacia sensu lato* was *polyphyletic* consisting of 5 monophyletic groups which each warrant recognition as a distinct genus. These 5 genera are :

1. Two small endemic genera of new worlds, *Acaciatella* (resurrected by Rico Arce and Bachman, 2006) and *Mariosousa* (new genus described by Siegler et al, 2006).
2. Two relatively large genera of pan-tropical region, *Senegalia* (resurrected by Pedley, 1986) and *Vachellia* (containing the species tipe, *Acacia nilotica* (L.) Willd ex Dell and
3. Enormous genus mostly Australian, *Acacia sensu stricto* (Syn. *Racosperma*)

Vachellia is taxonomically well removed from other genera in *Acacia sensu lato*, being nested within a paraphyletic of tribe Mimosease; there are 2 subclades in *Vachellia*, i.e. one species from Africa, and another from Amerika to which *V achellia farnesiana* (L.) Wight & Arn. belongs. The other four genera are in a paraphyletic grade within genera of *Ingeae* tribe. It is this 5-genera classification of *Acacia sensu lato* which is accepted to day In addition to phylogenetis evidence based on nucleotide data, a range of morphological, biochemicals, and palinologicals data and other data all supported the classification of 5 genera in *Acacia sensu lato*.(Maslin et al. 2003).

After a long debat toward the proposal of Orchad and Maslin (2003) to replace the original type of *Acacia,* African species *A.scorpioides* W.F. Wight & Arn.(= *A.nilotica*), with a new type, the Australian species, *A.penninervis* Sieber ex DC was approved under *Vienna Code*, during 17th International Botanical Congress in Melbourne, that the name of *Acacia* was conserved. A major changes occured in the classsification of *Acacia*. The name *Acacia* applies to a group of Australian Acacia previously called *Acacia* subgen *Phyllodineae*, now is called as *Acacia sensu stricto* and *Racosperma* is its synonim. While *Vachellia* is the correct name for small pantropical Acacia previously called *Acacia* subgen. *Acacia*. Almost all combination of species under *Acacia sensu lato* may be put into any one of genus *Acaciatella, Mariosousa, Senegalia* and *Vachellia*, accept 13 species (10 from Madagaskar, 3 from Afrika). However, it is still possible to have further generic segregaron in the *Acacia sensu lato*, as there have been new taxa being added to the genetic dataset. Miller &Siegler (2012) has suggested to have another genus separated from *Senegalia* to accomodate the group of *S. skleroxyla* (Tussac) Seigler & Ebinger for example.

In East and Southeast Asia there are 52 species *Acacia sensu lato*, under 3 genera , i.e. *Acasia sensu stricto, Senegalia* dan *Vachellia.*

In the world there are 1073 species of *Acacia sensu stricto*; 1063 of which are in Australia, forming the biggist genus of vascular plants in the continent. Nineteen taxa (representing 18 species) are outside Australia. Twelve in Asia and 7 in Pacific isalands. The distribution of 12 species in Asia geographically is very limited, only one in East Asia, *Acacia confusa* Merr from Taiwan Philippines and 11 in Southeast Asia. There are 4 endemic taxa, 3 in the southern region, where *Acacia sp* (Wetar) , and *A.wetarensis* Pedley exist only in the island of Wetar, Indonesia and *A.peregrinalis* M.W.McDonald & Maslin is only in Papua New Guinea and the other one *A. confusa* in northeast of the region the Philippines and Taiwan.

Eight other species grow outside but also do inside Australia. No *Acacia sensu lato* species grow in Cina exccept those under cultivation. *Acacia sensu lato* species indigenous to East and Southeast Asia are shrubs or trees with phillodinous foliage (bipinnate foliage on some introduced species). Like *Vachellia* they grow in drier habitat than that of *Senegalia*. Some taxa become a component of forest industry in Southeast Asia mainly Indonesia and Vietnam such as *Acacia crassicarpa* A.Cunn. ex Benth., *A.mangium* Willd. dan *A.auriculiformis* A.Cunn. ex Benth.*x mangium* planted for pulp production.

Senegalia in the world comprises 201 species, distributed pantropically. There are 45 species in Asia (India 19 species), Afrika 68, Amerika 102 and 2 species in Australia.Thirthy eight *Senegalia* are indigenous taxa for East and Southeast Asia reprsenting 32 species; these are the largest group of *Acacia sensu lato* in East and Southeast Asia. Thirteen taxa are in East Asia, 4 of which, *Senegalia delevayi* (Franch.) Maslin et al., (var. *delavayi* and var. *kunmingensis* .C.Chen & H.Hunsen) Maslin *et al.*, *S. teniana* (Harms) Maslin *et al.* and *S.yunannensis* (Franch) Maslin *et al* are endemic to Cina. Thirty three taxa occur in Southeast Asia, almost half of them are indigenous to the region, mainly *Senegalia borneensis* (I.C.Nelson) Maslin *et al*, *S.comosa* (Gagnep) Maslin *et a*l, *S. donnaiensis* (Gagnep) Maslin et al., *S. kekapur* (I.C.Nelson) Maslin *et al*, *S.kostermanii* (I.C.Nelson) Maslin et al., *S.meelbodi* (Craig) Maslin et al., *S.merrillii* (IC Nelson) Maslin eta al., *S.palawanensis* (IC Nelson) Maslin eta al., *S.pluriglandulosa* (Verde.) Maslin et al., *S.pseudoinstia* (Miq.) Maslin et al., *S.sulittii* (I.C.Nelson) Maslin et al, *S. tawitawiensis* (IC Nelson) Maslin eta al, *S. thailandika* (I.C.Nelson) Maslin et al, dan *S.verheiyenii* (I.C.Nelson) Maslin et al. These endemic taxa are scattered throughout the region, except in Brunei, no endemic *Acacia sensu lato* was recorded. A subregion with the highesst concentration of endemic *Senegalia* is Indonesia with 9 species although only *Senegalia kostermanii* and *S.verheiyenii* are found only in Indonesia, while the others are also found outside Indonesia. Almost all other 16 *Senegalia* species extend beyond the southeast Asian region, wesstward are found in India, and the surrounding countries, with 10 extend eastward to China. Only one taxon extends accros Australia *Senegalia pennata* subsp.*kerrii* (IC Nelson) Maslin et al, which has a very poor representation of indigenous *Senegalia* , only *S. albizioides* Pedley dan *S pennata* subsp.*kerrii.* Species *Senegalia* are characterised with bipinnate leaves and cauline prickles. Most of the indigenous *Senegalia* in East and Southeast Asian are woody lianas; those distributed widely are *S.catechu* (L.f) PJH Urter & Mabb.; *S.tundra* (Robx & Rottler) Maslin and *S.ferruginea* (DC) Pedley Maslin (all of which have prickles in their nodes), while those with limited geographic distribution are

S.kostermanii from Indonesia and *S.teniana* from China (both of which have sctattered prickles) seemingly obligate shrubs or trees which lack of scandent branches. Most species have broad, flat, chartaceous, and straight pods, but in *S.thailandica* , the pods are slightly inflated, and tightly curled. These species can be arranged in 4 subgroups (defined principlely by prickle distribution and leaflet characteristic) centred on *Senegalia pennata*, *S.caesia*, S. *andamanika* (IC Nelson) Maslin *et al* and *S.catechu* . The one species with different corpological feature is the widespread and variable *Senegalia rugata* (Lam.) Briton &Rose (syn. *Acacia concina*) . In this species the pods are smooth, thick and fleshy when fresh, but when dry become wrincled blackish with very hard texture. This species name has about 10 heterotypic synonyms and further study is needed to asses the taxonomic status of *Senegalia rugata*. *Senegalia albizioides* (in Australia) currently is the only recognised close relative to *S.rugata*

Vachellia genus in the world comprises 163 species, distributed pantropically and Asia has 30 species (India 11), Afrika 84, while America has 57 and 8 in Australia. In the Southeast asian region there are 8 species (non in East Asia), making it the least represented in the region among the 3 species (*Acacia sensu srticto, Senegalia* and *Vachellia*). The term East Asia is defined to include only Taiwan and China, while Southeast Asia is defined to cover Papua Timur, Indonesia, Filipina, Vietnam, Kamboja, Laos, Singapura, Malaysia, Thailand dan Myanmar. The are 5 endemic species in the northern region, 3 restricted to Myanmar, *Vachellia inopinata* (Prain) Muslin et al., *V. kingii* (Prain) Muslin et al., dan *V. Myangii* (Lace) Muslin et al, all of these species are poorly known, one in in Thailand V, siamensis (Craib) Maslin et al., and widely distributed *V. harmandiana* (Pierre) Maslin et al., in Camboja, Laos, Thailand dan Vietnam. The tree species that extend beyond the region are namely *Vachellia nilotica* subsp. *indica* (Benth.) Kyal. &Boatwr., *V. leucophloea* (Roxb) Maslin et al., dan *V. tomentosa* (Rottler) Maslin et al, that extends westward, where *V. nilotica* subsp. *indica* (Benth.) Kyal. &Boatwr., extends to Middle East. Excluding *Vachellia harmandiana* dan V. *leucophloea* only *V. tomentosa* that is widely distributed in the Southeast Asian region. *Vachellia farnesiana* (L.) Wight.Arn. was reported as introduction (somtimes naturalisation) in several countries this species is native to America. The indigenous Vachellia in the Sotheast Asian region is characterized by bipinnate foliage subtended by spiny stipule.

When scrutinised although it is stated that all combination under *Acacia sensu lato* may be accomodated in 5 genera (*Acaciatella*, *Mariosousa*, *Vachellia, Senegalia* and *Acacia sensu stricto*), but species of *Acacia* in Africa have not been treated accordingly, While the name of

Vachellia nilotica is utilised in this report, we are still utilising the term of *Acacia nilotica* growing in Baluran National Park as stated by Wuliarni-Sutjipto& Lemmens (1991) that it is *A.nilotica* (L.) Willd. Del subs. *indica* one of nine subspicies as reported by Africa (Brenan. 1983). The nine subspecies *Acacia nilotica* (L.) Willd. Ex Del are :

1. subsp. *nilotica*
2. subsp. *indica* (Benth.) Brenan (1957)
3. subsp. *cupressiformis* (J.L.Stewart) Ali &Faruki (1969)
4. subsp. *tomentosa* (Benth.)Brebab (1957)
5. subsp. *adstringens* (Schumach & Tonn) Huberty (1949)
6. subsp. *subalata* (Vatke) Brenan (1957)
7. subsp. *kraussiana* (Benth.) Brenan (1957)
8. subsp. *leiocarpa* Brenan (1957)
9. subsp. *hemispherica* Ali Faruqi (1969)

A.nilotica is a complex species, it may be may be due to a possible inbreed among the subspecies, for example *A.nilotica* subsp. *indica* may interbreed with *A.nilotica* subsp. *hemispherica* giving Acacia having characters similar to that of *A.nilotica* subsp.*adstringens* and that of *A.nilotica* subsp.*subalata* (Ali & Qaiser, 1980)

3.2. *CHIASMIA ASSIMILIS* (WARREN) (LEPIDOPTERA: GEOMETRIDAE)

Although *A.nilotica* was imported to Australia from India in 1840's the biological control of *Acacia nilotica* was initiated only in 1970's. Apparently in that period *A.nilotica* did not cause any problem to the farmers. The animal husbandary was dominated by sheep raising. *A.nilotica* did not grow excessively because *A.nilotica* seedlings were grazed by sheeps and sheep does not distribute *A.nilotica* seeds through the feces as extensive as cattle does. When the animal husbandary was shifted from that of sheep raising to that of cattle breeding, the spread *A.nilotica* become very rapid. Mature dropped *A.nilotica* pods were eaten by cattles, while the *A.nilotica* pod was nutricious, with high carbohydrate and protein content, the *A.nilotica* seed was not damaged in the digestative tract of cattles. The seeds were excreted through cattle dung. Those seeds enjoy a good media growth for germination and further growth, and *A.niltica* growth spread rapidly. In 1970an *A.nilotica* invaded more than 7 million ha of Mitchell pasture northeast od Queensland, and chemical control was becoming too expensive.

Biological control was initiated by Queensland Department of Land, with an extensive exploration to find natural enemies of *A.nilotica* since 1980's in Pakistan, 1987 in Kenya,

Afrika Selatan up to 2012 in India, by establishing local field laboratoiesy in each country. From Pakistan the exploration discovered 43 species of phytopgagous insects, and 16 was valued as stenophaous (Mahyudin, 1981). In Kenya the exploration collected insects from *A.nilotica* subsp. *subalata* (Vatke) Brenan (1957), and *A,nilotica* subsp. *leiocarpa* Brenan (1957) up to 1992. Here it was collected 90 phytophagous insect species that attack *A.nilotica* but only a few species considered to be suitable as a bilogical control agent. In 1990's the exploration was also conducted in South Africa and the faunal survey was directed toward *A.nilotica* subsp. *kraussiana* (Benth.) Brenan (1957). In cooperation with entomologist from ARC Plant Protection Research Institute of Pretoria the survey produced more than 600 arthropod species (Stals et al, 2004). In 1999 field laboratory of Queensland Department of Land was constructed to strengten the survey of biological control candidate led by A.Witt. In India th survey was targeted on *A.nilotica* subsp. *indica* started in 2008, so far collected 77 insect species and 14 pathogens from Tamil Nadu dan Karnataka, as well as 14 insect species and 11 pathogens from Gujarat and Rajaratnam in Northwest India .

From those extensive exploration of biological control agents in India, Pakistan , South Africa and Kenya, and experinces from the field release in Australia (Queensland) the most successful biocontrol agent for *Acacia nilotica* is *Chiasmia assimilis* Warren) (Lepidoptera: Geometridae)

Chiasmia is a genus of the old world consisting of 265 species distributed in the eastern Palaearctic, Afrotropika and Indo-Australia region(Scoble& Kruger, 2002). Hosts are known for on;y 26 from 178 species from Afrotropika and mostly Leguminosae, mainly various terutama berbagai jenis *Acacia* species (Kruger, 2001). Other plants reported from Africa as its host plants are *Tamarix* (Tammaricaceae), *Sterculia* (Sterculaceae) dan *Mallotus* (Euphorbiaceae). No congender considered as a pest of Leguminosae plants (Singh, 1990) however C.*clathrata* is considered as a pest on *Medicago sativa* and *Trifolium spp*.di Europe (Zang, 1994). Four *Chiasmia spp*. are known associated with *Acacia nilotica*, (1). *Chiasmia streniata* (Guenee) known from i *A.nilotica* (presumably *A.nilotica* subsp.*indica*) in India (Browne, 1968) and also known to occur on *Acacia, Hippobromus* dan *Albizia* (1Kruger, 2001) in Afrika. (2). *Chiasmia farcata* (Warren) ia a species of Sothern Africa on *A.nilotica and A.karoo* Hayne (Kruger,2001). (3). *Chiasmia assimilis* (Warren) [= *Semiothesa assimilis* (Warren)] discovered in souther Africa from Dursban to norther Zimbabwe, East Afrika, to Cameroon. This was collected only from *A.nilotica* subsp. *kraussiana* (Benth.) Brenan, but all three are common here and are all attacked by C.assimilis, *A.nilotica* subsp. *subalata* (Vatke) Brenan and *A.nilotica* subsp. *leiocarpa* Brenan. (4). *Chiasmia inconspicua* (Warren) [=

Semiothesa inconspicua (Warren)]. *Chiasmia assimilis* dan *C.inconspicua* were collected from *A.nilotica* subsp. *subalata* dan *A.nilotica* sbsp. subsp. *leiocarpa* in Kenya, growing near Voi, Coast State, Kenya ($3^0$24'S; 38^0 35'E) in Mei 1997. The collected larvae were airfreighated to Brisbane where they were housed in the laboratorium fasilities of Alan Fletcher Research Station, with cut foliage *A.nilotica* or potted young *A.nilotica*. There were 20 generations successfully developed during the host specificity testing before being release to the field.

The second importation was carried out by collecting larva of *Chiasmia assimilis* in South Africa in 2002, from *A.nilotica* subsp, *kraussiana* (Benth) Brenan (Wardill et al, 2004). This secomd importation was established under Tropical Weed Research Center, constituting the second coloni of *Chiasmia assimilis* .

The performance of *C.assimilis* in the field reported from Australia is very imteresting. The first colony was collected from *A.nilotica* subsp *leiocarpa* dan *A.nilotica* subsp. *subalata* in Kenya. The female imago oviposited 400- 500 eggs. The larvae feed on cut foliage of *A.nilotica* in 2-3 weeks before pupating in soil or plant litter. The life cycle is completed approximaely within one month, so in one year it may have several generations. *C.assimilis* was tested for its host specificity on 74 local Australian plant species mainly *Acacia* genus, and shown a narrow range of host plants. It was shown that *C.assimlis* was able to complete its life cycle in 4 Australian *Acacia spp*. but the survival on these plants were insufficient to sustain population, and *C.assimilis* was approved for field release. There were about 74000 were released in the nortestern Queensland. However with those big number of biocontrol agent released , the agent failed to establish. That is why a second importation of *Chiasmia assimilis* was organised from South Africa collected from *A.nilotica* subsp. *kraussianan.* This colony when relesed in the field soon establised and its impact was observed not only attacking the mature plants but also attacking seedling of *A.nilotica.* juga menyerang semai , causing the semai succumb into slow death. It is this colony that has been imported by BIOTROP to Indonesia.

IV. METHODOLOGI

4.1. REQUEST OF PERMIT TO IMPORT *CHIASMIA ASSIMILIS* FROM AUSTRALIA

With a letter of SEAMEO BIOTROP dated 2 March 2017, No 194/DIPA.RES/III/2017 signed by the director of BIOTROP, Dr. Irdika Mansyur, a request was sent to Minister of Agriculture of Republic of Indonesia, for a permit to import the biological agent of *C. assimilis* from Australia. Agency of Plant Quarantine under the Ministry of Agriculture of Republic of Indonesia responded SEAMEO BIOTROP request with a letter dated 20 March 2017, No 4073/HM.110/K.3/03/2017 requiring further information needed for evaluating the request. Request for further information was soon fulfilled attached to a SEAMEO BIOTROP letter dated on 6 April 2017, No, 323/DIPA.RES/IV/2017. It seems the regulation to import biocontrol agent was slightly different from the prvious procedure, because currently bioagent is a commercial item, therefore , SEAMEO BIOTROP request was also considered as such. To our friends in the Agency of Plant Quarantine , we informed them that SEAMEO BIOTROP was nofor commercial purposes, the results of the research when proven specific will be released free in Baluran National Park , Situbondo, East Java. With the recommendtion of the Head of Commission of Biological Agent dated 8 May 2017, No.6042/KR.040/L/5/2017 the permit to import *Chiasmia assimilis* was was granted in the form of Ministerial decree by the Minister of Agriculture No. 358/Kpts/KR.040/6/2017 dated 6. Juni 2017. This permit is slightly different from the previous one dimanding that after the completion of research activities the colony of *C.assimilis* must be destroyed under the supervision of the quaraantine officer of Quarantine Agency of Sukarno Hatta Airport Cengkareng. It is rather disturbing because when arrived such insect colony may carry parasitoids which we the researchers have to clean them up before using for research activities. When importing again, a series of activities must be carried out again to ensure the colony is clean from unwanted contamination

4.2. PREPARATION TO RECEIVE THE PACKET OF *C.ASSIMILIS* COLONY

4.2.1.Laboratorium facilities

Research facilities comprise insectarium room and plants for host specificity testing. The insectarium is provided with an incinerator to destroy package materials for sending the colony of biocontrol agent. This special room is also provided with an air conditioning systems Panasonic 1,5 Pk and fan to keep the temperature optimum at around 25^0C ($23,5^0$- 32^0C. The door into the insectarium room is made dobble one is opened only if the other is closed.

4.2.2. The establishment of potted A.nilotica

Seed of *A.nilotica* collected from Baluran National Park is dorman, to germinate the seeds are dipped into a hot water for about 3 minutes and washed with cold water . The seed are then arranged in a big tray of rice husk cole saturated with water and covered with a sheet of plastic to keep the moisture approximately 100%. This tray is watered daily, and when the seeds start to lift their cotyledons the plastic coverage is removed and the seedlings are allowed to grow to reach 3 leaves. The seedling are then trasferred to plastic aqua glass containing a mixture of rice husk cole mixed with compost and put under the morning sun for 2 weeks before finally transferred to plastic pot having diameter of 17 cm and 14 cm height conaining a mixture of soil, compost and rice hust cole at the ratio of 3:1:1.

Figure 1. Plastic trays for germinating of *A.nilotica* after dipping into hot water and Plastic pots of growing seedling of *A.nilotica*

4..2.3. The process of receiving packet of biocontrol agent C.assimilis from Australia

The packet containg colony of biocontrol larvae *C.assimilis* was sent by Dr.M.Day, on Monday 11 Sept. 2017 from Townville to Brisbane. On 12 Sept.2017 the packet was sent to Indonesia through the expedition agent of Toll Priority Global (TPG), with an air bill No 813012682005. On Thusday 12 Sept. 2017 Saiful Bachri SSi. A member of the research team went to Sukarno Hatta international airport to tech the consignment. He was told that there was no Qantas flight from Australia on 12.09.2017. On the following day Wednesday . 13 Spt.2017 once again S.Bachri SSi went to Sukarno Hatta international airport to fetch the packet containing the colony of *Chiasmia assimilis* . Together with the staf of Quarantine Office at Sukarno Hatta international airport, Ir. Habihi, S. Bachri Ssi, were tracking the where about of the consignme17 nt; they found out that Quantas flight did not caary that consignment. Through internet tracking, https://online.toll.com.au/trackandtrace/ it was known that the consigment was carried by Singapore airlinr flight, and the packet was taken off and kept at the

store house DPEX at Changi airport with its status as “waiting for further instruction”. This unfavourable information was sent to Dr.M.Day, who contacted TPG to send the packet to Bogor, under a name of Dr Soekisman Tjitrosoedirdjo, mobilephone 0812 9843097. Subsequently the packet was sent to the international airport of Suta, Indonesia on Friday 15.Sept. 2017, with Air Asia flight , and the consignment was handled by the expedition agent of DPEX the cooperator of TGP. S. Bachri SSi together with Sukarno Hatta International Airport staf. Ir. Habibi contacted the Custom Officer and with his assistance they were able to locate the position of the consignment, i,e, at the store House of PT GARUDA under the management of TABITA. Considering that the content of the consignment was a colony of *Chiasmia assimilis* an insect surviving on fresh leaves of *A.nilotica*, it would be imperative to get the consignment out, to address the colony immediately to ensure its survival. Although it was very already late, 22.30 pm, S.Bachri Ssi and Ir Habibi come to the store house and asked earnestly to the officer to let them tahe the packet out, and the officer agreed. The packet was brought to the Quarantine Office at the international airport and after recorded was brought and transferred to BIOTROP insectatium at that night.

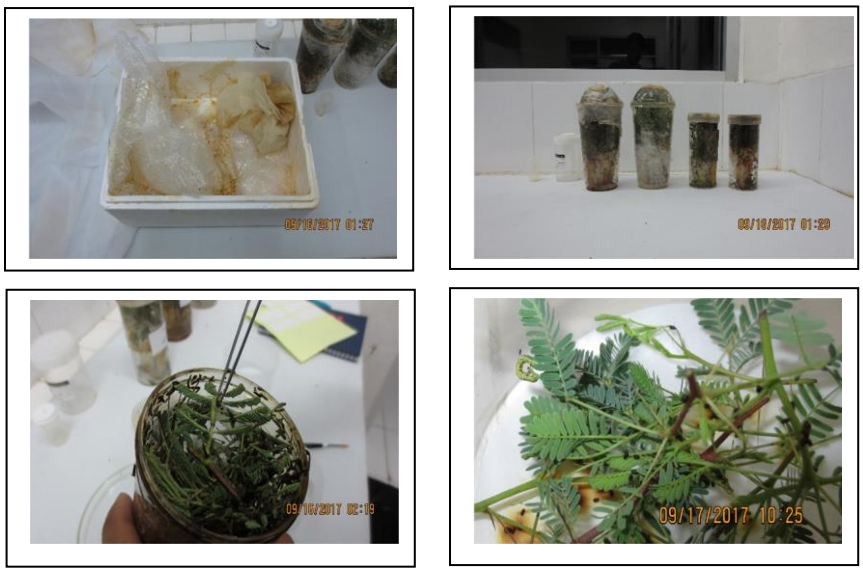

Figure 2. A. Sterofom box carrying the colony of *C.assimilis* sent through airfreightpertama. B. Glass plastic contianing *C. assimilis* larva from Australia. C. *C.assimilis* larva found still alive in the consignment; D *C.assimilis* larva maintained inthe glass plantic. When opened from the box

The packet, one sterofom box measuring 38 cm x 31 cm x, 22 cm , was opened at BIOTROP insectarium containing 2 big drinking plastic glass mesuring 19.5 cm height, and diameter of 9.25 cm and 2 small ones measuring approximately hal of the big one. One of the big glass was broken releasing water all over the whole box, however the cut leaves of *A.nilotica* was surprisingly remain fresh and the larvae were bussy eating the cut leaves.

Apparently the cut shoot of *A.nilotica* was dipped into a small plastic tube of film roller filled up with water and sealed. After a week of ordeal during transportation the casualty was 11 larvae died, 3 pupa damaged, and one imago died, while the rest of the container was burn in the incenerator..The larva were of various ages . Another larvae died, and the living larva were then transferred to the big plastic glass, before being transferred to the growing *A.nilotica* in pot. The 10 living larva left were kept on the young *A.nilotica* growing in pots, inside small cage and th results were presented in Tabel 1.

Tabel 1. The content of Colony *Chiasmia assimilis* under the first consignment

No	Glass	Larva content	Pupa	Notes
1	1	4	1	7 days later died , leaving 1 pupa
2	2	2	1	3 days later died , leaving 1 pupa
3	3	4	2	1 days later died, leaving 2 pupa
4	4	1(2)	2	4 days later died, leaving 1 pupa.

On 23 Sept. 2017 from the first packet, 5 pupa were obtained, after emerging 2 imago died leaving only 3 imago survived .

The second packet was hand carried by Dr. M.Day himself flying with Singspore airline flight that arrived at Sukarno Hatta International Airport at 7.45 am on 22 Spt. 2017. After going through. The custom procedure the packet was fetched by S.Bachri Ssi assisted by the quarantine officer at The international Airpot of Sukarno Hatta, Ir. Habibi and with Dr. M.Day were brought to the Insectarium, BIOTROP, Bogor. The second packet of *Chiasmia* colony, was packed differeently, in the first packet the big plastic glasses was broken, releasing water all over the whole box. The young shoot of *A.nilotica* was not dipped into a tube of film roller, but only wrapped with moisted tissue paper, and contained in an ordinary plastic boxes used to wrap cakes. The following day the larva were transferred to glasses of fresh young *A.nilotica* leaves and the rest of the packet were burned down in the incinerator.

From 88 larva 16 was found dead, and the rest of the larva were transferred to glasses of cut *A.nilotica* shoot and the following day were trasferred to living young *A.nilotica* growing on plastic pot.

Figure 3. rearing larva in the cages in Quarantine facilities SEAMEO BIOTROP

4.3.3.HOST SPECIFICITY TESTING .

4.3.1. Selection of test plants.

The host specificity testing in the framework of invasive alien plant species has undergoe a significant changes to be more efficient. In principle insect (biocontrol agent) for biological control purposes is monophagous so in the framework of host specificity testing it is examined if the insect is monophagous or specific attacking only the target species, however in practice it is more of investigating on the range of host plants. The interpretation whether the insect is sufficiently specific is if it is able to complete the life cycle only on the test plants, when it is able to complete the life cycle on other species it will be judged if the population developed can sustain on the species . It is understood that an interaction between insect and its host plant has gone very long along the evolution processes, where a particular insect has spent a considerable energy to be monophagous than polyphagous. In modern biosystematic a genus should be monophyletic, therefore, if a monophagous insect has an alternative host it will come from the same genus. Therefore, the selection of plants for host speccificity testing first will come from the close taxonomic relative to target species and systematically away and it is described as the centrifugal approach starting from the species outward. It is also important

to include plants living in the same ecosystems, or plants that are attacked by the same pathogens and of course crop plants that are planted in the sama area.

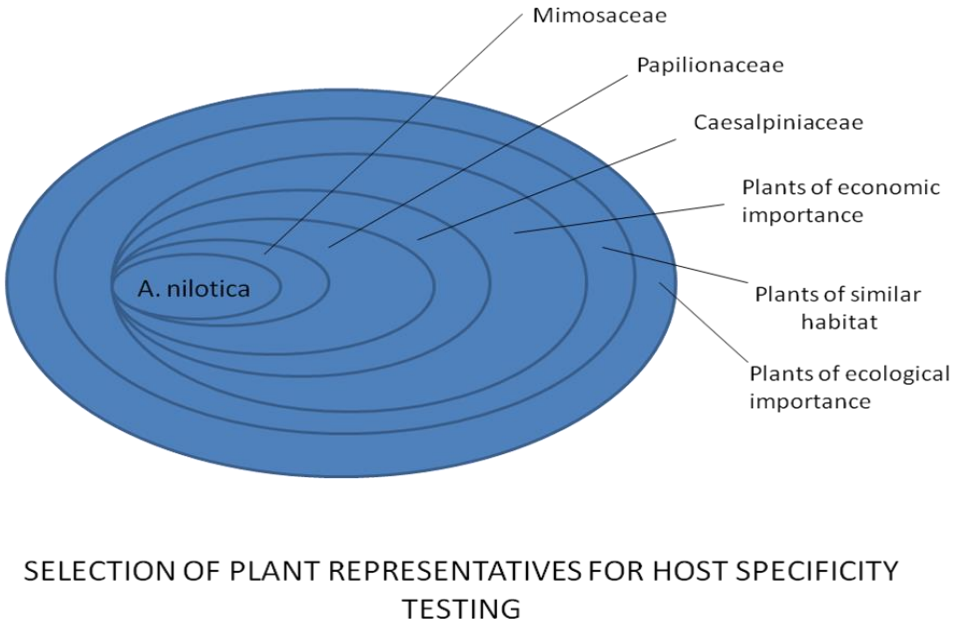

Figure 4. Diagram of centrifugal approach for the selection of test plant for host specificity testing.

4.3.2. Host specificity testing with choices.

The host specificity testing with choices was carried out using cut foliage of test plants in trays measuring 28 cm length x 20.5 cm width x 7 cm thick . The tray was covered with a lit that was holed at 6 x 13.5 cm^2 and covered with plastic gauze to facilitate exchane of air. This tray was lined with 1.5 cm thick moisted rockwool to provide enough moisture for the cut foliage and covered with thin plastic layer. The cut foliage tested were inserted into the moistened rockwoll arrange in such a way to be equal distance from the site of release of the larva. Nine tests were carried out, in the first 6 tests cut foliage of 10 different plant species one of which was *A.nilotica* as the control were arranged inside the tray, and replicated 3x. The three more tests only cut foliage of 5 different species and one of which *A.nilotica* as the control, were arranged in the tray and again replicated 3x. Five larva of 5 days old of *Chiasmia assimilis* were released into the centre of the tray. The evaluation was carried out at 6 days after after the release of the larva to see how severe the leaf damage on test plant compared to that on *A.nilotica*. If there was an indication of leaf damage from being eaten by the larva the experiment is continued to the experiment without choice.

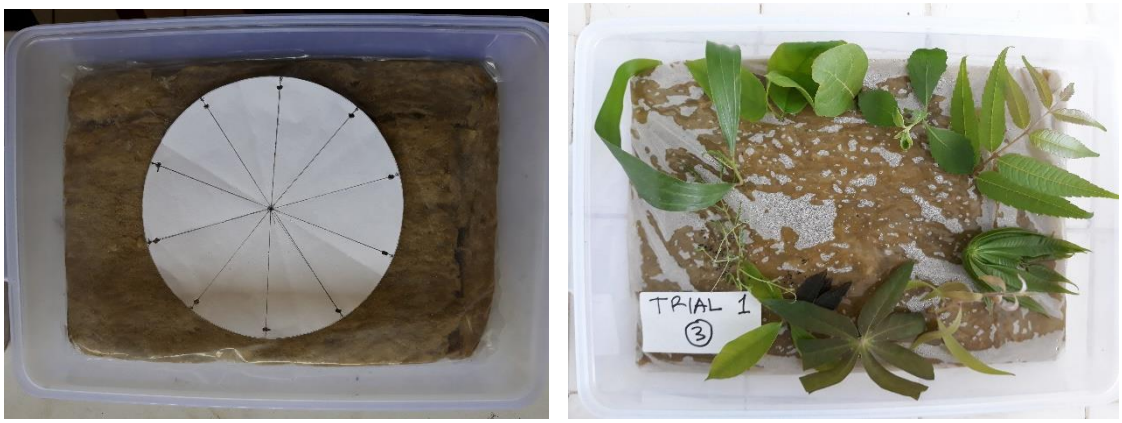

Figure 5. A.The position of cut foliage in the tray for host specificity testing with choices. B. The cut foliage have undergound selection of feeding by the larvae of *C.assimilis*, attacking only on *A.niotica*

4.3.3. Host specificity testing without choice

When there is an indication that the cut foliar is eaten by the larva of *C.assimilis* the Related plants are once again tested without host specificity testing without choices. This host specificity testing without choice uses potted living plants instead of cut foliage of test plants. The experimental design is similar to that of host specificity testing with choices, i.e. using 6 different plant species and replicated four time, and releasing 10 larva and kept until these larva are able to complete the cycle or dead along the life cycle. The evluation is carried out by comparing their survival of *C.assimilis* against its survival in *A.nilotica*.

Figure 5. set up for No choice using potted plants secured with plastic with 0.5 mm thickness

5. RESULTS AND DISCUSSSION

Data of the host specificity test of *C.assimilis* is presented in Table 2, it appered that the larva thrived on cut foliage of *Vachellia nilotica* (syn.*Acacia nilotica*), but refuced to eat most of the leaves of tested plant species. And died.

Tabel 2. Survival abiity of 3 days old *C.assimlis* larva on test plants. (n=3)

N0	Species Names	Local Name	D1	D 2	D 3	D 4	D5	D 6
Mimosceae								
1	*Vachellia indica* (L.) P.J.H. Hurter & Mabb. (Benth.) Kyal Boatwtr. (syn. *Acacia nilotica* subsp. *indica* (Benth) Brenan		+++	+++	+++	+++	+++	+++
2	*Vachellia leucophloea* (Roxb.) Maslin, Siegler Ebinger, syn. *Acacia leucophloea* (Roxb) Willd.	Pilang	0	0	0	0	0	0
3	*Vachellia tomentosa* (Rottler) Maslin, Siegler Ebinger, syn. *Acacia tomentosa* Willd.		0	0	0	0	0	0
4	*Acacia auriculiformis* Benth.		0	0	0	0	0	0
5	*Acacia mangium* Willd.	Mangium	0	0	0	0	0	0
6	*Adenanthera pavonina* L.	Saga	0	0	0	0	0	0
7	*Calliandra calothyrsus* Meisn.	Kaliandra	0	0	0	0	+	+
8	*Leucaena leucocephala* (Lam.) de Wit	Petai cina	0	0	0	0	0	0
9	*Parkia speciosa* Hassk.	Petai	0	0	0	0	+	+
10	*Pterocarpus indicus* Willd.	Angsana	0					
11	*Samanea saman* (Jacq.) Merr.	Ki hujan	0	0	0	0	0	0
12	*Arachis hypogaea* L.	Kac.tanah	0	0	0	0	0	0
13	*Glycine max* (L.) Merr.	Kac.kedelai	0	0	0	0	0	0
Papilionaceae								
14	*Vigna radiata* (L.) R.Wilczek	Kc. Ijo	0	0	0	0	0	0
15	*Phaseolus vulgaris* L.	Buncis	0	0	0	0	0	0
16	*Vigna unguiculata* (L.) Walp.	Kc panjang	0	0	0	0	0	0
17	*Cajanus cajan* (L.) Millsp.	Gude	0	0	0	0	0	0
18	*Pisum sativum* L.	Buncis	0	0	0	0	0	0
19	*Colopogonium cerelum*	Kalopo	0	0	0	0	0	0
20	*Clitoria ternatea* L.	Kembang Telang	0	0	0	0	0	0
21	*Pueraria phaseoloides* var. *javanica* (Benth.) Baker		0	0	0	0	0	0
22	*Centrosema pubescens* Benth.	Centro	0	0	0	0	0	0
23	*Mucuna bracteata* DC.	Benguk	0	0	0	0	0	0
24	*Flemingia macrophylla* (Willd.) Merr.	Otok otok	0	0	0	0	0	0

25	*Erythrina spermatica*	Kc.dadap	0	0	0	0	0	0
26	*Gliricidia sepium* (Jacq.) Walp.	Gamal	0	0	0	0	0	0
Caesalpiniaceae								
27	*Bauhinia purpurea* L.	Bunga kupu-kupu		0	0	0	0	0
28	*Caesalpinia pulcherrima* (L.) Sw.	Kembang merak		0	0	0	0	0
29	*Cassia fistulosa*			0	0	0	0	0
30	*Dalbergia latifolia* Roxb.	Sonokeling		0	0	0	0	0
31	*Delonix regia* (Hook.) Raf.	Flamboyan		0	0	0	0	0
32	*Tamarindus indica* L.	Asam jawa		0	0	0	0	0
33	*Caesalpinia sappan* L.	Secang		0	0	0	0	0
34	*Senna siamea* (Lam.) H.S.Irwin & Barneby	Johar		0	0	0	0	0
Asteraceae								
35	*Pluchea indica* (L.) Less.	Beluntas		0	0	0	0	0
Gombretaceae								
36	Terminalia catappa L.	Ketapang		0	0	0	0	0
Sterculiaceae								
37	*Sterculia foetida* L.	Kepuh		0	0	0	+	+
38	*Theobroma cacao* L.	Kakao		0	0	0	0	0
Meliaceae								
39	*Azadirachta indica* A.Juss.	Mimba		0	0	0	0	0
40	*Swietenia macrophylla* King	Mahoni		0	0	0	0	0
41	*Toona sureni* (Blume) Merr.	Suren		0	0	0	0	0
Acanthaceae								
42	*Avicennia marina* (Forssk.) Vierh.	Api api		0	0	0	0	0
Euphorbiaceae								
43	*Aleurites moluccana* (L.) Willd.			0	0	0	0	0
44	*Manihot esculenta* Crantz	Ubi kayu		0	0	0	0	0
45	*Hevea brasiliensis* (Willd. ex A.Juss.) Müll.Arg.	Karet		0	0	0	0	0
Boraginaceae								
46	*Cordia obliqua*	Kendal		0	0	0	0	0
Ramnaceae								
47	*Ziziphus jujuba*	Widoro bekol		0	0	0	0	0
Myrtaceae								
48	*Syzygium polyanthum* (Wight) Walp.	Salam		0	0	0	+	+
49	*Psidium guajava* L.	Jambu air		0	0	0	+	+
50	*Eucalyptus* sp			0	0	0	0	0
Sapindaceae								
51	*Schleichera oleosa* (Lour.) Merr.	Kesambi		0	0	0	0	0
52	*Nephelium lappaceum* L.	Rambutan		0	0	0	0	0

Malvaceae								
53	*Gossypium hirsutum* L.	Kapas		0	0	0	0	0
54	*Corchorus capsularis* L.			0	0	0	0	0
55	*Hibiscus rosa-sinensis* L.	Kembang sepatu		0	0	0	0	0
56	*Hibiscus tiliaceus* L.	Waru		0	0	0	0	0
57	*Thespesia lampas* (Cav.) Dalzell & A. Gibson	Kapasan		0	0	0	0	0
Rubiaceae								
58	*Coffea arabica* L.	Kopi		0	0	0	0	0
Verbenaceae								
59	*Tectona grandis* L.f.	Jati		0	0	0	0	0
Bombacaceae								
60	*Ceiba pentandra* (L.) Gaertn.	Randu		0	0	0	0	0
Poaceae								
61	*Saccharum officinarum* L.	Tebu		0	0	0	0	0

From the above showed that all 5 larvas were found dead on species test plants, except on 5 plant species: *Calliandra calothyrsus* Meisn., *Parkia speciosa* Hassk., *Sterculia foetida* L., *Syzygium polyantum* (Wight) Walp.and *Psidium guayava L.* the cut foliage of which showed an indication of damage eaten by the larva of *C.assimilis*. Therefore, these 5 plant species were again subjected to further host specificity testing without choice, with its control *Vachellia nilotica* (syn. *A.nilotica*). Each pot of these 6 young plants was wrapped tightly with stiff transparant plastic forming a tube at an height suited to the height of the young plant. At the top of the tube a plstic gauze was fitted and this arrangement was replicated 4x, Five 3 days old larva *C.assimilis* were released on each tube; the evaluation was carried out to investigate the development of these larva up to imago.

Tabel 3. The performance of 3 day old larva of *C.assimilis* on 5 test plant species

No	Nama Species	Nm lokal	6 hari	Fase Pupa	Fase Imago
1	*Calliandra calothyrsus* Meisn	kaliandra	0	0	0
2	*Parkia speciosa* Hassk	petai	0	0	0
3	*Sterculia foetida* L	kepuh	0	0	0
4	*Syzygium polyantum* (Wight) Walp	salam	0	0	0
5	*Psidium guayava L*	Jambu air	0	0	0
6	*Vachellia nilotica (A.nilotica*)	nilotika	86.67%	(see below)	(see below)

In the 6th day 5 larva of *C.assimilis* were found dead on *Calliandra calothyrsus* Meisn, *Parkia speciosa* Hassk, *Sterculia foetida* L., *Syzygium polyantum* (Wight) Walp, and *Psidium guayava*

L. while under *Vachellia nilotica* as the control, 86.67 was still survived It seems that the range of host plant of *Chiasmia assimilis* is very narrow infact according to this test was only on *Vachellia nilotica (* syn. *Acacia nilotica).*

The details of *Chiasmia assimilis* development at the insectarium of BIOTROP were as follows .

LARVA

Greenish *C.assimilis* larva at its early state of development will change to brownish before pupation. The larvae moves with 3 pairs of front legs and 2 pairs of hind legs. This larva may be mistaken as "ulat jengkal" commonly found in Indonesia, also known as "looper" The maintenance of *C.assimilis* larva is easy; may be done using feeding cut young shoot of *A.nilotica* or in a jar, or just let the lava graze the shoot of living *A.nilotica* in pots. When using cut foliage you have to add new shoot to frequently the jar. Maintaining the larva on living *A.nilotica* is much easier, but you have to make sure you do not have the intrution of ant or other animals to your set up.

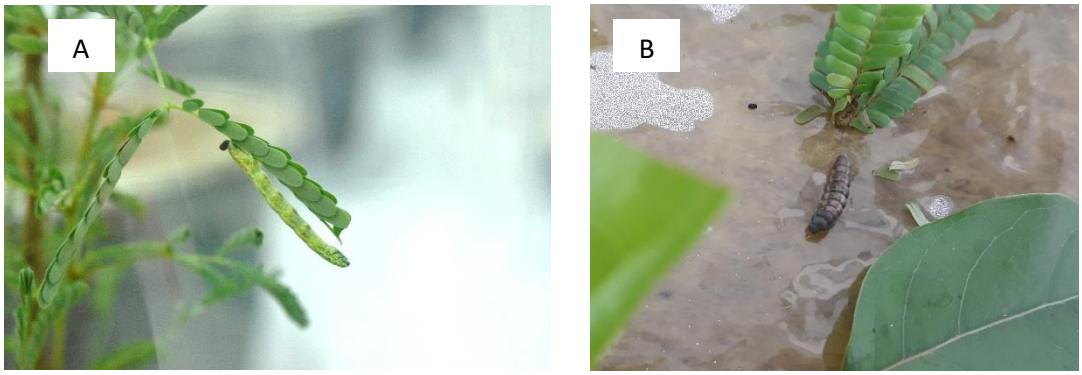

Figure 6. A. green larva of *C. assimilis* and B. brownish larva before pupating

The consignment of *C.assimilis* larva, was dismantled opened soon and transferred the larva into a jar containing young shoot of *A.niltotica* ; living and dead larva were counted, the dead ones were burn in incenerator. The larva were observed eating the provided young leaves of *A.nilotica* from Indonesia. It was discovered that an insect other than *C.assimilis* was found and it was a parasitoid. The insect was carefully preserve for future identification. After preparing cages with potted good *A.nilotica* having growth green and healthy leaves those larva were released into the cages with potted *A.nilotica*

Food quality effects larva growth, and development, healthy larva will look greenish while unhealthy and sick larva may be yellowish and shrinking. Potted *A.nilotica* that has been

defoliated soon was replaced after trasfering the larva to new potted *A.nilotica.* The pots of defoliated *A.nilotica* were one fertilized with NPK and put outside the insectarium under the morning sun. Cages with defoliated of potted *A.nilotica* also be simply added with new potted *A.nilotica* close enough to those already defoliated, so that those hungry larva can move and reach the new potted *A.nilotica* to eat. The speed of consuming leaves of *A,nilotica* will slow down with time, not besause that they do not want to eat, but because they want to pupate, their colour is also chanching to brownish look.

PUPA.

Larva usualy moves downward to pupate and these larva will find the most covenient place for pupating, sometimes hidden in cracks of soil down to 1-5 cm deep or betwwen wall of plastic pot and soil, or even between pots. In the early work with poor experience about *Chiasmia assimilis* life, we were surprised to see the moth or imago of this *C.assimilis* was flying around in the insectarium room, Apparently those larva moved at night , hidden, pupated, and emerged into imago without the staff knowledge. This prepupation stage was considered a susceptible stage to attack of ants, and contributing to a high proportion of larva failure to develop into pupa. Pupa hiding in the soil were collcted from the soil one by one using a spoon made from a piece of filter paper to avoid damaging this very soft pupa, when damages the emerging imago deveoped abnormaly or even did not emerge all together.

The collected pupa were counted and appropriately put into cages where a sheet of stiff transparant plastic wrapped around the platic pot to form a tube having a diameter of that pot, 15 cm and the height was 70 cm. the top of which was fitted with plastic gauze for aeraton. Almost every day imago emerged , caunted, and differentiated into male and female and kept until they die.

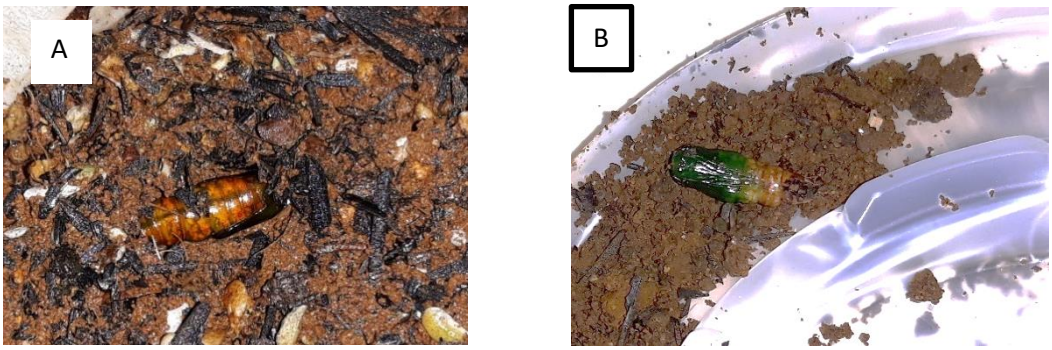

Figure 7. A. pupating larvae was layed down in soil. B. Initial larva becoming a pupa look greenish

IMAGO

The cage was provided with a cotton ball dipped into a soltion of 10% honey and hang with a piece of nylon threat from the gauze at the top of the tube (made of sheet of tranparant, stiff plastic, wrapped around the plastic pot of *A.nilotica* plant forming a big tube with the variable height suited to that of test plants),

Figure 8. A. Tubes made of stiff transparant platic sheet wrapped around the plastic pot to house the imago of *C.assimilis*. B. The mated imago of *C.assimilis*.

The imago of *C.assimilis* was small with nice brownnish colour with blackish patter in the lower part of its wings, with serrated edge The female imago was bigger than male imago, the shloud be paired to mate to have fertile eggs. They need 1-2 days to mate and the female will start ovipositing. When not mated, hoever, the female will also oviposit but infertile eggs. It happened in the early maintenance of this *C.assimilis*.

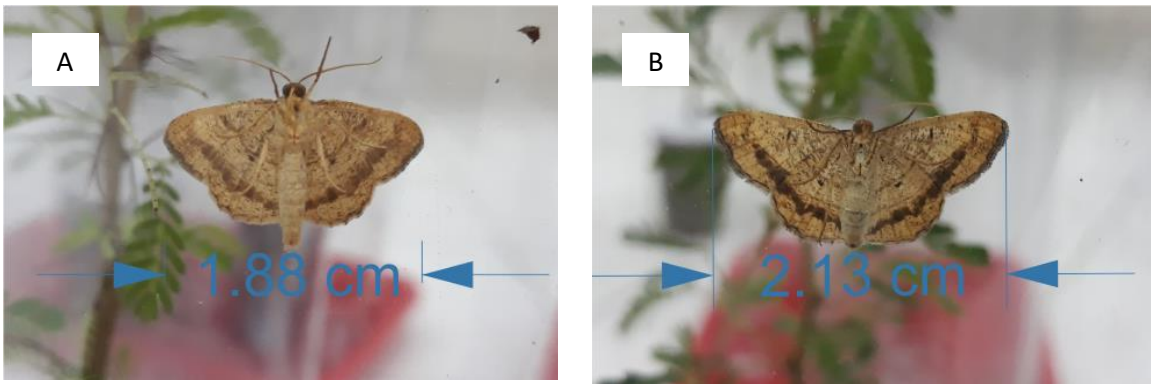

Figure 9. Imago of *Chiasmia assimilis* A. male imago. B. female imago

EGGS

The eggs of *C.assimilis* are green and became white transparent after hatched. shape are oval shaped, eggs appear one days after imago mating, female moth usually lays ths eggs randomly on the leaves, twigs, shoots, and thorns of *A.nilotica*, eggs are also found attached on the gauze or strip of sticky/duck tape, it should be considered that the cages for eggs rearing always tight and ventilation alywas good. Every day, female adult can produce the eggs, and it makes eggs hatched in different days. New larvae can be found after 4 days since the eggs appeared. Sterile eggs can be recognize after 4 days, the shape became doughnout with transparent filment in the middle of the eggs. From the above practical works the following ouput of *Chiasmia assimilis* life cycle was presented in Table 4.

Figure 10. eggs of *C.assimilis* on tips of *A.nilotica*.

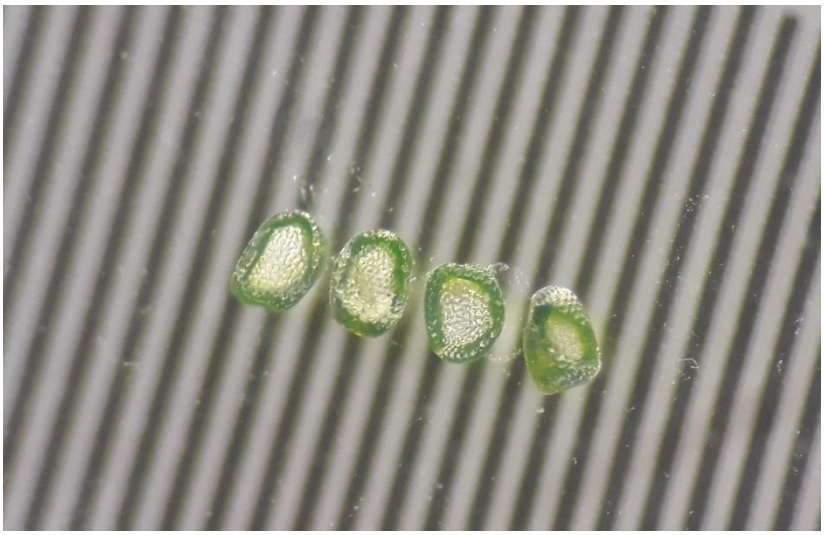

Figure 11. sterile eggs observed under microscope stereo, collected from the gauze.

Table 4. The performance of *Chiasmia assimilis* growth and development under the laboratory condition

No	Growth Stage	Number	Percentage
	From imago to eggs and to larva		
1	Pupa from the 1sr and 2nd shipment	11	
2	Imago	5	
3	Eggs produced	453	
4	Hached	355	65.38
5	Sterile	188	
From larva to pupa			
	Total larva	355	
	Total pupa	137	38.59
From pupa to imago			
	Total pupa	137	
	Total imago	89	64.96

The biggest casualties were attributed to transportation, the first shipment of larva sent from Australia to Indonesia was about 40-50 larva and the second shipment was a bout 80-100 larva, and the pupa collected was 11 and the imago was only 5. We did not know which one was male and which one was female. We thought would be good to have pairs of imagoes mated, but not so easy. However, the eggs produced were quite substansial , 453 eggs, probably we have only 2 female, one female oviposited its eggs attached to the plastic gauze and proven to be sterile. A poor knowledge of larva behaviour also contributed to confusion, because the larva sudently disappear from the cage, which in fact they were pupating in the soil, it was unknowingly discovered when the imagoes were flaying around the insectarium room, furtunately we have doubble door preventing them from flying out of the room.

From these host specificity testing againts 60 plant species tested, using centrifugal approach of selecting samples of tested species with *Acacia nilotica* as a member of Mimosaceae at the center followed by Papillionaceae, Caesalpiniaceae, plants at Baluran NP, and crops from Legumnosae as well as Poaceae, no indication other than the target plant *A.nilotica* as the host plant of *C.assimilis*. It is condluded that *Chiasmia assimilis* may be applied as the bioconrol agent of *A.nilotica* which now is *Vachellia nilotica*. Host specificity testing in Australia (Palmer *et al.,* 2007), carried out on 2 species of biocontrol agents *C.insconspicua* and *C.assimilis* towards species of *Acacia sensu stricto ,* although the taxonomical distance between *Acacia sensu stricto* and *Vachellia nilotica* or *Acacia nilotica* was quite far. *C.assimilis* newly hatched larva were allowed to graze on cut foliage of many Acacia sensu stricto, the results indicated those on the cut foliage of *A. nilotica,* 68% successfuly developed into imago, but on *A.decurrens, A. pulchella* and A. *deanei,* only 3%,

7% dan 3% each manage to complete the life cycle . With these data the decission maker of the Australian government decided to accept *Chiasmia assimilis* as a biological control agent for *A.nilotica* and allow *C.assimilis* to be released in the field.

The field performance of *C.assimilis* is very impresive, a substnsial area of *A.nilotica* has been deffoliated and it will reduce the population of *A.nilotica* in the future. It is interesting to study this developmental concept of biocontrol though. In the past it was believed that successful biocontrol agent would be operational when the agent was collected from the same subspecies as the target species. However for this time, it is not so, *Chiasmia assimilis* (Warren.)(*Lepidoptera, Geometridae*), previously collected from *A,nilotica* subsp. s*ubalata*, and *A.nilotica* subsp. *leiocarpa* in Kenya and when rleased in Australia it did not establish. However when the same species of *C.assimilis* but collected from *A.nilotica* subsp. *kraussiana* in South Africa which also attacking *A.nilotica* subsp. *subalata* as well as *A.nilotica* subsp. *leiocarpa* which are also around in the area, when imported to Australian and released in the field, established well, and impacted the growth of *A.nilotica* impressively. This species that BIOTROP was importing last September.

6. COORDINATOR AND PERSONAL OF THE RESEARCH

I. Coordinator

Dr. Soekisman Tjitrosemito, MSc.

II. Personal of the Research

Dr. Sri S. Tjitrosoedirdo, MSc.

Setiabudi, S.Hut.

Drs. Imam Mawardi

Indah Wahyuni, M.Si.

Saiful Bachri

7. REFERENCES

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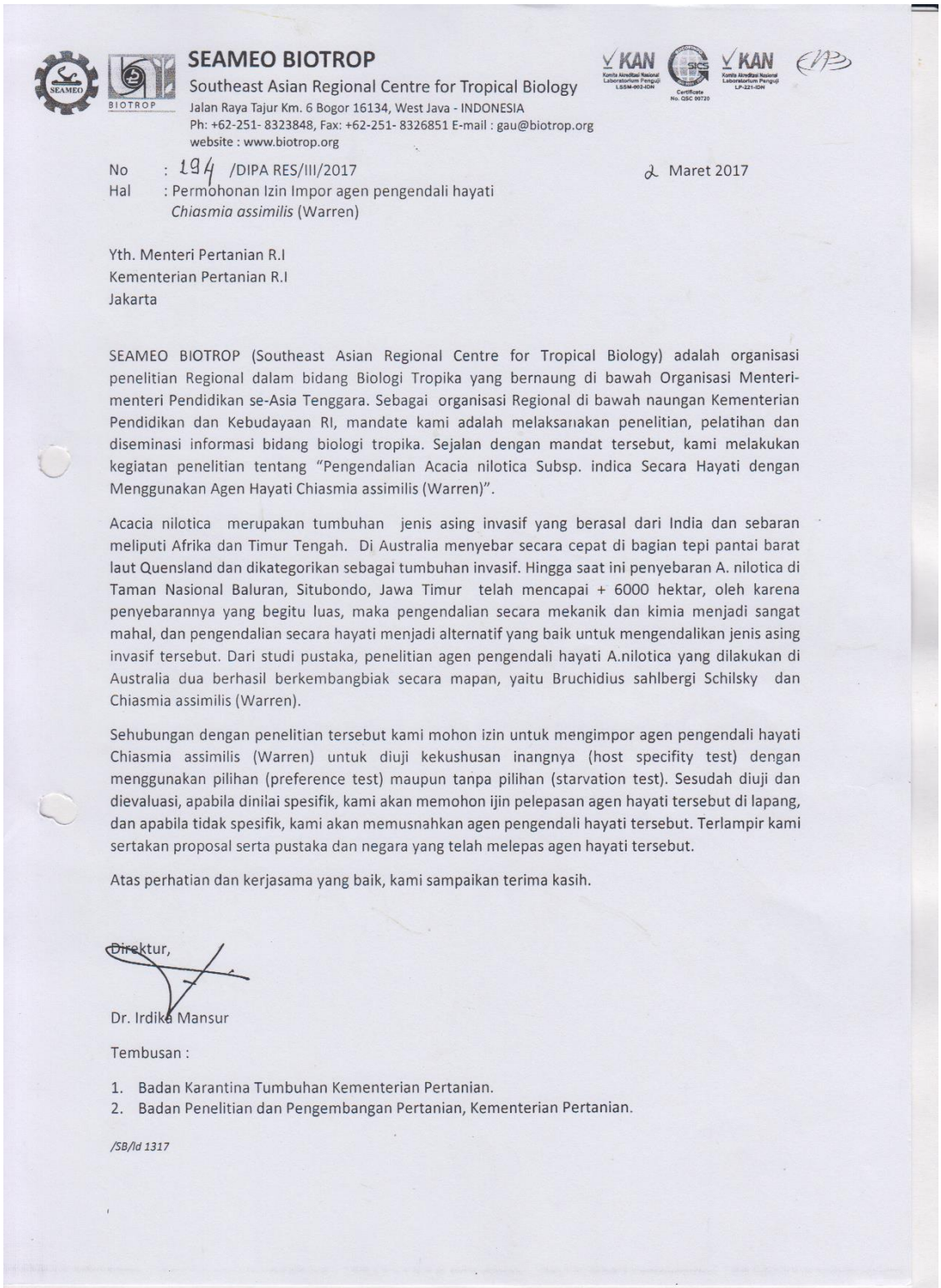
8. APPENDICES

1. Letter to Ministry of Agriculture of Indonesia for Import permit bioagens *Chiasmia assimilis* Warren Lepidoptera-Geometridae



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website : www.biotrop.org

No : 194 /DIPA RES/III/2017 2 Maret 2017
Hal : Permohonan Izin Impor agen pengendali hayati *Chiasmia assimilis* (Warren)

Yth. Menteri Pertanian R.I
Kementerian Pertanian R.I
Jakarta

SEAMEO BIOTROP (Southeast Asian Regional Centre for Tropical Biology) adalah organisasi penelitian Regional dalam bidang Biologi Tropika yang bernaung di bawah Organisasi Menteri-menteri Pendidikan se-Asia Tenggara. Sebagai organisasi Regional di bawah naungan Kementerian Pendidikan dan Kebudayaan RI, mandate kami adalah melaksanakan penelitian, pelatihan dan diseminasi informasi bidang biologi tropika. Sejalan dengan mandat tersebut, kami melakukan kegiatan penelitian tentang "Pengendalian Acacia nilotica Subsp. indica Secara Hayati dengan Menggunakan Agen Hayati Chiasmia assimilis (Warren)".

Acacia nilotica merupakan tumbuhan jenis asing invasif yang berasal dari India dan sebaran meliputi Afrika dan Timur Tengah. Di Australia menyebar secara cepat di bagian tepi pantai barat laut Queensland dan dikategorikan sebagai tumbuhan invasif. Hingga saat ini penyebaran A. nilotica di Taman Nasional Baluran, Situbondo, Jawa Timur telah mencapai + 6000 hektar, oleh karena penyebarannya yang begitu luas, maka pengendalian secara mekanik dan kimia menjadi sangat mahal, dan pengendalian secara hayati menjadi alternatif yang baik untuk mengendalikan jenis asing invasif tersebut. Dari studi pustaka, penelitian agen pengendali hayati A.nilotica yang dilakukan di Australia dua berhasil berkembangbiak secara mapan, yaitu Bruchidius sahlbergi Schilsky dan Chiasmia assimilis (Warren).

Sehubungan dengan penelitian tersebut kami mohon izin untuk mengimpor agen pengendali hayati Chiasmia assimilis (Warren) untuk diuji kekushusan inangnya (host specifity test) dengan menggunakan pilihan (preference test) maupun tanpa pilihan (starvation test). Sesudah diuji dan dievaluasi, apabila dinilai spesifik, kami akan memohon ijin pelepasan agen hayati tersebut di lapang, dan apabila tidak spesifik, kami akan memusnahkan agen pengendali hayati tersebut. Terlampir kami sertakan proposal serta pustaka dan negara yang telah melepas agen hayati tersebut.

Atas perhatian dan kerjasama yang baik, kami sampaikan terima kasih.

Direktur,

Dr. Irdika Mansur

Tembusan :
1. Badan Karantina Tumbuhan Kementerian Pertanian.
2. Badan Penelitian dan Pengembangan Pertanian, Kementerian Pertanian.

/SB/Id 1317

2. Letter of response from Ministry of Agriculture of Indonesia

KEMENTERIAN PERTANIAN
BADAN KARANTINA PERTANIAN
JALAN HARSONO RM NOMOR 3 RAGUNAN, PASAR MINGGU JAKARTA 12550
GEDUNG E Lt. 1, 5, 7 TELEPON/FAKSIMILI (021) 7816484, 7816483, 7816482, 7816481
Website:http://www.karantina.deptan.go.id
Email:infokarantina@deptan.go.id

Nomor : 4073 /HM.110/K.3/03/2017 — 20. Maret 2017
Lampiran : 1 (satu) eksemplar
Perihal : Permohonan ijin pemasukan agens hayati *Chiasmia assimilis* (Warren)

Kepada Yth.
Direktur SEAMEO BIOTROP
di
Bogor.

Sehubungan dengan surat Saudara No. 194/DIPA RES/III/2017 tanggal 2 Maret 2017 tentang permohonan izin impor agens pengendali hayati *Chiasmia assimilis* (Warren), yang telah kami tindaklanjuti dengan audit kecukupan dokumen, diinformasikan bahwa permohonan Saudara belum mengikuti ketentuan administratif sebagaimana tercantum dalam Keputusan Menteri Pertanian Nomor 411 Tahun 1995. Saudara dapat melakukan perbaikan dengan melengkapi informasi lainnya mengikuti prosedur sebagaimana terlampir.

Untuk kelancaran pendistribusian dokumen sebagai bahan kajian oleh Komisi Agens Hayati, selain penyampaian permohonan beserta dokumen lainnya dalam bentuk *hard copy*, kami menyarankan agar disediakan pula dalam bentuk *soft file*. Seluruh dokumen tersebut dapat dikirimkan melalui alamat *e-mail*: kehati.nabati@gmail.com sesegera mungkin dalam rangka percepatan layanan.

Demikian kami sampaikan, atas perhatiannya diucapkan terima kasih.

Kepala Pusat Karantina Pertanian
dan Keamanan Hayati Nabati
selaku Wakil Ketua Komisi Agens Hayati

Dr. Ir. Antarjo Dikin, M.Sc
NIP. 19630206 198303 1 002

Tembusan kepada Yth.:
Kepala Badan Karantina Pertanian selaku Ketua Komisi Agens Hayati (sebagai laporan).

Lampiran surat nomor : 4073/HM.110/K.3/03/2017

PROSEDUR PENYAMPAIAN PERMOHONAN IJIN PEMASUKAN AGENS HAYATI

1) Pemohon menyampaikan surat permohonan ijin pemasukan agens hayati yang ditujukan kepada **Menteri Pertanian c.q. Kepala Badan Karantina Pertanian selaku Ketua Komisi Agens Hayati.** Dalam surat permohonan izin harus termuat informasi antara lain mengenai :
   a. nama dan alamat orang atau badan hukum yang akan memasukkan agens hayati;
   b. nama dan alamat pengirim dan/atau produsen agens hayati di luar negeri;
   c. tujuan pemasukan;
   d. negara asal agens hayati;
   e. nama umum, nama ilmiah dan nama dagang agens hayati;
   f. jumlah agens hayati yang akan dimasukkan;
   g. sarana, peralatan dan kualifikasi tenaga yang dimiliki oleh orang atau badan hukum tersebut;
   h. wadah atau kemasan yang digunakan;
   i. cara pengangkutan;
   j. perkiraan tanggal pemasukan;
   k. tempat pemasukan;
   l. tindakan-tindakan pengamanan yang akan dilakukan untuk mencegah terjadinya kontaminasi dan/atau terlepasnya agens hayati;
   m. stadia perkembangan.

2) Surat permohonan ijin harus disertai dengan lampiran :
   a. informasi mengenai :
      1. biologi agens hayati;
      2. hasil penelitian yang pernah dilakukan di negara asalnya dan/atau negara lain;
      3. manfaat dan laporan pengkajian tentang dampak negatif yang ditimbulkan dalam penggunaan agens hayati tersebut di negara asalnya dan/atau negara lain;
      4. langkah-langkah penanggulangan yang telah dilakukan untuk mengatasi dampak negatif dari penggunaan agens hayati tersebut di negara asalnya dan/atau negara lain;
      5. musuh alami, antagonis serta kompetitor agens hayati tersebut;
      6. habitat asal, karakteristik serta spesifikasi agens hayati tersebut
      7. informasi dari produsen bahwa agens hayati tersebut diperoleh dan/atau diproduksi menurut cara-cara yang dapat dipertanggungjawabkan kebenarannya
   b. surat keterangan yang menyatakan bahwa agens hayati tersebut diproduksi dan/atau dikirim oleh orang atau badan hukum yang diberi izin untuk itu oleh lembaga yang berwenang di negara asalnya;

3. Reply letter for Import Permit to Ministry of Agriculture of Indonesia

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Southeast Asian Regional Centre for Tropical Biology
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Ph: +62-251- 8323848, Fax: +62-251- 8326851 E-mail : gau@biotrop.org
website : www.biotrop.org

Nomor : 323 /DIPA RES/IV/2017 6 April 2017
Hal : Permohonan Izin Impor agen pengendali hayati *Chiasmia assimilis* (Warren)

Yth. Menteri Pertanian
c.q. Kepala Badan Karantinan Pertanian
selaku Ketua Komisi Agen Hayati
Jakarta

Menindak lanjuti surat kepala pusat Karantina Pertanian dan Keamanan hayati Nabati selaku wakil ketua Komisi Agen Hayati nomor 4073/HM.110/K.3/03/2017 tertanggal 20 Maret 2017 perihal Permohonan izin Pemasukan Agen Hayati *Chiasmia assimilis* (Warren), dengan hormat kami sampaikan ketentuan administratif sebagaimana yang tercantum dalam keputusan Menteri Pertanian nomor 411 tahun 1995. Berikut ini informasi yang kami tambahkan (sesuai dengan urutan dalam surat jawaban tersebut) dalam lampiran terpisah beserta *reference* untuk informasi biologi agens hayati dan hasil penelitiannya. Sebagai informasi agens hayati ini diimport dalam rangka penelitian pengendalian hayati tumbuhan asing invasif, di Taman Nasional Baluran.

Atas perhatian dan kerjasama yang baik, kami sampaikan terima kasih.

Direktur,

Dr. Irdika Mansur

SB/Id 5417

4. Detail documents for PERMENTAN NO. 411 tahun 1995

PROSEDUR PENYAMPAIAN

PERMOHONAN IJIN PEMASUKAN AGENS HAYATI

1. Pemohon menyampaikan surat permohonan ijin pemasukan agens hayati yang ditujukan kepada Menteri Pertanian c.q. Kepala Badan Karantina Pertanian selaku Ketua Komisi Agens Hayati. Dalam surat permohonan izin harus termuat informasi antara lain mengenai :

No	Item	Keterangan
A	Nama dan alamat orang atau badan hukum yang akan memasukkan agens hayati	SEAMEO BIOTROP. (Southeast Asian Regional Centre for Tropical Biology) Jl. Raya Tajur Km. 6 Bogor 16134, Indonesia Office: +62-251-8323848 Office: +62-251-8326851 Email : gau@biotrop.org
b	Nama dan alamat pengirim dan/atau produsen agens hayati di luar negeri.	Dr. Michael Day Senior Entomologist Biosecurity Queensland Department of Agriculture and Fisheries Level 3C West Ecosciences Precinct GPO Box 267, Brisbane, Qld 4001 AUSTRALIA Ph: (07) 3708 8553 International: 61 7 3708 8553 Fax: (07) 3708 8429 International: 61 7 3708 8429 Email to: Michael.Day@daf.qld.gov.au
C	Tujuan pemasukan	Penelitian pengendalian hayati *Acacia nilotica*, tumbuhan asing invasif yang telah menginvasi sebagian besar dari savana di TN. Baluran
D	Negara asal agens hayati	Australia
E	Nama umum, nama ilmian dan nama dagang agens hayati	Nama umum tidak dikenal, nama alamiah *Chiasmia assimilis* (Warren) (Lepidoptera: Geometridae), tidak ada nama dagang, karena tidak diperdagangkan.
F	Jumlah agens hayati yang akan dimasukkan	200 larva

G	Sarana, peralatan dan kualifikasi tenaga yang dimiliki oleh orang atau badan hukum tersebut.	Lembaga penelitian pengendalian hayati *A. nilotica* dengan memakai berbagai agens hayati dengan fasilitas penangkaran insectarium dan rumah kaca uji efikasi tertutup dengan incinerator untuk memusnahkan sisa/sampah penelitian
H	Wadah atau kemasan yang digunakan	Dari pengalaman sebelumnya biasanya dikirim dalam karton yg diperkuat dengan kayu penguat dengan daun *A.nilotica* didalamnya
I	Cara pengangkutan	Dengan penerbangan komersial, langsung dari Brisbane, diharapkan dalam kurun waktu 24 jam sampai di Jakarta.
J	Perkiraan tanggal masuk	Belum tahu, Karena koloni akan dikirim jika surat izin import sudah dikirim ke Biosecurity Queensland Department of Agriculture and Fisheries, Australia.
K	Tempat pemasukan	Bandara Sukarno Hatta
L	Tindakan-tindakan pengamanan yang akan dilakukan untuk mencegah terjadinya kontaminasi dan/atau terlepasnya agens hayati.	Dengan petugas karantina Bandara, consignment itu dibawa ke BIOTROP langsung, segera dibuka dan semua pembungkus dibakar di incenator fasilitas yang ada di insectarium BIOTROP, sedang serangganya dilepas di dalam kurungan dalam insectarium dengan pintu rangkap untuk mengurangi resiko serangga dewasa terbang keluar.
M	Stadia perkembangan.	Dikirim dalam stadia larva

2. Surat permohonan ijin harus disertai dengan lampiran :

a. Informasi mengenai :

No	Item	Keterangan
A	Biologi agens hayati	Palmer et al (2007)
B	Hasil penelitian yang pernah dilakukan di negara asalnya/atau negara lain	Palmer et al (2007)
C	Manfaat dan laporan pengkajian tentang dampak negatif yang ditimbulkan dalam penggunaan agens hayati tersebut di negara asalnya dan/atau negara lain.	Belum ada laporan dampak negatif dari pemakaian agens hayati

D	Langkah-langkah penanggulangan yang telah dilakukan untuk mengatasi dampak negative dari penggunaan agens hayati tersebut di negara asalnya dan/atau negara lain.	Belum ada laporan langkah2 untuk mengatasi dampak negatif agens hayati.
E	Musuh alami, antagonis serta kompetitor agens hayati tersebut.	Belum pernah dilaporkan, kalau di Indonesia bisa saja *Apantheles* sp. menjadi parasitoidnya.
F	Habitat asal, karakteristik serta spesifikasi agens hayati tersebut.	Habitat asal adalah savana kering di Afrika, dimana *A. nilotica* tumbuh, agen hayati ini makan daun terutama *A. nilotica* yang masih muda.
G	Informasi dari produsen bahwa agens hayati tersebut diperoleh dan/atau diproduksi menurut cara-cara yang dapat dipertanggungjawabkan kebenarannya.	Metoda penangkaran, uji efikasi dan pelepasan dikerjakan oleh peneliti berpengalaman

b. Surat keterangan yang menyatakan bahwa agens hayati tersebut diproduksi dan/atau dikirim oleh orang atau badan hokum yang diberi izin untuk itu oleh lembanga yang berwenang di negara asalnya.

Agen hayati itu dikirim oleh oleh ahli yang kompeten melakukan penelitian pengendalian hayati pada lembaga yang memang sejak awal dibangun untuk mengendalikan species tumbuhan asing invasif dengan agens hayatinya.

5. Import permit from Ministry of Agriculture of Indonesia

MENTERI PERTANIAN
REPUBLIK INDONESIA

KEPUTUSAN MENTERI PERTANIAN REPUBLIK INDONESIA
NOMOR : 358/Kpts/KR.040/6/2017
TENTANG
PEMBERIAN IZIN PEMASUKAN AGENS HAYATI *CHIASMIA ASSIMILIS (WARREN)* DARI AUSTRALIA KEPADA *SOUTHEAST ASIAN REGIONAL CENTER FOR TROPICAL BIOLOGY* UNTUK KEPERLUAN PENELITIAN

DENGAN RAHMAT TUHAN YANG MAHA ESA

MENTERI PERTANIAN REPUBLIK INDONESIA,

Menimbang : a. bahwa dalam rangka pengendalian jenis asing invasif *Acacia nilotica, Southeast Asian Regional Center for Tropical Biology* (SEAMEO BIOTROP) akan memasukkan *Chiasmia assimilis (Warren)* dari Australia untuk keperluan penelitian;

b. bahwa berdasarkan hasil kajian teknis oleh Komisi Agens Hayati, *Chiasmia assimilis (Warren)* dari Australia, dapat disetujui dan diberikan rekomendasi untuk dimasukkan ke dalam wilayah Negara Republik Indonesia;

c. bahwa berdasarkan pertimbangan sebagaimana dimaksud dalam huruf a dan huruf b, perlu menetapkan Keputusan Menteri Pertanian tentang Pemberian Izin Pemasukan Agens Hayati *Chiasmia assimilis* (*Warren*) dari Australia kepada *Southeast Asian Regional Center for Tropical Biology* untuk Keperluan Penelitian;

Mengingat : 1. Undang-Undang Nomor 12 Tahun 1992 tentang Sistem Budidaya Tanaman (Lembaran Negara Republik Indonesia Tahun 1992 Nomor 46, Tambahan Lembaran Negara Republik Indonesia Nomor 3478);

2. Undang-Undang Nomor 16 Tahun 1992 tentang Karantina Hewan, Ikan, dan Tumbuhan (Lembaran Negara Republik Indonesia Tahun 1992 Nomor 56, Tambahan Lembaran Negara Republik Indonesia Nomor 3482);
3. Undang-Undang Nomor 5 Tahun 1994 tentang Pengesahan Konvensi Perserikatan Bangsa-bangsa Mengenai Keanekaragaman Hayati (Lembaran Negara Republik Indonesia Tahun 1994 Nomor 41, Tambahan Lembaran Negara Republik Indonesia Nomor 3556);
4. Undang-Undang Nomor 32 Tahun 2009 tentang Perlindungan dan Pengelolaan Lingkungan Hidup (Lembaran Negara Republik Indonesia Tahun 2009 Nomor 140, Tambahan Lembaran Negara Republik Indonesia Nomor 5059);
5. Peraturan Pemerintah Nomor 6 Tahun 1995 tentang Perlindungan Tanaman (Lembaran Negara Republik Indonesia Tahun 2009 Nomor 140, Tambahan Lembaran Negara Republik Indonesia Nomor 5059);
6. Peraturan Pemerintah Nomor 14 Tahun 2002 tentang Karantina Tumbuhan (Lembaran Negara Republik Indonesia Tahun 2002 Nomor 35, Tambahan Lembaran Negara Republik Indonesia Nomor 4196);
7. Peraturan Presiden Nomor 7 Tahun 2015 tentang Organisasi Kementerian Negara (Lembaran Negara Republik Indonesia Tahun 2015 Nomor 8);
8. Peraturan Presiden Nomor 45 Tahun 2015 tentang Kementerian Pertanian (Lembaran Negara Republik Indonesia Tahun 2015 Nomor 85);

9. Keputusan Menteri Pertanian Nomor 411/Kpts/TP.120/6/1995 tentang Pemasukan Agens Hayati ke dalam Wilayah Negara Republik Indonesia;
10. Keputusan Menteri Pertanian Nomor 1481/Kpts/OT.160/4/2012 tentang Komisi Agens Hayati;
11. Peraturan Menteri Pertanian Nomor 22/Permentan/OT.140/4/2008 tentang Organisasi dan Tata Kerja Unit Pelaksana Teknis Karantina Pertanian;
12. Peraturan Menteri Pertanian Nomor 94/Permentan/OT.140/12/2011 tentang Tempat Pemasukan dan Pengeluaran Media Pembawa Penyakit Hewan Karantina dan Organisme Pengganggu Tumbuhan Karantina (Berita Negara Republik Indonesia Tahun 2012 Nomor 7) sebagaimana telah diubah dengan Peraturan Menteri Pertanian Nomor 44/Permentan/OT.140/3/2014 tentang Perubahan atas Peraturan Menteri Pertanian Nomor 94/Permentan/OT.140/12/2011 tentang Tempat Pemasukan dan Pengeluaran Media Pembawa Penyakit Hewan Karantina dan Organisme Pengganggu Tumbuhan Karantina (Berita Negara Republik Indonesia Tahun 2014 Nomor 428);
13. Peraturan Menteri Pertanian Nomor 43/Permentan/OT.140/10/2015 tentang Organisasi dan Tata Kerja Kementerian Pertanian (Berita Negara Republik Indonesia Tahun 2015 Nomor 1243);

Memperhatikan :

1. Permohonan *Southeast Asian Regional Center for Tropical Biology* (SEAMEO BIOTROP) Nomor: 194/DIPA RES/III/2017 tanggal 2 Maret 2017;
2. Rekomendasi Ketua Komisi Agens Hayati Nomor 6042/KR.040/L/5/2017 tanggal 8 Mei 2017;

MEMUTUSKAN:

Menetapkan :

KESATU : Memberikan izin pemasukan kepada *Southeast Asian Regional Center for Tropical Biology* (SEAMEO BIOTROP) beralamat di Jalan Raya Tajur Km. 6 Bogor, Jawa Barat untuk memasukkan *Chiasmia assimilis* (*Warren*) sebanyak 200 (dua ratus) larva.

KEDUA : Izin Pemasukan sebagaimana dimaksud dalam diktum KESATU:

a. hanya untuk keperluan penelitian; dan
b. diberikan untuk jangka waktu 6 (enam) bulan sejak tanggal ditetapkannya Keputusan Menteri ini.

KETIGA : Pemasukan agens hayati sebagaimana dimaksud dalam diktum KESATU harus dilakukan sesuai dengan ketentuan sebagai berikut:

a. mengikuti ketentuan sebagaimana diatur dalam Pasal 13 sampai dengan Pasal 28 Keputusan Menteri Pertanian Nomor 411/Kpts/TP.120/6/1995 tentang Pemasukan Agens Hayati ke dalam Wilayah Negara Republik Indonesia;
b. dikemas sedemikian rupa sehingga dapat menjamin tidak ada agens hayati yang terlepas selama perjalanan;
c. melalui Bandar Udara Soekarno-Hatta, Cengkareng;
d. segera dibawa ke laboratorium *Southeast Asian Regional Center for Tropical Biology* (SEAMEO BIOTROP) dengan pengawalan petugas karantina dari Balai Besar Karantina Pertanian Soekarno-Hatta, Cengkareng; dan

e. ditempatkan dalam ruangan tertutup yang kedap serangga, tidak tercampur dengan barang lain, dan terhindar dari aktifitas orang yang tidak berkepentingan, setelah kedatangan agens hayati di laboratorium.

KEEMPAT : Penelitian sebagaimana dimaksud dalam diktum KEDUA huruf a dilaksanakan dengan ketentuan sebagai berikut:

a. penelitian dilakukan oleh peneliti pada *Southeast Asian Regional Center for Tropical Biology* (SEAMEO BIOTROP);
b. pihak pengelola laboratorium wajib bertanggung jawab atas penanganan agens hayati agar tidak terlepas keluar;
c. selama penelitian berlangsung diawasi oleh petugas karantina Balai Besar Karantina Pertanian Soekarno-Hatta, Cengkareng;
d. paling kurang satu kali selama periode penelitian dilakukan monitoring oleh Komisi Agens Hayati;
e. pada akhir kegiatan penelitian, seluruh sisa bahan penelitian harus dimusnahkan di bawah pengawasan petugas karantina Balai Besar Karantina Pertanian Soekarno-Hatta, Cengkareng; dan
f. setelah penelitian selesai dilakukan, *Southeast Asian Regional Center for Tropical Biology* (SEAMEO BIOTROP) wajib menyampaikan laporan tertulis hasil penelitian kepada Menteri Pertanian melalui Kepala Badan Karantina Pertanian selaku Ketua Komisi Agens Hayati.

KELIMA : Keputusan Menteri ini mulai berlaku pada tanggal ditetapkan.

Ditetapkan di Jakarta
pada tanggal 5 Juni 2017

MENTERI PERTANIAN
REPUBLIK INDONESIA,

AMRAN SULAIMAN

Salinan Keputusan Menteri ini disampaikan Kepada Yth.:

1. Menteri Lingkungan Hidup dan Kehutanan;
2. Direktur Jenderal Bea dan Cukai, Kementerian Keuangan;
3. Pimpinan Unit Kerja Eselon I di lingkungan Kementerian Pertanian; dan
4. Kepala Balai Besar Karantina Pertanian Soekarno-Hatta, Cengkareng.