Bacillus thuringiensis, TERACON-1 (TI) FOR BIOLOGICAL CONTROL OF BAGWORMS

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Bacillus thuringiensis (Bt.) produces protein inclusions (parasporal inclusions) adjacent to the endospore during sporulation. The parasporal inclusions consist of one or more insecticidal proteins in the form of a crystal or crystal complex. These insecticidal proteins are commonly known as insecticidal crystal proteins (ICP) or delta (δ) endotoxin. The primary insecticidal activity of *Bt.* is due to ICP (Espinasse *et al.*, 2002). It comprised of 90% of microbial insecticides produced in the world (Bajwa and Kogan, 2001). *Bt.* typically produces δ-endotoxins in parallel with spore formation during the stationary phase of the cell growth cycle (NPTN, 2004). The Cry1 proteins are a group of delta endotoxins that principally target lepidopteran species, including several important crop pests (Swadener, 1994).

PCR screening for gene content of MPOB indigenous isolates showed that several of these isolates harbour toxins effective for oil palm lepidopteran pest (Ramlah Ali and Basri, 2002). In oil palm industry, defoliators such as bagworms, nettle caterpillars (Siti Ramlah *et al.*, 2003) and bunch moth can be effectively controlled using Bt product (Mohd Basri *et al.*, 1994; Siti Ramlah and Mohd Basri, 1997). Indigenous *Bt.* isolates containing the right Cry proteins and devoid of β-exotoxin can be propagated at pilot-scale as ideal alternative to chemical. The undesirable effect of chemical pesticides on the environment and biodiversity has create a niche for the use of environmentally safe alternative, the *Bt.*

MODE OF ACTION

The active ingredients of *Bt.* are the parasporal crystalliferous δ-endotoxins and the spores. The toxins are stomach poison, therefore must be ingested by the pest. Several Cry proteins or δ-endotoxins have receptor proteins in gut lining of *M. plana* (Siti Ramlah, 2000). Toxin receptor binding complex leads to insertion of activated toxin into receptor (Siti Ramlah, 2000), creation of pores, osmotic lysis and insect death.

FERMENTATION / UPSTREAM

Indigenous MPOB Bt1 culture and the high magnifying microgarph of cells are as shown in *Figures 1a* and *1b*, respectively. It contains Cry proteins effective for the control of bagworms. MPOB fermentation medium, Agronat 1 was developed using agricultural raw materials. The Agronat 1 gave high yield of cell and crystal proteins. As the bacteria sporulates, it produces crystalliferous toxins within the cell.

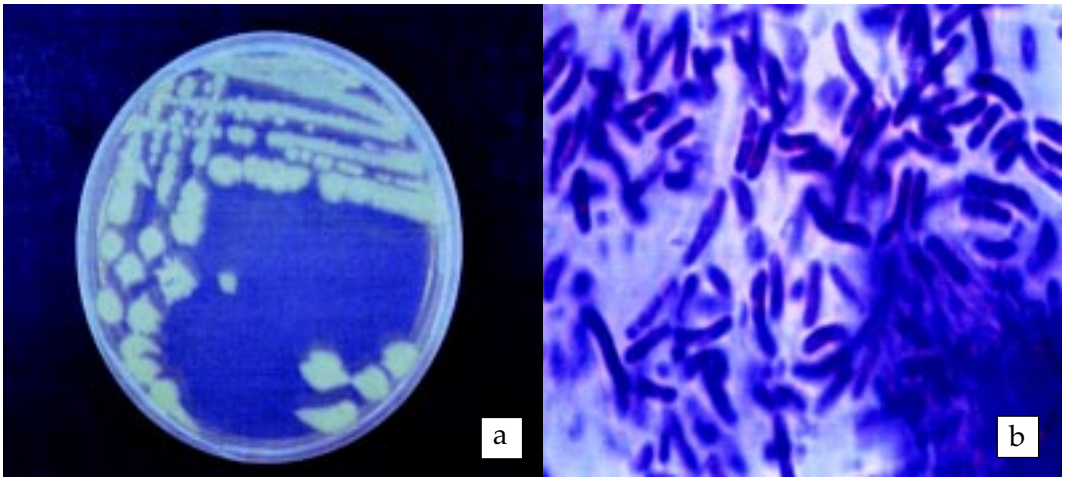

Figure 1. The indigenous MPOB BT1 culture on (a) nutrient agar and (b) cell micrograph with magnification 4000X.

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DOWNSTREAM PROCESS

The downstream process involved centrifugation and filtration using several cut-off cartridges. During downstream process, the cell were subjected to high centrifugation forces. Fermented liquid culture were spun and the concentrate was stored in storage tank. The concentrate was then spray-dried using fluidized spray-dryer to form wettable powder, Terakil-1. The effluent was filtered through 0.45 µm cartridge to get rid off debris and remaining cells. Subsequently, the filtrate was passed through two different cartridges and the retentate was collected aseptically as shown in *Figure 2*.

Figure 2. Teracon-1 collected aseptically using filtration system.

CAPACITY OF Bt PRODUCTION

The Microbial Technology Engineering Centre (MICROTEC) has the capacity to produce approximately 2.712 t of insoluble δ-endotoxins annually to be formulated as Terakil-1. The light liquid or effluent is filtered to retrieve soluble proteins concentrate called Teracon-1. The annual production of Teracon-1 is estimated to be 1.05 t. The laboratory analyses of toxins was conducted using SDS-PAGE, HPLC prior to laboratory and field bioassay.

LABORATORY BIOASSAY

Preliminary bioassay indicated that the Teracon-1 at 0.086 µg ml^{-1} and 0.86 µg ml^{-1} resulted in 70% to 80% mortality of third larval instar of *M. plana*, given via foliar spray and foliar capillary technique, as seen in *Figures 3* and *4*, respectively.

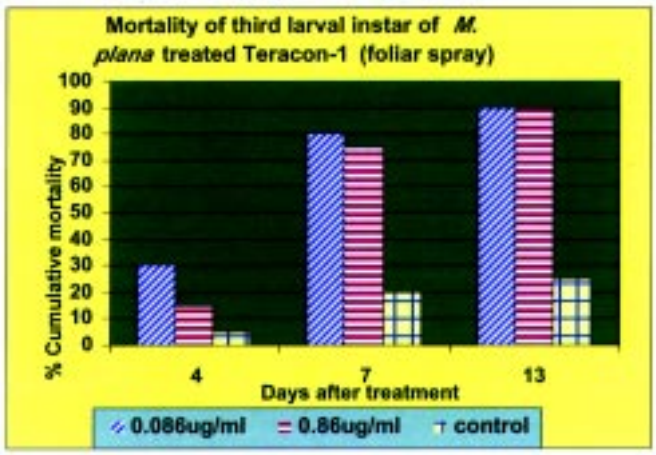

Figure 3. Mortality of M. plana *treated with Teracon-1 delivered via foliar spray.*

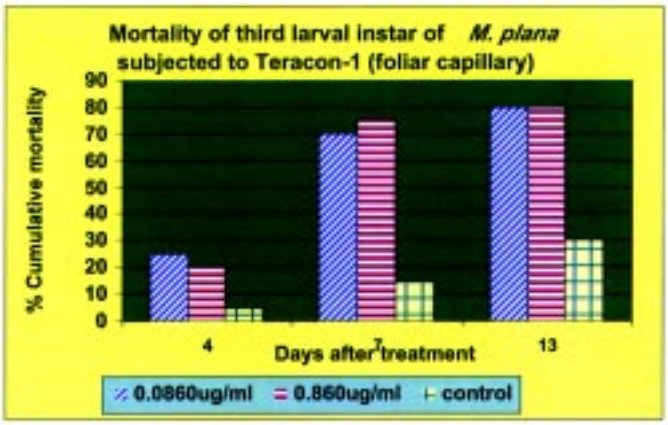

Figure 4. Mortality of M. plana *treated with Teracon-1 delivered via foliar capillary technique.*

FIELD TRIAL

Field application of Teracon-1 against *M. plana* using trunk injection technique was undertaken at MPOB Teluk Intan on mature palms (*Figure 5*). Two different doses of Teracon-1 namely 10 ml and 20 ml were used for the first generation application which was conducted on 12 April 2005. Controls include monocrotophos and untreated. The field trial was conducted in replicates of four, with plots sized 12 x 12 palms and three rows of guard palms. Central 6 x 6 palms were used as recording palms. The data gathered from the first generation application indicated that the rate of Teracon-1 need to be increased (*Figure 6*). The rate of Teracon-1 applied was increased to 20 and 30 ml for the second generation of *Pteroma pendula* larvae which hatched from the unaffected pupae. The second generation application was given via trunk injection on 11 May 2005. Teracon-1 at 30 ml per mature palm substantially reduced the bagworm population as compared to untreated.

Figure 5. Trunk injection of Teracon-1 at MPOB Teluk Intan for control of bagworm.

BENEFIT OF THE PRODUCT

The Teracon-1 is non-toxic and free from undesirable beta-exotoxin. It is a cell free concentrate of soluble Cry proteins, easily taken up to the foliage via capillary force for the control of bagworms and nettle caterpillars. It is a product from MICROTEC effluent or waste. Teracon-1 generates revenue for MICROTEC and solves the problem of effluent treatment. Bt application is made easier and there is no need for turbo-mist blower.

ECONOMIC ANALYSIS

The fixed cost for MICROTEC is RM 10 000 000. The pay back period is seven years, with internal rate of return (IRR) of 16% and the net present value (NPV) at 10% discount rate is RM4 729 893. The benefit cost ratio (BCR) for discount rate of 10% is 1.38.

RECOMMENDATION

This form of Bt product is recommended for estates which do not have turbo mist blower powerful enough for spraying the mature palms. It can be trunk injected using conventional trunk injecting device. It is also recommended that the trunk injection be conducted against early larval instars of bagworm.

CONCLUSION

The use of Teracon-1 is recommended to reduce reliance on chemicals. No extra device is required for delivering it to the palm foliage.

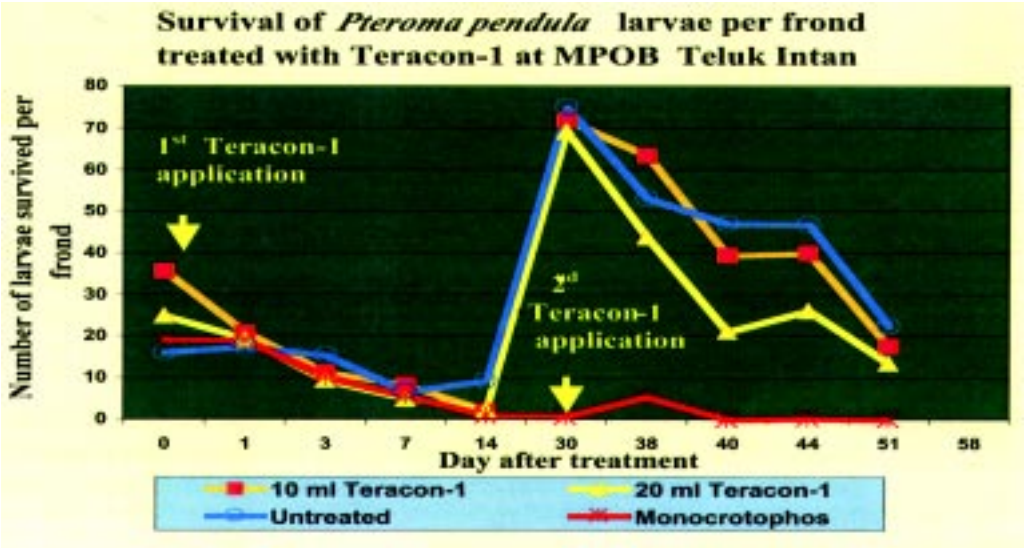

Figure 6. Teracon-1 for the controlling multi-stages infestation of Pteroma pendula *at MPOB Teluk Intan.*

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