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Significance of bacterial bioluminescence in agriculture

Introduction:

Production and emission of light from a living organism is called Bioluminescence. Important bacterial genera having luminescence activity are Vibrio, Photobacterium, Xenorhabdus and Photorhabdus. These bacteria glows in dark when grown on artificial biochemical media.

Biochemistry of bioluminescence:

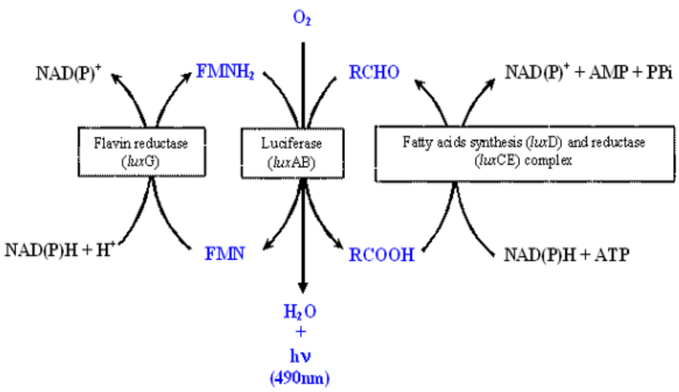

Fig 1: Bioluminescence reaction.

Luciferase enzyme catalyze the bioluminescence reaction. LuxAB genes operates this bioluminescence pathway. Three main substrates mediate this light reaction: the dissolved oxygen supplied through the surrounding environment, bacterial metabolic product (reduced flavine mononucleotide) and aldehyde coded by the luxCDE genes.

Genetics of bioluminescence:

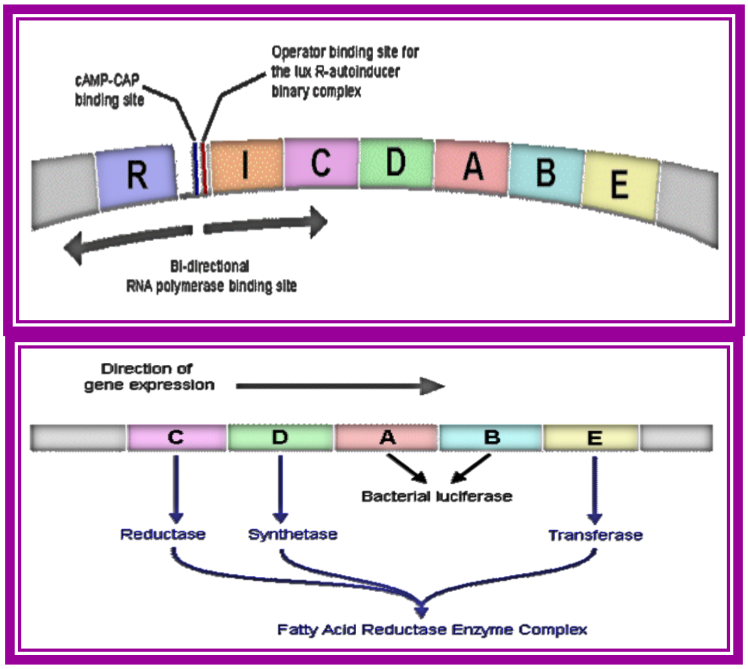

Fig 2: Lux operon: Genes encoding key components of bacterial bioluminescence.

The enzyme luciferase catalyse reaction between luciferin and oxygen which in turn produce light. The genes responsible for bioluminescence gathered on an inducible lux operon. The luxI gene directs bacteria to synthesize the autoinducer Homoserine lactone. The luxR gene product is believed to function as a receptor for the autoinducer to activate the expression of the lux operon that results in luminescence. The luxA codes for alpha subunit of luciferase enzyme and The luxB codes for beta subunit of luciferase enzyme. Both the alpha and beta subunit combines with each other and form active luciferase enzyme. The luxC, luxD and luxE combinely codes for fatty acid reductase enzyme complex.

Applications in agriculture: a) Biocontrol, b) Biosensor, c) Development of autoluminescent plants

a) Biocontrol:

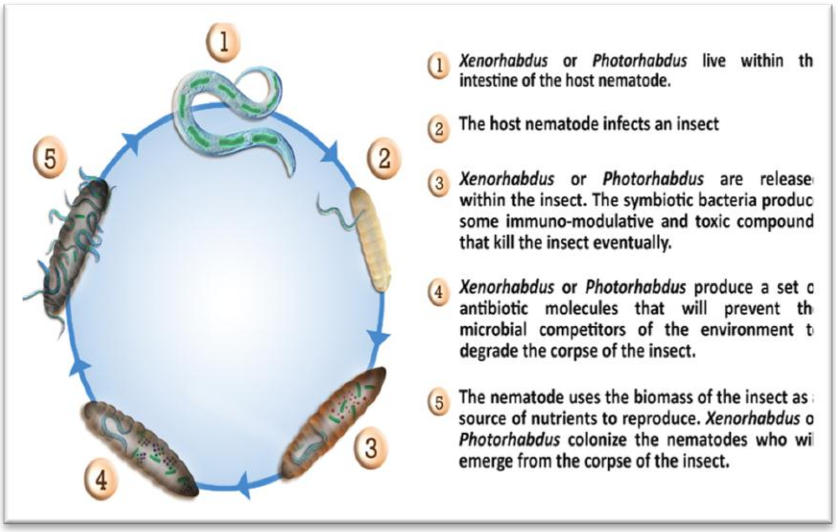

Fig. 3:Life cycle of bioluminescence bacteria with entomopathogenic nematodes EPN

The life cycle of bioluminescence bacteria Photorhabdus and Xenorhabdus having main role in biocontrol of plant pathogens begins and ends with the colonization of the intestinal tract of a soil-dwelling and non-feeding stage of the entomopathogenic nematodes (EPN) from the Heterorhabditidae and Steinernematidae families respectively. The bacteria are normally found colonizing the gut of infective juvenile (IJ)-stage nematodes. After inflowing within insect host, IJs move around the haemolymph where bacterial symbionts are released in the haemolymph, here the bacteria duplicate and destroy the insect while converting the insect dead body into a nutrient rich material that supports growth and development of nematode. (Goodrich-Blair and Clarke, 2007).

1) Phyto-pathogenic insects

Wang et al. (2012) isolated a protein complex from X. nematophila and confirmed it as Xnpt protein complex by peptide sequencing and confirmed it by bioinformatics tools i.e. Mascot Protein Search Program. The protein complex showed very high insecticidal activity which is established by antifeedent and LC50 assay showed very high oral virulence to diamondback moth (DBM) larvae, Plutella xylostella L. by inhibiting the activities of various proteases of insect midgut.

2) Phyto-pathogenic nematodes

Vyas et al. (2008) reported that in case of root knot disease caused by M. Javanica, with high shoot and root parameters in exo and endo-toxin factor (2% v/w) treatments while average galls in groundnut root was considerably little with different Xenorhabdus isolates.

3) Phyto-pathogenic fungi

Fang et al. (2016) reported that cell-free filtrate of X. nematophila TB culture showed powerful (90%) inhibitory effect on Phytophthora capsici and Botrytis cinerea mycelia growth. The cell-free filtrate and bioactive compounds (1000 μg/mL) of TB culture showed protective and therapeutic effects (70% decrease in disease) on grey mold both in plants and detached tomato fruits.

b) Biosensors:

Bioluminescent biosensors are constructed using colonies of live bacteria that have a luciferase (Lux) gene bound to another gene that is activated during an environmental alteration and light is produced, physically signaling the change.

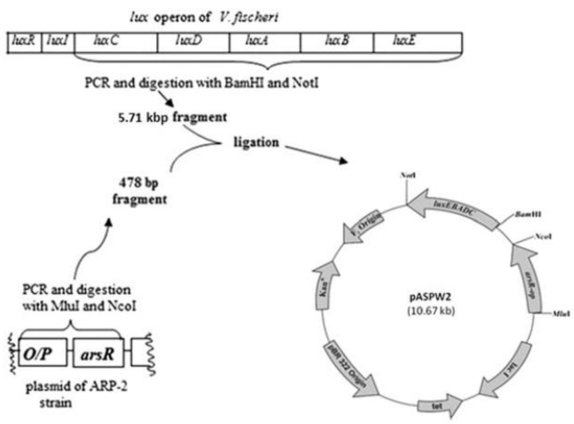

Fig. 4: Construction of plasmid pASPW2 (10.67 kb). luxCDABE (5.7 kb) genes from genomic DNA of V. fischeri was amplified and digested by BamHI and NotI. A fragment of 478 bp was amplified from plasmid DNA of wild-type E. coli strain and digested with NcoI and MluI.

Sharma et al. (2017) developed bioreporter E. coli pASPW2 sensor specific for arsenite and arsenate detection and showed increased luminescence as the concentration of both the metal increases.

b) Development of plants with autoluminiscence:

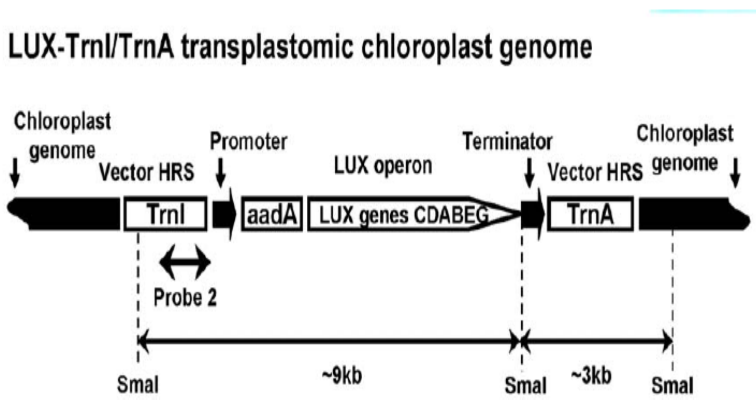

Fig. 5: Vector pCAS3-LUX-trnI/trnA transformed within plastid

Krichevsky et al. (2010) produced Nicotiana tabacum transplastomic plants, having the lux operon from P. leiognathi. The transplastomic tissues emitted 82 x 10^6 photons/min of visible light. Upon autoradiography of the transformed LUX-TrnI/TrnA shoots produced focused and defined image spots around the transplastomic tissue, while no such emission was detected with the wild-type tissue.

Recent research & development in India on lux operon:

- Plutella and Helicoverpa are showing resistant to transgenic to some extent in view of this problem the futuristic transgenic crop will be based cry + lux gene (multiply gene stacking technology) in chloroplast expressing new generation plant are under way to develop by ICGEB (international center for genetic engineering and biotechnology), New Delhi.
- Gus genes are commonly used as marker for transgenic crop but presently lux gene is being promoted as reporter gene.

Conclusions:

- Emission of light by biological system is known as bioluminescence which is a trait of bacterial genera Vibrio, Photobacterium, Photorhabdus and Xenorhabdus due to reaction between luciferin and oxygen mediated by luciferase enzyme regulated by inducible lux operon
- Most familiar bioluminescence bacteria for biocontrol include Xenorhabdus spp. and Photorhabdus spp. which live in symbiosis with entomopathogenic nematodes Hetrorhabdtis and Steinernema. which attract insect by emitting light and producing various toxins thereby inhibiting phyto-pathogenic insects, nematodes and fungi
- Genetically engineered bioluminescence bacteria very well employed as biosensors for monitoring pollution of soil and water as well as generate autoluminescent plants

Future thrusts:

- Efforts are required to explore novel genera of bioluminescent bacteria useful in agricultural sector
- Biology and Genetic engineering should join hands to explore bioluminescent bacteria for development of biosensors to monitor soil, water and air pollution as well as detoxification of the contaminant from the polluted sites
- Research is to be focused to introduce bioluminescence genes in to ornamental plants and road side trees which can light up houses as well as streets with saving in electricity and environment.

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