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Biopesticides and Their Role in Sustainable Agricultural Production

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Abstract

Biopesticides are derivatives of plants, microorganisms and insects. Substances from plants and animals have been used to manage diseases in crops, animals and humans. Reliance on nature to heal nature is a practise for many people around the world. Use of natural products was overtaken by synthetic chemicals due to their efficacy, reliability and quick knock down effect. However, synthetic pesticides have become a health hazards for humans and environment due to their toxicity and pollution. Biopesticides are potential alternatives to synthetic pesticides. Sources of biopesticides are readily available, easily biodegradable, exhibit various modes of action, are less expensive and have low toxicity to humans and non-target organisms. Neem, pyrethrum, cotton and tobacco are known sources of botanical pesticides and have already been commercialized. Other sources of botanical pesticides include garlic, euphorbia, citrus, pepper among others. Species of *Trichoderma*, *Bacillus*, *Pseudomonas*, *Beauveria* have been commercialized as microbial pesticides. Biopesticides are however faced with challenges of formulation, registration, commercialization, acceptance and adoption. This paper describes several aspects of biopesticide development, including but not limited to, their sources, production, formulation, commercialization, efficacy and role in sustainable agriculture.

Keywords

Biopesticides, Conservation Agriculture, Integrated Pest Management, Environmental Safety

1. Introduction

Biopesticides are products and by-products of naturally occurring substances such as insects, nematodes, microorganisms, plants as well as semiochemicals [1]. Based on the nature and origin of the active ingredients, biopesticides fall into several categories such as botanicals, antagonists, compost teas, growth promoters, predators and pheromones [2]. Plants and microorganisms are the major sources of biopesticides due to the high components of bioactive compounds and antimicrobial agents [3]. The active compounds in plants include phenols, quinones, alkaloids, steroids, terpenes, alcohols and saponins [4]. Different plant families have varied antimicrobial bioactive compounds which include oil components such as α- and β-phillandrene, limonene, camphor, linalool, β-caryophyellene and linalyl acetate depending on the plant family [5] [6]. Microbial biopesticides include bacteria species such as *Pseudomonas*, *Bacillus*, *Xanthomonas*, *Rahnella* and *Serratia* or fungi such as *Trichoderma*, *Verticillium* and *Beauveria* species [7]. Biopesticides exhibit different modes of action against pathogens such as hyperparasitism, competition, lysis and predation [8].

Plant growth promoting rhizobacteria protect plants from biotic and abiotic stresses and they also enhance plant growth and enhance formation of root hairs [9]. The most common species of plant growth promoting rhizobacteria include *Agrobacterium*, *Ensifer*, *Microbacterium*, *Bacillus*, *Rhizobium*, *Pseudomonas*, *Chryseobacterion* and *Rhodococcus* [10]. They colonize the environment around the plant roots, fix nitrogen, increase phosphate solubilisation and result in general increase in plant yield [11]. Species of *Pseudomonas* and *Bacillus* have been used as biofertilizers with reports showing increase in plant growth, yield and phosphorous and zinc content in fruits and soils [12]. Natural enemies including predators, pathogens and some insects are also used as biopesticides in management of insect pests. Parasitoids, wasps, beetles, lace wings, bugs and lady birds are used in management of destructive pests such as boll worms (*Helicoverpa armigera*) in important crops such as cotton [13] [14]. Compost teas are filtrates of compost extracts and are similarly used as biopesticides [15].

This review discusses the current status of knowledge on biopesticides including their sources, production, formulation, commercialization, role in sustainable agriculture and their limitations. It also brings together the different types of biopesticides and evidences of their use against important pests in different crops.

2. Limitations and Challenges in the Use of Conventional Pesticides

There are harmful effects associated with the use of synthetic pesticides such as toxicity and poisoning [16]. Synthetic pesticides also lead to environmental pollution due to the unbiodegradable nature of their constituent compounds [17]. According to Parliman [18], degradation of metham sodium and other fumigants was reported to last up to over six months after application. In a report by PAN [19], metham sodium pollutes the air and soil thereby affecting the population of natural enemies in the soil. Methyl bromide has been banned from agricultural use due to its negative impact on the environment. It is associated with depletion of ozone layer which contributes significantly to climate change [20]. The constituent compounds of chemical pesticides contaminate soils rendering them unsuitable for crop production [21]. They also pollute surface and ground water, killing aqua life after inhalation and consumption [22]. Use of dichloro diphenyl trichloroethane (DDT) for instance led to poisoning of birds, marine species and humans. It has been reported to have carcinogenic properties leading to its ban from agricultural use [23].

Continuous use of synthetic pesticides leads to development of resistant plant pathogen strains leading to their resurgence. Farmers apply more chemicals in an effort to eradicate such pests [24] [25]. In the process of managing target pests, synthetic pesticides kill non-target beneficial organisms such as pollinators, predators and antagonists thereby disrupting biodiversity [26] [27]. After application, the active compounds of the synthetic pesticides are taken up and retained by crops. Consumption of such crops poses chronic health problems to humans due to the accumulated toxic chemical residues [28]. Exposure to pesticides adversely affects the human population, directly or indirectly. For example, pesticides containing Malathion and Trichlorfon have been reported to cause reproductive complications in humans [29]. Exposure to some pesticides have also been reported to retard growth, induce chemical and structural changes in body organs as well as disturb immune responses. They also reduce resistance of animals to disease-causing pathogen infections [22]. Continuous exposure to pesticides such as chloropyrifos cause gene mutations, genetic damages, reproductive health problems and chronic diseases such as asthma, hypertension and cancer [30] [31].

In a study by Xavier *et al.*, [32], application of Fenpyroximate on chilli peppers (*Capsicum annum* L) resulted in retention of its residues even after sun drying and processing. Similarly, spinosad (spinosyn A and spinosyn D), Indoxacarb and Deltamethrin containing insecticides used to control *Rhizoctonia dominica*, *Sitophilus oryzae* and *Trogoderma granarium* were found to be persistent for up to 120 days after application [33].

The horticulture sector in many developing countries has been particularly adversely affected by the use of synthetic pesticides. The European Union (EU) set out strict regulations regarding levels of pesticide residues and safety of agricultural produce exported to their markets. The use of pesticides containing Dimethoate on vegetables was banned by EU. Failure to comply with this regulation led to rejection and destruction of fresh vegetable consignments containing chemical residues above the required limits [34]. Residues of the restricted chemicals should not exceed 0.02 parts per million (ppm) in a sample of vegetables. The percentage of inspection was increased to 10% on fresh produce at ports of entry into the European Union [35]. According to European Commission [36], Maximum Residue Levels (MRLs) of unknown pesticides should not exceed 0.01mg/Kg and there was imposed a 10% sampling per consignment in fresh beans and pods. Interceptions of fresh produce almost ruined Kenya's export market reputation due to presence of traces of banned pesticides [37]. Following the guidelines made by the EU and the losses incurred due to rejection and destruction of fresh vegetable consignments, there was a reduction in volumes of horticultural exports. This negatively affected the livelihoods of small holder farmers who are the major producers of vegetable crops [38]. This led to introduction of a cloud-based traceability system which uses a quick reference (QR) code and GPS coordinates to pinpoint the individual farmer whose consignment fails to comply with regulations [39]. This has resulted in increase of the cost of production and several farmers opted out of the export business.

3. Biopesticides of Botanical Origin

Based on the method of extraction, botanical pesticides can either be plant extracts or essential oils [6]. They are obtained from plants parts such as leaves, barks, flowers, roots, rhizomes, bulbs, seeds, cloves or fruits which are either fresh or dried. Dried plant parts are preferred as this reduces water concentration resulting in higher yield of active ingredient [40]. A Gas Chromatography-Mass Spectrometry (GC-MS) analysis was carried out on *Citrus sinensis* and d-limonene and myrcene were reported as the major constituents of the oil component. The products were tested against a cereal leaf beetle (*Oulema melanopus*) on wheat and a mortality of up to 85% was reported on larvae observed in 48 hours [41]. Aqueous fruit extracts of *Withania somnifera* were tested for activity against *Fusarium oxysporum* f.sp. *radicis-lycopersici*, the causal agent of fusarium crown and root rot disease in tomatoes. At a concentration of 2% the extracts inhibited growth of the fungal pathogen by up to 56% *in vitro* [3].

Table 1 shows examples of plants reported to have potential as sources of botanical pesticides and respective target pests. *In vitro* experiments involving several ethanolic plant extracts, turmeric (*Curcuma longa*), lemon (*Citrus limon*), garlic (*Allium sativum*), pepper (*Capsicum frutescens*) and ginger (*Zingiber officinale*) were reported to be significantly effective against *Alternaria solani*, *Pythium ultimum*, *Rhizoctonia solani* and *Fusarium oxysporum* f. sp. *lycopersici* [42]. Turmeric (*Curcuma longa*) was reported to be the most effective with a growth inhibition of up to 73% against *Alternaria solani*. *Pseudomonas syringae* p.v. *tomato* was effectively managed *in vitro* by *Rhus coriaria*, *Eucalyptus globulus* and *Rosmarinus officinalis* [43]. *Eucalyptus globulus* was reported efficacious in preventing the bacterial speck of tomato (*Pseudominas syringae* p.v. *tomato*) by up to 65% under greenhouse conditions. At 5% concentration, a mortality rate of up to 78% was reported on juveniles of root knot nematodes (*Meloidogyne* sp) by extracts of *Nerium oleander*. When the concentration was increased to 10% a mortality of between 65% - 100% was observed on second stage juveniles treated with extracts of *Eucalyptus* sp, *Cinnamomum verum*, *Nerium oleander*, *Azadirachta indica*, *Zingiber officinale* and *Allium sativum* [44]. The most common and already commercialized botanical pesticides are derived from neem (*Azadirachta indica*), pyrethrum (*Chrysanthemum cinerariifolium*), sabadilla *(Schoenocaulon officinale*) and tobacco (*Nicotiana tabacum*) [45].

The quality of extracts and oils is highly dependent on the solvent used and method of extraction [46]. The solvents should be of low toxicity, able to dissolve as many compounds as possible, evaporate easily at low temperatures and

Table 1. Plants with potential as botanical pesticides and respective target pests.

Source plant	Target pest	Host	Reference
Azadirachta indica	*Aphis craccivora* *Aphis gossypii* *Amrasca devastans* *Myzus persicae* *Sitobion avenae* *Lipaphis erysimi* *Bemisia tabaci* *Sciothrips cardamomi* *Rhizopus* sp. *Aspergillus* sp. *Monilinia fructicola* *Trichothecium roseum* *Pythium aphanidermatum* *Alternaria alternata* *Helminthosporium* sp. *Vibrio cholerae* *Bacillus subtilis* *Meloidogyne javanica* *Meloidogyne ingognita*	*Vigna unguiculate*; *Gossypium hirsutum*; *Solanum tuberosum*; *Triticum* sp.; *Brassica* sp.; *Prunus salicina*; *Lycopersicon esculentum*; *Capsicum chinense*; *Cardamomum* sp.;	[53]-[65]
Allium sativum	*Curvularia lunata* *Aspergillus niger* *Candida albicans* *Trichophyton rubrum* *Drechslera tritici-repetis* *Bipolaris sorokiniana* *Alternaria raphanin* *Fusarium flocciferum* *Fusarium graminearum* *Rhizoctonia solani* *Colletotrichum* sp. *Bacillus subtilis* *Salmonella senflenberg* *Staphylococcus aureus* *Staphylococcus epidermidis* *Sitotroga cerealella* *Spodoptera littorals* *Tenebrio molitor* *Callosobruchus maculatus* *Plutella xylostella* *Brevicoryne brassicae*	*Triticum* sp.; *Lycopersicon esculentum*; *Abelmoschus esculentus*; *Panax* sp.; *Capsicum* sp.; *Human and animal* sp.; *Oryza* sp.; *Gossypium hirsutum*; Stored grain products; *Vigna unguiculate*; *Brassica oleracea*;	[66]-[77]
Euphorbia sp.	*Pseudomonas aeruginosa* *Enterobacter aerogenes* *Escherichia coli* *Salmonella typhi* *Aspergillus flavus*	*Human and animal* sp.; *Arachis hypogaea*;	[78] [79] [80] [81]
Curcuma longa	*Tribolium castaneum* *Bactrocera zonata* *Trichoplusia ni* *Alternaria solani* *Streptococcus pyogenes* *Streptococcus mutants* *Ralstonia solanacearum* *Escherichia coli* *Listeria monocytogenes* *Bacillus subtilis*	*Triticum aestivum*; *Prunus persica*; *Brassica oleracea*; *Solanum lycopersicum*; Human sp.; Animal sp.;	[42] [82]-[89]

Continued

Tagetes spp.	*Fusarium oxysporum* f.sp. *gladioli* *Fusarium oxysporum* *Klebsiella pneumoniae* *Brevicoryne brassicae* *Plutella xylostella* *Mamestra brassicae* *Meloidogyne incognita*	*Gladiolus grandifloras*; *Leucadendron*; Human and animals spp.; *Brassica oleracea*;	[90] [91] [92] [93] [94]
Cinnamomum zeylanicum	*Botrytis cinereal* *Penicillium expansum* *Aspergillus oryzae* *Fusarium solani* *Escherichia coli* *Staphylococcus aureus* *Bacillus subtilis* *Salmonella typhimurium* *Bursaphelenchus xylophilus* *Meloidogyne* sp.	*Zea mays*; *Pinus densiflora*;	[43] [95] [96] [97] [98] [99]
Thymus vulgaris	*Penicillium* spp. *Saccharomyces* spp. *Aspergillus niger* *Tilletia tritici* *Xanthomonas vesicatoria*, *Escherichia coli* *Salmonella typhimurium* *Diaphorina citri* *Megalurothrips sjostedti* *Eloidogyne incognita* *Helicotylenchus dihystera* *Pratylenchus brachyurus*	*Gallus gallus domesticus*; *Triticum aestivum*; *Solanum lycopersicum*; *Citrus aurantium*; *Cajanus cajan*; *Gossypium hirsutum*;	[2] [100]-[107]
Jatropha spp.	*Aphis fabae* *Sitophilus zeamais* *Rhyzorpertha dominica* *Tribolium castaneum* *Oryzaephilus surimanensis* *Aspergillus flavus* *Alternaria alternate* *Penicillium glabrum* *Bactrocera cucurbitae* *Meloidogyne incognita*	*Vigna unguiculate*; *Zea mays*; *Triticum aestivum*; *Solanum melongena*;	[108]-[113]
Zingiber officinale	*Fusarium oxysporum*, *Pythium aphanidermatum* *Rhizoctonia solani* *Aspergillus flavus* *Aspergillus parasiticus* *Escherichia coli* *Salmonella typhi* *Tribolium castaneum* *Drosicha mangiferae* *Trichoplusia binotalis* *Necrobial rufipes* *Dermestes maculatus*	*Solanum lycopersicum*; *Arachis hypogaea*; *Coffea* spp.; *Mangifera indica*; *Oyza sativa*; *Brassica oleracea*; *Clarias gariepinus*;	[114]-[119]

should preferably possess preservative properties [47]. The choice of solvent is dictated by the target active compounds. Although water is the universal solvent, it extracts less antimicrobial compounds compared to other solvents [48]. Organic solvents such as ethanol and methanol yield better extracts and their results are consistent. Other extraction solvents include dichlomethane, acetone and hexane [49]. According to a study by Wetungu *et al.*, [50] methanol and hexane extracts from *Tarchonanthus camphoratus* gave higher growth inhibition capacity against *Escherichia coli*, *Salmonella typhi*, *Staphylocccus aureus*, *Proteus mirabilis*, *Candida albicans* and *Klebsiella pneumoniae.* Methanol was also reported to be efficient in extracting seed components of *Morinda citrofolia* and the extracts showed antibacterial activity of up to 63% against *Escherichia coli* and *Pseudomonas* spp [51]. The differences among the different solvents are attributed to their polarity [52]. The plant parts from which the extracts are obtained also influences the quality of the extracts.

4. Microorganisms as Sources of Biopesticides

Microorganism-based biocontrol agents form the bulk of commercialized biopesticides and they include bacteria, viruses, fungi, nematodes and protozoa [120]. There are up to 175 reported microbial based biopesticide active agents and they have been used in management of pathogens, weeds, insects and nematodes [121]. Examples are illustrated in **Table 2**. Majority of the microbial biopesticides are used to manage soil borne pathogens [8]. Bacterial species that have been utilised as biopesticides include *Bacillus*, *Pseudomonas*, *Burkholderia*, *Xanthomonas*, *Enterobacter*, *Streptomyces*, *Serratia* and these are either obligate facultative or crystalliferous. Fungi used as biopesticides include species of *Trichoderma*, *Beauveria*, *Metarhizium*, *Paecilomyces*, *Fusarium*, *Pythium*, *Penicillim* and *Verticillium. Steinernama* and *Heterarhabditis* are nematode species used to make biopesticides [7]. The mechanisms of action exhibited by microorganisms against plant pathogens include hyperparasitism, competition, secretion of volatile compounds, antibiosis and parasitism [122].

The major sources of microorganisms with pesticidal activity are agricultural fields where they co-exist with other microorganisms including pathogens and beneficial species [123]. The rhizosphere is usually concentrated with various classes of important microorganisms. Other rich sources of microorganisms include hay, manure, cow shed, as well as straw [124]. Formulation of the microbial pesticides has a great contribution to the effectiveness of the resultant product and it is usually dependent on the substrate used. A study done by Adan *et al.*, [125] showed that a formulation of *Trichoderma harzianum* prepared in black gram bran, peat soil and water had a high level of activity against damping off of eggplant seedlings caused by *Sclerotium rolfsii*. The activity was attributed to the high number of spores produced by the fungus.

According to Prasad and Syed [26], exposure of *Helicoverpa armigera* to conidial suspension of *Beauveria bassiana* resulted to antifeeding habits, blackening

Table 2. Examples of microorganisms with potential as biopesticides.

Microorganism	Target pest	Host	Reference
Trichoderma spp.	*Penicillium stekii* *Fusarium* spp. *Ceratocystis radicicola* *Pythium* spp. *Alternaria* spp. *Rhizoctonia* spp. *Verticillium* spp. *Mcrophomina phaseolina* *Botrytis cinereal* *Phytotphtora* spp. *Meloidogyne* spp.	*Lycopersicon esculentum*; *Phoenix dactylifera*; *Zea mays*; *Gossypium hirsutum*; *Solanum lycopersicon*; *Solanum tuberosum*; *Capsicum annum*; *Solanum melongena*;	[127]-[126]
Beauveria spp.	*Dendrolimus tabulaeformis* *Spodoptera litura* *Uvarovistia zebra* *Cyclocephala lurida* *Frankliniella occidentalis* *Hylobius abietis* *Oryzaephilus surinamensis* *Tuta absoluta* *Polyphylla fullo*	*Pinus tabulaeformis*; *Phaseolus vulgaris*; *Brassica oleracea*; *Avena sativa*; *Triticum aestivum*	[137]-[146]
Paecilomyces spp.	*Meloidogyne javanica* *Phytophthora palmivora*	*Solanum lycopersicon*; *Vigna mungo*; *Theobroma cacao*;	[147]-[152]
Bacillus spp.	*Macrophomina phaseolina* *Fusarium graminearum* *Colletotrichum* spp. *Phaeomoniella chlamydospore* *Fusarium solani* *Meloidogyne incognita* *Spodoptera frugiperda* *Xanthomonas campestris* pv. *campestris*	*Glycine max*; *Triticum aetivum*; *Capsicum annuum*; *Vitis vinifera*; *Vicia faba*; *Vigna mungo*; *Solanum lycopersicon*;	[153]-[163]
Metarhizium spp.	*Musca domestica* *Spodoptera litura* *Ceratitis capitate* *Polyphylla fullo* *Tetranuchus urticae* *Maruca vitrata* *Tuta absoluta* *Callosobruchus maculatus*	*Citrus sinensis*; *Polyphylla fullo*; *Phaseolus vulgaris*; *Solanum lycopersicon*;	[146] [164]-[170]
Pseudomonas spp.	*Vibrio harveyi* *Fusarium oxysporum* *Alternaria alternata* *Sclerotium rolfsii* *Botryodiplodia theobromae* *Rhizoctonia solani* *Aspergillus aculatus* *Dreshlera biseptata*	*Mangifera indica*; *Solanum lycopersicon*; *Solanum melongena*; *Vitis vinifera*; *Oryza sativa*; *Triticum aetivum*;	[171] [172] [173] [174] [175]
Verticillium spp.	*Trialeurodes vaporariorum* *Bemicia tabaci*	*Phaseolus vulgaris*; *Euphorbia ulcherrima*;	[176] [177]

of the body and the larvae becoming sluggish and morbid. The fungus finally consumes the entire larval tissues resulting in its death. Beric *et al.*, [124] reported that isolates of *Bacillus* showed antagonistic activity against rice pathogen, *Xanthomonas oryzae* p.v. *oryzae*, and the activity was attributed to production of a bacteriocin by the bacterium. Treatment of wheat and rice plants with concentrations of *Chaetomium globosum* reduced the severity of wheat rust (*Puccinia recondite*) and rice blast (*Magnaporthe grisea*) by up to 80% [126]. Late blight (*Phytophthora infestans*) on tomato was also controlled by *Chaetomium globosum* by up to 50% while mycelial growth of *Pythium ultimum* was inhibited *in vitro* in well diffusion assays. The activity of the fungus was attributed to production of two types of chaetoviridins, A and B [126]. These studies indicate that microbial biopesticides can be incorporated in integrated pest management for sustainable agriculture.

5. Predators and Parasitoids as Biopesticides in Agriculture

A predator kills and feeds on prey while parasitoids grow on or inside their hosts and eventually kill them [178]. The predators include beetles (Carabidae), ladybirds (Coccinellidae), spiders, lacewings (Chrysopidae) and true bugs while parasitoids mainly consist of wasps and other hemipterans [13]. Examples of predator organisms used in management of crop pests are presented in **Table 3**. These natural enemies are mainly found in the environment and are not evenly distributed. In order to have them in large numbers, they are either reared under controlled conditions and released into the fields or are multiplied in open fields containing the prey [179]. The most common way of rearing these predators is by growing them on their preferred hosts. This is either done in screen houses or growth chambers where the host plants are first grown and then exposed to pest infestation [180]. The predators are then introduced and they are maintained by growing on the prey [181]. Alternatively, the predators can be grown in cylinders where they are supplied with the prey and all other necessary conditions for growth are provided. An example is the mass rearing of *Phytoseiulus persililis* on *Tetranychus urticae* Koch [179]. The optimum growth of predator mite, *Neoseiulus californicus*, was observed when grown on an artificial diet supplemented with eggs of *Ephestia kuehniella*, *Artemia franciscana* cysts and maize bran [182]. Such artificial diets are important in reproduction, development and survival of the predators during rearing as well as reduction of production costs.

Predators can also be grown on egg masses of their prey or other suitable hosts which gives them a longer storage capacity. This has been employed in management of mealy bugs using parasitoids [183]. The predators can also be grown on other feeds such as rice bran as long as it provides the necessary nutrients to the insects [184]. Due to economic concerns, these organisms are reared on artificial media with carefully evaluated nutritional needs and requirements. Their growth media ranges from beef and liver to crushed lepidopteran pupae. These provide a combination of hormones and nutrients needed by

Table 3. Examples of predator organisms used in management of crop pests.

Predator	Target pest	Host	References
Phytoseiulus spp.	*Tetranychus urticae* *Tetranychus evansi*	*Phaseolus vulgaris*; *Solanum lycopersicon*; *Fragaria ananassa*;	[180] [189] [190]
Neoseiulus spp.	*Tetranychus urticae* *Oligonychus perseae*	*Vicia faba*; *Capsicum annuum*; *Persea americana*;	[191] [192]
Amblyseius swirskii	*Scirtothrips dorsalis*	*Capsicum* sp.;	[193]
Aphidius colemani	*Aphis gossypii*	*Dendranthema grandiflora*;	[194]
Diglyphus isaea	*Liriomyza huidobrensis*	*Solanum tuberosum*;	[195]
Encarsia formosa	*Bemisia tabaci*	*Solanum lycopersicon*;	[196] [197]

the predators for growth [185]. Artificial media provides as good nutrients as the host plants and reduces the cost of growing the plants. The artificial media is mostly used in laboratories and has been used for rearing *Trichogramma* and *Anastatus* spp [186].

An *in vitro* study by Xu and Enkegaard [187] showed that *Amblyseius swirskii* predated on *Frankliniella occidentalis* and *Tetranychus urticae* nymphs with preference to their first instars. The predation rate on *T. urticae* was 4 - 6 nymphs in 12 hours. The authors reasoned that the outcome of the predation is highly dependent on several factors among them being the host plant traits. A synergistic effect on predation between *Amblyseius swirskii* and *Phytoseiulus persimilis* against two-spotted spider mite (*Tetranychus urticae*) with a mortality rate of up to 86% was reported by Fiedler [188]. Introduction of *Amblyseius californicus and Amblyseius degenerans* into a population of *Tetranychus urticae* under laboratory conditions recorded a mortality of up to 72% within 15 days.

6. Production, Formulation and Commercialization of Biopesticides

Botanical pesticides are prepared from plants and plant parts obtained from the environment, natural or man-made [198]. The materials are cleaned of dirt or foreign materials and then extracted either using solvents or distillation to obtain extracts or essential oils, respectively [199]. The resultant extracts are then subjected to screening for activity *in vitro* against different pests using different methods such as disc diffusion, agar well diffusion, agar dilution and poisoned food technique [200] [201]. The most active botanicals are then evaluated for efficacy in managing pests and diseases under field conditions. The active constituents of the selected extracts are then identified for optimum formulation [202]. Intensive laboratory and field trials conditions are carried out to ensure that the most efficacious combination of the active compounds, carrier materials, emulsifiers, surfactants and other components used in pesticide development are optimized. The efficacy report from the laboratory and field trials is used to request for registration of the product from the pest control products body. (**Figure 1**)

Production of microbial pesticides follows the same procedure as botanicals, except the that the antagonistic microorganisms are collected from sources like the cowshed, hay fields, rhizosphere, compost and manure [203]. They are isolated in to pure cultures in the laboratory and maintained in agar slants [204]. *In vitro* efficacy trials are carried out following methods such as dual culture, agar discs diffusion and agar well diffusion [205]. The active microorganisms are multiplied on a suitable substrate in the laboratory and mixed with carrier materials, enhancers and stabilizers for field application [206]. Repeated laboratory and field efficacy trials are conducted until registration process begins (**Figure 1**).

Before the natural products are commercialized, they are usually tested in the laboratory and under field conditions for efficacy against the target pests. The active compounds are also identified using techniques such as thin layer chromatography (TLC), high-performance liquid chromatography (HPLC) and gas chromatography-mass spectrometry (GC-MS) [207] [208]. Stabilizers and carrier materials are always added to the active compounds to enhance their applicability and longevity. Formulation of the active compounds should improve the stability of the compound as well as increase its efficiency and applicability. It should also reduce its degradability due to climatic factors such as heat, water and acids. Carrier materials majorly used include petroleum distillates, corn starch, talc, clays, and water. Emulsifiers such as soap are also added to the compounds during formulation and they are optimized to ensure effectiveness is

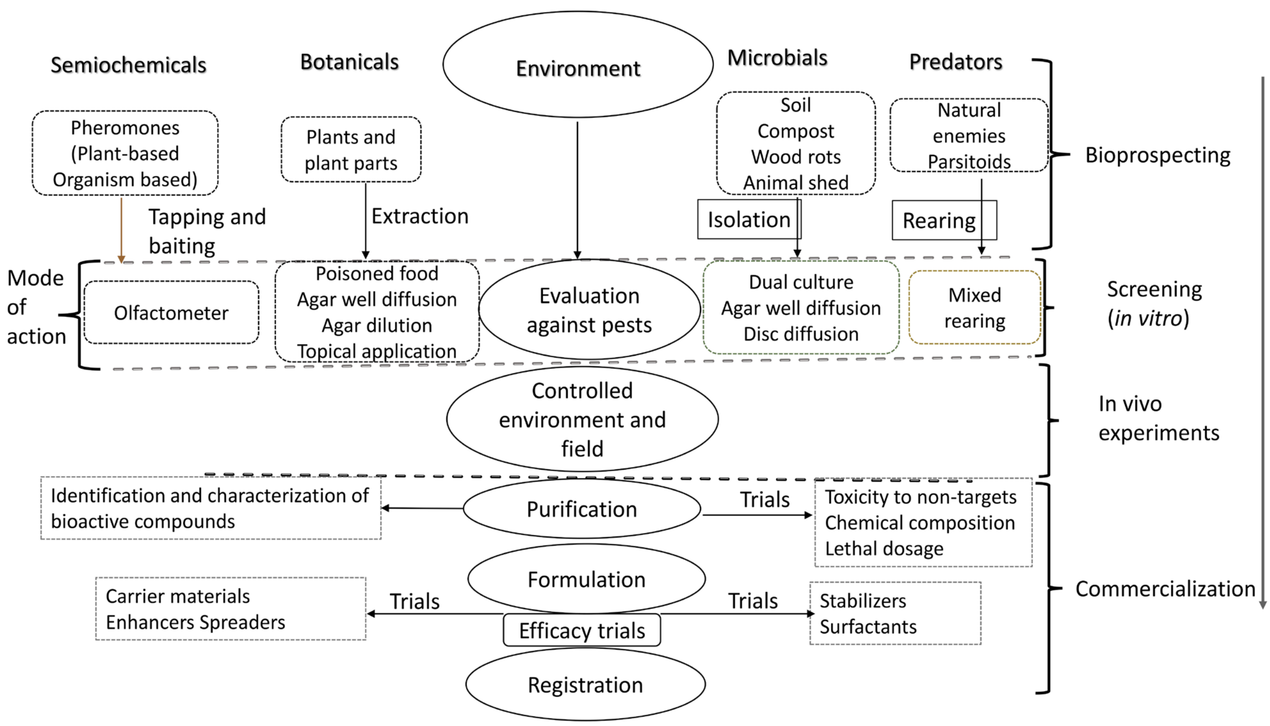

Figure 1. Steps in production and evaluation of different types of biopesticides.

not lost. Biopesticides of plant origin that have been formulated and commercialized for agricultural use include neem and pyrethrum [209] while microorganisms include species of *Bacillus* and *Trichoderma* [210].

7. Modes of Action of Biopesticides

Each type of biopesticide exhibits varied modes of action. Microbial pesticides act on pathogens by antagonism, hyperparasitism, antibiosis and predation. Botanical pesticides inhibit growth of pathogens, modify their cellular structures and morphology and exhibit neurotoxicity on insects. Botanicals also repel insects, suppress oviposition and feeding. Predators mainly kill the prey through parasitization or injection of toxic substances which eventually kill the prey. Semiochemicals are used to lure the target pests and they can then be managed through other means such as sterilization or death.

Extracts from plants belonging to Asteraceae family have been reported to inhibit hyphal growth and induce structural modifications on mycelia of plant pathogenic fungi [6]. Asteraceae plants contain compounds such as flavanoids, coumarins alkaloids and terpenoids which could lead to absolute fungal toxicity. Some compounds lead to changes in the cell wall as well as the morphology of celleular organelles [211]. In some instances, the bioactive compounds cause partitioning of fungal cell membranes making them permeable leading to leakage of cell contents. Plant bioactive compounds also lead to separation of the cytoplasmic membrane which leads to damage of the intracellular components and swelling of cells leading to eventual death [212]. Compounds such as allicin found in garlic (*Allium sativum*) bulbs lead to suffocation of the pest due to effects on receptors of neurotransmitters [77]. Phenolics and terpenoids build hydrophobic and ionic bonds which attack a multiple of proteins in the insects leading to physiological malfunction [213]. Compounds in plant extracts and essential oils also interfere with receptor cells leading to malfunctioning of the nervous system and failure of coordination leading to death of the insect [214].

Different classes of microorganisms have different modes of actions. Hyperparasitism is one of the most reported mode of action on many biocontrol agents [215]. The antagonist kills the pathogen or its propagules while some attack the sclerotia or the hypha of the fungal pathogen. A single pathogen could be attacked by a number of biocontrol agents [216]. *Pasteuria penetrans* is an example of a biocontrol agent that parasitizes on root-knot nematodes of *Meloidogyne* spp. [217]. Species of genus *Trichoderma* exhibit a predation mode of action by producing enzymes that directly kill cell walls of the pathogens and colonize the environment therein.

Some microorganisms produce compounds that kill other microorganisms, a mechanism called antibiosis. This is most common with bacteria belonging to species of *Pseudomonas*, *Agrobacterium*, *Bacillus*, *Burkholderia*, *Pantoea* and it has also been reported in the fungus *Trichoderma* spp. [218]. Sufficient quantities of antibiotics need to be produced for enhanced biocontrol. Some microbial species such as *Bacillus cereus* produce multiple compounds that could suppress more than two pathogens and this is effective in crop disease management [219]. Other classes of microorganisms such as *Lysobacter* and *Myxobacteria* produce lytic enzymes which hydrolyze compounds leading to suppression of pathogens [220]. *Beauveria bassiana* inhibits chitin development in insects by conidia attaching to the body of insects. After germination the hypha penetrates through the cuticle and grows throughout the insect body eventually killing it [26].

Semicochemicals such as female sex pheromones are used to lure the male insect pests which are then sterilized thereby decreasing their effectiveness. Upon mating with the sterile male insects, the females lay unfertilized eggs thereby reducing harmful insect populations [221]. Host location pheromeones lure insects into sites with mass traps from where they may be sterilized or starved to death [222]. Predators may feed on the prey or a particular life stage of the prey such as nymphs or larvae [187]. The predator prey ratio is of importance in balancing the populations of the pests as well as biodiversity [223].

8. Efficacy of Different Types of Biopesticides

Synthetic pesticides are considered more effective than biopesticides in managing crop pests [224]. Their effectiveness sometimes has nonetheless no much significance in managing a particular population of pest as would the biopesticides [225]. Biopesticides in other instances perform better than synthetic pesticides when applied in the right regimes, concetrations and appropriate frequencies [226]. Research reports across the world have presented different plants, microorganisms and predators with potential as biopesticides. Natural enemies predate on insect pests which balances their population in the ecosystem. Such predators are important in agricultural systems [223] [227]. The mechanisms used by predators to lure insects include scents and other attractants. Some of these scents, called pheromones, have been commercialized and are being used in management of important crop pests such as *Tuta absoluta* [221]. The commercial pheromones are baited to aid in luring the adult insects and then deactivating them by sterilization or startvation to death [228].

Certain plants contain compounds which they use to protect themselves against pests and this ability has been explored by researchers in an effort to manage different crop pests [118]. Some plants have been found to contain compounds that are effective against several pests including fungi and nematodes [229] [230]. Some species of microorganisms have antagonistic properties towards other species and are therefore effective as biopesticides [231].

Ngegba *et al.*, [232] reported that extracts of neem (*Azadirachta indica*) and Mexican sunflower (*Tithonia diversifolia*) inhibited growth of rotting disease pathogens of tomato, *Aspergillus niger*, *Fusarium oxysporum* and *Geotrichum candidium* by up to 100%. Extracts of castor seeds (*Ricinus communis*) effectively inhibited growth of post-harvest pathogens *Penicillium oxalicum* and *Aspergillus niger* of yams (*Dioscorea alata*) in a dose dependent poisoned food technique experiment [233]. Similar effects were reported by Devi *et al.*, [234] on post-harvest fungi including *Fusarium solani*, *Rhizopus arrhizus* and *Sclerotium rolfsii* after using extracts from *Duranta erecta* and *Lasonia ineruis*. Methanolic extracts of *Chenopodium ambrosioides* exhibited antifungal activity against *Fusarium oxysporum* f.sp. *ciceris* a pathogen that causes wilt of chick pea (*Cicer arietinum*) by up to 50% [235].

A biopesticide formulation containing onion (*Allium cepa*) and ginger (*Zingiber officinale*) was evaluated for efficacy against tomato fruit worm (*Helicoverpa armigera*) and registered a 70% - 80% control [236]. During the study, yield increment was also observed on plants treated with the formulation compared to the untreated controls. Muzemu *et al.*, [237] reported over 50% reduction of rape aphids (*Brevicoryne brassicae*) and tomato red spider mites (*Tetranychus evansi*) by powder extracts of *Lippia javanica* and *Solanum delaguense.* Populations of *Megalurothrips sjostedti* were reduced by extracts of *Piper nigrum*, *Cinnamomum zeylanium* and *Cinnamomum cassia* and were reported to be strong repellents [105]. Number of larvae and pupa of *Helicoverpa armigera* were effectively reduced by extracts of C*urcuma longa*, *Allium sativum* and henge (*Ferula assa-foetida*) in a study by Shah *et al.*, [226]. Extracts of *Artemisia herbaalba*, *Eucalyptus camaldulensis* and *Rosmarinus officinalis* soaked on leaves of broad bean (*Vicia faba)* caused a mortality of 60% - 100% of green peach aphid (*Myzus persicae*) after 24 hours of exposure in dose dependent *in vitro* experiments [238]. In another study, topical application of *Azadirachta indica*, *Mangifera indica*, *Polyalthia longifolia*, *Annona squamosa* and *Ficus benghalensis* caused a 100% mortality of bed bugs (*Cimex lectularius*) after 19 seconds of contact [239]. The effectiveness of plant extracts on insects is credited to the solvents used and their ability to extract major compounds with insecticidal properties [240] [241].

Bacillus subtilis, *Pseudomonas putida* and *Pseudomonas aeruginosa* were evaluated against *Fusarium oxysporum* f.sp. *ciceris* and reported to have better control in seed treatment and resulted in an increment in growth parameters [205]. Species of *Bacillus* have been reported to produce compounds effective against important fungal pathogens including *Rhizoctonia solani* and *Xanthomonas oryzae* pv. *oryzae* [124] [242]. Compounds from *Chaetomium globosum* have also been reported effective against important fungal pathogens of rice such as *Magnaporthe grisea* and *Puccinia recondita* [126]. Anitha and Rabeeth [243], reported reduced severity of fusarium wilt of tomato after using *Streptomyces griseus*. In a seeded media experiment, *Stenotrophomnas maltophilia*, *Bacillus subtilis* and *Pseudomonas aeruginosa* exhibited antagonism against *Erwinina carotovora* [244].

A formulation consisting of compost tea extracts and poultry litter reduced severity of bacterial wilt (*Ralstonia solanacearum)* of brinjals (*Solanum melongena*) [245]. Higher efficacy was observed when the compost tea extract was applied as a soil drench and the poultry litter applied on the soil which resulted in healthy plants and improved yield. A similar formulation reduced the incidence and severity of late blight (*Phytophtora infestans*) of potato (*Solanum tuberosum*) when the compost tea was applied as a foliar spray [246]. Research by Pane *et al.*, [247] showed that a compost tea formulation containing wood chips improved yield of lettuce (*Lactuca sativa* var. *gentilina*) and Kohlrabi or German turnip (*Brassica oleracea* var. *gongylodes*) when applied as a foliar spray.

Semiochemicals have been employed in management of insect pests. As reported by Chermiti and Abbes [222], mass trapping by use of sex pheromones with water traps has been used in management of *Tuta absoluta* by delaying initial attacks on tomato plants. Similarly, fruit flies (*Rhagoletis cingulata*) have been managed through semiochemicals such as sex pheromones, oviposition, host location and mating pheromones [248]. According to Powell and Pickett [249], these semiochemicals could be insect-plant induced or insect induced and the end result is enhanced parasitizing of the insect populations.

Predators of insect and microbial nature have been effectively used in management of insect pests. Species of *Amblyseius swirskii* have been used in the management of thrips, *Frankliniella occidentalis* and *Scirtothrips dorsalis* and spider mites (*Tetranychus urticae*) [187] [193]. *Phytoseiulus persimilis* is an effective predator mite against spider mites (*Tetranyshus* spp.) [188]. According to a study by Vá Squez *et al.*, [194], aphid predators (*Aphidius colemani*) were reported to be effective on *Aphis gossypii* on crysanthemums (*Dendranthema grandiflora)*.

9. Why Biopesticides in Sustainable Agricultural Production

Biopesticides are as effective as synthetic pesticides in management of crop pests [24]. Natural products are also eco-friendly since they are easily biodegradable and therefore do not pollute the environment [250]. Consumer tastes and preferences fluctuate over time and following the demand for organically produced food, this makes biopesticides suitable alternatives to synthetic pesticides [251]. Biopesticides have very short pre-harvest intervals and are therefore safe to use on fresh fruits and vegetables [209]. They are also target specific and hence do not affect the beneficial organisms such as the natural enemies [252]. They are effective in small quantities and their use promotes sustainable pest management and hence contribute towards sustainable agriculture [253].

Natural pesticides do not cause resistance build up among pests [145]. Availability of their source materials makes them inexpensive to attain since they are found within the natural environment and some of them are used for other purposes like food and feed [254]. Biopesticides are safe products both for the applicant and the consumer since they have no toxicity [16]. Therefore, biopesticides can suitably be incorporated in integrated pest management (IPM) which helps reduce the amounts of chemical pesticides used in management of crop pests [255]. Natural products decompose quickly which makes them safer for use in the environment [256]. Pesticides from natural sources have very short re-entry intervals which guarantee safety for the applicant [257]. Biopesticides are also used in decontamination of agricultural soils through introduction of important microbial species [258].

10. Limitations Facing Use of Biopesticides

While biopesticides provide such advantages as safe environment and healthy food for human consumption, there are factors that limit their full adoption as pest and disease management options. High doses of the constituent compounds are needed for efficacy under field conditions [252]. The concentration of the bioactive compounds in plants is dictated by the environment under which they grow [15]. The constituent active compounds are also dictated by the diversity of plants and their varieties resulting in differences in the responses to pathogens [259]. The quality of botanical extracts is also dependednt on the method of extraction used [255]. During formulation, it is sometimes challenging to get the right proportions of the active and inert ingredients needed. There are also no standard preparation methods and guidelines for efficacy testing especially under field conditions [251]. While the *in vitro* tests produce excellent results, there are always inconsistencies at the field due to low shelf life and sometimes poor quality of source materials or preparation methods.

Adoption of biopesticides of predatory nature need a lot of consideration such as host crops and dispersal capability [260]. Crop coverage and exposure time are essential and for a small acreage this could prove expensive since application may be manual [261]. Registration of the products requires data on chemistry, toxicity, packaging and formulation which is not always readily available [262]. The cost of producing a new pesticide product is usually high and has a lot of resource limitations [257]. Lack of a readily available market makes it hard to invest in biopesticides [257]. There are insufficient facilities and capital for production of biopesticides especially in the slowly developing countries. The shelf life of natural products is dependent on many factors such as temeperatures and moisture which are sometimes difficult to control [120]. Biopesticides also face high competition from synthetic pesticides and if the former were produced for a small agricultural activity, the costs may be relatively high and therefore not feasible. There is insufficient awareness about biopesticides especially among the small-scale growers, stake holders and policy makers. In the case of microbial pesticides, there is usually no trust in the value and use chain between producers, buyers and users and considering the risk of importation, synthetic pesticides appear reliable [263].

11. Conclusion and Future Prospects

Despite the many challenges facing the adoption of biopesticides, they still remain suitable alternatives to conventional pesticides. Use of synthetic chemicals has raised numerous concerns due to their negative effects on the environmental, human health, natural enemies and ecosystem balance. Some of the active ingredients of synthetic pesticides have been found to be carcinogenic thus posing a threat to human life. Biopesticides offer better alternative to synthetic pesticides due to their low toxicity, biodegradability and low persistence in the environment. The base materials for biopesticides are readily available and inexpensive. Data on toxicity levels, chemistry, active compounds and their compatibility with other methods of pests and disease management is needed to aid in formulaton and commercialization. Globally, researchers have conducted studies on effectiveness of natural plant protection products with significant results being from *in vitro* experiments. There are also studies on effectiveness of biopesticides under controlled environments and field conditions with varying results. Further research is recommended to close the gaps in formulation of biopesticides. Stable products under field conditions will be a guarantee of utter effectiveness of biopesticides in crop pest management. Researchers should therefore work together with engineers in the government and industry as well as farmers to provide stable, durable formulations of biopesticides.

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