

7. Biological and Genetic Pollution

Deepali Rajwade

Assistant professor,
Department of Biotechnology,
Govt. N. P. G. College of Science,
Raipur.

Preeti Mehta, Sadhana Jaiswal

Assistant professor,
Department of Microbiology,
Govt. N. P. G. College of Science,
Raipur.

Abstract:

Non-indigenous species are the ones which reach to the regions outside their natural habitats & their normal range of dispersal. It can be any part of the species or its gametes which may propagate in the new environment. It also includes hybrid between an exotic and an indigenous species. This transition may be result of natural phenomenon, climate changes or it can be due to human intervention. Invasive alien species is a subclass of non-indigenous species which have the potential to spread to new regions and adversely affect the biological diversity and equilibrium of that ecosystem. Biological pollution can be defined as the degradation of ecological balance due to change in biological organization resulting from invasion of alien species. The technique of genetic engineering refers to the modification of an organisms DNA for a purpose. The transfer of altered genes from a genetically engineered organism to the wild population is termed as genetic pollution. Although this gene flow may at times be beneficial for the native species but it is termed as pollution when it negatively impacts the natural population. As the introduction of new varieties by conventional & recombinant DNA techniques is beneficial for the fulfilment of societal needs, it cannot be totally curbed. Scientists are working on this challenge for development of new technologies that will minimize the risk of gene mixing between non-indigenous species or genetically engineered species and the wild type. By the joint effort of political & scientific community, policies can be formulated to check the harmful impacts of this gene mixing.

7.1 Introduction:

Natural resources, gene pools, and genetic variety are all declining alarmingly as a result of anthropogenic activity and non-sustainable use. Many species' natural populations have declined dramatically as a result of overexploitation, habitat changes such as physiographic, abiotic, and biotic aspects, and the introduction of exotic species. Individuals' short-term fitness and the population's long-term survival are both aided by genetic variety, which allows for adaptation to changing environmental conditions.

Pollution is the process of rendering land, water, air, or other components of the environment unclean, unsafe, or unsuitable for use. It is the introduction of substances or energy into the environment by man, either directly or indirectly, with such negative consequences as harm to living resources, hazards to human health, impediment to natural activities, impairment of land and water quality, and diminution of amenities (Joint Group of Experts on the Scientific Aspects of Marine Environmental Protection -GESAMP). When purposely brought into an area, even simple things like light, sound, and temperature can be deemed pollutants. It reduces the fitness of individuals, populations, species, and communities to survive.

7.2 Biological Pollution:

The impact of humanity's actions on the quality of the aquatic and terrestrial environment is referred to as biological pollution which can be specifically defined as the introduction of non-native and invasive organisms (Elliott, 2003). It includes the impact of imported, invasive species on an individual (internal biological pollution by parasites or viruses), a population (by genetic alteration), or a community (by expanding or lowering the species complement), as well as the generation of negative economic implications.

According to Elliot, the adverse effects of bio pollution can be observed at various levels:

- a particular organism (internal pollution by parasites or diseases),
- a population or community (through genetic alteration, such as hybridization of IAS with a native species, domination of IAS, replacement or eradication of native species)
- a habitat (by altering physical and chemical factors)
- An ecosystem (by alteration of energy and organic material flow).
- Ecological – by alteration of energy and organic material flow

7.2.1 Biological Pollution by Pathogens & Parasites-

This includes the compounds in our environment that originate from living beings and have the potential to harm our health. Pollen from trees and plants, insects or insect parts, some fungi, germs and viruses, and even animal hair, skin scales, saliva, and urine are all examples of bio pollutants.

7.2.2 Sources:

- Pollens from plants
- Microorganisms like bacteria, viruses & molds transmitted by people, animals, soil, and plant waste
- Saliva and animal dander (skin flakes) from home pets
- Cockroaches, rodents, and other pests or insects
- Droppings and body parts from cockroaches, rodents, and other pests or insects
- The protein found in rat and mouse urine is a powerful allergen. It has the potential to become airborne when it dries.

Biological pollutants are the living creatures or are created by them. They are frequently found in regions where food, moisture, or water are provided like mold can grow in damp or wet spaces like cooling coils, humidifiers, condensate pans, or unvented restrooms. They also tend to accumulate in drapes, bedding, carpets, and other areas where dust settles.

7.2.3 Health Effects from Biological Contaminants:

Biological contaminants may trigger allergic reactions like hypersensitivity pneumonitis, allergic rhinitis & some types of asthma. Infectious diseases like influenza, measles, and chicken pox are spread via the air borne pathogens. Molds and mildews produce poisonous substances that cause diseases. Disease-causing biological agents in the indoor air are especially dangerous to children, the elderly, and persons with breathing issues, allergies, or lung disorders.

- Biological contaminants can cause a variety of health concerns, including:
- sneezing
- watery eyes
- coughing
- shortness of breath
- dizziness
- lethargy
- fever
- and digestive problems

7.2.4 Remedial measures

Biological contaminants can accumulate in environment causing detrimental ecological consequences. Indoor allergens can be tackled with cleaning and following hygienic practices. Various physical, chemical & biological approaches exploiting new technologies can be utilized for the detection, and removal or reduction of contaminant. For example contaminants in water bodies can be identified and quantified *in situ* using microbial whole-cell biosensors, negating the need for water samples to be tested off-site. Similarly, by manipulation of the composition and function of the indigenous microbial communities present in contaminated environments, we might improve the innate bio degradative processes. Likewise bacteriophages can be used to target and reduce the quantity of biological pollutants such as harmful & pathogenic bacteria.

7.3 Biological pollution by invasive species:

The introduction of alien species (AS) into ecosystems is a source of disruption that can also be considered a pollutant. The phrase "biological pollution" has recently been used to describe the issues produced by alien species (AS) (Boudouresque, 2002). An alien species also referred to as non-native, non-indigenous, exotic or introduced, is a species that has been brought by humans outside of its natural range and dispersal capabilities, either purposefully or unintentionally (IUCN, 1999; Occhipinti-Ambrogi, 2004; Colautti, 2004). Natural changes in distribution range (for example, due to climatic change or ocean current dispersal) do not define a species as an alien. If an alien species "population has reached an

exponential growth stage and is rapidly expanding its range"(Occhipinti-Ambrogi, 2004) or its" introduction causes or is expected to cause economic or environmental impact or harm to human health (IUCN, 1999), it is deemed invasive. Alien species cause structural and functional diversity at multiple levels of biological organisation, including genetic, population, community, and habitat/ecosystem (Reise, 2006). An effect of alien species can be seen as a decline in ecological quality caused by the changes in biological, chemical, and physical aspects of the ecosystems. Elimination or extinction of sensitive or rare species; alteration of native communities; algal blooms; modification of substrate conditions and shore zones; alteration of oxygen and nutrient content, pH, and transparency of water; accumulation of synthetic pollutants, and so on are just a few examples of these changes. As a result, the definition of bio pollution could be expanded to include effects on the invaded ecosystems' structural components (both biotic and abiotic) and functioning. Many invasive species are unintentionally introduced into new areas. Zebra mussels are native to Central Asia's Black Sea and Caspian Sea. Zebra mussels landed in North America's Great Lakes by chance, clinging to big ships travelling between the two continents. Zebra mussels have become so abundant in the Great Lakes that they have become a threat to native species. Some species are purposefully introduced to a new environment. These species are frequently introduced as a pest control measure. Introduced species are sometimes brought in as pets or as aesthetic displays. Importers of these species have no idea what will happen if they do so. Even scientists can't predict how a species will adapt to a new environment.

Invasive species multiply too quickly after being introduced. Five cats, for example, were sent to Marion Island, a portion of South Africa in the southern Indian Ocean, in 1949. Mice were given to the cats as a pest control measure. By 1977, the island had roughly 3,400 cats, putting the native bird population in jeopardy. Many invasive species thrive because they compete for food with native species. Bighead and silver carp are two huge fish species that escaped from fish farms in the 1990s and are now widespread in North America's Missouri River. These fish eat plankton, which are microscopic organisms that float in the water. Plankton is also consumed by several native fish species, such as paddlefish. The paddlefish's feeding cycle is slower than that of the carp. In the lower Missouri River, there are now so many carp that paddlefish are starving.

Losses in US \$			Species in the US & India				
Introduced pests	US	India	Category	United States		India	
				Total	Alien	Total	Alien
Weeds	27.9	37.8	Plants	42000	25000	45000	18000
Crops	6.0	0.92	Mammals	346	20	316	30
Pastures			Birds	650	97	1221	4
Vertebrates			Reptiles & Amphibians	247	53	741	NA
Crops	1.0	---	Fishes (freshwater)	938	138	2546	300
Arthropods	15.9	16.8	Arthropods	650000	4500	54430	1100
Crops	2.1		Microbes	134644	20000	NA	NA
Forests							
Plant pathogens							
Crops	23.5	35.5					
Forests	2.1	---					
Total	78.5	91.02					

Figure 7.1: Economic Disasters Due to Invasive

<https://www.downtoearth.org.in/coverage/invasive-alien-species-10861>

7.3.1 Impact on Habitats:

Many invasive species devastate habitat, the natural homes of other plants and animals. Large rodents native to South America known as Nutria, were transported in the early 1900s to North America by ranchers in the hopes of raising them for their fur. When the ranchers failed, some nutria were released into the wild. They are becoming a major problem in the United States' Gulf Coast and Chesapeake Bay regions. Tall grasses and rushes are Nutria's favorite foods. These plants are essential to the marshy wetlands of the region. Many organisms rely on them for food, nesting grounds, and shelter. They also aid in the stabilization of silt and soil, avoiding land erosion. By devouring wetland grasses, Nutria devastates the area's food web and habitat.

7.3.2 Impact on Economy:

Some invasive species have a significant economic impact. Water hyacinth is a South American plant that has spread throughout the world as an invasive species. Because of its lovely blossoms, people frequently introduce the plant that grows in water. However, the plant spreads swiftly, suffocating natural fauna. Water hyacinth grew so heavily on Lake Victoria, Uganda, that boats couldn't get through it. Some ports were shut down. The water hyacinth blocked sunlight from reaching the seafloor. Plants and algae were unable to develop, restricting the feeding and reproduction of fish. The fishing industry on Lake Victoria has deteriorated. Property can also be harmed by invasive species. Small zebra mussels jam boat engine cooling systems, while larger ones have damaged water pipes at power stations around the Great Lakes region.

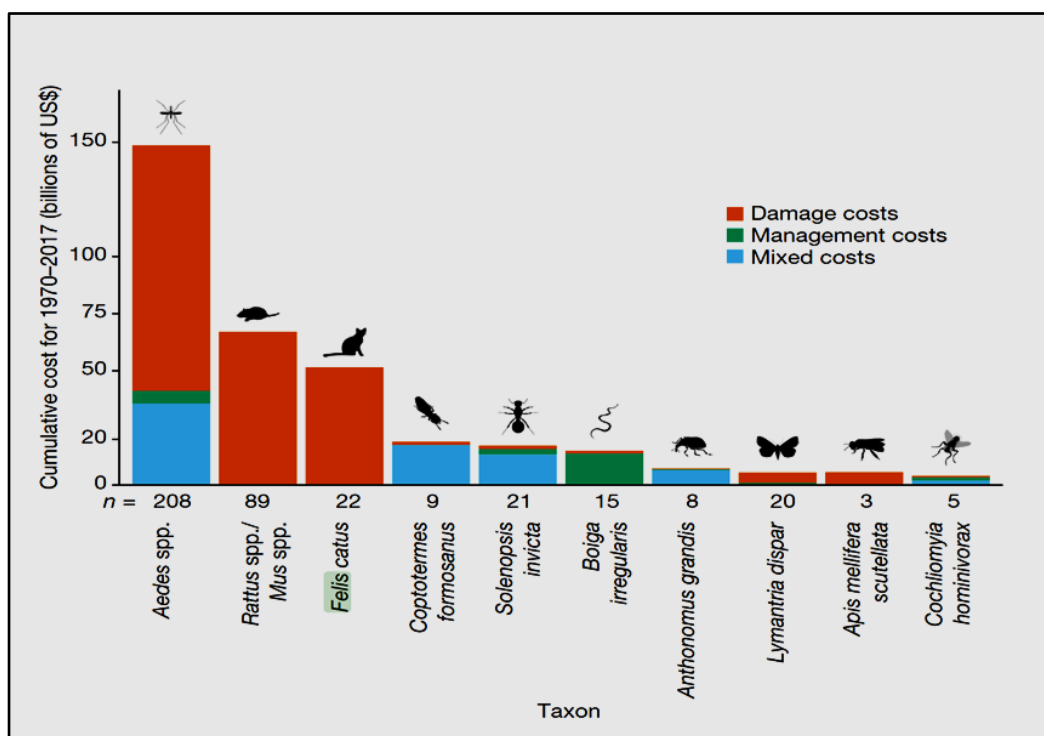


Figure 7.2 between 1970 and 2017, the ten most expensive taxes in terms of both cumulative damage and administration costs (in billions of US dollars) a species or a group of species is represented by each bar. The number of cost estimates is indicated by the numbers beneath the bars. This rating highlights the limitations of current data and the need for more comprehensive and consistent expense disclosures. An open source platform (<http://phylopic.org/>) was used to create all of the animal silhouettes. Melissa Broussard designed the Coptotermes silhouette. (Diagne, 2021).

7.3.3 Impact on Ecosystem Functioning:

Invasive alien species (IAS) can have devastating ecological consequences in invaded areas. In their new settings, they may have fewer natural predators, allowing them to quickly increase their population and spread. They can spread illnesses, outcompete or prey on local species, disrupt food chains, and even modify ecosystems by changing soil composition or creating environments that support wildfires, for example. These effects may result in the extinction of native species on a local or global scale, as well as ecological destruction may result in trophic net shifts and changes in energy and organic material flow. Some incursions can have a variety of implications on ecosystem functioning (material flow between trophic groups, primary production, and the relative degree of organic material degradation). IAS can also have significant socioeconomic consequences. As a result of IAS effects on human health, infrastructural damage, and agricultural losses, the European Union (EU) suffers annual damages totaling EUR 12 billion. There are around 12,000 foreign species in Europe, with 15% of them being invasive. IAS are the third most serious threat to endangered species in Europe. According to a 2015 assessment, IAS has a direct impact on 354 vulnerable species (229 animals, 124 plants, and 1 fungus), accounting for 19% of all threatened species in Europe. The newly adopted EU Biodiversity Strategy emphasizes the necessity of addressing this danger, suggesting that by 2030, "established invasive alien species be managed and the number of Red List species they threaten be reduced by 50%."

The European Commission (EC) proposed legislation in the form of an EU Regulation on IAS in 2013, with the goal of preventing their entry, providing early warning/rapid response, and ensuring effective and coordinated management. Since 2016, IUCN has provided technical and scientific support for the implementation of the EU IAS Regulation through a series of service contracts with the European Commission and collaboration with the IUCN Invasive Species Specialist Group (ISSG). The Invasive Species Specialist Group of the IUCN's Species Survival Commission is a global network of science and policy professionals. By raising awareness of IAS and looking into strategies to prevent, contain, or eradicate them, this Specialist Group hopes to lessen threats to natural systems and native species.

7.3.4 Solutions:

The majority of experts feel that the most efficient strategy to prevent future exotic species invasions and contribute to biodiversity protection is to prevent new species introductions in the first place. Although "exotic stowaways" continue to exist as a result of international trade and travel, ecologists point out that governments and citizens have the authority to limit the risk of such creatures being released into new settings. At ports of departure and

arrival, closer screening of pallets, containers, and other international shipping goods could reveal insects, seeds, and other stowaway species. To prevent purchasers, sellers, and carriers of illegal exotic pets, several ecologists and government officials have urged for harsher fines and the possibility of incarceration.

Increased port controls, on the other hand, will not help invasive species that have already established themselves. Furthermore, climate change may provide new chances for some invasive species. Photosynthesis (and thus growth and reproductive success) in some plants has been demonstrated to be fueled by persistent increase in atmospheric carbon dioxide concentrations. Climate warming associated with increases in atmospheric carbon will likely allow botanical invaders, such as kudzu and another ornamental plant from Asia called Oriental bittersweet (*Celastrus orbiculatus*), and insect invaders, as well as the diseases they may carry, to gain footholds in habitats previously off-limits to them. Some ecologists have suggested that strong monitoring and eradication initiatives be implemented to avoid such scenarios from occurring. Many habitat restoration programs, for example, priorities replanting and reinstalling native species while also eradicating invasive species. Ecologists think that by combining these activities with effective education programs that equip individuals with the knowledge and resources to deal with foreign plants, animals, and other species in their communities, invasive species will be able to prevent further biodiversity loss.

7.4 Biocontrol:

Other species are sometimes introduced to assist in the control of invasive species. The prickly pear cactus, which is endemic to the Americas, was out of control in Australia. Rangeland, where ranchers raised livestock, was being destroyed by the cactus. To consume the cactuses, the government imported cactus moth caterpillars. Cactus caterpillars are natural cactus predators.

Biological control is promoted as a safer alternative. However, under actual field situations, it has not been very successful. The principle behind biological management is simple: find a pest or weed predator in its natural habitat and introduce it to the invaded area. However, several stringent requirements must be met. For example, the predator must be host-specific and not predate on other plants or animals. The Australian Vedalia lady beetle was introduced to California, USA, in 1889 to suppress the citrus orchid-threatening cottony-cushion scale insect. The pest was fully eliminated by the beetle. The introduction of *Dactylopius opuntiae* to manage the prickly pear has also been a huge success in India. However, determining host-specificity is problematic. There are numerous examples of biocontrol agents turning into pests. The most notable is the release of the predatory snail *Euglandina rosea* in the Hawaiian Islands to control the foreign African giant snail. *E. rosea* ate up the majority of Hawaii's 41 native snail species, but the African giant snail still crawls through the island's forest floors. The Project Directorate of Biological Control (PDBC), which is situated in Bangalore, is a nodal body in India. A scientist who wants to import a biological controlling agent must first provide all relevant information about the organism, such as if it is unique to a specific weed and whether it can be replicated in laboratory settings. Biological control agents must undergo extensive testing. The organism should have all of the qualities that biological regulating agents should have. Their handling and discharge should be risk-free as well.

7.4.1 Ecological History of Parthenium:

Parthenium is claimed to have arrived with a shipment of wheat from Mexico. In addition, parthenium seeds are too tiny to be intercepted. Pune was the source of the first parthenium report. Mechanical and chemical technologies initially failed to eradicate the weed. It is dangerous to persons who have asthma. Pollenises are caused by its pollen. There have also been reports of neurological problems. It causes dermatitis in rodents such as rabbits in woodland environments. The fact that zycogramma bicolorata only consumes parthenium weed is a plus as a biocontrol agent against Parthenium. It attacks the plant's leaf, which the plant can't survive without. There have also been instances of the insect consuming sunflowers. However, a government fact-finding commission discovered that the zycogramma bicolorata could not develop on sunflowers after an investigation.



Figure 7.3: A Roadside Clearance Overtaken by Profusely-Growing Parthenium Plants. Photo by Aathira Perinchery



Figure 7.4: Zygogramma Bicolorata (Parthenium beetle) Source- Wikipedia

7.5 Genetic pollution:

7.5.1 Genetic modification:

The fundamental goal of genetic modification is to locate genes in an organism that can be tweaked and then passed on to a target organism to produce the desired characteristic bearing species. It might be viewed as a challenge to nature's core realities, with negative consequences on a microscopic level that add up to genetically generated pollution. Plants and other species can now be genetically modified thanks to genetic engineering (Barta, 2006; Beyer, 2002; Bryan, 2004). New genes from any other species can be injected into the subject species to give it desirable features. Transgenes are the genes that have been inserted. In a few circumstances, the subject specie's gene is removed in order to compare the organism's nature and properties. To examine changes in behaviour and attributes, several genetic alterations are made. Organizations like Greepeace⁶ and TRAFFIC⁷ have labelled the changes brought about by genetic manipulation as "undesirable."

7.5.2 Health and Safety Concern from Genetically Modified Plants:

Plants contain various metabolites. Genetic engineering may cause unanticipated changes in metabolism, which could modify such compounds or develop new toxicants. This is especially concerning for pest-resistant and herbicide-tolerant crops.

7.5.3 Toxicity Potential:

Arpad Pusztai, a scientist at the Rowett Institute in Aberdeen, Scotland, revealed unpublished data of experimental rats given GM potato designed to carry the *Galanthus nivalis* agglutinin (GNA) lectin gene for insecticidal capabilities in a televised interview in 1998. The rats appeared to have stunted growth, abnormalities in the gut lining, and immune system impairment. Because rats fed unmodified potatoes with GNA had identical effects, the effect was attributed to the GNA. However, it was claimed that increases in jejenum crypt length and decreases in cecal mucosa thickness were caused by the potato's genetic change, not by the presence of GNA (Ewen and Pusztai, 1999). Following that, it was proposed that the viral origin of the Ca MV promoter utilized to drive gene expression could be to blame for the modifications. Because the information was not immediately published in peer-reviewed journals, there was some uncertainty about his assertions at first. Pusztai was fired as a result of the publicity, and his unpublished work at Rowett Institute was evaluated and judged to be defective by the Royal Society of the United Kingdom. In 1999, the work was ultimately published in *The Lancet* as a letter. However, coming so soon after Europe's "mad-cow" BSE catastrophe, the issue did nothing to alleviate public fears about food safety and the legitimacy of scientific advice.

7.5.4 Allergenicity:

The possible allergenicity of novel proteins generated by the transgene put into the plant is a serious food safety problem, particularly in insect-resistant crops. The Star Link GM maize variety, for example, was shown to have allergenic qualities in the United States, the European Union, and Japan, and its usage was restricted to feed by regulatory agencies.

7.5.5 Contamination of Food:

Crop plants including maize, canola, and soybean are being genetically modified to create medications, vaccines, and industrial materials like plastics, lubricants, and nonfood polymers. There is a risk of contamination of food and feed with these products in the absence of suitable labelling and segregation systems. As evidenced by the instance of Agro Evo's Star Link corn, ensuring regulation of these crops is an issue (Aventis). Because it had been modified with a gene that encodes the BT protein Cry9c, the variety was resistant to corn borer. The US Environmental Protection Agency had approved the variety's production in 1998, but had limited its usage to animal feed due to allergic concerns. In 2000, however, residues of Star Link corn were discovered in Kraft Foods taco shells, prompting in food recalls and major disruption of the food supply (Pollack, 2000).

7.5.5 Nutritional Composition:

Plants that have been genetically modified may experience changes in metabolism as a result of the introduction of new genes. This, in turn, may have an impact on the nutritional value of food. For example, GM rice has been found to accumulate xanthophylls and prolamines, which could lead to nutritional imbalances in consumers.

7.5.6 Antibiotic Resistance:

To make transgenic plants, antibiotic genes are used as selectable markers. This raises the question of whether these genes could be conveyed to gut microorganisms via GM foods, resulting in antibiotic-resistant diseases.

7.6 Environmental Concerns:

By wandering pollen grains, genes from the GM crop can be transferred to wild and weedy relatives. This could result in things like

7.6.1 Genetic pollution:

Because a large fraction of GM types in numerous crops have genes providing herbicide tolerance, "super weeds" could emerge. Percy Schmeiser, a Canadian farmer whose fields were polluted with "Round-up Ready" canola by pollen from a nearby GMO farm, is a well-known example of genetic pollution. The event gained media attention after Monsanto won a lawsuit alleging Schmeiser had infringed on their intellectual rights.

7.6.2 Loss of Biodiversity:

When GM cultivars are introduced to new settings, new genes may be transferred to traditional varieties and wild relatives of the crop. It may potentially have an ecological impact by impacting non-target creatures. The findings of Cornell University scientists published in the journal *Nature* in 1999 showed that monarch butterfly larvae fed on milkweed leaves covered with pollen from BT maize did not grow as well as those fed on

control milkweed leaves (Losey et al., 1999). Following the public outrage, a large research cooperation between six groups in the United States and Canada was formed to investigate this. Their findings, which were reported in PNAS (USA) in 2001, showed that pollen toxicity was dependent on the Bt gene used: expression of Bt in pollen from event Bt176 was higher than in pollen from other events carrying the cry1Ab gene (Sears et al., 2001). Because Bt176 is no longer available in maize varieties grown in the United States, the overall risk to Monarch butterfly populations is thought to be negligible.

7.6.3 Emergence of Resistance:

Toxin genes from the *Bacillus thuringiensis* (BT) bacteria have been inserted into numerous crops to make them insect-resistant. Insects could acquire tolerance to the poison if proper steps are not taken, rendering the entire plant-incorporated pesticide technique useless.

7.7 Transgene Escape & Genetic Pollution:

Vertical gene flow, or transgene escape, is a reality that is usually limited to within species or closely related species. Horizontal gene flow, or gene movement between species, is extremely rare. Diagonal gene flow, on the other hand, occurs between closely related species (Gressel, 2015). In order to avoid transgene escape, genetically modified (GE) crops have been cultivated for commercial and research reasons with specific constraints. However, we have failed to manage gene flow in a systematic manner after 22 years of GE crops (Ryffel, 2014). Cotton, maize, soybean, oilseed rape, rice, and wheat have all been reported to exhibit convincing evidence of transgene escape (Baltazar et al., 2015; Dong et al., 2016; Londo et al., 2011; Mizuguti et al., 2010; Ramzan et al., 2014; Serrat et al., 2013). Transgene flow may occur not just around the GM field, but also further away. These conclusions are not limited to a single region of the world; rather, the instances demonstrate the problem's global scope. Hybridization of GM plants with their traditional parents has been reported, as has the occurrence of seed. Cotton is an interesting example, where gene flow has been documented both vertically and diagonally in various investigations.

The infection of unaltered or natural organisms with changed genes from genetically hybridised organisms is known as genetic pollution. Genetic pollution, according to environmentalists¹ and various organisations, is a bad thing. It was previously known as gene flow from domestic, non-native sub species to wild native populations, but it is now known as gene flow from genetically engineered species to non-GE species. This transformation is referred to by a variety of labels, including genetic degradation, genetic aggression, and genetic mixing. However, no one has come to a consensus on any of them. As a result, the meaning of genetic pollution is still up for debate.

The first genetically modified plant was an antibiotic tobacco plant which was introduced as the first GMO in 1983. Gene flow from genetically modified organisms to non-genetically modified organisms can occur unintentionally. Cross pollination, water pollination, and animal pollination are all possible ways for genes to travel. Animals, water, and cross-pollination are all possible ways for genetically modified organism seeds to reach non-modified species. Genetic pollution can also occur when wild and modified organisms mate, resulting in hybrids.

By interacting with the DNA of other creatures, this unintended mixing could result in genetic pollution. Although it is not as widespread as genetic pollution in plants, the process of gene flow in animals is similar to that of plants. The transfer of genes from one animal to another can result in genetic pollution by changing the animal's characteristics. Animal genetic pollution is a major problem since it can have a negative impact on a particular breed of organism and perhaps lead to extinction.

Table 7.1: Examples of Natural Transgene Escape in Various Crops (Rizwan et al.)

From	To	Transgene escaped	Trait	Type of flow	Medium of escape	Region	Reference
Cotton <i>Gossypium hirsutum</i>	Non-Bt. cotton	MON-531, Cry1Ac, Cry2A	Insect resistance	Vertical	Pollen and seed	Pakistan	Ramzan et al. (2014)
	Non-Bt. cotton	Cry1Ac and CP4 EPSPS	Insect and herbicide resistance	Vertical	Pollen	Beijing China	Yan et al. (2015)
Maize <i>Zeamays</i>	Non-GM maize	MON-810	Insect resistance	Vertical	Pollen	Slovakia, Spain	Mihalčik et al. (2012),
	Non-GM maize	MON-89Ø34-3, MON-88Ø17-3, MON-ØØ6Ø3-6	Insect resistance and herbicide tolerance	Vertical	Pollen	Mexico	Baltazar et al. (2015)
Soybean <i>Glycine max</i>	Conventional soybean	EPSPS	Herbicide resistance	Vertical	Outcrossing by honeybees	Brazil	Chiari et al. (2011)
	Glycine soja	EPSPS	Glyphosate tolerance	Diagonal	Pollen	Japan	Mizuguti et al. (2010)
	Dwarf male sterile line	Nib8	Wheat yellow mosaic virus resistance	Vertical	Pollination by wind	China	Dong et al. (2016)
	Non GM wheat	Bar and gfp	Herbicide resistance	Vertical	Pollen	Russia	Miroshnichenko et al. (2016)
Rice <i>Oryza sativa</i>	Weedy rice	ALS	Imidazolinone resistance	Vertical	Pollen	USA	Valverde (2013)
	O. sativa F. spontanea	Bar	Herbicide resistance	Diagonal	Cross pollination	Spain	Serrat et al. (2013)

7.7.1 Transgene Containment and Mitigation Strategies:

When dealing with trans-gene flow, it's important to keep the situation of transgenic crops in mind. While combating gene flow, studies on risk assessment during gene escape could be valuable. In general, there are two options: keeping the gene in its original GMO or mitigating the consequences (Gressel and Al-Ahmad, 2006).

7.7.2 Physical Containments:

Because most gene flow occurs through pollen or seed, one strategy to keep transgenes in check is to prevent seeds and pollen from dispersing (Linder et al., 1998).

This dissemination can be avoided by isolating the GM crop and using various physical barriers, as well as careful seed preparation.

Positive features- It's easy and simple to use for all crops & economical.

Challenges- It's unable to contain transgene completely & flow of gene through seed based products is almost impossible to stop (Arriola, 1997; Linder et al, 1998)

7.7.3 Biological/Molecular Containments:

- **Sterility:**

Positive features – It gives good results using complete sterility.

Challenges-It's not feasible in all crops as vegetative propagation is necessary for complete sterility.

Also it may lead to monopoly of seed companies as the farmer will not be able to produce own seeds (Daniell, 2002; Schernthaner et al., 2003)

- **Cleistogamy:**

Positive features- Its a mode of biological control without causing any threat to gene pool.

Challenges – It may lead to inbreeding depression. It's not applicable to all crops & some leakage has been observed (Husken et al., 2010; Gealy, 2005)

- **Apomixes:**

Positive features-It's good for fixing heterosis.

Challenges- It's a difficult process to attain & can cause dispersal through pollen if not complemented by sterility (Bicknell and Kultunow, 2004; Bhat et al., 2005)

- **Maternal transformation:**

Positive features-It can effectively hinder the dispersal through pollen & can be a good option if complemented with female sterility.

Challenges –It's not applicable in all crops due to biparental inheritance. Also backcrossing of hybrid with GM crop can disperse the trait (Maliga ,2004; Haider et al., 2009)

- **Incompatible genome:**

Positive features-No extra labor is required in this.

Challenges-It can only be possible in crops having multiple genomes. Compatibility with homologous genomes have been reported (Lu, 2003).

- **Gene splitting:**

Positive aspects features-It can be effective if complimented with other techniques

Challenges- It alone can lead up to 25% gene flow in segregating generation (Dong et al., 2015; Wang et al., 2014).

- **Expression in virus:**

Positive features-It Alone can manage transgene flow effectively.

Challenges- Transgene will be good only for single generation (Kelloniemi et al., 2008).

- **GURTs (Genetic use restriction technology)**

Positive feature-It's an effective technique.

Challenges-It's not been evaluated yet. Issues regarding monopoly of seed companies may occur (Swanson and Goschl, 2000).

- **Transgenic mitigation:**

Positive feature -It disables transgene irrespective of flow & delivers good results.

Challenges- If the transgene is not deleted, expression can be restored at any time. Different blocking genes utilized can be a novel hazard to biosafety. A very little amount of transgene flow is still present as this can slow down the process but not totally stop it.

7.8 Conclusion:

The biology of invasion should be a priority research area to study the effect on communities over a period of time. There is a challenge to segregate natural invaders from the man made ones. A clear assessment of the effect of anthropogenic activities at large and small scale will help in devising strategies to combat invasion. In terms of genetic pollution, transgene escape from GM crop plants into non-GM and wild cousins could represent a threat to the ecosystem. Understanding how transgenes escape will make it easier to grow GM crops in a sustainable and safe manner. Furthermore, varied production processes in the agriculture sector, as well as farmers' choices to plant GM, conventional, or organic crops, require a perception of food safety and identity maintenance.

As a result, in order to promote GM technology, bio safety precautions as well as prospective strategies to contain or attenuate the transgene effect should be considered.

7.9 References:

1. Arriola, P. E. (1997). Risks of escape and spread of engineered genes from transgenic crops to wild relatives. *Ag Biotech News and Information (United Kingdom)*.
2. Baltazar, B. M., Castro Espinoza, L., Espinoza Banda, A., de la Fuente Martínez, J. M., Garzón Tiznado, J. A., González García, J., & Zavala García, F. (2015). Pollen-mediated gene flow in maize: implications for isolation requirements and coexistence in Mexico, the center of origin of maize. *PLoS One*, 10(7), e0131549.
3. Barta, A., Sommer Gruber, K., Thompson, D., Hartmuth, K., Matzke, M. A., & Matzke, A. J. (1986). The expression of a nopaline synthase—human growth hormone chimaeric gene in transformed tobacco and sunflower callus tissue. *Plant molecular biology*, 6(5), 347-357.
4. Beyer, P., Al-Babili, S., Ye, X., Lucca, P., Schaub, P., Welsch, R., & Potrykus, I. (2002). Golden rice: introducing the β -carotene biosynthesis pathway into rice endosperm by genetic engineering to defeat vitamin A deficiency. *The Journal of nutrition*, 132(3), 506S-510S.
5. Bicknell, R. A., & Koltunow, A. M. (2004). Understanding apomixis: recent advances and remaining conundrums. *The Plant Cell*, 16(suppl_1), S228-S245.
6. Boudouresque, C. F., & Verlaque, M. (2002). Biological pollution in the Mediterranean Sea: invasive versus introduced macrophytes. *Marine pollution bulletin*, 44(1), 32-38.
7. Chiari, W. C., Ruvolo-Takasusuki, M. C. C., Chambó, E. D., Arias, C. A., Hoffmann-Campo, C. B., & Toledo, V. A. A. (2011). Gene flow between conventional and transgenic soybean pollinated by honeybees. *Herbicides: mechanisms and mode of action, Rijeka: In Tech, Croatia*, 137-152.
8. Colautti, R. I., & MacIsaac, H. J. (2004). A neutral terminology to define ‘invasive’ species. *Diversity and distributions*, 10(2), 135-141.
9. Daniell, H. (2002). Molecular strategies for gene containment in transgenic crops. *Nature biotechnology*, 20(6), 581-586.
10. Diagne, C., Leroy, B., Vaissière, A. C., Gozlan, R. E., Roiz, D., Jarić, I., & Courchamp, F. (2021). High and rising economic costs of biological invasions worldwide. *Nature*, 592(7855), 571-576.
11. Dong, Y., Wang, X., Tang, Q., & Wang, Z. (2015). Theoretical basis of gene splitting technique and its application in the control of transgene flow. *Agricultural Biotechnology*, 4(3), 1.
12. Dong, S., Liu, Y., Yu, C., Zhang, Z., Chen, M., & Wang, C. (2016). Investigating pollen and gene flow of WYMV-resistant transgenic wheat N12-1 using a dwarf male-sterile line as the pollen receptor. *PLoS One*, 11(3), e0151373.
13. Elliott, M. (2003). Biological pollutants and biological pollution—an increasing cause for concern. *Marine Pollution Bulletin*, 46(3), 275-280.
14. Ewen, S. W., & Pusztai, A. (1999). Effect of diets containing genetically modified potatoes expressing *Galanthus nivalis* lectin on rat small intestine. *The Lancet*, 354(9187), 1353-1354.
15. Gealy, D. R. (2005). Gene movement between rice (*Oryza sativa*) and weedy rice (*Oryza sativa*)-a US temperate rice perspective. *Crop ferality and volunteerism*, 323-354.

16. Gressel, J., & Al-Ahmad, H. (2006). Mitigating transgene flow from crops. *ASB News Report*.
17. Gressel, J. (2015). Dealing with transgene flow of crop protection traits from crops to their relatives. *Pest management science*, 71(5), 658-667.
18. Haider, N., Allainguillaume, J., & Wilkinson, M. J. (2009). Spontaneous capture of oilseed rape (*Brassica napus*) chloroplasts by wild B. Rapa: implications for the use of chloroplast transformation for biocontainment. *Current Genetics*, 55(2), 139-150.
19. Hüskens, A., Prescher, S., & Schiemann, J. (2010). Evaluating biological containment strategies for pollen-mediated gene flow. *Environmental Biosafety Research*, 9(2), 67-73.35.
IUCN, 1999. IUCN guidelines for the prevention of biodiversity loss due to biological invasions. Newsletter of the Species Survival Commission IUCN – The World Conservation Union 31, pp. 28–42.
20. Kelloniemi, J., Mäkinen, K., & Valkonen, J. P. (2008). Three heterologous proteins simultaneously expressed from a chimeric potyvirus: infectivity, stability and the correlation of genome and virion lengths. *Virus research*, 135(2), 282-291.
21. Leppäkoski, E., & Olenin, S. (1999, January). Xenodiversity of the European brackish water seas: the North American contribution. In *Marine Bioinvasions. Proceedings of the First National Conference* (pp. 107-119).
22. Linder, C. R., Taha, I., Rieseberg, L. H., Seiler, G. J., & Snow, A. A. (1998). Long-term introgression of crop genes into wild sunflower populations. *Theoretical and Applied Genetics*, 96(3), 339-347.
23. Losey, J. E., Rayor, L. S., & Carter, M. E. (1999). Transgenic pollen harms monarch larvae. *Nature*, 399(6733), 214-214.
24. Lu, B. R. (2003). Transgene containment by molecular means-is it possible and cost effective?. *Environmental Biosafety Research*, 2(1), 3-8.
25. Maliga, P. (2004). Plastid transformation in higher plants. *Annual review of plant biology*, 55, 289.
26. Mihalčík, P., Hrkčová, K., Singer, M., Plačková, A., & Kraic, J. (2012). Effect of MON 810 cultivation and prevention to adventitious presence in non-GM fields: A case study in Slovakia. *Plant Protection Science*, 48(Special Issue), S11-S15.
27. Mizuguti, A., Ohigashi, K., Yoshimura, Y., Kaga, A., Kuroda, Y., & Matsuo, K. (2010). Hybridization between GM soybeans (*Glycine max* (L.) Merr.) And wild soybean (*Glycine soja* Sieb. et Zucc.) under field conditions in Japan. *Environmental Biosafety Research*, 9(1), 13-23.
28. Ness, B. (2004). Encyclopedia of Genetics. Revised Edition.
29. Occhipinti-Ambrogi, A., & Galil, B. S. (2004). A uniform terminology on bioinvasions: a chimera or an operative tool?. *Marine pollution bulletin*, 49(9-10), 688-694.
30. Pollack, A. (2000). Kraft recalls taco shells with bioengineered corn.
31. Ramani, S., & Bhumannavar, B. S. (2004). Interaction of two indigenous predators of the spiralling whitefly, *Aleurodicus dispersus* Russell with the introduced parasitoid, *Encarsia guadeloupae* Viggiani in India. *Journal of Entomological Research*, 28(3), 199-203.
32. Ramzan, S., Shaheen, T., Hussain, K., Qasim, M., Asif, M., & Bukhari, S. A. (2014). Vertical flow of Bt genes in transgenic cotton (*Gossypium hirsutum* L.). *JAPS: Journal of Animal & Plant Sciences*, 24(6).
33. Reise, K., Olenin, S., & Thielges, D. W. (2006). Are aliens threatening European aquatic coastal ecosystems?. *Helgoland Marine Research*, 60(2), 77-83.

34. Rizwan, M., Hussain, M., Shimelis, H., Hameed, M. U., Atif, R. M., Azhar, M. T., & Asif, M. (2019). Gene flow from major genetically modified crops and strategies for containment and mitigation of transgene escape: A review. *Applied Ecology and Environmental Research*, 17(5), 11191-11208.
35. Ryffel, G. U. (2014). Transgene flow: Facts, speculations and possible countermeasures. *GM crops & food*, 5(4), 249-258.
36. Sears, M. K., Hellmich, R. L., Stanley-Horn, D. E., Oberhauser, K. S., Pleasants, J. M., Mattila, H. R., & Dively, G. P. (2001). Impact of Bt corn pollen on monarch butterfly populations: a risk assessment. *Proceedings of the National Academy of Sciences*, 98(21), 11937-11942.
37. Serrat, X., Esteban, R., Peñas, G., Català, M. M., Melé, E., & Messeguer, J. (2013). Direct and reverse pollen-mediated gene flow between GM rice and red rice weed. *AoB Plants*.
38. Swanson, T., & Goschl, T. (2000). Genetic use restriction technologies (GURTs): impacts on developing countries. *International Journal of Biotechnology*, 2(1-3), 56-84.
39. Valverde, B. E. (2013). 2. *Korean Journal of Weed Science*, 33(1), 11-23.
40. Wang, X. J., Jin, X., Dun, B. Q., Kong, N., Jia, S. R., Tang, Q. L., & Wang, Z. X. (2014). Gene-splitting technology: a novel approach for the containment of transgene flow in *Nicotiana tabacum*. *PLoS One*, 9(6), e99651.
41. Yan, S., Zhu, J., Zhu, W., Li, Z., Shelton, A. M., Luo, J., & Liu, X. (2015). Pollen-mediated gene flow from transgenic cotton under greenhouse conditions is dependent on different pollinators. *Scientific Reports*, 5(1), 1-9.
42. Yoshimura, Y., Matsuo, K., & Yasuda, K. (2006). Gene flow from GM glyphosate-tolerant to conventional soybeans under field conditions in Japan. *Environmental Biosafety Research*, 5(3), 169-173.
43. <https://www.downtoearth.org.in/coverage/invasive-alien-species-10861>.