



## Know the Pheromones: Basics and Its Application

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### ABSTRACTS

Pheromones are the chemicals secreted out side the environment by a living organism for sustaining biological activities. The origin, first isolation, uses in life stages of living organisms are enumerated. The classification of the so far reported pheromones based on their activities are given.

**Keywords :** Pheromones, pests, management

### INTRODUCTION

Living organisms use chemicals for communication with each other as well as with their surrounding environment. Species of both Plant Kingdom and Animal Kingdom secrete or release chemicals voluntarily and some times involuntarily which elicit a particular behavioural response. EMITTER and RECEIVER relationship during the process and both species undergo behavioural changes (Modified after Yadav *et al.*, 2004). Chemicals used for communication between the organisms are known as semio-chemicals or info-chemicals. These chemicals are also called “assembling scents” by Kettlewell (1942,1943,1946,1955) from the Greek *Pherein* ( to carry) and hormone (to excite, stimulate) by Karlson and Butenandt (1959). Further Kirschenblatt (1952) proposed the term “*telegones*”. Micklem (1959) termed it as “*pherormone*” instead of pheromone. Karlson and Luscher (1959) have given a more scientific clarification for the use of the term *pheromone*.

### SEMIOCHEMICALS

Behaviour Modifying Chemicals are called **Semiochemicals**, which can be classified into **two** Categories viz., **Allelochemicals and Pheromones**

#### **Allelochemicals (interspecific) encompasses:**

- a) Kairomones (benefit:emitter)
- b) Allomones (benefit: receiver)
- c) Synomones (benefit:both emitter & receiver)

#### **Pheromones (intraspecific- in most cases)**

Pheromones are ectohormones (secretion out side the animal/insect body) unlike hormones which are released internally by the ductless glands. A pheromone is a mixture of the chemicals released by an individual in to the environment for specific reactions. Pheromones are produced by ectodermal glands on the abdomen and associated with mandibles of hymenopterans and wings

of lepidopteran insects. The higher animals use diverse modes of communication such as visual, acoustic, tactile and olfactory during their interactions either for every day life activity or for social progeny sustainability. Among all the phyla, chemical communication system has been extensively studied in Phylum Arthropoda, and considerable progress has been made particularly in class Insecta, the largest group of the animal kingdom covering about 52% of total animal fauna recorded so far. Chemical communication is a well-known mode of information exchange in the class Insecta.

#### **Who could attempt the pheromone work first**

The presence of pheromone was demonstrated for the first time by Jacentkovski (1932) by placing virgin females in the traps located in gypsy moth populations infested woods. He was surprised to see many males in the trap.

#### **Discovery and Definition of pheromones**

Later Butenandt *et al.* (1959) first discovered and named as the sex pheromone in the silk worm (*Bombyx mori*). Unlike higher animals, the insects communicate between sexes of their own species (Karlson and Luscher, 1959; Karlson and Butenandt, 1959) or with its sub-species (David *et al.*, 1985) or very rarely, with different species of a genus or family or species of a different order (Kydonieus and Berosa, 1982). In the present day concern for minimising the use of synthetic pesticides in agrosystem and to save earth from the polluting toxic chemicals, the need of developing eco-friendly Integrated Pest Management (IPM) technologies is the foremost issue of both WHO and FAO. Use of natural products like botanicals and pheromones is few of the important components to be integrated with other IPM components like biological control agents and cultural practices in addition to develop pest and disease tolerant /resistant varieties for the agricultural crops. Use of pheromones is one of such potential options, which can be utilized to

Eggwash Con.(ee/ml)	No.of exp.	No. of eggs laid on leaves		% deterrence
		Control	Treated	
8	20	93/4126 (44.4)a	74/2643 (35.7)a	11.4/21.9
32	23	149/4571 (30.7)a	71/2159 (30.4)b	35.5/35.8
128	37	274/10,223 (37.3)a	125/3394 (27.2)b	37.3/50.2
512	3612	318/10,237 (32.2)a	27/1858	60.6/69.3 (23.8)b

minimize the pesticide load on the precious earth. Intensive work has been done on the basic aspects of the sex pheromones in various insects and mammals in India since 1980 in India. Majority of the information available on the pheromones related to the occurrence of sex pheromones and a few on the location of sex pheromone glands, etc. These basic studies have given a fillip to strengthen the research on the pheromones.

A large quantum of research has gone into devising ways of application of pheromones in the field. The pheromones being volatile substances, the main efforts of the researchers are to produce a regulated release of these compounds to enable them for longer stay and in amounts sufficient to affect the insects. Of all the pheromone release delivery system (media), rubber septa (sleeve stoppers) are the most widely used as trapping baits by researchers, since they can easily be obtained and loaded with pheromones and handled conveniently. Similarly, pheromones can be impregnated to: Cigarette filters, Polyethylene vial caps, PVC pellets, Control release formulations like: Hollow fibre, Twist-tie rope, Plastic laminated flake, Microcapsules etc.

#### Monitoring of insect-pests with pheromones

It is difficult to forecast the occurrence of the major insect pests unless it is noticed in a damaging proportion. Hence, monitoring of the insect pests using various gadgets becomes imperative. Timely and efficient monitoring should be the foundation of sound IPM programs, without which no proper pest management decision can be made. A few of the examples of application of pheromones employed for monitoring purposes are as follows (modified after Yadav *et al.*, 2004). Early detection of pest, mapping pest distribution, quarantine inspection, pest monitoring for action thresholds / decision support, mapping insecticide resistance frequency, monitoring parasites / predators, estimation of population dynamics, estimation of population densities, timing of management procedures based on the threshold and efficacy of management procedures based on the threshold

#### Mass Trapping.

Mass trapping is a direct method of managing the insects. In India many researchers have attempted for mass trapping of the target insects using pheromones. Based on the number of insects trapped, a decision can be taken to continue the recommended number of traps/unit area for mass trapping or to remove all the traps and use only for monitoring. If the anticipated insects are going to occur only below ET levels, there is no need for continuing the traps meant for mass trapping thus reducing the costs.

#### Mating disruption

With the commercial availability of insect sex pheromones for several agricultural pests in the 1990s, scientists and entrepreneur turned their attention to mating disruption as a “biorational” approach for insect control in the developed countries. Emission of pheromone is relatively low from each source such that a down wind trail is created and not lost in the background of released pheromone. Males following these trails are thought to spend their mating energies in pursuit of artificial pheromone sources.

#### Mechanism of mating disruption

Male confusion is thought to be the result of ambient pheromone concentrations sufficient to hide the trails of the calling female to the large doses from diffuse sources such as microcapsules or larger doses of pheromone at the point source dispensers such as tie-on polyethylene rope. Added to the effect, is the adaptation of antennal receptor sites and for habituation of the insect’s central nervous system. Specific receptor sites on the antennae respond to the pheromone molecules. When a receptor site is continually activated by high ambient concentrations of the pheromone, the resulting electrical signal diminishes (measured by an electroantennogram). The receptor site becomes unresponsive and the insect becomes navigationally blink. When the insect’s central nervous system is inundated with signals from the receptor sites, it becomes habituated and no longer remains capable to provide the directed behaviour. The net result of confusion is that the male is unable to orient to any pheromone source and follow the upwind trail to a mate (Flint and Doane, 1996).

Some possibilities of the mechanism of the confusion approach as put forth by Pedigo (2002) include:

1. Camouflage or covering up the natural pheromone scent of females
  2. Misdirection of males to scents from multiple point sources of synthetic pheromone, and
  3. Adaptation/habituation by desensitizing male antennal receptors through constant pheromone-like exposure
- Similarly, Bartell (1982) delineated five mechanisms by which mating disruption could be successful as below:
1. Sensory adaptation of the pheromone receptors and central nerve habituation,

2. False trail following caused by a multiplicity of the trails produced by synthetic odour source,
3. Inability of insects to discriminate odour trails from background,
4. Imbalance of sensory input caused by the release of an unnatural ratio of pheromone components,
5. Peripheral sensilla blocked by pheromone analogues

The first preliminary field test demonstrating the potential of this approach was conducted in 1967 with the cabbage looper, *Trichoplusia ni*. In this test, pheromone concentrations were shown to thwart males from being lured to female moths.

Insects communicate through six different modes for life activities, are as follows:

- i) Hormones, ii) Sound, iii) Pheromone, iv) Motion, v) Exocrine glands (responsible for secretions to the exterior of the body or into the lumen of body cavity) and vi) Enzymes

On the basis of interaction mediated, pheromones are sub-divided into ten categories which are as follows:

#### **i) Territorial Pheromones**

The territorial pheromones can be classified based on purpose and characteristics interactive evolution of behavior and ecology:

Type A: Large defended area: Courtship, mating, nesting and food gathering

Type B: Large defended area: Used for breeding but not foraging

Type C: Small defended area around nest

Type D: Mating territory for courtship activities only

Type E: Roosting or shelter positions

In dogs, these pheromones are present in the urine, which they deposit on landmarks serving to mark the perimeter of the claimed territory. Boars, cats do possess territorial pheromones.

#### **ii) Trail Pheromones**

These pheromones are common in social insects. For example, ants mark their paths with these pheromones, which are non-volatile hydrocarbons (Brian *et al.*, 1993). Certain ants lay down an initial trail of pheromones as they return to the nest with food. This trail attracts other ants and serves as a guide. As long as the food source remains, the pheromone trail will be continually renewed. The pheromone must be continually renewed because it evaporates quickly. When the supply begins to dwindle, the trailmaking ceases.

#### **iii) Alarm Pheromones**

Alarm pheromones serve to rapidly disperse a group of insects usually as a response to predation. These kinds

of pheromones are usually of short duration and the dispersed individuals usually reform aggregations. Some individuals release this pheromone to exhibit aggressive behaviour in the presence of predators. Alarm pheromones have been recorded in the mites, tree hoppers, aphids and the true bugs etc.

#### **iv) Aggregation Pheromones**

Aggregation pheromones may or may not be produced by either sex to congregate the species for feeding or reproduction. In India, a few of the researchers have reported the occurrence of the aggregation pheromones. Mondal (1990) revealed that when larvae of rust red flour beetle, *Tribolium castaneum* were reared on an artificial diet containing 200 ppm, 2-methyl-1,4-benzoquinone and/or 1 ppm of the aggregation pheromone 4,8-dimethyldecanal, there was a significant reduction in adult emergence, and adults exhibited elytral deformities. Singh (1993) has reported on the release of both aggregation and sex pheromones by coffee bean weevil, *Araecerus fasciculatus*. Narula and Gandhi (1995) have reported on the possible aggregation pheromones present in fruitfly, *Dacus cucurbitae*.

#### **v) Sex Pheromone**

Sex pheromones generally originate from females and attract males for mating. It is the sex pheromone of insects that is of particular interest as a component to be used in IPM in agriculture both field and storage. Pheromone is originally defined as “substances, which is secreted outside by an individual and received by a second individual of the same species, which shows a specific reaction”. This definition clearly indicates that a chemical or a mixture of chemicals that is secreted outside by an individual and triggers an immediate response of sexual behaviour in the receiving individual, called as sex pheromone. Ethology, the study of animal behaviour has made a considerable progress during the last 4-5 decades, integrating their ecology, neurophysiology, endocrinology, and sensory physiology. A few well known researcher, who considerably contributed to such studies are Tinbergen (1951), Karlson and Luschar (1955), Karlson and Butenandt (1959), Frisch (1967), Muller Schwar (1969), Hinde (1970), Lorenz (1970,1971), Thorpe (1979), Yoshio Tamaki (1985). In fact they have widened the concepts and open new frontiers in the ever continuing strides of ethological investigations related to pheromones. Z-9-tetradecen-1-yl acetate, Z-11-tetradecan-1-01 and Z-11-tetradecanal have been registered for codling moth and leaf roller moths control in orchard crops (Rosell *et al.*, 2008).

#### **a) Attract and Kill**

There is a new generation pheromone called “attract & kill” which includes an attractant and a killing agent. Not much work has been done on this type of pheromone in

India. Selective attraction + insecticide treatment is the basis of this technique. Attract and kill involves an insecticide for killing the target insects. The inherent problem with this technique is the escape of the resistant strains to many insecticides even though they may be attracted. Some of the species *viz.*, *Anthonomus grandis*, *Bactrocera spp.*, *Ceratitidis sp.*, *Cydia pomonella*, *Ephestia kuehniella*, *Grapholita molesta*, *Musca domestica*, *Mythimna separata*, *Odontotermes formosanus*, *Pectinophora gossypiella*, *Plodia interpunctella*, and *Spodoptera spp.* that occur commonly both in India and other countries have been tested the efficiency of attract and kill pheromone in foreign countries.

#### **b) Lure and Infect – Auto- Infection System**

In this case the attracted insects only are auto infected. Virtually no work has been undertaken on this aspect in India. However in Japan an interesting work has been reported (Anonymous, 2000).

#### **vi) Epideictic (spacing) Pheromones/Oviposition Deterrent Pheromone (ODP) / Mating deterrent pheromones**

This pheromone is otherwise called spacing pheromone. There are pheromone mediated distribution of insects. Oviposition deterrence by various concentration of methanol-eggwash: More the concentration more was the deterrence.

#### **vii) Nasonov Pheromones**

The Nasonov gland pheromone of *Apis cerana indica* was analysed for the first time and reported to contain neral ((Z)-citral) as the main component (Naik *et al.*, 1988). Nasonov gland pheromone (NGP) having a mixture of components produced in the Nasonov gland of worker honey bees. These worker bees use Nasonov pheromone as an assembly pheromone. NGP of citral and geraniol (2:1 ratio) can be useful for assemblage of the worker bees. Traps containing insecticide, dichlorvos strips reported to be repellent to the bees.

#### **viii) Royal Pheromones**

The control of the workers bee by the queen through royal pheromones may not be as complete as many text books would have us believe. This is not surprising. Any apiculturist knows that exceptions are the rule by which honey bees and beekeepers must live. *Sciences et Avenir*, Number 603, May 1997, published in Paris carries an article that appears to show this all too well. Called "Revolt in the Hive," the article describes research done in Melbourne, Australia (Oldroyd *et al.*, 1997- Full reference not given; refer-Shelley *et al.*, 2003). According to them a rebellion is carried on by worker honey bees in some colonies in the quest to make their own genetic

contribution to the species. In spite of the queen's royal pheromonal control, the article says, DNA study confirms that workers continually are laying eggs in colonies that have many drones. This gives credence to Dr. Winston's doubts concerning inhibition of worker ovarian development. To avoid detection by their sisters, these rebellious egg-layers have covered up their deeds by ensuring their eggs marked with royal pheromone. The authors further suggest that some workers have even compounded their crime by replacing royal eggs with their own. Such anarchy, the article concludes, carries a virus of its own perpetuation, the production of drones, which pass the tendency on to other colonies through queens they mate with.

#### **ix) Calming (appeasement) Pheromones**

A recent innovation that has come on the market is offering a new way of helping the dogs suffering from all kinds of problems ((Jacqui Neilson, 2003). Among them are attention seeking, excessive licking, destruction, noisiness, and fears and phobias, such as gun-shyness and firework phobia and fear of thunder. It is known as a "Dog Appeasing Pheromone" (DAP). A bitch exudes a special pheromone when she has pups. Once the pups begin to explore, they can easily become terrified, as everything is so enormous, especially in today's homes, with people and furniture. Wild pups may meet predators and have to get used to wind and weather and their own surroundings, such as enormous trees and bushes. The DAP pheromone calms them, tells them everything is OK, not to worry, Mum's here, and they rush back to the bitch for safety, and are re-assured. Inevitably, they then leave her, and go into an environment where there is nothing like that to help them.

#### **x) Primer Pheromones**

Isabelle Leoncini *et al.* (2004) have given a comprehensive account of the typical primer pheromone of honey bee for easy understanding of the subject. The same has been given. Many animal species communicate via pheromones. Releaser pheromones cause rapid, transient changes in behavior, whereas primer pheromones cause more long-term changes in both behavior and physiology (Wyatt, 2003). These pheromones trigger a change of developmental events. Hundreds of releaser pheromones are known; in contrast, very few primer pheromones have been identified, primarily because they are much more difficult to assay (Slessor *et al.*, 1988). Research published in 1992 led to the hypothesis that the age at onset of foraging in honey bee colonies is regulated by worker-worker interactions (Huang and Robinson, 1992). Old bees inhibit the maturation of younger bees. For example, when a portion of a colony's foragers is removed to simulate predation, young bees develop faster compared with

those in a control colony in which the same number of individuals is depleted, but evenly across different age classes (Huang and Robinson, 1996). Conversely, when foragers are confined to their hive by artificial rain young bees delay, rather than accelerate, their maturation (Huang and Robinson, 1996). The feasibility of the worker social inhibition concept has been supported both by empirical findings (Robinson, 2002; Huang and Robinson, 1992; Huang and Robinson, 1996; Huang *et al.*, 1998; Naug and Gadagkar, 1999; Leoncini *et al.*, 2004) and theoretical models (Naug and Gadagkar, 1999; Beshers *et al.*, 2001; Amdam and Omholt, 2003).

### Pheromones in Human

Pheromone-induced responses are mediated primarily by the vomeronasal organ (VNO). The VNO, also known as “Jacobson’s organ”, is part of an accessory olfactory system. It is present in a variety of non-human vertebrates but its existence in the human has been open to question until recently. The VNO was first discovered by Ruysch (1703), a military doctor, in a soldier with a facial wound. The organ was named after Jacobson who published his findings on animals, but not humans, in 1811. Various other investigators have published studies. Its occurrence in many animals, in human embryos and mentioned that it may be found occasionally in the adult human and more detail can be found in Moran *et al.* (1994) and Stensaas *et al.* (1991). No research work on the existence of the pheromones in human beings from India is on record. However, a lot of information is published on the human pheromones from other countries. By definition, a human pheromone elicits changes in the physiology and/or behavior of a con-specific. Stern and McClintock (1998) showed that the pheromones of women regulate ovulation, presumably by affecting levels of Luteinising hormone (LH) and Follicle-stimulating hormone (FSH). Minimally, human pheromones appear to alter both physiology and behavior in other humans. It is still unknown how many different pheromones are produced in human but some of them have been investigated in recent years. Most studies focused on the 16-androstenes, metabolites of the characteristically male sexual hormones, the androgens, which are secreted by the apocrine glands.

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