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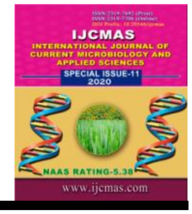

Review Article

Colony Collapse Disorder and their Causes

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A B S T R A C T

Honeybees play crucial role in boosting agricultural production by significantly increasing the yields by helping in pollination. A third of all our food depends on their pollination. However, in recent years, honey bee colony declines in Europe, United States and Middle East, but such declines are not apparent in South America, Africa, Asia and Australia. Current technologies have become a source of omnipresent electromagnetic pollution from generated electromagnetic fields and resulting electromagnetic radiation. Electromagnetic radiation causes harm not only honeybees but also potentially affect the human being, birds, bats and plants etc. This is despite the fact that extremely low frequency electromagnetic fields were classified as potentially carcinogenic. For these reasons, in recent decades there is a need for scientific research in order to observe and understand the influence of electromagnetic radiation on living organisms.

Introduction

Colony Collapse Disorder and honey bee population decline are very recent developments in the agricultural world and their causes are highly contested. Some of these debated causes include cell phone use, diseases, monoculture crops, intensive agriculture, as well as the introduction of invasive bee species in new habitats. Honey bees play a vital role in the pollination of agricultural crops (Patel *et al.*, 2016). A third of all our food depends on their pollination. Many agricultural crops such as *Anacardium occidentale*, *Castanopsis acuminatissima*, *Cinnamomum kerrii*, *Coccinia grandis*, *Cocos nucifera*, *Coffea Arabica* are pollinated by honeybees (Suwannapong *et al.*, 2011). Along with pear *Pyrus communis* L., Apple *Malus domestica*, Japanese Plum *Prunus salicina*, Blueberry *Vaccinium ashei* (Dedej and Delaplane, 2003). Tomato *Solanum lycopersicum*, Sunflower *Helianthus annuus* (Greenleaf and Kremen, 2006). Self-pollinated crops also benefit from insect pollination, that increase yield up to 30% from pollinator visits and also collection of nectar or pollen and benefit farmers from pollinator's service. Lack of pollinators causes decline in fruit and seed production (Partap, 1992). Several studies have stressed the importance of honeybees for fruit and seed yields in different crops and cultivars (Delaplane and Mayer, 2000; Free, 1993 and Klein *et al.*, 2007).

A world without pollinators would be devastating for food production. The economic value of bees' pollination work has been estimated around € 265 billion annually, worldwide. So, also from a purely economic point of view, it pays to protect the bees. However, in recent years, honey bee colony declines have reached 10-30% in Europe, 30% in the United States and up to 85% in Middle East, but such declines are not apparent in South America, Africa, Asia and Australia (Kluser, 2010). The syndrome behind honeybee population decline is known generally as Colony Collapse Disorder (CCD), which is characterized by a rapid loss of adult worker bees (VanEngelsdorp, 2009).

Colony Collapse Disorder

Colonies with CCD can appear healthy just weeks prior to collapse. However, the adult bees soon "disappear" (hence its historic nickname "disappearing disease") from the colonies, leaving behind a hive full of honey, pollen, open and capped brood, a queen and maybe a few worker bees. The phenomenon was first called "Fall-Dwindle Disease," but was renamed because of the unusual characteristics of the colony declines. Moreover, the condition is not only seasonal but manifests itself throughout the year. The term "dwindle" implies a gradual loss, whereas CCD onset is sudden. Beekeepers report that colonies with CCD do not contain any dead bees, nor are there dead bees on the ground outside of the colonies. The adult bees simply vanish. The final symptom is that small hive beetles, wax moths and other nearby honey bees ignore the empty hives even though the hives contain foodstuffs on which they ordinarily feed. Researchers suspect many factors to be responsible for the killing of the bees, of which the varroa mite, pesticides, viruses, farming practices, monoculture, hygiene in the hive and climatic factors are the most widely cited possibilities (U.S.D.A. 2007). Other causes might include genetically modified crops (Malone and Pham-Delegue, 2001) and exceptionally cold winters. CCD initially affects the worker bees, which abscond the hive. The queen bee is usually abandoned in the hive with the young brood with ample of honey. However, without the worker bee population, the colony becomes unsustainable and dies out.

Disorder is characterized by:

An absence of adult bees in colonies with few dead bees in the hive or in front of the hive.

Frequent presence of capped brood in colonies.

Presence of food reserves (honey, pollen) that had not been robbed out and hives not attacked by pests such as wax moths, hawk moth and *Vespa* species

In colonies that still have bees, characterized by small clusters with a laying workers

Colony losses are caused by a variety of factors acting together. These include the ecto-parasite mite *Varroa destructer*, environmental stress, transgenic crops, lack of food variety a toxic ingredient (hydroxyl furfural) in high fructose corn syrup which is commonly fed to bees in the USA, electromagnetic radiation from cell-phone towers, antimicrobial and acaricidal chemicals used by beekeepers to control infectious diseases, mites and the widely used agrochemicals including the neonicotinoid insecticides.

In some countries there are reports of mysteriously dying bees. It appears as if the losses are at their worst in the northern American states and neighbouring Canada. The American bee-keepers reported 25% - 50% losses through "Colony Collapse Disorder" (New Scientist, 2007). They also reported that 50-90% of their bees to have disappeared within the previous six months and the remaining bee colonies were said to be so weak they can produce hardly any honey (CNN, 2007).

What's Causing Colony Collapse Disorder

Many possible causes of CCD have been put forward like diseases, pests, pesticides, genetically modified organisms (GMO) farming, electromagnetic radiation, changes in brood temperature, nanotechnology, food deficiencies and loss of forage. Currently, interactions between some of these factors, especially the pests and pathogens are regarded as the most likely explanation (Neumann and Carreck, 2010).

Electromagnetic radiation caused by mobile phones is thought to have effects on the ability of bees to navigate causing whole colonies of bees to disappear (Warnke, 2009). Also HAARP (high-frequency active auroral research project), a military project with many antennas based in Alaska, is mentioned as a cause of CCD in 2006 (VanEngelsdorp, 2009, Warnke, 2009).

Colony Collapse Disorder may be caused by a number of interwoven factors:

Global warming

Global warming, which has caused flowers to bloom earlier or later than usual. When pollinators come out of hibernation or aestivation the flowers that provide the food they need to start the season have already bloomed.

Pesticide use on farms

Some toxic pesticides meant to kill pests can harm the honey bees needed for pollination. Many pesticides banned by other countries because they harm bees.

Habitat loss brought about by development, abandoned farms, growing crops or flowers in gardens without leaving habitat for wildlife that are not friendly to pollinators.

Parasites such as ecto and endo parasitic mites

Pathogens of bees or brood: viruses (ABPV, KBV, IAPV, BQCV, DWV, SBV), fungus (Stone and Chalk brood) and protozoan (*Nosema ceranae* and *Nosema apis)*

Queen failure (relative risk colony mortality increases)

Colony stress (compromises immune system may disrupt social system) - possible causes include poor nutrition, movement (migratory stress), environmental factors (drought, cold temperatures) and pesticides

Possible Causes of Colony Collapse Disorder

There are several factors currently being explored and debated to explain CCD and they fall into two main classes: living organisms and human activity.

Living Organisms

Most of the living organism threats are invasive species. Small hive beetles which come from South Africa are harmful too. They lay their eggs in beehives, which damages honeycombs and makes bees flee the hive Another invasive species threat is the mite *Varroa destructor*, since "it feeds on bee hemolymph (insect "blood") and wreaks havoc in hives" (Watanabe, 2008). They attack both the adults and the brood (Bessin, 2001). There also exist two species of microsporidia, the *Nosema apis* and *Nosema ceranae*, which damage the internal organs of bees. This causes bees to become much more vulnerable to bacteria and viruses, including chemical agents and pesticides. Research has also discovered a new virus, "Invertebrate Iridescent Virus "IIV" in North America, that when combined with the aforementioned *microsporidia*, decimates hives very efficiently**.**

Human Activity

The second major set of threats to honeybees is from human activity itself. In the US, scientists have found traces of more than 120 different pesticides in beehives (Valo, 2011). According to the ARS Bee Research Laboratory, "some subtle interactions between nutrition, pesticide exposure and other stressors are converging to kill colonies (Benjamin, 2010).

Another anthropogenic cause is the abundance of genetically modified organisms (GMOs). GMOs produce their own insecticides, which can harm bees who, while gathering pollen and nectar, ingesting those artificial proteins. It is also important to consider what "GM pollens in honey have to do with food safety and possible horizontal gene transfer to human cells (Sanford)". Crop monocultures are one of the other causes of bee decline since the honeybee immune system needs a diversity of pollen from the plants and flowers that bees are in contact with (Black, 2010). Their immune system produces glucose oxidase, which is used to sterilize food for the whole colony and to protect the honey from microbes. When there are less than 5 different kinds of pollen around the hive, bee immune systems are weakened and they become more susceptible to diseases and death.

Finally, the last cause examined is the influence of electromagnetic radiations and cell phone use on bees. Researchers in India studied the effects of cell phone radiation on honeybees by sending electromagnetic waves through the hives twice a day for fifteen minutes. As a result, after three months, "bees stopped producing honey, egg production by the queen bee halved and the size of the hive dramatically reduced (Herriman, 2010).

India is one of the fastest growing mobile telephony industries in the world. The first mobile telephone service started on the non-commercial basis on 15 August, 1995 in Delhi. During the last 25 years, India has seen exponential growth of mobile telephoning. At present The 4.5 lakh mobile towers in India are turning the country into an open microwave. Nearly 1,172.44 million Indians have mobile phones, making it the second largest mobile subscribers in the world after China. At present, there are nearly top five companies providing mobile telephoning constituted 98.98% market share. According to the Telecom Regulatory Authority of India (TRAI, 2019), the composition of telephone subscribers using wireless form of communication in urban area is 55.93% and rural area is 44.07%. To support this growth of cell phone subscriber in the country, there has also been a tremendous growth of infrastructure in the form of mobile phone towers. The transmission towers are based on the electromagnetic waves, which over prolonged usage have adverse impacts on humans as well as on other fauna.

Effect on navigation

Animals that depend on the natural electrical, magnetic and electromagnetic fields for their orientation and navigation through earth's atmosphere are confused by the much stronger and constantly changing artificial fields created by technology and fail to navigate back to their home environments. Most people would probably shrug this off, but it affects among other one of the most important insect species: the honeybee. Because the bee happens to be the indispensable prerequisite for fructification: without bees, the fruit, vegetable and agricultural crops will fall short.

Magnetoreception refers to the way which organisms can perceive natural magnetic fields (like the Earth magnetic field and use them for different purposes like orientation. In bees, magnetoreception is thought to act through magnetite and cryptochromes (Edmonds, 1996; Winklhofer and Kirschvink, 2010; Gegear *et al.,* 2010). Bees sense the Earth magnetic field and use this field to determine the direction in which new honeycombs should be built in the dark in nesting cavities (Gould and Gould, 1988).

This magnetoreception system is highly sensitive, because honeybees can detect magnetic fields as low as 26 nanoTesla (nT), compared to the 45000 nT of Earth's magnetic field (Kirschvink *et al.,* 1997). In case man-made EMF disturbs the way honeybees sense the Earth magnetic field, they could be affected in their homing ability and/or the accuracy of the waggle dance. However, they could in many situations perhaps still rely fully on other navigation systems, like the sun's position.

According to Dr Ulrich Warnke, he concludes that the orientation and navigation of bees may be disturbed by man-made technical communications fields. It may be due to the effects on their cryptochrome pigments, which they use for both solar and magnetic navigation, and is highly sensitive to radio frequency radiation. Cryptochromes absorb light and use its energy to repair damaged DNA. They also regulate the timing of their natural circadian rhythms. Animals that navigate using the earth's magnetic field also use cryptochromes to sense the direction of the field. Cryptochrome can detect the direction of the field because it uses the energy of light to flip an electron between two parts of the molecule to generate a pair of unstable magnetic free radicals.

The electron tries to return to its original position, but the rate at which it does so depends on the direction of the earth's field relative to the molecule, and gives an indication of the direction of the field. Bees use internal 'clocks' for navigation. Mobile phone radiation can disrupt these biological clocks. Once disrupted in bees, they are no longer able to compensate for the changing position of the sun throughout the day, causing them to fly in the wrong direction, away from the hive (Andrew Goldsworthy). (Rubin *et al.,* 2006) also discovered that the molecular structure of the biological clock of the honey bee is more similar to the biological clock of mammals than that of other insects, including the circadian production of melatonin.

The clock is also essential for navigation that uses the sun as a compass because the sun moves during the day from east to west. EMFs from telecommunications infrastructures could interfere with bees' biological clocks that enable them to compensate properly for the sun's movements and may fly in the wrong direction when attempting to return to the hive. They could disappear mysteriously.

This phenomenon has been widely reported in the past years. In contrary, none of the apiculturist in India reported the term "CCD" and their bee hives facing this phenomenon. In India, bee colonies decline only due to floral dearth if not migrated to bee floral regions. There is no impact of radiation on the foraging activity of *A. mellifera* L which were under the influence of mobile tower radiation (Patel and Mall, 2019).

Effects of EMF on other organisms

A lot of studies have been done on the effects of EMF on mammals, insects and some on plants as well. Studies on the effects of EMF on the fruit fly (*Drosophila melanogaster*) will be highlighted as this species is relatively closely related to the honeybee (both are holometabolous insects). Studies about the effects of EMF on orientation will also be discussed, as honeybees and other animals which make use of a magnetic compass have a similar mechanism for navigation.

Studies on animals confirmed that the most consistent and reproducible responses to acute radio frequency (RF) exposure were caused by thermal effects. When animals' core temperatures are increased by about 1 °C due to thermal RF exposure, deficits in learning behaviour occur.

A significant increase in body temperature caused by RF radiation can impair male fertility and increase incidences of foetal malformations and anomalies as well as embryo and foetal losses (Vecchia *et al.,* 2009). According to a report by Dr. R.K Kohli, the Director of the Centre for Environmental Studies, India, "The electromagnetic radiations from the towers generate heat and kill microorganisms present in the soil near it. This in turn harms those organisms that feed on them and it disturbs the ecological cycle".

Studies on EMF impacts

Impacts on Human

EMRs and their negative impacts on biological systems and environment have already been reported by several studies. Some earlier investigators also have contended that there is no measurable risk of reproductive failure and birth defects from EMF exposures in humans (Brent *et al.*, 1993). Epidemiological studies - Sleep disruption, Headache, Depression, discomfort, irritability, nausea, dizziness, appetite loss, muscle spasms, numbness, tingling, altered reflexes, 4 times incidence of Alzheimer's disease (Hakansson *et al.*, 2003). 3 times amyotrophic lateral sclerosis (ALS) (Savitz *et al.*, 2013)

Bird species are disappearing

There are in total 47 species, including honey bees, that make use of a magnetic compass (Wiltschko and Wiltschko, 2005). The house sparrow, for instance, has become clearly scarcer in England and some western European countries.

An investigation carried out between October 2002 and May 2006 in Valladolid in Spain, was launched to examine whether this decline in the sparrow population was related to electromagnetic radiation by mobile base stations. The result showed with a high degree of statistical confidence that the number of sparrows was reduced when the electrical field strengths of the antennae exceeded certain values (Balmori and Hallberg, 2007).

Effects on plants

Tkalec (2008) exposed duckweed (*Lemna minor L.)* to radiofrequency radiation at 400 and 900 MHz and concluded that non-thermal exposure to RF fields induced oxidative stress in duckweed as well as unspecific stress responses, especially of antioxidative enzymes.

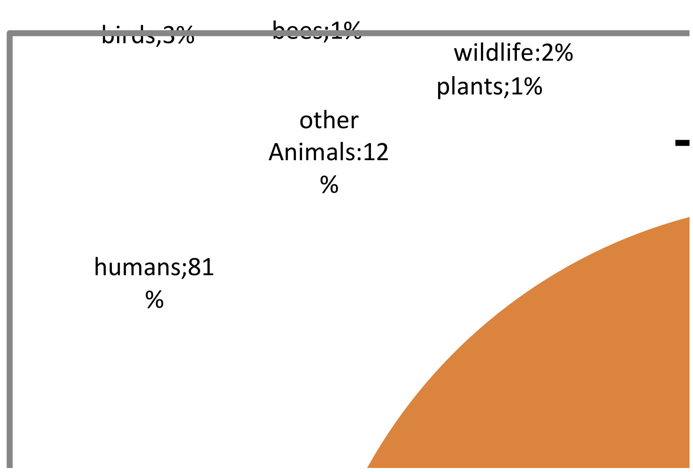
He also found that the observed effects markedly depended on the field frequencies applied as well as on other exposure parameters, such as strength, modulation and exposure time.

Table.1 Percentage of production dependent on honey bee pollination

Crop	Dependence%	Crop	Dependence%	Crop	Dependence%
Almond	100	Grapefruit	72	Peanut	2
Apple	81	Kiwifruit	72	Pear	45
Apricot	56	Lemon & Lime	18	Plum & Prune	63
Avocado	90	Lucerne seeds	90	Pumpkin	10
Bean-Soybean	5	Lupin	10	Raspberry	90
Blueberry	90	Macadamia nut	81	Rockmelon	90
Canola seed	90	Mandarin	27	Strawberry	4
Canola	14	Mango	72	Sunflower	90
Cherries	81	Nectarine	48	Watermelon	63
Cotton lint	8	Orange	27	Zucchini	90
Cucumber	90	Papaya	16	Vegetable seed	90
Field pea	45	Peach	48		

Sources: ABARES calculations based on Cook et al. (2007); Cunningham, Fitzgibbon and Heard (2002); Gordon and Davas (2003): Keogh, Robinson and Mullins (2010);Monck, Gordon and Hanslow (2008); Morse and Caldefone(2000)

Fig.1 Studies on EMF impacts



Roux (2008) exposed tomato plants to low level RF radiation and reported that the accumulation of stress-related mRNA was typical of an environmental stress response and a direct consequence of RF radiation. They concluded that the plants perceived the radiation as an injurious stimulus. Effect on bats: Activity of bats seems to be much reduced in areas with electromagnetic fields with densities more than 2V/m (Balmori, 2009). Based on this fact it was recommended to use EMR to repel bats from wind farms (Nicholls and Racey, 2007).

In another study in a Free tailed bat colony (*Tadarida teniotis*) the number of bats decreased when several phone were placed 80m from the colony (Balmori *et al.,* 2007).

The following areas for specific studies are suggested to be taken up

Field studies on impact of cell towers on bee colonies and apiculture,

Bird/bat/insect mortalities at mobile phone towers with special reference to towers along bird migratory paths.

Studies on birds / bats / bees to find the effect of EMR on their communication, orientation and co-ordination.

Effect of EMF on amphibian metamorphosis and sex determination in reptiles.

Laboratory studies to develop an understanding on certain species, on their physiological and behavioural aspects, making use of the techniques of bioassay/bio-monitoring.

Measurement, monitoring and mapping of background EMF levels and power density across India involving independent research agencies.

Regulations/standards to include the ecological characteristics of an area while determining the location of transmission towers, relay stations etc.

Regulations to control installation of transmission towers in human residences/hospitals/dense habitations.

Conduct ecological impact assessment of transmission towers and base stations, with standardised protocols/parameters.

Neonicotinoid Problems

Neonicotinoids are highly toxic to bees and have been blamed as a leading cause of CCD and colony decline. Neonicotinoids are systemic and can lead to contamination of pollen and nectar can lead to residues in the hive in pollen (~3ppb) and wax (0.1 ppb) - levels below acute and chronic toxicity levels may be released by guttation in seedlings.

Use banned in France, Italy and Germany and for some uses in England. In 2008, large number of bee kills in Germany associated with contaminated dust released during the planting of treated corn seed. In 2011, bee kills due to thiamethoxam and clothianidin detected in corn plants in India. Other concerns for neonicotinoid toxicity, synergistic effects and interactions with pathogens.

Pesticide Effects on Colony Health

Fungicides can reduce microflora in beebread and may affect natural resistance mechanisms in colony, especially to fungal disease.

Pesticides (including antibiotics) can affect the microbiota of the honey bee, compromising the immune system and overall bee health.

Pesticide spray adjuvants (such as organo siloxane surfactants) may affect learning and increase bee mortality.

Protect bees when using pesticides

Adopt an IPM approach for controlling pests in areas where bees are present.

If possible, choose pesticides that will specifically target your pest and not harm bees.

Keep in mind that dusts and microencapsulated pesticides may be similar to pollen in size and shape. Do not over-apply pesticides, use only the amount directed on the label.

If possible, avoid applying pesticides during at mid-day when bees are most likely to be out foraging for nectar and pollen.

Farmers must be rewarded for practices that help wild bee populations thrive, such as leaving habitat for bees in their surrounding fields, alternating crops so bees have food all year long and not using harmful pesticides.

Economic Impacts

CCD should theoretically have an effect on food prices. To begin with, the unit price to rent a colony of bees has increased from $45-65 to $170 dollars, almost three times as much, in the United States. This should logically correlate to higher prices in our supermarkets. Constant or increased demand for pollinator crops coupled with a reduced supply will also result in higher prices.

Ecosystems

Honeybees play a vital role in the pollination of agricultural crops. Bees are estimated to pollinate over 66% of the world's 1,500 crop species. They also contribute directly or indirectly to 15-30% of global food production (Patel and Mall, 2015). However, their positive effect on natural ecosystems is often overlooked and the declining population of honeybees will have drastic long-term effects on the biodiversity of ecosystems. Although it is nearly impossible to determine the economic value of honeybees on biodiversity and ecosystems, they undoubtedly play a central role in many ecosystems (Brown and Cuthbertson, 2009). The drastic decline in honeybee populations and the resulting consequences of reduced crop yields will have dire effects on the world's ecosystems and biodiversity. It is a well-known fact that more than 35% of global crop production depends on insect pollination. Most of these pollinators are wild bees (Klein *et al.,* 2008).

Agricultural Practice

Current agriculture practices in the developed world use immense amounts of herbicides and insecticides to maximize yields in largely monoculture crops. It is possible that the chronic use of these poisons has lasting effects on pollinator health, even though they are not the intended targets. One change to modern agriculture would be to reduce the amount of chemicals used in order to improve pollinator health.

The seriousness of the health hazards due to radiation from the cell phones and cell towers has not been realized among the common man. Cell operators continue to claim that there are no health issues. The Department of Telecommunication (DoT) in India has set new norms for cell phone towers with effect from September 1, 2012 (The Hindu, 2012). Exposure standards for RF-EMF radiation has been reduced to one-tenth of the existing level and SAR from 2 to 1.6 W/kg. This came after the Ministry of Environment and Forests (MOEF) set up an Inter-Ministerial Committee (IMC) to study the effects of RF-EMF radiations on wildlife and concluded that out of the 919 research papers collected on birds, bees, plants, other animals, and humans, 593 showed impacts, 180 showed no impacts, and 196 were inconclusive studies.

Studies on impact of cell phone tower radiation on birds and wildlife are almost non-existent from India. There is an urgent need for taking up well designed studies to look into this aspect. Available information from the country on the subject of EMF impacts is restricted to few reports from honey-bees. However, these studies are not representative of the real life situations or natural levels of EMF exposure. More studies need to be taken up to scientifically establish if any, the link between the observed abnormalities and disorders in bee hives such as Colony Collapse Disorder (CCD).

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