METHODS OF STUDYING ANIMAL BEHAVIOR

Ethology is the study of animal behaviour to find out natural responses of animals to various environmental stimuli. Some studies are also done in laboratory conditions to elicit measured responses. Therefore, ethology involves laboratory as well as field studies and has strong relationship with other sciences such as ecology, environmental science, neurology, physiology, psychology and evolution.

The beginning of modern ethology commenced with the experimental as well as field studies done by the Dutch biologist Nikolas Tinbergen, Austrian biologists Konrad Lorenz and the German Karl von Frisch, who were jointly awarded Nobel Prize in 1973 for their contribution to this new science.

LABORATORY STUDIES

Neuroanatomical techniques

Different types of behaviour are controlled by specific regions of the brain. If a particular part of the brain is damaged, the behaviour of the animal is altered. Broca (1861) identified speech area on the cerebral cortex by the slurring of speech of a patient as a result of injury to the brain. Brain parts can be damaged by making cuts with a knife or by the neurotoxic kainic acid and behaviour is observed.

Carl Lashley (1938) conducted his studies on memory by ablation on different brain parts of rats which were trained to running maze. Which area of the body is affected by damaging which part of the brain was studied on rats by De Groot (1959); on cats by Jasper & Marsan (1954); and on dogs by Lin et al. (1961).

Stereotaxic equipment can be used to place small and precise injuries in brain. Micropipettes can be used to inject minute quantities of chemicals in precise locations of brain, such as limbic system, and behaviour can be recorded.

Studies can also be done by training the animals in skinner box, in which a lever can be pressed by the animal to get reward.

Neurophysiological techniques

Physiological studies can be done by recording electrical activity of brain by EEG or by stimulating different areas of brain by planting electrodes. Alpha, Beta, Theta and Delta waves are recorded by EEG. Alpha waves that are believed to emanate from the parietal and occipital lobes of brain reveal resting and peaceful relaxed state of brain that is otherwise alert. Beta waves are produced in frontal lobes and indicate the daily mental activity, concentration and thought. Theta waves denote emotional stress and sometimes hallucination. Delta waves are generated in deep sleep.

Neurochemical techniques

These techniques involve stimulation of parts of brain by drugs such as alcohol, opium, hashish, bhang etc. which alter the behaviour of the animal. Tranquilizers, barbiturates and drugs like calmpose, larpose etc. are phychoactive drugs which affect the brain and change the behaviour of animals.

Hormones such as estrogen and testosterone can be introduced into hypothalamus through canulation and the behaviour changes can be recorded. Adrenalin, histamine, testosterone and dopamine stimulate different parts of the limbic system. For example, stimulation of amygdala brings about aggressive behaviour and stimulation of septum pellucidum gives immense pleasure to the animal.

The modern techniques, e.g. PET scanning, CT scans, MRI etc. detect glucose utilization in different parts of brain, which is an indication of activity of that part.

FIELD STUDIES

BEHAVIORAL SAMPLING METHODS

Focal Animal Sampling

In this sampling method an individual from a group of animals is selected and all behaviours are recorded for a specified time period. During the specific period, all activities that the animal performs are recorded, while the activities of the other animals of the group are not recorded.

After the time period is over, the observer moves to another individual of the group to record its activities. This continues until all animals of the group have been observed for the specified time period. Individuals are identified by marks and named. Jane Goodall conducted such studies on chimpanzees. This method provides unbiased data on a wide variety of questions about the animals and is generally considered most satisfactory approach to studying animal behaviour.

Ad Libitum sampling

Ad libitum in Latin means "at one's pleasure". A group of animals is selected and the observer remains with this group for a considerable period of time to observe all activities of the group. No constraints are placed as to what should be recorded and when.

All behaviours including interactions among the individuals are recorded in field notes. For instance Diana Fossy observed gorillas by living with the group whole day and observing all kinds of behaviour. Because the observer can never keep track of every minute activity of animals, the results of these observations can be biased depending on the situations that attract the observer's attention.

Instantaneous Sampling

In this method the observer records the behaviour of an individual in a group at predetermined time intervals, e.g. hourly or half hourly or per minute. The observer records the state of the animal rather than events. The sample interval should be as short as possible and behaviours should be easily identifiable.

Continuous Sampling

In this method, the observer simply records all the activities of the animals while they are being watched. This sampling method is very helpful in recording the sequence of activities that make the behaviour, such as courtship display in birds or fighting sequences in deer or moose.

Scan Sampling

In scan sampling the behaviour of all individuals of a group of animals are recorded at fixed time intervals. This involves rapid scanning of the whole group of subjects at regular interval and behaviour of each individual is recorded. Usually the observer restricts himself to recording of few categories of behaviour. An example of scan sampling would be to observe a group of animals and record the behaviour of each animal per unit of time. This provides data on the distribution of behaviour states in group for a long time period.

All Occurrence sampling

Sometimes certain behaviours are performed by several animals at the same time. For example, one animal starts the alarm call and other animals follow. In such cases the behaviour can be recorded as one event. One can record the number of alarm calls per unit of time, which will provide rate of occurrence of the behaviour in a fixed period of time.

Sequence Sampling

In this method, focus is on an interaction instead of on the individuals. The whole sequence is recorded from beginning to end. For example, the aggressive and submissive behaviours can be recorded in social primates. Various types of interactions in social insects such as ants, termites and honey bees can be recorded by sequence sampling.

One-zero sampling

Recording of the occurrence is done in "Yes" or "No", depending on the activity performed or not. The results can be presented in frequencies. For example attack behaviour in territorial animals or infant-killing in monkeys.

TRACKING ANIMALS IN THE WILD

In order to study the behaviour of animals in their natural environment it is important that the animals are spotted in the wild, identified and studied without disturbing them. Animals can be identified by natural marks such as broken horn or tail, body scars or ear notches. Size and shape of the horns and tusks, facial features or pattern of stripes and spots can also be of help in identification. Photographs can be taken or drawings are made to record the identifications.

Wherever it is possible, animals can be captured, marked and then released in the wild for easy identification while studying their behaviour. Ringing of birds is a common method of identifying birds and to trace their migratory route. Fishes can be collected by net, marked and released. Birds can be collected by fog nets which are invisible to the flying birds. GPS tracking systems are used these days to track the animals in the wild, in which animal's movements can be continuously tracked for several days.

KINSHIP SELFISHNESS AND ALTRUISM

There are four possible types of interactions among individuals living together in a population. First, *cooperation* or *mutualism,* in which both the participants gain from the act as in the nest building by both male and female birds, or cooperation in the colony of social insects.

Second, *altruism* in which the actor (individual that carries out the action) pays fitness cost to the recipient that gets the benefit as in social insects.

Third is the *selfishness,* in which the actor gains but the recipient loses in terms of fitness. Fourth interaction, which is rather rare in nature, is *spite* in which both the participants lose in terms of fitness. As for example in the case of two eagles fighting in the air for the possession of a killed prey, which ultimately falls down and is taken away by a fox.

KINSHIP & ALTRUISM

Kinship is a phenomenon that occurs in social animals or in closely knit populations which are genetically related to one another. In these populations kin selection operates and traits that result in decreased personal fitness but increase the survival and reproductive fitness of the species are favoured by natural selection. Kin selection works not on individuals but on genotypes.

Altruism evolved in colonies that show kinship. An altruist by way of helping other individuals increases the fitness of its own genome. A honey bee worker is a sterile female and shares at least 50% of its genotype with its sisters even when its mother and father are unrelated. If a worker decides to breed on its own, its diploid daughters and haploid sons will never be more than 50% related to it. So, the workers choose to become sterile and ensure survival of their genetically identical sisters, because the queen can produce more offspring than workers can do individually.

Kin selection leads to altruism in a colony and fitness is direct when it gives the individual personal benefit and reproductive advantage, and indirect when the reproductive benefit goes to the colony or relatives. Kinship favours the spread of an allele that increases the indirect component of fitness of an individual and in most instances it gives rise to altruism.

The gene that favours altruism could spread when participants are related and the cost to the individuals is low as compared to the benefit to recipient. Therefore, altruism is promoted by kin selection and close genetic kinship.

In a large number of bird species, especially those in which nesting opportunities are limited, young ones help their parents in rearing their own sisters and brothers by way of nest building, nest defence and feeding the chicks, although they are themselves capable of breeding. In such birds, as for example in bee-eaters help is always given to their kin.

RECIPROCAL ALTRUISM WITHOUT KINSHIP

The theory of group selection was championed by Wynne-Edwards (1962). Altruism has evolved among the related individuals by means of kin selection. But there are also instances of cooperation among the unrelated individuals. Altruistic act towards non-kin is possible only if the recipient is likely to return the favour at a later date, in a *'Tit for Tat'* manner.

Natural selection will favour altruism among unrelated individuals only if they reciprocate. Non-reciprocating or selfish individuals of the population are selected out. Robert Trivers (1971) proposed that reciprocal altruism can develop in the following conditions:

1. If interacting individuals remain together for considerably longer period of time.
2. If frequency of altruistic attempts is high.
3. If the cost and benefit to both individuals are more or less equal.
4. If selfish individuals that fail to reciprocate are punished in some way, such as withdrawing the benefits in future.

Species which have mutual dependence in defence, foraging, territoriality etc. are most likely to develop reciprocal altruism, as in monkeys, baboons, chimpanzees and man. Kin selection and reciprocal altruism are sometimes found to coexist in many social groups of animals and at times it is difficult to distinguish between the two or measure them independently. Altruism is promoted by group selection but when it benefits close relatives it is promoted by kin selection.

TROPHOLAXIS

This is a phenomenon in which food is offered by one individual to the other which is not its own offspring. This is very common in social insects where feeding is done by specialized individuals of the colony called workers. In chimpanzees distribution of meat among individuals after collective hunting of monkey has been recorded.

Wilkinson (1984) studied blood-sharing in vampire bats *(Desmodus rotundus)* in Costa Rica. The bats demonstrated altruistic behaviour by regurgitating blood meal and sharing it with others. Trophollaxis is essential for the survival of species which do not find enough food and starving individuals must be helped for the benefit of the species. Wilkinson also found that bats regurgitate food more frequently to relatives and rarely to non-relatives, since relatives are likely to reciprocate when they do not find meal themselves. This is called reciprocal altruism.

COST OF SELFISHNESS

Individuals living in groups enjoy the advantage of protection from the predators as some members out of hundreds will spot the predator and give alarm calls to alert the others. Similarly, prey hunted by a predator group can be shared by all members and sometimes even injured individuals that cannot hunt themselves can also get food. But this favour must be returned by individuals when their turn comes.

If the favour is not returned then the individuals are labelled *selfish* and will be selected out of the group. Herding protects the herbivore animals from predators but an individual straying away from the group will be killed by predators and eliminated from the population. Selfishness is therefore punished by natural selection. Selfishness is also found in the prides of lions where male lion kills all the cubs after dethroning a lion and taking over his pride.

This is done to bring the lionesses to oestrus so that he can have his own progeny quickly. Selfishness is also seen in protocooperation in which only one individual derives benefit, as in the case of suckerfish attached to the shark. Shark does not get any benefit but the suckerfish gets leftover food from shark's mouth. Group selection and kin selection, therefore, demand faithfulness to the society and selfish individuals are selected out and eliminated from the population.

ORIENTATION, NAVIGATION AND HOMING

Orientation is the position of the animal with reference to gravity or resource. This is the position the animal maintains in order to reach the resource. Positional orientation is to maintain upright posture against gravity for which vertebrate have membranous labyrinth and invertebrate statocyst.

Object orientation takes place when the animal tries to approach an object which may be food or water. Aquatic animals move vertically in pond or lake which is called strato-orientation. When the animals try to move from grassland to forests, deserts or mountains it is called zonal orientation. Animals which migrate long distances generally possess topographical or geographical orientation.

KINESIS

Kinesis is the movement of an animal in response to stimuli. It may be oriented or undirected movement depending on the source of stimulus. The response may be proportional to the intensity of stimulus.

Klinokinesis is the change of direction during movement which may increase or decrease in the light of intensity. Generally the animal moves right and left alternately to compare the direction of stimulus to gain correct orientation. Animals having single receptor show alternate movements. Caterpillar and maggots looking for the sites of pupation vacillate while moving.

Orthokinesis depicts speed of locomotion which s related to the intensity of stimulus and accumulation of action specific energy in the animal. Whole body of the animal is involved. For example, burrowing animals such as *Ammocoete* larvae of lampreys burrows in sand away from light. Cockroaches move from brighter areas to darkness.

Different types of kineses are termed with respect to the stimulus, e.g. hygrokinsis is with respect to humidity as in isopods; photokinesis in which stimulus is gradient of light and chemokinesis is with respect to chemical stimuli.

TAXIS

Taxis is the orientation of the animal with reference to the direction of stimulus in space. Movement can be towards or away from the stimulus and depending upon the stimulus it can be names as follows: hygrotaxis (humidity); geotaxis (gravity); chemotaxis (taste or odor); thermotaxis (temperature); anemotaxis (air current); rheotaxis (water current); phototaxis (light intensity); phonotaxis (sound waves); astrotaxis (sun, moon and stars); menotaxis (angle to the stimulus); mnemotaxis (based on memory).

Klinotaxis occurs in those animals which have single receptor, as in *Euglena,* which compares the intensity of stimulus by alternate lateral movements. Similarly in the maggots of Diptera the light sensitive organ is a cluster of cells above and behind the mouth and the negative response to light is compared by flexing movement of body.

Tropotaxis is found in animals which have paired receptors as eyes in Planaria. Animal gets equal inputs on both the receptors and hence it can move in straight line towards or away from light. If one eye of an insect is painted black it makes circling movements towards the side of painted eye.

Telotaxis is found when animal has a choice between the positive and negative stimuli or when the animal does not have a balanced input on the two receptors. Orientation is effected by fixing the image on one side by moving the head and making a choice. Honey bee seeing two light sources flies to any one by making a choice.

Menotaxis involves maintaining a constant angle in relation to the source of stimulus. Nocturnal moths have a habit of flying by keeping the light source (usually stars and moon) at right angle to the body so that they can fly parallel to the ground. But when they do the same with artificial light that is to closer, they are forced to fly in circles. Honey bees fly from their hives to the flowers by maintaining a constant angle to the sun as revealed by the wagging dace of the scout bees. The angle to the sun is remembered by foraging bees while watching the dance on the vertical surface of the comb. Foraging bees then fly towards the food source maintaining the same angle to the sun.

Mnemotaxis was first described by Kuhn (1919). This is orientation based on memory that was studied by Niko Tinbergen (1951) with his experiment on digger wasp. Wasp circles around the nest and carries a memory map of the nest and its surroundings, which helps it to accurately orient itself and return to the nest. This is also called zonal orientation and geographical orientation which involves distance, direction and landmarks that make topography of the area and help the animal in homing to its nest.

NAVIGATION & HOMING

Migratory animals which cover long distances either to reproduce or to escape from the harsh climate must find their way accurately over oceans, deserts, forest and mountains. Fishes, birds and many invertebrates possess extraordinary capabilities to cross oceans, deserts and mountains in order to reach their destination.

Invertebrates such as crustaceans, amphipods, ants, bees and wasps possess strong homing and navigational instinct and are guided by the sun, moon, stars and topography of the area in following accurate route. Monarch butterflies migrate thousands of kilometers from Canada to Mexico to escape harsh winter and return back accurately to the same place.

FISH NAVIGATION

How fishes find their way in huge expanses of sea and reach their destinations which lie thousands of kilometers away has been a mystery. It is believed that they orient by the positions of stars and moon in the night sky and sun in daytime to find the direction of swimming.

They also make use of temperature gradients and ocean currents which help them in swimming and also in navigation. However, it has been experimentally proven by A.S. Hasler that salmons are guided by the odour of their parent stream during return journey.

Odour map gets imprinted in their brains when they migrate as larvae from tributaries to the sea and they can navigate back from the sea using this odour map when they become adults. Eels can also migrate to Sargasso Sea using similar odour maps but how their larvae, *leptocephali* find their way back to the river mouths, crossing vast stretches of Atlantic Sea is a mystery. Probably their parents leave some kind of odour trails during their journey.

NAVIGATION IN BIRDS

Birds use a number of methods to find their way during migration. Many use celestial navigation, a method of orienting the body to the arc of the sun, to the phases of the moon, or to the pattern of stars in a particular season, which is called menotaxis. Hummingbirds and pigeons are able to determine the position of the sun even on overcast days because they can detect the ultraviolet radiation it emits.

Experiments in planetarium on night migrant birds, such as white throated warblers and indigo buntings reveal that they orient themselves by the position of stars in the night sky.

Some birds are sensitive to coriolis force that arises by deflection of winds in the northern hemisphere by earth's rotation.

Some diurnal birds use topographical landmarks such as mountains, river valleys, and forests to orient themselves on the migration route. Some are able to detect infrasound or low-frequency sounds that are produced by ocean waves. Many birds, particularly seabirds, identify their destinations by characteristic odours.

Many birds possess instinct or some kind of internal compass or biological clock that guide them through the route of migration. Young birds follow the migration route accurately without previous training or experience by their inherent capacity to navigate.

Some birds such as oil birds of South America and Himalayan cave swift possess echolocation and can be guided by it.

The classic experiment proving the internal-clock theory was done by German Gustav Kramer during the early 1950's. He placed Starlings wanting to migrate in a cage from which they could see the sun. The birds would sit looking in the direction toward which they wanted to fly. Significantly, if the Starlings couldn't see the sun, they didn't face in any particular direction.

Also during the 1950's, the German Franz and Eleonore Sauer did a similar experiment with birds that could and could not see the night stars and arrived at similar results. Certain species can orient themselves according to the sky's major stars. In fact, an experiment with Mallard Ducks found that if the moon is so bright that important stars are hidden by glare, released ducks can't orient themselves as well as on darker, moonless nights.

Some birds, such as pigeons, are sensitive to changes in the earth's magnetic field because of the presence of magnetite in their head and neck muscles. During early 1970's, W.T. Keeton tied small, bar magnets on the backs of pigeons. When released at locations the birds had never seen before, the pigeons with non-magnetic bars found their ways home but those with bar magnets got confused.

In a 2007 German scientists found tiny iron oxide crystals in the skin lining of the upper beak of pigeons, which might be of help to the birds to sense the earth's magnetic field and assist them to identify their geographical position.

The researchers also discovered cryptochromes, which change their chemistry in the presence of a magnetic field, in the retinas of migratory birds' eyes. The molecules might then affect light-sensing cells in the retina to create images due to magnetic field and help the bird to navigate during flight.

Infrasound travels much farther than ordinary sound and it comes from many different natural sources, including ocean waves, surf, winds, storms, earthquakes and other geologic events. It is believed that birds can hear infrasounds that we cannot hear and hence they possess this accessory navigational capability.

COURTSHIP BEHAVIOUR AND ANIMALS

Courtship is a social behaviour in which there is an interaction between the male and female members of a species leading to mating and reproduction. Courtship evolved due to the fact that very large number of sperms is produced which must search and fertilise few ova leading to competition among sperms.

Since males possess sperms, they must compete with one another in order to win over the female to fertilise her ova which are a limited resource. The gametic selection has translated into sexual selection among males and females, leading to male-male competition and female choice. Courtship display is an extension of this male-male competition in which males evolved various devices and techniques to persuade female to reproduce.

IN DROSOPHILA

Courtship behaviour in vinegar fly *(Drosophila)* was described by Bastock & Manning (1968). Male and female come together within 2 mm of each other and then male circles around her. Female is discriminatory and wrongly approaching males are kicked off. Male vibrates one wing during circling which stimulates the female.

Vibration of the wing produces sound as well as air current which act on the antenna of female. This is followed by touching with front tarsi and genitalia licking. Mating occurs after about 3 minutes by male mounting the female. Often mounting occurs but mating is unsuccessful. Courtship of wingless male is not accepted by female.

IN 3-SPINED STICKLEBACK FISH

Three-spined stickleback fish *(Gasterosteus aculeatus)* is found in ponds and rivers of Europe. Male is bluish-black in colour with bright red belly while female is silvery in colour. Male finds a place in sandy bottom where there are weeds. Male builds a tunnel-like nest in sand among weeds and defends territory around the nest. Then male swims near the surface over the nest to invite females.

Other males are attacked and chased away aggressively. Male swims upward from below and stabs the female from below with his dorsal spine. When response of female is positive both of them swim in zig-zag fashion towards the nest. If female likes the nest it enters inside and male follows.

Male places his head against the tail fin of female and quivers, which provokes the female to release eggs. Male then deposits his sperms over the eggs and female is chased away. Male then swims again to the surface to solicit another female. Up to 5 females can be made to lay eggs in his nest by the male. Male then guards the eggs and oxygenates them by fanning with fins till they hatch.

IN BIRDS

Courtship behaviour has evolved in birds to the highest level in which auditory as well as visual displays are used by males to impress females. Shape, size and colour of feathers have evolved for displaying and dancing.

Singing generally has evolved in male birds living in dense forests where there is limitation of visual distance but sound can travel to long distances. For example, males of cuckoos, starling, lapwings, larks, grackles, nightingales and bulbuls are accomplished singers and use these auditory stimuli to attract females.

Some birds imitate other animals to impress females, e.g. grackles, parakeets, starlings, magpies and shrikes. Lyre bird of Australia is a celebrated mimic whose male can imitate the sounds of mobile phones, alarm clocks, tweets of reversing vehicles and bike engines only to impress female of his extraordinary capabilities.

Nest building is also used as a means of visual stimulus to attract female. In weaver birds and bower birds male builds a nest and invites females to inspect it. If female likes the nest mating occurs. In the case of oropendolas *(Zarhynchus)* it is the female that builds the nest and invites males into it.

Feather display by male is a common phenomenon in birds' courtship, e.g. birds of paradise, peacock, pheasants, grouse etc. in which length and brilliance of feathers is the deciding factor to attract female as well as to warn other intruding males.

Dancing is also a stimulus used by males to woo females in a large number of species of birds. Dancing and cooing in pigeons and doves is a courtship behaviour. Peacocks and birds of paradise males not only display their feathers but also dance and show different tactics to attract females.

Aerial displays in flight and aerobatics have been recorded in pigeons, kites, buzzards and doves. In buzzard (Buteo) male and female hook their claws and fly in circles before mating. Lek birds such as grouse clear an area of weeds in the forest where all males and females of the area gather. All males dance and display their feathers in this mating arena, while the females passively watch the proceedings. Mating takes place after several hours of dancing.

In the case of crested grebe *(Podiceps cristatus)* both male and female come together and do head-shaking ceremony after which both carry out diving displays. Then both rise vertically to the surface of water and do penguin dance after which nest material is exchanged. Mating takes place after a considerably long courtship display.

Jackson's whydah *(Drepanoplectes jacksoni)* male prepares a display arena by clearing grasses and then dances around the central grass tuft and jumps into air frequently while the female watches. Mating takes place but female raises her family alone without any help from male.

Lesser florican jumps above the tall grasses and floats down with outstretched wings and tail, loudly calling all the time. Females are attracted by this display.

SOCIAL LIFE IN PRMATES

Primates were not social animals when they evolved from the primitive insectivore ancestor in Palaeocene epoch. However, gradually primates became gregarious and social interactions developed in them leading to highly developed social life as in humans.

SOLITARY PROSIMIANS

Prosimians such as tarsiers, bush babies and lorises are mostly nocturnal and highly arboreal primates. Males are found solitary or in pairs with females in breeding. Females live with infants till they become independent. Prosimians are shy animals and hiding in foliage is their means of defence. They rarely come down from trees. The black coloured Aye-aye is nocturnal and lives singly or in pairs.

MONOGAMOUS PROSIMIANS

Tree shrews *(Tupaia* species) are the most primitive primates alive today and live singly or in pairs in the forests of S.E. Asia. As they are highly territorial and defend their territory with aggression, their social units are broken up into nuclear families, e.g. male, female and juveniles. Males mark their territory by urine and defend it by threat-call and tail-flicking. Intruders are quickly attacked and chased away.

Lemurs occur in the tropical rain forests of Madagascar. They are also monogamous and live in a group male, female and up to 4 young ones. Male and female marry for life and live together. Some species mark territory.

Tamarins are South American monkeys which are also monogamous and live in nuclear families of male, female and juveniles.

SINGLE MALE BISEXUAL GROUPS

Some monkeys, e.g. hanuman langur, howler monkey, red-tailed monkey and blue monkey, live in a social group in which there is a single dominant male having a harem of several females. Young males are chased away by this dominant male and hence they form all male groups outside the other groups. Harem is protected by the overlord male but is constantly attacked by males from the all male groups to unseat him from the dominant position.

MULTIMALE BISEXUAL GROUPS

Baboons *(Papio)* are terrestrial primates which are found in large groups that may include thousands of individuals. There are several units in which one male and several females with young ones form a small group. These small units forage together. Male protects the group and herds females together and prevents them from meeting other males. Hierarchy is maintained in females for access to male. These small units together form large bands that live together, sleep together and defend them collectively from predators. All males collect together to attack a predator which is usually a leopard.

Rhesus monkeys also form multimale bisexual groups that form large foraging units. One male is dominant and others subordinate. A foraging unit is formed by 3-8 males, each having 5-7 bonded females. Many units form large groups of hundreds of individuals for foraging and for defending.

SOLITARY APES

Orang-Utan lives in the dense forest of Sumatra and Borneo and is completely arboreal, feeding on a diet of fruits and leaves. Males are solitary, seeking females only for mating and shares no family responsibilities. Females are found with the young ones, usually only one young is found with female. For sleeping on the tree female makes a nest with branches and leaves in the fork of tree.

MONOGAMOUS APES

The lesser apes, white handed gibbon and hoolock gibbon are found in the dense forests of eastern India, China and Burma. They are highly arboreal and swing under the branches with the help of excessively long arms. Male, female and up to 4 young are found in one family unit. The units communicate with one another by loud hooting calls. Both male and female share family responsibility together.

MULTIMALE BISEXUAL APES

Gorilla is the largest ape living in the dense forests of Cameroon, Gabon, Congo and Uganda. Males are terrestrial as they are too heavy to be arboreal. Females and young make nests among branches on trees for sleeping. They are found in groups of about 20 individuals. Old dominant males are called "silver backs" which dominate over other males and females. Hierarchy is observed among males as well as females while feeding, drinking or access to females.

DIFFUSED SOCIAL APES

Chimpanzees form diffused social groups of up to 50 individuals. Females are silent and shy. There is hierarchical ranking among males as well as females but females accept several males and there is no conflict. They are omnivorous and sometimes hunt monkeys and share its meat. They also make nests in the fork of trees for sleeping. Both males and females defend their group by screaming, gesturing and by throwing sticks and stones at the intruder.

CRYPSIS

In nature predator and prey have evolved together for millions of years. Prey must deceive the predator in order to escape getting killed, while the predator must also use deception to catch the prey unawares. Crypsis is also a form of mimicry but the former has a wider meaning that includes mimicking even non-living objects such as stones, rocks, twigs and even the background.

Protective colouration: Majority of the animals match the background in colour to escape the attention of the predator. For example, hares and rabbits are earth-coloured, grasshoppers are green and beach crabs have the same colour as pebbles.

Counter-shading: This is also called natural shading, in which the protectively coloured animals have darker shade on the dorsal side and lighter on the ventral side of body. This is to neutralise the sunlight falling from above, which lightens the upper side of the body, while the shade below darkens the colour. Some animals press their bodies against the ground and remain motionless when in danger in order to eliminate the shadow. Flatfish, cuttlefish and chameleon can even change the body colour to neutralise the effect of light and shade. Squid *(Abraliopsis)* possesses light-producing organs on the ventral side, which can be switched on when there is sunlight from above and they become invisible from below.

Disruptive coloration: Another method to enhance the effect of natural shading is obliteration of the outline of body by having spots, patches or stripes, which break the continuity of the outline and thus help the animal to camouflage much more effectively. For example, salamanders, deers, leopards, tigers, fishes, rays all have patches on the body.

Protective resemblance: Some animals resemble their habitat not only in colour but also in structure, so that the camouflage is complete and they become completely inconspicuous in their environment. Caterpillars of the family Geometridae resemble the twig of the plant on which they feed in colour as well as shape. Similarly, sea horse possesses thread-like processes emanating from body, which make

it invisible among seaweeds where it lives. Stick insects of the order Phasmida are found on grasses and have thin and elongated body and appendages, which make them look like strands of grasses.

Indian Leaf insect which feeds on broad-leaved plants in eastern Himalayas, has the entire body flattened and wings and legs shaped like leaves. The green, leaf-like insect is impossible to detect on green plants even by trained entomologists.

Aggressive resemblance: Predators also use crypsis so as to approach the prey undetected to mount a sudden attack. Many predators camouflage to ambush an unsuspecting prey that may wander near them. Tigers have stripes that help them to hide in tall grasses. Slow moving predators like praying mantis and ambush bug have the same colour and shape as the plant on which they sit and attack by surprise. Crab spiders are known to resemble flowers on which they wait for the prey, not only in colour but also in appearance.

The American hawk *(Buteo albonotata)* resembles vultures in general appearance. Since vultures are scavengers, birds are not scared of them and venture close to them, when the hawk in the garb of vultures attacks them.

Cryptic actions: Feigning death or injury is sometimes adopted as a measure to distract the attention of predator. Many beetles curl up like pebbles and remain motionless to escape attention of the predator. South American opossum also feigns death when alarmed. Echidnas, hedgehogs and porcupines also curl up when alarmed and feign death. Their body spines become erect as defence structures. Some birds, which make their nests on the ground, e.g. ostriches, lapwings and prancticoles feign injury on the wing or leg and struggle to run or fly to distract the attention of the predator from its nests.

Dymantism: Some butterflies, namely, pansies, *colites* and *Melanitis,* expose bright colours of wings when they suddenly fly, which temporarily frightens the predator and gives them time to escape. Some birds also expose flashes of bright plumage when they take to flight. Emperor moth is nocturnal and rests in shady places in daytime. Its wings possess large eyespots, which give a false impression of a hiding cat, which is enough for birds to avoid them. The butterfly, *Nymphalis,* has two pairs of eyespots on wings, which are revealed by lowering its wings when it is attacked by a predator.

Aposematism: Majority of protected species sport bright colours on the body to advertise their presence, so that the predators are warned not to attack them. The bright colours help in quick learning by the predators. For example, wasps that are protected by a sting are brightly coloured. Unpalatable butterflies, such as *Danaus,* are brightly coloured and are carefully avoided by the birds.

H.A. Ford demonstrated aposematism by conducting an experiment in which he used red and blue artificial caterpillars. Birds avoided the red ones.

Cryptic organs to misdirect attack: Swallowtail butterflies have long brightly coloured tail on the hind wing while the rest of the body is dull coloured. When the predators attack, they tend to catch the brightly coloured tail while the vital parts of the body escape damage. The small lantern butterfly *(Thecla)* has posterior extension of hind wing resembling head, while the actual head is small and inconspicuous. Bird attack is always misdirected on the false head.

DRIVE

The term Drive was introduced by Woodworth (1918) as motivational concept. Animals experienced drive as biological needs such as eating and drinking and alteration in their behaviour. Drive theories were later given by Sigmund Freud (1915) and Clark Hull (1943). Freud, who was physiologist by

training, believed that drives and urges such as hunger were recurring conditions in the body of animal that produced energy build up in the nervous system.

This energy build up caused psychological discomfort and restlessness that kept on increasing unless the urge was satisfied. Drive arose from a range of bodily disturbances, such as deprivation of food, water, air, sleep or temperature regulation, injury or activities like nest building. Freudian drive theory was based on the following three principles:

- Drive emerged from bodily needs of the animal.
- Drive energized the behaviour of animal due to restlessness.
- Reduction of drive by satisfying needs produced learning.

Konrad Lorenz (1950) proposed the **Psycho-hydraulic model** or **Flush toilet model** to explain the drive and consummatory behaviour, which has three steps:

1. Drive causes action specific energy to accumulate with time and causes increased restlessness in the animal, which results in searching behaviour for food, water, mate etc.
2. Consummatory behaviour starts after achieving the goal such as food or any other sign stimuli. The innate release mechanism releases the accumulated energy in the animal.
3. After consummatory behaviour there is a quiescent period in the animal as the accumulated energy has been released and the action stoped. This is called refractory behaviour of the animal.

HUNGER & THIRST DRIVE

Hunger drive is controlled by lateral hypothalamus and ventro-median nucleus, the former is stimulatory in function while the latter is inhibitory. Glucocorticoids inhibit the hunger drive. Lateral hypothalamus can be stimulated by epinephrine. The hunger and thirst drives depend on hours of deprivation of feeding on dry food.

HOARDING DRIVE

Many mammals such as male gerbil and squirrels possess hoarding drive as the lean season approaches. Low estrogens and testosterone levels stimulate hoarding drive in mammals. Castrated individuals show increased hoarding drive, which can be reduced by giving testosterone treatment.

MIGRATORY DRIVE

Migratory drive occurs in fishes and birds and may be seasonal or related to reproduction. Pineal glands, which is affected by day-light hours, affects migration in birds. In warblers pituitary gland influences migratory urge as well as excessive eating to deposit fat energy in the body. In stickleback fish thyroxin injection caused them to migrate. In *Salmon* and *Anguilla*, maturation of gonads produces migratory drive, so much so that they stop eating and set out to the course of migration crossing all obstacles on the way.

AGGRESSION DRIVE

Aggression is controlled by amygdala of the limbic system of brain and posterior hypothalamus is also involved to some extent. In most of the male mammals testosterone causes aggression while in females high oestrogen levels reduce aggression and make the female peaceful. Hydrocortisone also

increases aggression while hydroxydione decreases it. In ringed dove implants of testosterone propionate at specific sites of hypothalamus causes aggressiveness.

TERRITORIAL DRIVE

Many vertebrates mark and defend their territory. Dogs, hyenas and some prosimians mark their territory by their own urine. Monotremes and marsupials have anal glands which they rub on the ground to mark territory. In tigers and cheetahs also there are anal glands which spray the secretion on the trees to mark their territory. Gazelles possess orbital glands below the eyes which secrete a tar-like substance that they apply on grasses and bushes. Territorial behaviour is also hormone dependent. Yahr & Thiessen (1972) isolated 11 different hormones that influence territorial behaviour in vertebrates.

HORMONES IN SEXUAL DRIVE

Sexual drive involves courtship behaviour such as singing and dancing in birds, croaking in frogs and fighting in males of many vertebrates. In insects courtship behaviour is stopped if corpora allata are removed.

Hormonal levels increase in breeding season. Castrated males and females do not show sexual behaviour in vertebrates while testosterone injections elicit sexual behaviour. According to Johnson (1976) oestrogen enhances female attractiveness and receptivity and causes oestrous in females.

Hypothalamic Releasing Factor (LH-RF) and ACTH are known to affect copulatory behaviour in many animals.

PARENTAL CARE DRIVE

Gonadotropin secretion by pituitary gland cause not only courtship display but also parental care in birds. Progesteron injections made the birds sit on the eggs to incubate within 20 minutes.

In pigeons, secretion of prolactin from pituitary causes enlargement of crop to produce pigeon-milk which is fed to the chicks. Prolactin also acts directly on brain and makes the preoptic nucleus of hypothalamus in birds to respond to chicks calls.

LEARNING

Learning is the ability of the individual to remember and change one's behaviour in response to earlier experiences. Animals learn a great deal from their surroundings and also from their experiences, particularly during the growing period. Latent learning provides animals with knowledge about their surroundings and escape routes, and also areas where food and water is available. Niko Timbergen (1951) demonstrated by experiments on digger wasp *(Philanthus)* that it could remember its nesting site by landmarks and got confused when landmarks were changes.

W.H. Thorpe (1951) defines learning as an internal change in the animal causing adaptive changes in the behaviour as a result of experience.

N.E. Miller (1967) called learning a permanent tendency for a stimulus to elicit a response that can be reversed by training.

S.A. Barnett (1981) defined learning as any adaptive change in behaviour as a consequence of experience of repeated stimuli.

There are many types of learning in the animals as given below.

INSTINCT

Instinct is the innate behaviour of the animal which is a heritable characteristic. It is the inborn capacity of the animal to perform certain functions. This is also called species memory as it is learned by all members of the species through many generations.

Instinct is build up in the nervous system that controls and modifies the behaviour of the animal and it takes long time to be able to get executed.

Instinct is advantageous to animals that have short life span and no time to learn and no time for parents to teach the juveniles. Sometimes there is no parental care and juveniles are left on their own. Such animals must carry out their activities through instinct in order to survive.

For example, invertebrates in general perform complex activities through instinct. Nectar collecting and hive building by honey bees are done by instinct. Migration of many vertebrates such as sea turtles is by instinct. The catadromous eels and anadromous salmons migrate thousands of kilometres for spawning by instinct.

IMPRINTING

Imprinting is strongly controlled by genes. Young chicks must learn from parents' activities such as singing, nest building or following their mother. There are three types of imprinting:

Filial imprinting involves activities that are learned by the young ones from their parents in early stages of life. Hunting by cheetahs, leopards and tigers is learned from parents in younger age.

Sexual imprinting is the recognition of the opposite sex in the adult stage. Courtship display, such as singing, feather display and dancing must be learned during the growing stage by watching the adults. Peter Marler of the University of California by experiment demonstrated that chicks reared in isolation failed to develop courtship songs which they tried in adult stage. The intricate nest building by weaver birds must be learned by the individuals in the younger stage by watching the older individuals.

Social imprinting happens during the brief but sensitive early period of life but it has great stability and influences the behaviour of the animals towards the others for the rest of life. Adult behaviour is the result of learning during the early stages of life.

HABITUATION

Habituation is the decrease of responsiveness upon repeated exposure to a stimulus. This is stimulus-specific decline in the response resulting from repeated stimulation. For instance, animals in urban areas are not alarmed by the loud sound of vehicular traffic as they get accustomed to it, while their wild counterparts get extremely agitated by the same stimulus.

Habituation is not fatigue but learning not to respond to stimulus that is neither rewarding nor punitive to the animal. Habituation saves time and energy of the individual for more important activities.

SENSITIZATION

This is increased responsiveness to a repeated stimulus. Sensitization is opposite of habituation. Here the stimulus irritates or annoys the animal and hence the animal shows increased responsiveness each time the stimulus is applied. For example, if frog's skin is touched with a needle, it will wipe the area with its hind legs. This response will keep on increasing in intensity every time the needle is touched on body.

CONDITIONING

Conditioning is flexible learning in which a stimulus elicits a specific response from the animal. I.P. Pavlov (1927) conducted experiments with a dog, in which dog was given food after the gong of a bell. The dog got conditioned to the gong of bell and food and salivated immediately.

Later, the dog salivated at the gong of the bell even if no food was presented to it. If animals are presented with a choice of two or more stimuli, the animal gets conditioned to chose the best option. Birds get conditioned to choose the edible butterflies from the unpleasant ones.

SIGN STIMULI

Sign stimuli, also called **releasers** or **key stimuli,** are those stimuli that are capable of releasing Fixed Action Pattern (FAP) or consummatory behaviour of the animal. They are signals that evoke instinctive patterns of behaviour in animals, such as fighting behaviour in the territorial animals, triggered by the entry of another male. Lehrman found that courting male dove began to bow and coo to a stuffed model of female in the absence of a living female.

Konrad Lorenz (1972) was the first biologist to identify sign stimuli which he called **key stimuli** because they function as keys to release and unlock the fixed action pattern of the animal. He proposed the Innate Release Mechanism in response to sign stimuli.

Niko Tingbergen (1952) conducted experiments on stickleback fish in which bright red colour of the belly and neck is a sign stimulus for the other males to attack it, while it elicits attraction in female fish.

Bird chicks respond to jerks in the nest, which is a sign stimulus for them to open their beak for eating food. Similarly distress calls given by chicks are a sign stimuli for hens to release rescuing behaviour.

Bright red colour of the oral cavity of a cuckoo chick is a sign stimulus for the foster parent, warbler to feed it. Otherwise the chick is distinctly different from the foster parents.

Sign stimulus is often not one character but a combination of many stimuli, namely, shape of the bill, colour and patches on body or even actions and auditory signals.

Sign stimuli or releasers can be of three types:

Visual Releasers. They are morphological characters that are displayed to elicit response, as display of feathers or dancing in birds. Nest making behaviour in birds such as weaver bird, not only attracts females but also repels other males. Fire flies emit light signals that bring about response from females. In three-spined stickleback fish *(Gasterosteus)* males get nuptial coloration during breeding season in which neck and belly become bright red in colour to attract females and also to warn other males not to enter the territory.

Auditory Releasers. The sign stimulus is auditory signal like humming sound of male insects. Song birds such as cuckoos, nightingales, starlings establish their territory and attract the female for mating by singing, often for several days. The song is a signature stimulus of individual bird as it is specific. Birds of different areas sing in different dialect and elicit different response.

Stridulatory organs of some insects, such as cicadas and crickets are strong sign stimuli during breeding season.

Co-qui calls given by the tree frog, *Eleutherodactylus* are strong auditory releaser for males as well as female frogs of the same species.

Chemical Releasers. Pheromones are different types of volatile chemicals released in the atmosphere that act as releasers on the individuals of the same species or other species. Pheromones affect the individuals of the same species while allomones affect other species.

Sex pheromones in insects are produced by the females and perceived by the enlarged antennae of males of the same species, sometimes from the distance of a couple of kilometres.

In vertebrates, the sex pheromone, **Copulin** is secreted by the female in estrus.

Alarm pheromones are produced by skin cells of several species of fishes that have schooling or shoaling behaviour. Alarm pheromones are also released by the stinging honey bees and wasps which attract other individuals of hive to attack.

Ants and termites produce trail pheromones from the posterior end of abdomen which help them to follow each other.

Sign stimuli bring about responsiveness in target individuals which show consummatory behaviour. The responsiveness diminishes as the consumption proceeds and energy is released.

SENSORY FILTERING

Animals receive much more sensory information than they could possibly register in their brain and respond to. Therefore brain has to be selective and filter out certain information that is not so necessary. Sensory filtering or stimulus filtering takes place at several levels, namely, at the level of sense organs, nerves or different parts of brain.

Sensation is the basic data sent by sense organ to brain, and sense organs have their limitation and hence filter out much of the information. For example, human eye filters out ultraviolet and infrared rays from the spectrum.

Peripheral filtering is done by receptors because of their mechanical ability to receive and transmit information. Receptors are often highly specialized and respond to a narrow range of stimulus. For example caloreceptors can perceive sense of heat but not cold and frigidireceptors can only transmit the sense of cold. Bats can perceive ultrasonic sounds for echolocation but sense organs of other mammals do not possess that ability.

CNS filtering is done by different parts of brain by selective attention or because the part is not well developed. Perception is the interpretation by brain of sensory information in the light of earlier experience. A lot of information is received by brain but is not perceived.

Reticular Activating System located inside the medulla oblongata if inactivated, stops lots of nerve impulses coming through the cranial nerves.

Stimuli that reach respective areas of brain such as optic lobes, auditory lobes etc. can get filtered out if not important. Epithalamus, which functions as the central switch board, selects and sends only necessary nerve impulses to cerebral hemispheres.

When the nerve impulses arrive in different areas of cerebral cortex, they are analysed and interpreted and if found worthless can be rejected without perception.

Only information that is considered important is selected by the areas of cerebral cortex and interpreted, and motor action travels through the nerves to muscles to act. Neurochemical information coming from cerebral cortex affects hypothalamus, which stimulates endocrine system to alter the behaviour of the animal.

Muller's Law of specific nerve energies: Sensation perceived depends on the part of nervous system activated, and not on the sense organ stimulated.

Examples: The male of the South American tree frog *(Eleutherodactylus coqui)* produces ***co-qui*** call to attract female and also to repel other competing males. The tympanic membranes of male and female are adapted differently. Males can hear only the ***co*** note and get warned and repelled. Males cannot hear the **qui** part of the call, while females hear only the **qui** part of the call and therefore get attracted to males.

Olfactory cells located on the antennae of male moths (Lepidoptera) can perceive only specific pheromones which are released by the female of the same species. These pheromones cannot be perceived by the receptors of males of another species.

European Robin *(Erithacus rubicula)* attack red-breasted male robins. Only red feathers of the competitors are perceived and the other colours are filtered out for attack and for defending the territory.

BIOLOGICAL RHYTHMS

Biological rhythms are self –sustaining natural cycles of animal life history which maintain themselves regardless of the environmental factors. All animals possess innate biological clocks which are driven by the biochemical mechanisms. Erwin Bunning (1936) was the first biologist to carry out extensive work on biological rhythms.

CIRCANNUAL RHYTHMS

They show one-year periodicity, e.g. a large number of animals reproduce once in a year. Flowering in plants also takes place once a year. Insects and amphibians follow a cycle of hibernation and activity. Hummingbirds in South America move to the caves and become inactive in winter in Andes. Famous migration of Monarch butterflies from North America to Mexico and back follows annual cycle. Millions of these butterflies cover a distance of 3200 km to hibernate on trees in San Francisco. Many beetle species hibernate under the snow in Himalaya. Arctic and Antarctic animals generally follow annual cycles of activity.

CIRCALUNAR RHYTHMS

These rhythms synchronise with the 28 day phases of moon and tidal rhythms. Palolo worm lives in deep sea but swims to surface on the first day of the first quarter of moon in November in Fiji. The sea hare *(Aplysia)* shows periodicity which is exactly half of the lunar cycle.

TIDAL RHYTHMS

They are synchronised with the periodic rise and fall in sea level due to gravitational pull of sun and moon and centrifugal force of the earth. There are daily tides due to earth's rotation on its axis. Spring tides cause maximum rise and fall in sea level because moon and sun are on the same side of earth. Neap tides occur when sun and moon are on opposite sides of earth at full moon stage.

Circasyzygic Rhythms

They follow fortnightly cycle of 14.7 days of high tide after new moon or full moon. Molluscs exhibit egg laying behaviour according to this periodicity. Periwinkle also comes out of burrows on sea shores during high tides.

Circatidal rhythms

These follow 12.4 or 24.8 hour cycle that is synchronised with low and high tides twice a day. Animals living in burrows, such as polychaetes, planarians, crab etc. are submerged and exposed alternately and in the process get food brought by water currents. Bivalves such as *Mytilus* showed shell opening rhythm according to circatidal rhythms even when kept in the lab. Grunion fish spawns precisely at high tides.

CIRCADIAN RHYTHMS

These rhythms follow 24 hour cycle of activity and sleeping synchronised with light and darkness. So, the animals can be classified as nocturnal, diurnal and crepuscular, the last ones are active at sunrise and sunset. Birds are mostly diurnal and bats nocturnal which find their way by echolocation. Body metabolism and release of hormones are synchronised with 24 hour cycle.

Honey bees are known to have time memory. In experiments, honeybees after 5 hours of freezing came to food 5 hours late. Human beings experience jet lag when their circadian rhythm is disturbed while travelling in aeroplanes.

Larvae of *Wuchereria bancrofti* move to peripheral blood in the night but go to deeper blood vessels in daytime, which is synchronised with the blood-sucking habit of *Culex* mosquito.

Brady (1969) thought that optic lobes play an important role in controlling circadian rhythms in cockroach. Corpora allata and corpora cardiaca also release hormones that control day-night cycle. Cyclic AMP and serotonin are involved in biochemical events that control circadian oscillations.

In vertebrates, neural connections exist between retina and hypothalamus and pacemaker may be located in ventromedian nucleus of hypothalamus. In amniotes, the pineal and parietal bodies regulate photoperiodism. Melatonin secreted by pineal gland has anti-gonadotropic effect. Turtles synthesize serotonin during day and melatonin at night. However, this cycle disappears during hibernation.

DESIGINING AN EXPERIMENT

An experiment imposes an action treatment on a group of objects for observing the reaction. Because the validity of an experiment is directly affected by planning and execution, experimental design is extremely crucial for the validity of any experiment.

In experiments the researcher directs an action on to experimental units. These experimental units can be objects like any things or even subjects like humans or any organisms. For example, a population of certain illness is divided into few groups. Each group is 'treated' with a different dose of a medicine to checkout which dose is effective.

Biostatisticians study and analyse data generated from experiments. They have to plan their experiments properly to ensure that the right type of data is available to answer the questions. Moreover the data must be sufficient enough to be clear and efficient. This process of planning of the experiment is called experimental design.

Before carrying out the experiment, the researched must have a clear understanding of the data which he needs to collect and the end result which must be achieved. Also the researcher must be aware of the possible variations in the result. Thus, designing the experiment is very important in order to improve the precision of answers.

History of development of Designing of experiments

James Lind performed well-organized experiments in 1747 and he developed a cure for the disease scurvy. He was serving as a surgeon on a ship and he selected 12 men from the same ship. All these 12 men were suffering from scurvy. He then divided these men six groups of two each. For two weeks, he served various supplements to these people, along with their basic diet. These supplements were the treatments for scurvy as proposed by various people. He then compared the groups to find the efficacy.

The one thing missing out from his experiments was randomization. Randomization is very crucial in modern day experiments. Lind could get good and supportive data from his experimental design and he could find that one of the remedy worked very well.

Principles of experimental design

In 1935, Ronald A. Fisher, proposed a methodology for designing experiments, in his book "The Design of Experiments". Design of experiments is basically the process of planning, designing and analysing the experiment in such a way to effectively draw valid conclusions. Powerful statistical methods are very important to get statistically sound results. The three important principles of experimental design are randomization, replication and blocking. These principles play a very crucial role in decreasing or removing experimental bias.

Randomization: A Randomized experimental design assigns the randomly to an experimental group. As it is very difficult to reduce bias just be expert judgment, randomization is a common practice. With the help of randomization most consistent and trustful treatment groups can be created. Sometimes it is difficult to completely randomize an experiment, due to lack of time and cost.

Replication: Replication is the process of repeatedly running a part of the experiment or a part o it under varying conditions. Repetition has three main properties.

1. It allows the researcher to get more accurate estimate of the experimental error.
2. It can give the more accurate estimate of the interaction effect
3. It can help in decreasing the experimental error and increase precision.

Though randomization insures treatment of different experimental groups is as similar as possible, the results of a single experiment cannot be easily accepted. Randomly picking three objects from a group of five and experimenting on them may not be effective. To increase the effectiveness repetition of an experiment on a large group of objects is needed.

Blocking: Elimination of all the possible unnecessary deviations is very critical for any experiment. Arrangement of experimental units into blocks of similar entities is called as blocking. Observations collected under similar experimental conditions are grouped in the same block. Blocking reduces sources of deviations and thereby bringing more precision to the experimental design.

Steps for designing an experiment

Analysis of the design of experiments was built on the foundation of the Analysis Of Variance (ANOVA). The following are the steps for designing an experiment. Of the seven steps given below, step 1 and 2 are pre-experimental planning.

1. Recognition of the problem
2. Selection of response variable

3. Selection of factors, levels and ranges
4. Selection of experiment design
5. Performing the experiment
6. Analysis of the data statistically
7. Final conclusions and recommendations

Step 1: Recognition of the problem- Before designing the experiment it is very important to recognise and clearly define the problem based on evidences. A list prepared with specific problems to be dealt in the experiment can be of great help.

Step 2: Selection of response variable- A variable which can give useful information about the problem to be studied must be selected. Generally standard deviations and average deviations of measured characteristics are taken as response variable. These response variables should be measures before and after conducting the experiment.

Step 3: Selection of factors - The researcher should know about the factors which may influence the performance of the experiment. These factors which may have an effect on the experiment are called trouble factors or nuisance factors. These are the factors which the researcher does not want to deal with or wants to fluctuate in the experiment. There are also some other factors which the researcher wants to be constant which are called as design factors. The trouble factors are of three types namely controllable factors, uncontrollable factors and noise factors.

Step 4: Selection of experiment design- While selecting experiment design the researcher must consider inferences from statistics and sequential analysis. Based on these the experimental design must be randomized.

Step 5: Performing the experiment- When all the above steps are well planned, there is no doubt about the smooth flow of the experiment. Also the results and data obtained from the experiment must be properly recorded and analysed statistically.

Step 6: Statistical analysis of the data- To arrive at valid conclusions, the data recorded must be analysed statistically. Though non-statistical methods are also followed statistical methods are more reliable.

Step 7: Final conclusions and recommendations- Depending on the conclusions drawn from the statistical analysis of the experimental data further recommendations are made.

Applications of designing the experiment

- The main aim of the designing of the experiments was limited to agricultural needs. But the modern designing of the experiments has spread across various streams.
- Designing of experiments finds place in almost all the areas of science and engineering. Mainly the statistical analyses carried out are very helpful.
- In recent years, there has been a considerable utilization of designed experiments in many other Designing of experiments has also been used in service sectors areas like business, finance and government operations.