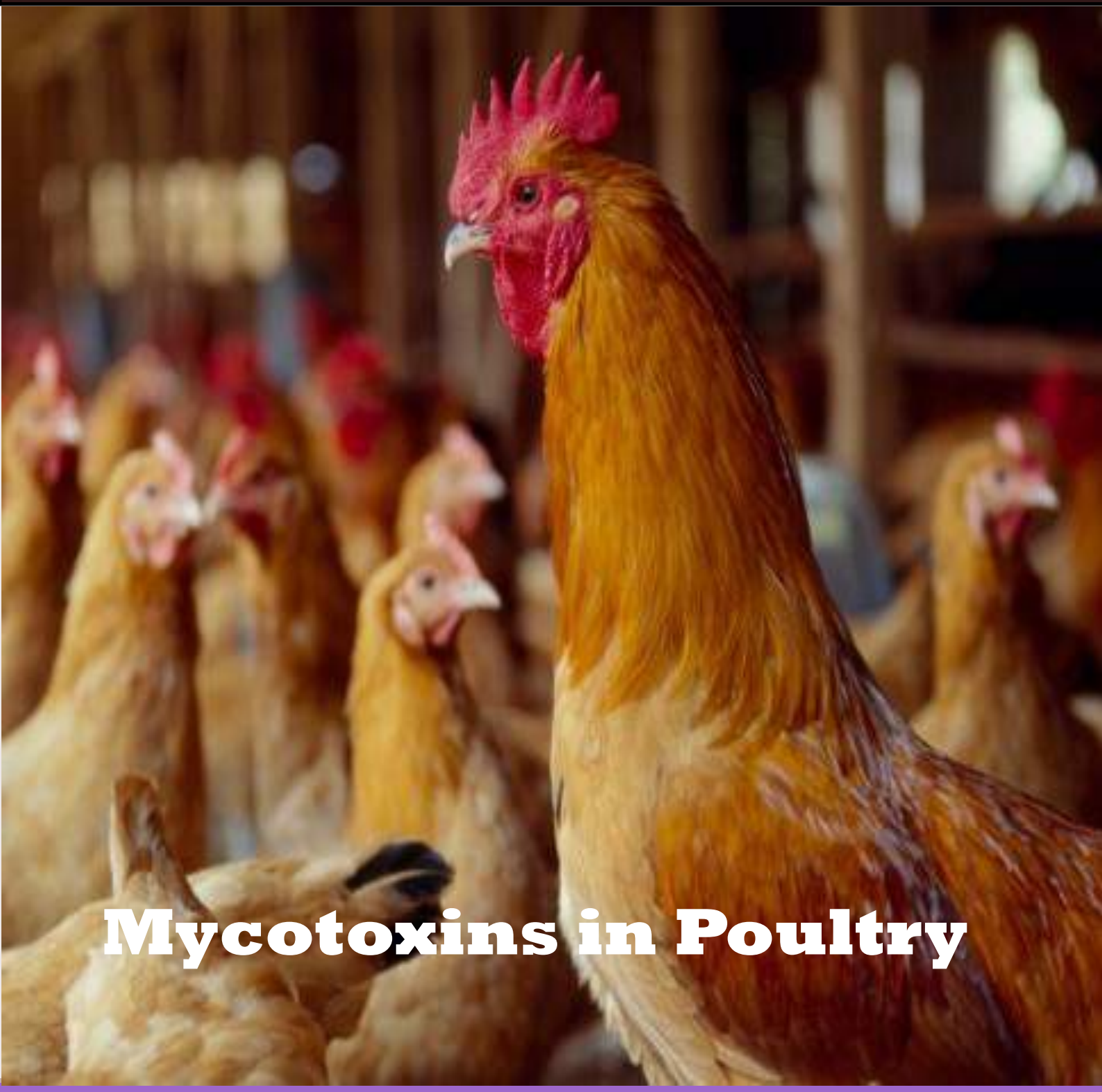
Indian Farmer

ISSN 2394-1227

A Monthly Magazine

Volume: 3 | Issue-4 | April- 2016 | Pages - 68



Mycotoxins in Poultry

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INDIAN FARMER

A Monthly Magazine

Volume: 3, Issue-4 April -2016

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Embryo Transfer Technology in Domestic Animals

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What is embryo transfer technology (ETT)?

It is a process by which an embryo is collected from a donor female and then transferred into a recipient female where the embryo completes its development. The expanding field of ETT can be viewed as the female counterpart of artificial insemination (AI). Females of superior genetic merit are superovulated with gonadotropic hormones (FSH, eCG), their eggs fertilized in *vivo* or oocytes fertilized in *vitro,* then the resultant embryos are transferred to recipients, genetically less distinguished surrogate mothers.With the help of ETT, a genetically superior female produces more offspring than she could by natural reproduction. The increased number of offspring thus maximizes the donor female's genetic abilities.

It is used in several species of domestic animals, namely cows, horses, goats and sheep and was first successfully accomplished by Walter Heape in 1890, started as a research tool and became a commercial enterprise in cattle in the early 1970s. Since then, the popularity of the procedure has increased among producers of different animal species. Breeding beef cattle interests offer large economic incentives to veterinarians and animal scientists to make this technology an efficient and effective tool. The commercial embryo transfer industry has grown steadily and will continue to do so as long as embryo transfer remains a key step in many of the developing technologies,such as prenatal sex selection.

What is the procedure for embryo transfer technology?

A. Selection of donor animal

As mentioned earlier, the donor selected for embryo transfer should have outstanding genotype. Donor cows should be selected considering the following criteria:

1. Animal should be between the age of 3-10 years
2. Should have regular heat cycles
3. Should be free genetic diseases and conformational abnormalities
4. Should not have history of reproductive difficulties
5. Should not have lesions in ovaries, cervix and uterus
6. Previous sound reproductive performance including not more than two inseminations per conception

B. Superovulation of the donor cow

"Superovulation" refers to the release of many oocytes (eggs) during a single estrus period. Superovulation of the donor can be achieved through treatment with gonadotropins. Most embryo transfer donors are treated with follicle stimulating hormone (FSH) to induce the maturation and ovulation of a larger than normal number of oocytes. Less labor is involved with use of superovulation procedures. Following embryo transfer, superovulated ova result in normal offspring with success rates similar to those achieved with normally ovulated ova.PMSG or eCG is used at the rate of 2500-3000IU and FSH is used at the rate of 20- 50 mg per donor.

C. Insemination of the donor

Because superovulation of the donor female causes the release of a large number of eggs over a 24-hour period, so it is better to inseminate the donor several times to achieve optimal fertilization.

D. Embryo collection from the donor

Success of embryo collection depends not only on the age of the embryos but also on the technical procedure used and skill level of the technician. Embryos can be collected by surgical and non-surgical methods.

1. Non-Surgical Method

Advances in technology have made embryo collection a non-surgical procedure with cattle, buffaloes and horses. A Foley's catheter (14-18G; 67cm long) with 30 ml inflatable balloon is fastened at the base of horn under epidural anesthesia. The catheter may be 2 or 3 way catheter. In 2 way catheter one is for inflation of balloon and other for inflowand outflow. The inflow is connected to a bottle of flushing medium (DPBS) and outflow is passed through embryo concentrator having pore size 70µ to filter out embryos. Each horn is flushed by 30-60ml of DPBS fluid 5-10 times to ensure all embryos are collected. This method for collection of embryos is desirable because all surgical techniques invariably lead to the formation of adhesions and there is less risk to the life and health of the donor.

2. Surgical method

This method is preferred most in sheep, goat and pigs. In this method the reproductive tract is exposed by mid-ventrally incision under general anesthesia. Flushing is carried out from oviduct by collecting embryos from Ostia and flushing media is introduced via utero-tubal junction. A volume of 2-20 ml flushing media is used to flush the oviducts. Unfertilized oocytes for specialized applications (such as *in vitro* fertilization) must be collected near the timeof ovulation.

E. Embryo evaluation

After collection, the embryos are evaluated for quality using a stereoscopic microscope. Embryos are evaluated for the quality and stage based on no.of blastomeres, size of blastocoels, compactness of blastomeres, color and appearance of cytoplasm and state of zona pellucida and perivitelline space. Embryos are graded on a scale from one(excellent) to four (poor).

F. Transfer of embryos to recipient

The recipient should be healthy, fertile, should be in good body condition and can be reasonably expected to deliver the transferred calf, trouble free at term.

Nutritional status should be good. The estrous cycle of donor and recipient needs to be essentially synchronized.Estrus synchronization is important to establish a uniform uterine environment for the embryo; therefore, only a one day or less difference in estrus between the donor and recipient is acceptable.Embryos that are perfectly symmetrical are having slight asymmetry, even granulation with no or slight blastomere extrusion is selected for transfer. Embryo is deposited at the middle of uterine horn ipsilateral to corpus luteum.

The survival rate of the embryo from implantation to term in the recipient female ranges from 55% to 70%.Recipient females are palpated within one to three months after embryo transfer to diagnose stage of pregnancy.

What are the advantages of embryo transfer technology?

1. Genetic conservation of endangered breeds or rear blood lines.
2. Genetic preselection and transfer of desired sex embryo.
3. Easy and hygienic transport of genetic material.
4. Fast genetic progress.
5. Creation of disease free herd.
6. Production of AI bulls from best proven bulls and cows available.
7. Production of twins.
8. Important for biotechnological procedures like production chimers, micromanipulation and transgenesis

What are the disadvantages of embryo transfer technology?

1. High costs of superovulation.
2. Time consuming.
3. Requires a higher level of management.
4. Specialized equipment and trained personnel.
5. Not all potential donors respond positively to treatment.

Calf Diarrhoea in Cattle

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It is rightly said that today's calf are tomorrow's high producing cow or breeding bull of high genetic merit. So proper care need to be given to calf health and management. Raising healthy calves with minimum mortality is crucial in successful dairying. Calf survival is an example of a trait that increasingly affects the revenue of modern dairy farms. Decreased calf survival causes huge economic loss to dairy farmers by means of loss of present value of the calf, higher replacement and veterinary cost and most importantly there is a reduced possibility for selection of superior animals for next generation, thus hampering the genetic progress.

Calfhood diseases:

Dairy calves are born with immature immune system, which increases their risk for infections and mortality during their first few weeks of life. Diseases of newborn and its mortality are a major cause of economic loss in livestock industry. The major infectious diseases of calves were diarrhoea, pneumonia, septicaemia, parasitic infection, ringworm etc. Among them, calf diarrhoea is a common disease affecting the newborn calf worldwide (Acha et al., 2004; Khan and Zaman, 2007; Ozkan et al., 2011). According to National Animal Health Monitoring System (NAHMS), USA

Figure 1: New born calf
(Photo courtesy: Dr.P.Mooventhan, Scientist, ICAR-NIBSM)

in 2007, 57% of the calf mortality was due to diarrhoea and most cases occurred in calves that are less than one month old (NAHMS, 2007).

Calf diarrhoea

Calf diarrhoea or 'scour' is the passing of abnormally high amounts of fluid in the faeces. It is usually a multi factorial disease involving an interaction between

microorganisms with the calf's immunity, nutrition and environment (Roy, 1990; Lorenz et al., 2011). Calf diarrhoea is the major cause of death in dairy heifer and beef calves aged less than four months old (Alam et al., 2012).This disease is associated with the loss of water and electrolytes that leads to an isotonic or hypotonic dehydration. A study was made by Malik and co-workers in the year 2013 on the month wise incidence of cattle calf diarrhoea cases in North West Uttar Pradesh. They found that there is a significant difference in the incidence of calf diarrhoea cases between summer and winter months. The maximum number of cases were reported after the rains and continued till the end of winter and autumn, which appears to be related to post calving season and climatic stress (Malik et al., 2013). Mortality rate of 53.4% for dairy calves due to calf diarrhoea was recently reported in Korea (Hur et al., 2013). Lorenzen et al., 2014 reported that calf diarrhoeic incidence was highest in the first three weeks of the calf's life, with the highest incidence between day 5 and 9. Economic losses in calf diarrhoea was due to morbidity and mortality in calves, treatment costs, and reduced growth rates in affected calves (Garaicoechea et al., 2006). Blood and Radostits, 1989 estimated 20% calf mortality may reduce net profit to 40%. The economic loss associated with calf death due to diarrhoea in Norway was estimated to be approximately 10 million US dollars in 2006 (Osteras et al., 2007).

ETIOLOGY

Escherichia coli were the main etiological agent responsible for either predisposing for viral infection or resulting in calf diarrhoea (Suresh et al., 2009).

The most common etiological agents, susceptible age of calves and common mode of transmission of calf diarrhoea were given below in the Table 1.

Infectious agent	**Age of affected calves**	**Most common methods of transmission**
Bacteria		
Escherichia coli	Usually less than 3 to 5 days	Fecal/oral
Salmonella (*S. typhimurium*)	Usually 2 to 6 weeks but can occur at any age	Fecal/oral, Colostrum or milk, Saliva/Nasal, In-utero
***Clostridium perfringens* type C**	Usually 5 to 10 days but can occur up to 2 months	Fecal/oral
Viruses		
Rotavirus	Usually 7 to 14 days	Fecal/oral
Coronavirus	Few days to several weeks	Fecal/oral
Bovine virus diarrhoea	Any age	Fecal/oral, Colostrum or milk, Saliva/nasal, In Utero
Protozoal parasites		
Coccidiosis (Genus Eimeria)	Usually from 17 days to 6 months	Fecal/oral
***Cryptosporidium* sps**	5 to 35 days	Fecal/oral


Fig.2: Thin and watery faeces (Photo courtesy: Dr.P.Mooventhan, Scientist, ICAR-NIBSM)

CLINICAL SIGNS

Typical signs include production of thin and watery faeces, abnormal colour of faeces (white to yellow green), signs of dehydration like sunken eyes, dry mucous membranes, rough hair appear, cold extremities, loss of appetite, difficulty in getting up or unable to rise the body, loss of weight rapidly, becoming lean and metabolic acidosis, loss of consciousness or even death (Tennant et al., 1972; Grove-White and White, 1999). Mostly mild to moderate dehydration was found to be constant feature in diarrhoeic calves (Singh et al., 2014).

Clinical signs	(%) dehydrated
Few clinical signs	Less than 5
Sunken eyes, skin tenting for 3 to 5 seconds	6 to 7
Depression, skin tenting for 8 to 10 seconds, dry mucous membranes	8 to 10
Recumbent, cool extremities, poor pulse	11 to 12
Death	Greater than 12%
Condition of dehydration	Skin tenting
Normal	Less than 2 seconds
Mild	2 to 3 seconds
Moderate	3 to 6 seconds
Severe	Greater than 6 seconds

Alam et al., 2012

Assessing dehydration:

Several methods like skin tenting test or based on clinical signs can be used for assessing dehydration in diarrhoeic calves.

Haematological and Biochemical variables:

Usually studying haematological and biochemical parameters helps in understanding the metabolic and health status of the animal. In diagnostic procedure, it is useful in comparing the

values obtained from ill animal to that of normal animal. Knowledge on changes in these variables associated with diarrhoea would be useful in developing preventive measures and assessing the prognosis of individual cases.

Alam et al., 2012 reported that haematological (increased tRBC, Hb and PCV), biochemical (increased BUN, creatinine and TBL) and plasma protein parameters (increased albumin, α-globulin and decreased γ-globulin) can reflect the severity of diarrhoea and dehydration in affected Hanwoo calves. Similarly in Bovine viral diarrhoea affected local iraqi calves, higher PCV values, increased serum fibrinogen and haptoglobin concentrations, leukopaenia, lymphopaenia and thrombocytopaenia were reported in diseased calves than in control animals (Alsaad et al., 2012). In Collibacillosis affected diarrhoeic calves, significant increase in PCV, total serum protein, serum urea nitrogen, serum albumin, creatinine and potassium, while significant decrease in serum glucose, sodium and chloride were recorded (Singh et al., 2014).

PREVENTION AND CONTROL

Minimizing the incidence of scours is a multi-step, management-reliant effort that involves three tiers: 1) the animal, 2) the infectious agents responsible for the illness and 3) the environment. The immunity of the calf is key in preventing calf scours. Make sure calves start nursing as soon as possible after calving to get adequate colostrum. It is important because it provides maternal immunity to the calves until its own immune system becomes functional. This is called passive transfer of immunity. Weaver et al., 2000 reported that calves are said to be failure of passive transfer (FPT), if the calf serum IgG concentration is less than 10 mg/mL when sampled between 24 and 48 hours of age.

Figure 3: Colostrum feeding Photo courtesy: Dr.P.Mooventhan, Scientist, ICAR-NIBSM

Vaccination of cows with any necessary calf scours vaccines well prior to calving helps in providing disease protection through the colostrum. Once the calf has acquired adequate immunity from the dam, management factors that prevent pathogen load from overwhelming the immune system should be implemented. Sanitation, animal management and vaccination are also considered to be important in scour prevention. Cleaning and disinfecting all calving areas prior to calving season, separating healthy pairs from sick calves immediately, equipment, boots and hands are thoroughly cleaned after handling sick animals will decrease the build-up of pathogens in the herd.

CONCLUSION

Diarrhoea in newborn calves is one of the most challenging clinical syndromes encountered by farmers in small and large scale dairy farms. Since calf is the back bone of dairy industry, White revolution will be meaningless if the calf

dies away unchecked in large numbers. Special attention such as following proper management practices and campaigning for creating awareness of dairy farmers towards this problem should be addressed, so that each calf must be healthy and remains that way throughout its growing life.

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Project Appraisal Systems for Livestock Management

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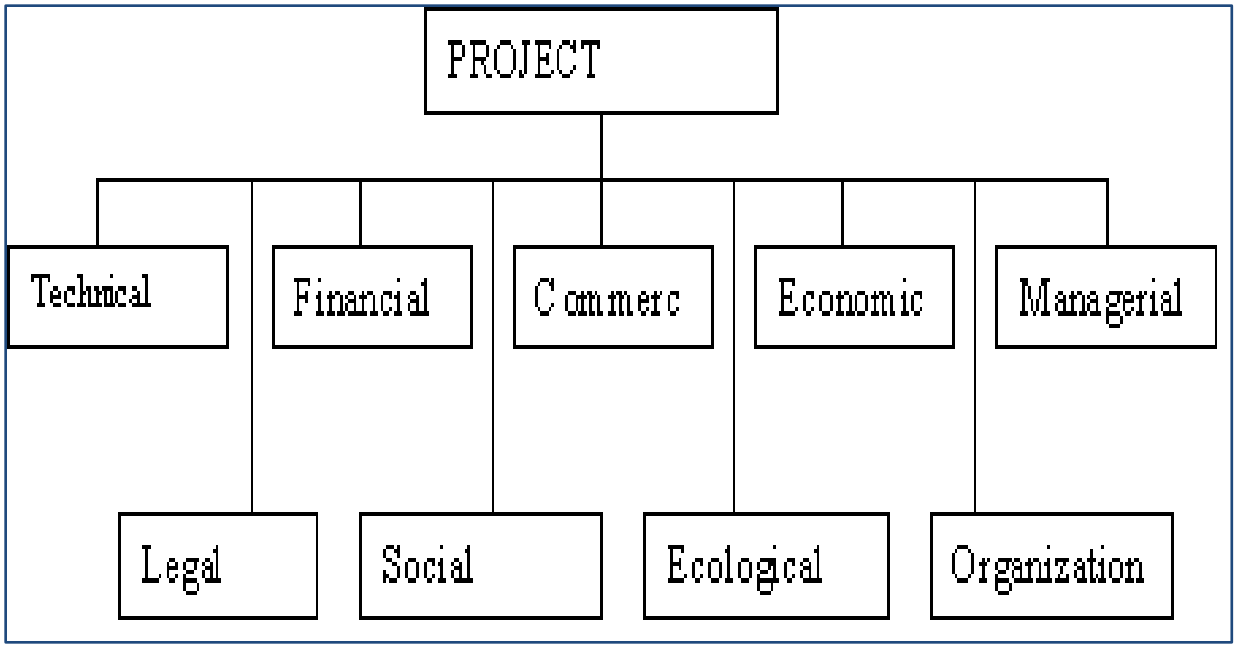
Project appraisal is a generic term that refers to the process of assessing, in a structured way, the case for proceeding with a project or proposal. In short, project appraisal is the effort of calculating a project's viability.

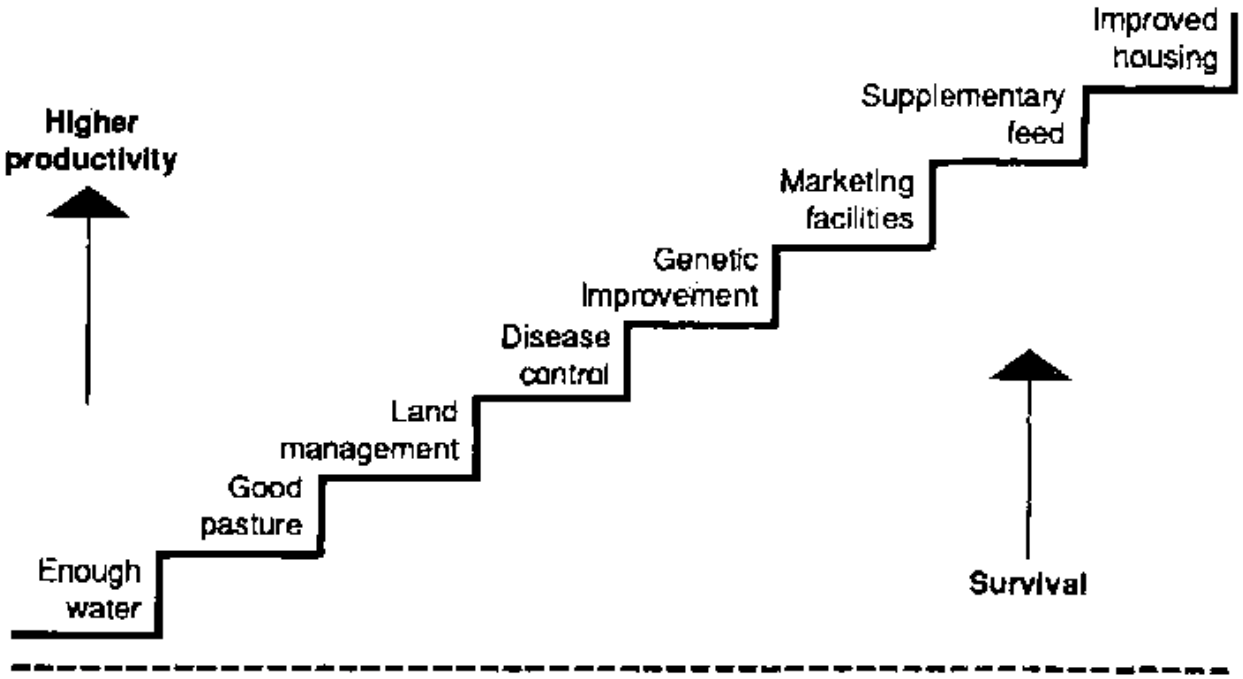
- Initial Assessment
- Define problem and long-list
- Consult and short-list
- Develop options
- Compare and select Project appraisal



Project appraisal

Types of appraisal

- Technical appraisal
- Project appraisal
- Commercial and marketing appraisal
- Financial/economic appraisal
- Organisational or management appraisal
- Economic appraisal
- Cost-effectiveness analysis
- Scoring and weighting



Treasury Appraisal/Monitoring For Construction Projects

Treasury's role in assessing and monitoring major capital works projects (those valued at $1 million or more) is dependent on the size and risk of the project, the risk being assessed using the Gateway Risk Profile Assessment tool. A strong emphasis is placed on the quality of the business case supporting any funding/ investment decision and service delivery objectives of the agency concerned.

AGENCIES NEED TO DEMONSTRATE CLEARLY THAT

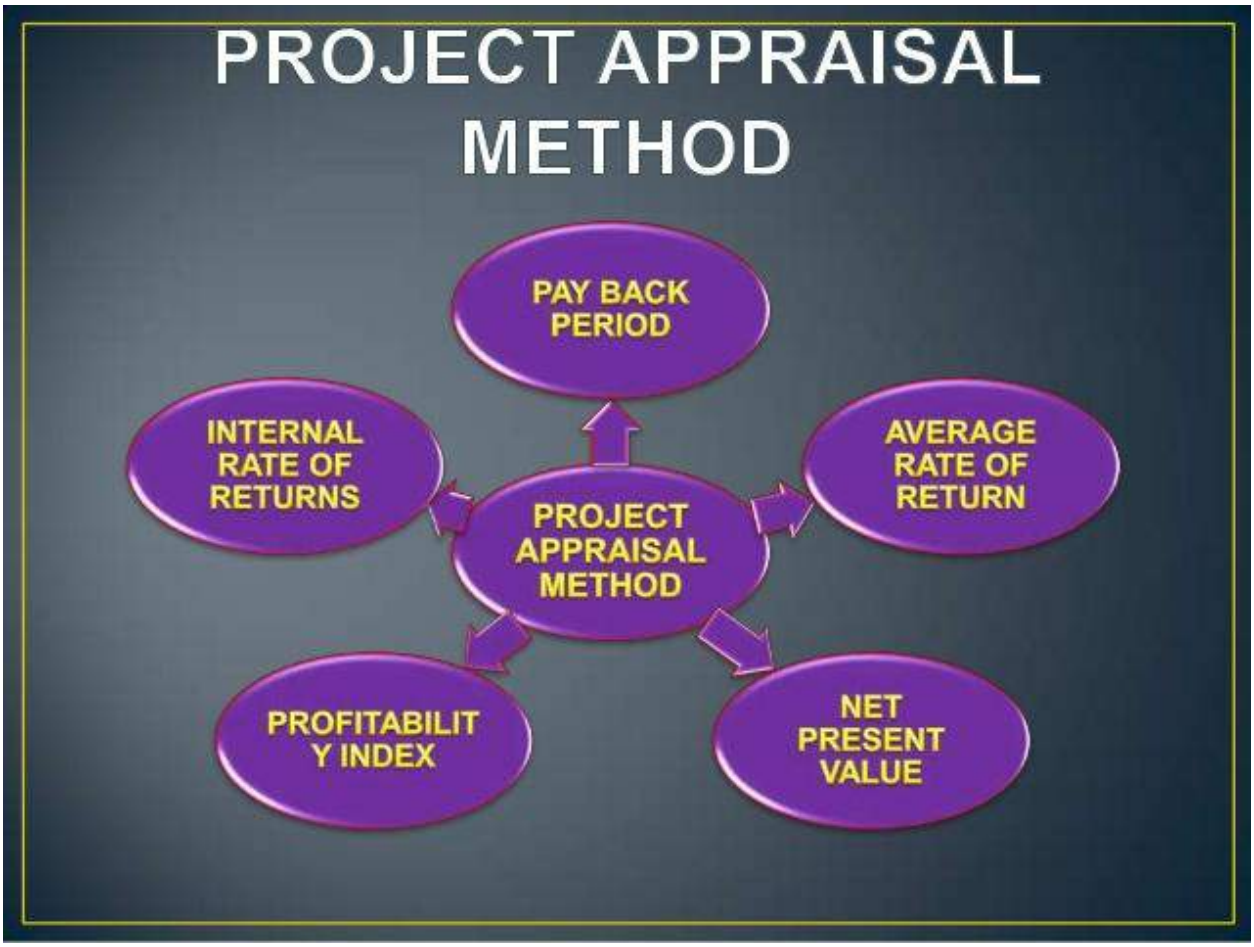
- The project supports its service delivery objectives
- The initial business case that triggers any funding is sound
- The project, as it is delivered, either remains consistent with the original business case or properly informed decisions are made in terms of the project's future if this is not the case.
- Project Appraisal Report to demonstrate the Business Case is properly developed prior to going to the Budget Committee of Cabinet or internal funding approval. This will include copies of the project Risk Profile Assessment using the Gateway tool, Mandatory Business Case Gateway Review and Economic Appraisal

- Procurement Strategy Report and Pre-Tender Estimate to reconfirm the Business Case prior to calling tenders
- Post Tender Review Report to reconfirm the Business Case prior to contract award
- Material Variations Report highlighting major changes to scope, cost and time after contract award as they occur.

Business Case (to be submitted to Treasury prior to Agency seeking Budget Committee approval of investment decision)

1) Scope and objectives of project, and analysis of how the project fits into:

- Government's strategic planning outcomes and priorities
- Agency's strategic objectives and Results and Services Plan (RSP) – e.g., explanation of the services the project will support; the results or outcomes these services will contribute to; and the gap in that service delivery at present



Typical Documentation Requirements From Agencies

- Agency's Total Asset Management Plans, an integrated set of five documents comprising
- Asset Strategy, which is supported by: a) Capital Investment Strategic Plan

b) Asset Maintenance Strategic Plan
c) Asset Disposal Strategic Plan
d) Office Accommodation Strategy.

2) Economic Appraisal report, including but not limited to:

- Demand studies, pricing analysis, surveys
- Costs – capital, operating, maintenance
- Benefits, where quantifiable
- Basis for assumptions, methodology - data sources, references.

3) Supporting information underlying the Economic Appraisal

- Land and/or property valuation reports
- Engineering reports
- Market studies, traffic sudies, other relevant material providing information on utilisation/demand/supply of the service
- Environmental reports
- Social impact studies

4) ***Financial Appraisal (Agency perspective)***

- Costs – capital, operating, maintenance; provision for contingencies
- Data sources, references for assumptions (e.g., CPI, building price index, wage increases; internal rate of return/hurdle rate; assumed Government contribution if any; and so on)
- Financial impacts, including the retiring of older assets and associated operating and maintenance savings
- Any third party revenues, source for revenue assumptions (e.g., for a road project, could include basis for traffic forecasts and alternative tolling regimes)
- Justification for assumed discount rate.

5) Financial Impact Statement (impact on the State Budget)

- Agency financial impact (savings and costs): broad implications for the agency, such as additional staff, equipment, and so on
- Financial impacts on other agencies
- Preliminary Accounting Treatment - Impacts on Statement of Financial Performance and Statement of Financial Position.

6) Risk Assessment, Mitigation and Valuation (if appropriate):

- Identify key sources of risk
- Information sources, including probability and impact of certain risks occurring or not occurring on similar projects.

Programming of Fetal Behavior in Cattle

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Abstract:
Understanding of behavior of fetus and its physical development help in proper management of livestock in post-natal life. High mortality can be minimized with considering the behavioral need of fetus. Fetal kinesis in animal is of two types i.e., simple fetal movement and complex fetal movement. The Jaw, neck forelimb and hind limb show simple fetal movement. Jaw movement represents, suckling behavior by fetus. Complex fetal movement is called "stepping", which develop when full term is approached. Righting is actually the active instances when fetus made attempts to raise head and neck against the gravity. Fetal bunting helps to orient the fetus towards the pelvic roof. Fetal play increases the vascularity to the musculo-skeletal system for its proper development. Fetal cardiogram and ultrasonographic study of fetus help to identify twin, triplets and cloned pregnancies with respect to fetal behavior. Mal-postured and dystocia showed deviated fetal behavior.

INTRODUCTION

Natural feel and behavior emerges and changes during development of animals. It reveal pattern of behavior in young one of farm animals and also show effect of time aspect in programming of behavior. Complete understanding of behavioral in addition to physical development in young one help in proper management of livestock in coming life. High mortality rate can not be controlled without taking in consideration the behavioral need of fetus. Common feature of intrauterine behavior are kinetic, postural, taxic and tropic. Sensation of position, gravity, touch, smell and taste developed in uterus is already proved in precocial neonates like calves and lambs (Vince et al., 1982). Progressive behavior development in the fetus explains that particular behavior occurs in precise stage of development in active or passive manner. Ultrasound, radiography and fluoroscopy are common aids for study of intrauterine fetal behavior. Fetal kinesis in animal is of two types i.e., simple fetal movement and complex fetal movement.

Stages of fetal development:

Fig 1: Stages of fetal development & Effects of nutrient restriction

Fetal development described in table-1 & fig-1. Gross fetal activity results from groups of complex fetal movements quickly following one another.

Movement of fetus is due to its own muscle activity**.** Muscle starts moving as soon as it is innervated. Fetal movement can be classified as either elicited or spontaneous. Spontaneous movement triggered by brain or spinal cord.

Table -1: Stages of pregnancy and fetal development:

Sr. No.	Pregnancy	Size	Development
1.	First month (28 days) Embryonic period	Embryo 9-10 mm long	Extremity appear
2.	Second month (30-60 days)	8 cm	Pharyngeal cleft close, hoof
3.	Third month (60-90 days)	14 cm	Four stomach, Scrotum
4.	Fourth month (90-120 days)	24 cm (2 kg)	Hoof mature yellow color
5.	Fifth month (120-150 days)	35 cm (2.5-3 kg)	Tactile hair on the lips, chin, upper eyelid, and orbital arch. Scrotum filled with testis
6.	Sixth month(150-180 days)	46 cm	Eye lashes more developed
7.	Seventh month(180-210 days)	60 cm	Hair on tail tip, hair on coronet and on horn point
8.	Eighth month(210-240 days)	65-75 cm	Back covered with hair
9.	Ninth month	80-100 cm	Whole body covered with hair
10.	Tenth month	>100 cm	Fetus mature in begening

SIMPLE FETAL MOVEMENT

The neck forelimb and hind limb show main movement of this type. Repetitive muscle activity has positive effect on improved muscular development and muscle tone. In case of reduced fetal energy such as in thyroid deficiency underdevelopment of muscle is prominent finding. In thyrodectomized fetuses a marked reduction was observed with development of appendicular skeleton compared with control. Simple fetal movement is directly related to physical maturation in general and muscular development in particular. This fetal activity can be controlled with nutritional management.

JAW MOVEMENT

Rapid jaw movement is seen in mature fetus with lower jaw moving in vertical axis in relation to skull. This type of jaw movement represents, suckling behavior by fetus. During last week of pregnancy lower jaw moves with mouth remain closed represent "chewing". Thus mature fetus engaged in oral behavior like suckling and swallowing. This shows that actual suckling behavior appear before parturition.

COMPLEX FETAL MOVEMENT

Complex fetal movement is called "stepping", which develop when full term is approached. Extremely intense fetal activity is frequently followed by an extended period of fetal quiescence. Coordination of complex fetal movements develops and leads to their grouping in phases. Mass fetal activity in cattle occurs two to three days prior to parturition however soon before parturition activity declines. Mass kinesis involve around 3500 movements match with righting action in uterus (Fraser and Broom, 1998). Activity and quietness for the fetus do not correspond to those of mother. Fetus is most active from 9.00 AM to 2.00 PM and again from 7.00 PM to 4.00 AM reported in human pregnancy.

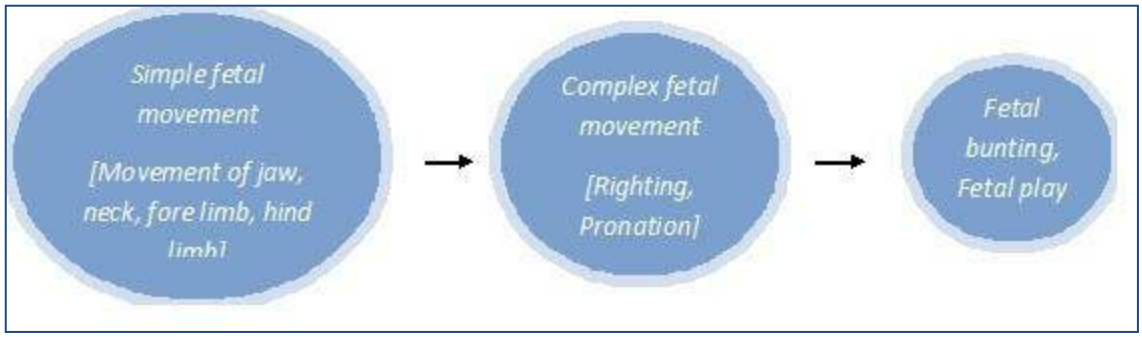
RIGHTING IN FETUS

Righting reflex also known as Labyrinthine righting reflex. It is a reflex that corrects the orientation of the body when it is taken out of its normal upright situation. This step is one of the component of complex fetal movement which involve change in posture and position of fetus. Righting reflex evidently occur during the days before parturition. It is actually the active instances when fetus made attempts to raise head and neck against the gravity.

PRONATION

As a result of series' of movement, the fetus terminally adopts the prone or squatting posture appropriate for birth. Pronation is a unique movement possible only in the forearms or hands, allowing the body to flip the palm face up. Fetal posture and engagement at pelvic inlet become fully organized in last hour of gestation in cattle. Behavior of fetus shown in Fig-2.

Fig: 2- Fetal behavior expression



Starting of parturition:

As the head progresses through birth canal the carpal joints fixed and hinged over pelvic inlet of dam this help in putting shoulder into birth canal. Typical birth posture is wedge like, should be attained before parturition. Fetal heart rate drop very suddenly when cow exerting full expulsive forces on fetus during expulsion of fetus. As the fetal abdomen reaches the birth canal hind limb extended and expulsion of fetus is complete. The first postural action of the expelled fetus is anti-gravity head elevation.

Fetal posture for normal birth:

Anterior presentation, dorso-sacral position, head extended, fore limb extended most of the time with head placed on fore limb. Postural defect especially the de-flexion of head and forelimb flexion appear to be associated with fetal inertia.

Fetal bunting:

This phenomenon occurs during pre-parturition period. Fetus jerks the anterior pole upward by extension at junction of the head and neck. During the pull, mouth remains closed. This behavior is similar to bunting while suckling in calf which helps to orient the fetus towards the pelvic roof.

Fetal play:

During last 6 week before birth in human fetus kicking and jabbing movement occur while sleeping lightly.

Whole body movement and movement of body parts are common in different stages of pregnancy. Moreover just before birth fetus changes its orientation actively and attains accurate position and posture of parturition. Pre-natal exercise help in pre-natal fetal modification i.e., fetus capable of performing the necessary movements will survive through birth. Pre-natal fetal play is similar to postnatal play of calf. The physiology of play behavior is to periodic increase in vascularity to the musculo-skeletal system for proper development of this system.

Maternal under-nutrition and over-nutrition reduces placental fetal blood flow and reduces fetal growth. Impaired placental synthesis of nitric oxide (angiogenesis, vasodilator) and ploy amines may provide a combined explanation for intrauterine growth retardation (Wu et al., 2004).

Fetal electro cardiogram in dairy cattle:

Fetal electro cardiogram i.e., fetal QRS pattern is detectable in pregnant cattle 5 months onward both in single and twin pregnancies (Too et al., 1966). A change in amplitude without changes in polarity is

common in later stage of pregnancy, however during earlier stages inversion in polarity was sometime observed.

Trans-abdominal ultrasonography of fetus:

Transabdominal ultrasonography help in diagnosis of large placentomes and hydrallantois that frequently accompany clone pregnancies. While examining heifer carrying 8 month old cloned fetus by trans-abdominal ultrasonography examination the fetal heart rate was 113 beats per minute. Hyper activity and imaging of hyperechoic particles in both allantoic and amniotic fluid was possible sign of fetal distress or death (Buczinski et al., 2009). Correlation was not observed between fetal heart rate and fetal activity. Measurement of different fetal parameters like thoracic aorta, metacarpal or metatarsal thickness could be reliable tool for early detection of the large offspring syndrome commonly found in cloned calves (Buczinski et al., 2007). Fetal activity was not associated with survival however it help in evaluation of fetal well being through assessment of fetoplacental unit (Buczinski et al., 2011).

Fetal kinesis and inertia:

Kinetic behavior in bovine fetuses can be studied in recent years using Doppler ultrasound. An apparently abnormal feature has been observed in bovine mal-posture and different cases of dystocia.

Muffled/faint memories of fetus:

Young ones are able to recognize pseudo-words while developing inside the utero as studied in human pregnancy.

CONCLUSION

Understanding fetal behavior is very much focused on acquisition of proper birth posture before parturition. Fetal position and posture in utero is result of fetal activity and maternal compressive forces. It is apparent that fetal behavior is related to position and postural requirement of birth processes and it is very clear that pre natal activity relates to some post natal practices like fetal suckling and butting. Righting and rotation in utero were active instances when fetus made attempts to raise head and neck against the gravity. This behavior of raising head against gravity is also seen in calf just after expusion. Fetal behavior plays a role in fetal kinesis, which ultimately establish muscular competence and help to acquire characteristic birth posture and survival in utero. Fetal cardiogram and ultrasonography further aid in study of behavior of fetus especially twin, triplets and cloned pregnancies.

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Rhizospheric Competency of Mycorrhiza with other Bio-Agents

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Mycorrhiza is a symbiotic association of fungi with roots of higher plants in which it helps the plant in many ways by enhancing phosphorous uptake, water uptake, nitrogen fixation, trace nutrients availability, antibiotics and hormone production and reduces pathogen inoculums in rhizospheric region of plant roots by applying many mechanisms such as physical, physiological, biological and biochemical mechanisms and protects plant roots from the attack of various soil-borne plant pathogens alone and in interaction with other bio-agents. This article will be helpful for providing knowledge about how mycorrhiza works in the rhizospheric region with other bio-agents in management of major soil-borne plant pathogens.

INTRODUCTION

Mycorrhiza is one of the major components of the agricultural natural resource and they are members of the Kingdom: Fungi; Phylum: Glomeromycota. Soil-borne plant pathogens such as fungi, bacteria and nematodes incurred a great economic loss to the agricultural productivity. For management of these problems, an extensive uses of chemicals for control of diseases pose a serious threat to the present day sustainable crop production systems (Dehne, 1982). The use of beneficial microorganisms is one of the alternative management strategies to have protective measures against soil-borne pathogens (Mukerji *et al.,* 2002). Therefore, many researchers are trying to use alternate approaches based on either manipulating or incorporating microorganisms to strengthen plant protection umbrella against soil-borne pathogens (Grosch *et al.,* 2005). When these bio-agents are applied along with arbuscular mycorrhizal (AM) fungi, it gives better reduction in soil-borne plant pathogens (Smith and Read, 2008) and boost up the crop growth (Schreiner and Bethlenfalvay, 1995) involving several mechanisms.

What is Mycorrhiza?

The word Mycorrhiza originated from the Greek word "*Mykes*" means "fungus" and "*Rhiza*" means "roots". Mycorrhiza is a symbiotic or feebly pathogenic association of fungus and roots of higher plants. The term "mycorrhiza" was coined by Albert Bernhard Frank to describe the symbiotic association of plant roots and fungi in 1885. It consists of vesicles, arbuscules, hyphae and spores. Vesicles are spherical or oval thick walled structures and acts as a storage

structures. Arbuscules are bush-like haustoria and performs function in absorbing the nutrients from soil. Hypha is of thick-walled or permanent and thin-walled or short-lived. It bears various kinds of spores (Dube, 2015).

Types of mycorrhiza: 1. Ectomycorrhiza (Ectomycorrhiza), 2. Endomycorrhiza (Erichoid, Orchid and Vesicular arbuscular mycorrhiza) and 3. Ectendomycorrhiza (Arbutoid and Monotropoid mycorrhiza) (Dube, 2015).

Hyphal network: Two types: The *Paris*-type (Intracellular) and *Arum*-type (Intercellular) (Dickson, 2004).

Functional diversity of AM fungi: It increases efficacy of N_2 fixation in legumes, overall absorption capacity of roots, mobilization and transfer of nutrients, tolerance against root pathogens by plants, production of plant growth hormones, and secretion of antibiotics and adaptation of plant to adverse environmental conditions (Garg and Chandel, 2010).

Mechanism

An AM fungus shows several mechanisms which are involved in the suppression of the soil-borne plant pathogens. 1. Physical 2. Physiological 3. Biochemical and 4. Biological mechanism (Sharma and Johri, 2002).

1. Physical mechanism: Lignifications of the cell wall and production of other polysaccharides has been reported to prevent penetration of *Fusarium oxysporum* (Dehne and Schoenbeck, 1979). It imparts great mechanical strength and diminishes the effect of vascular pathogens through a stronger vascular system of the mycorrhizal plants by increasing the nutrients flow (Schoenbeck, 1979).

2. Physiological mechanism: AM fungi indirectly affect host-pathogen relationship through competing for space or host resources (Smith and Gianinazzi, 1988). Higher levels of amino acids (arginine) in combination with root exudates of mycorrhizal plants have been reported to reduce chlamydospore production of *Thielaviopsis basicola* (Baltruschat and Schoenbeck, 1975). Higher concentrations of orthodihydroxyphenols in mycorrhizal plants resulted in suppression of *S. rolfsii* (Krishna and Bagyaraj, 1986).

3. Biochemical mechanism: Increased production of phytoalexins such as coumesterol supports mycorrhizal symbiosis and inhibits the activity of plant pathogenic fungi (Ross, 1972). Roots of host plants colonized by a mycorrhizal fungus exhibits high chitinolytic activity in which PR proteins like chitinase and ß 1-3 glucanases hydrolyzes the carbohydrate and chitin contents of the pathogenic fungi (Boller, 1993).

4. Biological mechanism: The interaction between AM fungi and other soil microbes in rhizosphere can be positive or negative (Mukerji *et al.,* 2002). The positive interaction of AM fungi with plant growth promoting rhizobacteria (PGPR) and N_2- fixing bacteria enhance the spore germination of AMF and the plant growth (Mayo *et al.,* 1986). Negative interaction is related to the ability of AMF to suppress and inhibit the various pathogens (Dehne, 1982).

Interaction of mycorrhiza with rhizobacteria in biological control

Several bacteria such as *Pseudomonas* spp., *Bacillus* spp., *Paenibacillus* spp. and *Rhizobium* spp. survive in rhizospheric region of host plants. In host plants, rhizosphere carbon flow is greatly affected by the presence of mutualistic arbuscular mycorrhizal fungi. Carbon from AM fungal mycelium is rapidly incorporated into microbial biomass and so these fungi have the potential to be important conduits of energy into rhizosphere bacteria that have bio-control potential (Siasou *et al.,* 2009). These rhizosphere bacteria have capacity to produce the antimicrobial secondary metabolite 2, 4- dicetyl-phloroglucinol (DAPG) (Cook, 2003). DAPG is broad range antibiotic with antibacterial and antifungal properties (Thomashow and Weller, 1996). Thus, it protects the roots of host plants from the infection caused by soil borne plant pathogens. Ambardar (2011) reported that application of *Glomus mossae* (Gm_1) with *P. fluorescens* and *B. cereus* recorded 19.99 and 20.00 per cent bacterial wilt incidence, respectively in tomato (cv. Solan Gola) compared to control (91.66%). While, in case of BWR-5 cultivar the same treatments found significantly superior over all other treatments in which no incidence of bacterial wilt was recorded. Zahid *et al.,* (2007) found that application of VAM fungi along with *Rhizobium* sp. gave significantly lowest mortality (20.84 %) of chickpea seedlings incited by *Sclerotium rolfsii* as compared to control (100%).

Interaction of mycorrhiza with *Trichoderma* spp. in biological control: Vazquee *et al.,* (2000) reported that AM fungi and *Trichoderma* spp. both promoted the plant growth and improved health when they colonized roots and facilitated them in a synergistic manner. *Trichoderma harzianum* induces the symbiotic association of AM fungi with vascular plants. Al-Asbahi (2012) showed that volatile biomolecules released by *T. harzianum* Rifai indirectly enhanced association of AM fungi with host plant roots. Haneefat *et al.,* (2012) reported that Auxin and Gibberllin levels were accumulated significantly in soybean plants when treated with *T. harzianum and Glomus mosseae*. Leta and Selvraj (2013) showed that application of *Glomus aggregatum* along with *T. harzianum* isolate (ATh1) resulted into lower white root rot (*Sclerotium cepivorum* Berk) incidence (33.81%) compared to control (90.50%) in onion. Dehariya *et al.,* (2015) concluded that application of *T. harzianum* and mycorrhiza in pre-inoculation method found effective in managing Fusarium wilt (*Fusarium udum*) of pigeonpea.

CONCLUSION

Mycorrhiza is ubiquitous in nature and can be effectively used for biological control of soil-borne plant pathogens as it has several mechanisms for suppression of the pathogen. Among all species of mycorrhiza, *Glomus* spp. is most widely used and found effective for biological control. Use of AM fungi is an effective alternative in biological control as it protects plants roots from soil-borne plant pathogens in rhizosphere. It gives better results in interaction with bio-agents such as *P. fluorescens* and *Trichoderma* spp. against soil-borne plant pathogens as compare to alone. It plays a major role in plant protection as well as in plant growth promotion.

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Ecological Weed Management: A Sustainable Way for Crop Production

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The word weed means any wild plant that grows at an unwanted place especially in fields or in gardens where it interferes with the growth of cultivated plants. A number of definitions of weeds have been proposed, but none has come to universal satisfaction. Weeds may be defined as plants with little economic value and possessing the potential to colonize disturbed habitats or those modified by human activities. Ecological weed management is a holistic system involving an entirely different approach to managing a farming system. The organic farmer is not interested in eliminating all weeds but wants to keep the weeds at a threshold that is both economical and manageable. Weeds can always be pulled or cut, but the question is simply how much time and money can a grower expend to reduce weed pressure. The more a grower is able to reduce weed pressure, the more economical it is to produce crops. Organic farming strongly relies on an integrated systems approach. Ecological weed management strategy is to integrate the options and tools, rather than on specific control practices which are available to make the crops and cropping system unfavorable for weeds and to minimize the impact of any weed that survive. Adoption of sustainable agricultural practices reduces the intensity of soil manipulation thereby creates an unfavorable condition for weed seed germination, reduces the organic matter depletion and soil erosion. Thus, the ecological approaches could be an option for weed and soil management which leads to sustainable crop production.

Approaches Involved in Ecological Weed Management

Ecological weed management promotes weed suppression, rather than weed elimination, by enhancing crop competition and phytotoxic effects on weeds.There are some different approaches are involved in ecological weed management are as follows.

(A) Preventive approach

Preventative approaches are used to reduce the spread of weeds. Sowing of clean seed is perhaps the most important weed management approach in any crops. In general, spread of weeds within country can be reduced by clean seed laws, cleaning farm equipment and produce, cleaning irrigation water, cleaning sand and gravel and reducing the number of weed seeds returned to the soil (Das., 2008). There are many ways to prevent weeds in agricultural activities which are well known including:-

- Control and manage the opportunity for new weeds to invade and spread
- Control and manage the spread of existing weed infestations
- Quarantine
- Monitor

(B) Cultural approaches

In Cultural practices such as tillage, mulching, and burning are used by certain traditional farmers to manage the weeds. Cultural practices are aimed to ensure better soil and crop management. Globally, cultural control has been one of the most widely used control options and includes stale seedbed techniques, crop rotation; increase the competitive ability of the crop, time of seeding and irrigation, inclusion of cover crops, and intercropping (Kumar and Rathore, 2014). Some of the cultural practices are given below:

- ***Selection of competitive cultivars***

Competitive crop cultivar offers a potentially cheap option to include in cultural weed management strategies. Select a weed-competitive cultivars with early seedling vigor, and high tillering to suppress weeds. Competitive cultivar can suppress weed seed production, limit future weed infestation, and become a safe, environmentally benign and low cost tool for weed management (Kumar *et al.,* 2013). Transplanted crops tend to have fewer weeds and less yield loss than direct seeded crops. Transplant healthy, vigorous seedlings that can better compete with weeds in early stages.

- ***Crop establishment method***

Crop establishment methods adversely affect the weed population and its dry weight. Over several years, the planting pattern/geometry of the crop has been

modified to increase yield, because these methods significantly influenced weed dynamics (Dev *et al*., 2013). Line sowing/drilling has replaced broadcasting to a large extent. High weed infestation is a major constraint for broader adoption of DSR (Rao *et al.,* 2007). Zero-till and FIRB sowing recorded lower weeds density with higher grain yield in wheat (Ahmed *et al.,* 2010; Jat, *et al.,* 2013). In zero till seeding by Happy Seeder machine with stubble mulching, undisturbed inter row space, where seeds lying at lower depths did not germinate (Bhullar *et al*., 2006) and it saves time and energy (Yadav *et al*., 2013).

❖ ***Intercropping***

Intercropping involves growing more than one crop in the same field at the same time. One main crop with one or more secondary crops interceded for weed suppression with the goal of maximizing yield of the main crop. Intercropping, preferentially spreading types of crops, legumes, cucurbits, sweet potatoes, contributes to a faster and denser ground cover suppresses weed growth and reduces erosion (Giri *et al*., 2006). In particular, crops with allelopathic potential when intercropped with other crop plants help to reduce weed intensity, and hence improve crop productivity. For instance, intercropping maize and cowpea on alternate ridges helped reduce weed [*Echinochloa colona* (L.) Link. *Portulaca oleracea* L., *Chorchorus olitorius* L., and *Dactyloctenium aegyptium* (L.) Willd.] intensity by 50% as well as improve land use efficiency (Saudy, 2015).

❖ ***Crop rotation***

Crop rotation is a planned sequence of crops growing in the same field year after year. Crop rotations can help in controlling weeds, supplying soil nutrients, improving soil tilth, and reducing soil erosion. The success of rotation systems for weed suppression appears to be based on the use of crop sequences that employ varying patterns of resource competition, allelopathic interference, soil disturbance, and mechanical damage to provide an unstable and frequently inhospitable environment that prevents the proliferation of a particular weed species (Liebman and Dyck 1993).

❖ ***Seed rate***

Variation in the seed rates and high seed rate significantly influenced weed population and their dry weight by securing an optimum plant population (Meena *et al.,* 2010), which shows excellent smothering effect on weeds (Sharma and Singh, 2011) and improving productivity and profitability of the crop.

❖ ***Cover crops/green manures***

Green manure crops are commonly associated with organic agriculture, and are considered essential for annual cropping systems that wish to be sustainable. One of the major benefits of green manures is their ability to suppress weeds (Blackshaw *et al*., 2001 and Mohler *et al*., 2012). Green manuring of Sesbania significantly suppresses weeds in rice due to shading and allelopathic effect (Yadav *et al.,* 2010).

❖ ***Nutrient management***

Proper crop nutrient management can play a pivotal role in weed management. The role of nutrient management through fertilizer application in crop production is substantial and very clear. Weeds take up significant amounts of nutrients, just like crops. Fertilizer application strategy to crop play important role in reducing weed population in crop as weeds are always strong competitor of crops for available resources (Blackshaw *et al*., 2005)

(C) Mechanical approaches

Most mechanical weed control methods, such as hoeing, tillage, harrowing, torsion weeding, finger weeding and brush weeding, are used at very early weed growth stages (Kewat, 2014). Summer ploughing increased the total buried weed seed population by 3-4 times compared to no ploughing. Offseason ploughing twice at 45 days interval was found to be superior in reducing the population of weeds; *Cyperus rotundus, C. difformis, Sphenoclea zeylanica* and *Fimbristylis littoralis* succeeding crops. Small farm implements and machine *i.e.* power tiller, marker and cono-weeder played very imperative role in controlling weeds, enhancement of productivity and reduction in drudgery (Deshmukh and Tiwari, 2011).

❖ ***Mulching***

Mulches are coverings placed on the surface of the soil. Mulching smothers the weeds by excluding light and providing a physical barrier to impede their emergence. Any material such as straw, plant residues, leaves, loose soil or plastic film can be used as a mulching material. Such materials as straw, bark, and composted material can provide effective weed control. The word mulch has been probably derived from the German word "*molsch*" means soft to decay, which apparently referred to the use of straw and leaves by gardeners as a spread over the ground as mulch.

❖ ***Hand pulling and digging***

Hand weeding is more effective for annual rather than perennial weeds due to its capacity of vegetative reproduction. Hand

hoes, push hoes and other traditional methods of hand weeding are still used worldwide in many agricultural crops. It is best to hand-pull weeds after rain, when soil is moist. Sturdy gloves should be worn to avoid prickles, blisters or sap burns to the skin. It is not appropriate for all weed species, such as those with underground bulbs. Hand tools such as broad knives and trowels can be used to remove underground parts of weeds (such as bulbs) that may reshoot.

❖ ***Flame weeding***

This method is a unique technique to kill weeds through the use of direct heat in the form of fire. Flame weeding is most prevalent in European countries (Bond and Grundy, 2001). Flame weeding uses the heat generated from one or more propane burners to kill weeds Flaming has shown good results after weed emergence but before crop emergence in potato, sugarbeet, carrot, and chilli (Melander, 1998).

❖ ***Use of weeders***

Use of mechanical weeders in agricultural operations is increasing because of non-availability of labours for weeding. The cost of the weeding operations is also reduced by using the machineries for weeding. The machineries like mini-weeders, power tillers, minitractor drawn rotavator are used for weeding in wider spaced crops like sugarcane, cotton, and orchards.

❖ ***Soil solarization***

Soil solarization is a special technique in which moist soil is covered by polyethylene film (usually black or clear plastic sheet) to trap solar radiation and cause an increase in soil temperatures for several weeks to levels that kill weeds, weed seeds, plant pathogens, and insects for economic crop production (Ascard *et al*., 2007; Singh, 2014). The process would raise the surface soil temperature by 8 to 12°C as compared to non-solarized soils. Transparent polyethylene was found highly effective for heating the soil than black polyethylene. Duration of 4 to 6 weeks is sufficient to give satisfactory control of most of the weeds.

(D) Allelopathy

The term allelopathy, from the Greek words 'allelon' and 'pathos' and meaning mutual harm or affection, is generally used to express growth inhibition of a plant through the release of chemicals into the environment from another plant. In agro ecosystems crops, weeds, trees and microbes constitute the biotic components, which not only interact among them but also with the abiotic environment. The allelopathic interactions among various

biotic components have a great potential in improving crop production, maintaining ecosystem stability, nutrient conservation, and above all in management of weeds and pests (Kong *et al.,* 2004).

(E) Biological approaches

Biological control involves the use of insects or pathogens (diseases) that affect the health of the weed. On the other hand biological weed control involves using living organisms, such as insects, nematodes, bacteria, pathogens, or fungi that affect the health of the weed or reduce weed populations. Usually, these biocontrol agents are from the same country of origin as the weed species. Plants become pests and are weeds when they run rampant because their natural enemies become ineffective or are nonexistent. The natural cycle may be interrupted when a plant is introduced into a new environment, or when men disrupt the ecological system. When we purposefully introduce biological control agents, we are attempting to restore or enhance nature's systems (Tiwari *et al*., 2013).

Integrated weed management

Integrated weed management includes more than one method of control viz., seed purity, crop varieties, spacing and methods of planting, cultivations, soil solarization, intercropping, crop rotation, water management, manure application, biological control and herbicides. Integrated weed management system is basically an integration of effective, dependable and workable weed management practices that can be used economically by the producers as a part of sound farm management system (Riemens et al., 2007).

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Salinity Stress In Plants

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Agricultural productivity in arid and semi-arid regionsof the world is very low. This is because, the crops in theseareas are naturally subjected to a multitude of abioticstresses. Of these, soil salinity limits crop production toa great extent.More than 800 million hectares of landthroughout the world is salt affected.This amount accounts for more than 6% ofthe world's total land area. Most of this saltaffectedland has arisen from natural causes,from the accumulation of salts over long periodsof time in arid and semiarid zones.Weathering of parental rocks releases solublesalts of various types, mainly chlorides ofsodium, calcium, and magnesium, and to a lesser extent, sulfates and carbonates.Sodium chloride is the most soluble and abundantsalt released. The other cause of accumulationis the deposition of oceanic saltscarried in wind and rain.Salinity is one of the most serious factors limiting the productivity of agricultural crops,with adverse effects on germination, plant vigour and crop yield.The saline growth medium causes many adverseeffects on plant growth, which are due to a low osmoticpotential of soil solution (osmotic stress), specific ioneffects (salt stress), nutritional imbalances, or a combinationof these factors. Osmotic balance is essential for plants growing in salinemedium. Failure of this balance results in loss of turgidity,cell dehydration and ultimately, the death of cells.On the other hand, adverse effects ofsalinity on plant growth may also result from impairment of the supply of photosynthetic assimilates or hormones to the growing tissues.

Water salinity

Water salinity is the amount of salt contained in the water. It is also called the "salt concentration" and may be expressed in grams of salt per litre of water (grams/litre).

Soil salinity

The salt concentration in the water extracted from a saturated soil (called saturation extract) defines the salinity of that soil.

Measurement of salinity

Salinity is the total concentration of water soluble salts in water and soil. Soil salinity can be measured as a concentration, but in most agricultural

situations it is measured as the electrical conductivity of a saturated soil paste (ECe) or a 1:5 (EC1:5) water extract in units of deci Siemens per meter (dS/m) at 25°C. In the past, salinity has been expressed as EC units (micro (µ) Siemens per centimeter) or parts per million (1dS/m= 1000EC units = 640ppm).The measurement of electrical conductivity does not indicate the types of salt present. Therefore, measurements are also made of specific salts such as chloride.

Salt concentration of the soil water (saturation extract)		Salinity
in g/l	in millimhos/cm	
0-3	0-4.5	Non saline
3-6	4.5-9	Slightly saline
6-12	9-18	Medium saline
>12	>18	Highly saline

Effects of salinity on plants

Soil salinity is a major factor that limits the yield of agricultural crops. At low salt concentrations, yields are mildly affected or not affected at all. As the concentrations increase, the yields move towards zero.High salinity affects plants in two main ways: First, osmotic stress and second, ionic stress (shown in Fig. 1)

Osmotic stress:

It affects growthimmediately and iscaused by the saltoutside the roots. Salt makes it more difficult for plants to withdraw water from the soil, even if the soil appears quite moist. In fact, the plant suffers from a form of drought which can result in retarded growth and reduced yield. High concentrations of salts in the soil disturb the capacity of roots to extract water.In cereals, the major effect of salinity on total leaf area is a reduction in the number of tillers; in dicotyledonous species, the major effect is the dramatic curtailing of the size of individual leaves or the numbers of branches. Curiously, shoot growth is more sensitive than root growth.

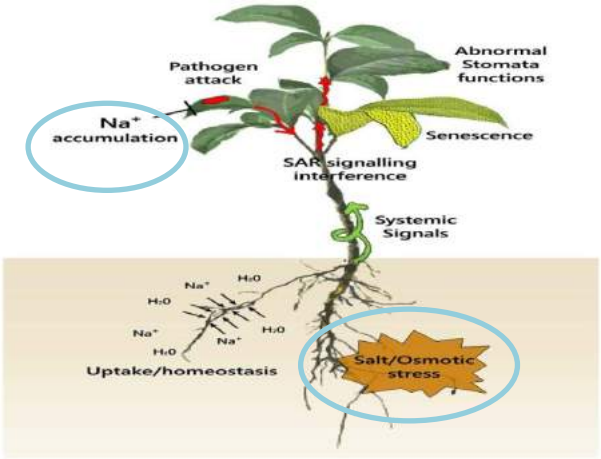

Figure 1:Osmotic stress and Ionic stress

Ionic stress:

It develops over time and is due to a combination of ion accumulation in the shoot and an inability to tolerate the ions that have accumulated. Some ions, such as Na^+ and Cl^- can be directly toxic to plants. When salt accumulates to toxic concentrations in the old leaves (which are no longer expanding and so, no longer diluting the salt arriving in them as younger growing leaves do), and they die. If the rate at which they die is greater than the rate at which new leaves are produced, the photosynthetic capacity of the plant will no longer be able to supply the carbohydrate requirement of the young leaves, which further reduces their growth

rate. Plants take up salts with the water thatthey use, and often these salts can damage the plant internally,affecting the plant's physiological processes,biochemical processes such as nutrient uptake and assimilation, andoften resulting in reduced growth, leaf burn and even plant death.

The osmotic stress not only has an immediate effect on growth (Table 1), but also has a greater effect on growth rates than the ionic stress. Ionic stress impacts on growth much later, and with less effect than the osmotic stress, especially at low to moderate salinity levels.

Table 1 Effects of salinity stress on plants

Effect of stress	Osmotic stress	Stress due to high leaf Na^+ (ionic stress)
Speed of onset	Rapid	Slow
Primary site of visible effect	Decreased new shoot growth	Increased senescence of older leaves

A two-phase model describing the osmotic and ionic effects of salt stress, was proposed by Munns (1995) (Fig. 2). During Phase 1, growth of both types of plants is reduced because of the osmotic effect of the saline solution outside the roots. Osmotic phase, which starts immediately after the salt concentration around the roots increases to a threshold level, making it harder for the roots to extract water; the rate of shoot growth falls significantly. During Phase 2, old leaves in

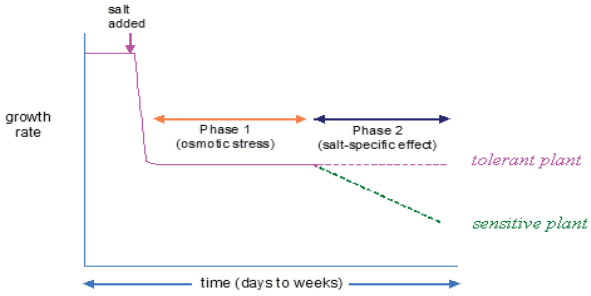

Figure. 2. Scheme of the two-phase growth response to salinity. Adapted from Munns (1995)

the sensitive plant die and reduce the photosynthetic capacity of the plant.

Salinity tolerance mechanisms

The mechanisms of salinity tolerance fall into three categories- a) the tolerance to osmotic stress, b) the Na^+ exclusion from leaf blades and c) tissue tolerance (shown in Fig. 3)

Tolerance to osmotic stress-The growth of salt-stressed plants is mostly limited by the osmotic effect of salinity, irrespective of their capacity to exclude salt,that results in

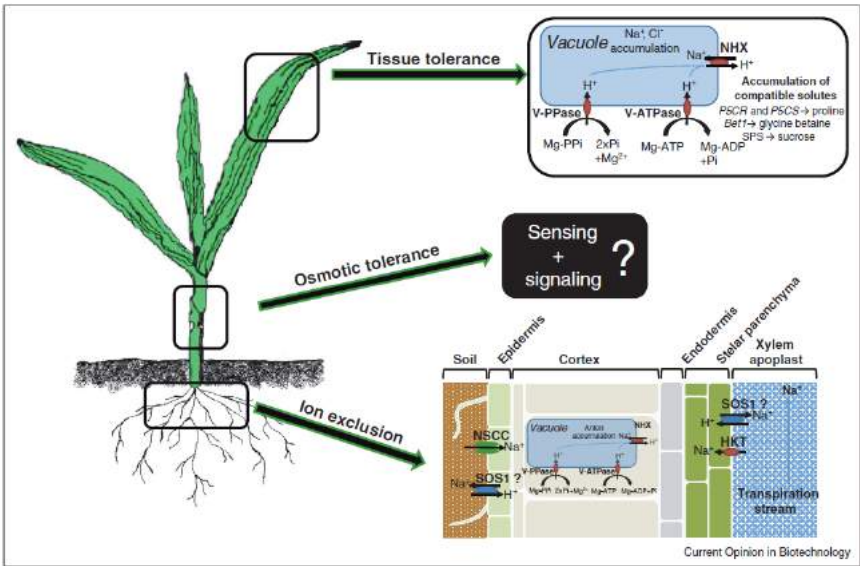

Figure 3:Mechanisms of salinity tolerance

reduced growth rates and stomatal conductance. In fact, osmotic tolerance involves the plant's ability to tolerate the drought aspect of salinity stress and to maintain leaf expansion and stomatal conductance. The osmotic stress immediately reduces cell expansion in root tips and young leaves, and causes stomatal closure. A reduced response to the osmotic stress would result in greater leaf growth and stomatalconductance, but the resulting increased leaf area would benefit only plants that have sufficient soil water. Greater leaf area expansion would be productive when a supply of water is ensured such as in irrigated food production systems, but could be undesirable in water-limited systems, and cause the soil water to be used up before the grain is fully matured.

Na^+exclusion from leaf blades-Ion exclusion, where Na^+ and Cl^- transport processes, predominantly in roots, prevent the accumulation of toxic concentrations of Na^+ and Cl^- within leaves. Na^+ exclusion by roots ensures that Na^+does not accumulate to toxic concentrations within leaves. A failure in Na^+exclusion manifests its toxic effect after days or weeks, depending on the species, and causes premature death of older leaves.Mechanisms may include retrieval of Na^+ from the xylem, compartmentation of ions in vacuoles of cortical cells and/or efflux of ions back to the soil.

Tissue tolerance-Tissue tolerance, where high salt concentrations are found in leaves, but are compartmentalized at the cellular and intracellular level (especially in the vacuole)to avoid toxic concentrations within the cytoplasm, especially in mesophyll cells in the leaf, a process involving ion transporters, proton pumps and synthesis of compatible solutes.Tolerance requires compartmentalization of $Na^+$and Cl^-at the cellular and intracellular level. Toxicity occurs with time, after leaf Na^+increases to high concentrations in the older leaves.

CONCLUSION

Most of this salt affected land has arisen from natural causes, from the accumulation of salts over long periods of time in arid and semiarid zones.Sodium chloride is the most soluble and abundant salt released. Salinity is one of the most serious factors limiting the productivity of agricultural crops, with adverse effects on germination, plant vigour and crop yield.At low salt concentrations, yields are mildly affected or not affected at all. High salinity affects plants by means of osmotic stress and ionic stress. Also, plants have developed three mechanisms of salinity tolerance- 1) the tolerance to osmotic stress, 2) the Na+exclusion from leaf blades and 3) tissue tolerance.

Ultrasonographic Appearance of Pathological Conditions of Bovine Uterus and Ovaries

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Ultrasonography (US) has been used in bovine practice since the 1980s.In bovine practice, ultrasonography has become an ideal complementary diagnostic tool for evaluating the female reproductive system in a non-invasive manner. US has also allowed for more accurate diagnosis compared with rectal palpation in reproductive management in cattle. Practical applications of US include early diagnosis of pregnancy, identification of twin foetuses, detection of ovarian and uterine pathologies and determination of foetal sex. Linear probes are preferred for transrectal ultrasound examinations of the ovaries and uterus. Linear-array transducers of 5.0 and 7.5 MHz frequency ranges are most commonly used in cattle.

INTERPRETATION OF IMAGES

It is based on an evaluation of the shape, contour, size, and position of the structure being studied, as well as its echogenicity, which depends on the amplitude of the echoes received. An echogenic structure reflects the majority of soundwaves back to the probe and thus appears from white to different shades of grey on the screen.

- Acoustic enhancement will appear as a hyperechoic region deep to the fluid (anechoic) area. This can often be seen in deep to large follicle.
- Gas and bone will totally reflect sound waves and produce strongest of the echo signals, leading to an image that appear on the screen as near white.
- Reverberation (rebounding of echoes back and forth between object and the transducer) is often encountered in transrectal scanning where gas-filled viscera are present.
- Heterogeneous echo texture (mixture of signals) is often characteristic of an individual structure, e.g. Corpus luteum and assist in the identification of that structure from the surrounding ovarian stroma.
- Changes in echo texture indicates a change in the physiological state of the organ due to an alteration in the nature of the cell or tissue type and thus in the tissue interfaces.

OVARIAN STRUCTURES IN THE COW

1. Follicles:

Its appear as anechoic circular structures surrounded by echogenic ovarian tissue. Their fluid content usually contains no reflections. The dividing walls of the two neighboring follicles of equal pressure

often form a straight line. The diameter in dominant follicle is 10 to 15 mm or greater.

2. Corpora lutea (CL):

A developing CL appears - poorly defined, irregular, greyish-black structure with echogenic spots within the ovary. A mid-cycle CLappears - well defined granular, greyish echogenic structure with a demarcation line visible between it and the ovarian stroma. A regressing CLappears -the demarcation line is faint, owing to the slight difference in echogenicity between the tissues. Many corpora lutea may have fluid-filled cavities. It ranging from less than 2 to greater than 10 mm in diameter at during the oestrous cycle and early pregnancy.

3. Ovarian cysts

In ultrasonographic appearance, ovarian cysts resemble large follicle. Differentiation between follicular and luteal cysts via rectal palpation is difficult, even for experienced practitioners

In case of follicular cyst - its appeared as a uniformly anechogenic ovarian structure >25 mm in diameter with a wall <3 mm thick and persists for > 10 days. The shape of cysts ranges from round to oval to polygonal, sometimes even angular.In case of luteal cyst – appeared as anechogenic structure >25 mm in diameter with grey patches within the antrum or along the inner cyst wall and a wall thickness >3 mm.

4. Cystic Corpora lutea:

Cystic corpora lutea are usually no larger than 3 cm in diameter and the wall is about 5–10 mm thick. Corpora lutea are rarely spherical usually presenting an oval shape. The image of the cavities depends on direction of the ultrasound beam and is either circular or oval. The fluid filled cavities of corpus luteum only seldom present reflections more often being homogeneous and near-black, whereas reflections are frequently observed in luteal cysts.

5. Ovarian tumors:

US image of the tumor contained two distinct regions. Hypoechoic transverse sections through numerous vessels were seen in the dorsal section of the tumor. The remainder of the tumor contained a coarsely echogenicity, producing an image of mixed tissue.

6. True anestrus ovary

The ovaries appear small, in which the small follicles arevisible on the ovarian surface with < 2-4 mm diameter of size.

UTERINE STRUCTURES IN THE COW

1. Embryonic death

First signs of an impending embryonic death are an under-sized embryo and a reduced amount of embryonic fluid. The death of a conceptus can be reliably diagnosed once the embryonic heart beat has stopped. As the resorption progresses the amount of embryonic fluid will decrease, while its echogenicity will increase. The embryo then losses its typical outline and becomes very indistinct.

2. Foetal mummification

Immediately below the uterine wall hyperechoic foetal mass can be detected. No uterine fluids could be seen between the surface of the mummified foetus and the uterine wall

3. Foetal maceration

The foetal bones were identified as echogenic structures in the uterine lumen suspended in anechogenic foetal fluids with a thickened uterine wall.

4. Hydrometra

The uterus was located in the caudal part of the abdominal cavity, filled with clear hypoecogenic fluid without placentomes or foetal membranes of foetus.

5. Postparum uterus

After calving, the most obvious US structure in the uterine lumen are the caruncles. Areas of differing echogenicity can be seen on the round or oval cross sections of caruncles. In many cases, lochial secretions can be seen inside the uterine lumen. They show the floccular echogenicities, which are typical of fluids that contains cellular components.

6. Pyometra

The pyometra is ultrasonographically characterized by a uterus which is considerably distended by an accumulation of fluid. If the secretions are very thick and contains many cellular elements its echogenicity may be same as that of the uterine wall, whereas a more liquid content will appear much darker than the surrounding wall of the uterus.

7. Puerperal Metritis

Enormous amount of anechoic fluid is present in the lumen along with echogenic particles. Distended uterine body with thickened uterine wallcan be seen. Mostly occurs within 10 days after parturition with clinical signs.

8. Clinical Endometritis

Moderate to large amount of anechoic fluid in lumen along with "snowy" echogenic particles. Which occurs more than 21 days after parturition. thickened uterine wall could be seen.

9. Subclinical endometritis

The local inflammatory response within the uterus would result in some degree of endometrial thickening and echogenic intra uterine fluid was related to higher neutrophil counts during examination.

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Sugar Industry By-Products: As a Livestock Feed

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Sugarcane may be a potential feed source for beef cattle in subtropical and tropical areas. Its advantages as a forage crop include adaptation to the tropical and subtropical environments, less sensitivity than other crops to poor soil fertility, the hot-humid climate, and insect and disease problems, existing technology for its production, high yield capability, and the unique ability to maintain consistent quality as a standing crop in the field.

Sugarcane industry by- products

1. Sugarcane tops
2. Bagasse
3. Sugar cane juice
4. Molasses
5. Press mud
6. Condensed molasses soluble/ dried yeast culture/ distillery residue/ yeast sludge.

1. Sugarcane tops

In South East Asian and South Asian countries, the small farmers feed their animals with cane tops either fresh or dried as a roughage source to meet the energy requirements for maintenance.

Cattle and buffaloes relish the chaffed sugar cane tops, the voluntary intake about 2.0 to 2.5% of the body weight. They can be maintained entirely on sugar cane tops with the supplementation of protein either through concentrate mixture of ground nut – cake or green leguminous forage crop like berseem and glyricidia. Sugar cane tops can also be converted in to silage of good quality and high palatability. Its acceptability is low in sheep and goat (Abrar *et al,* 2002)

Table 1: The chemical composition and Digestibility of fresh sugar cane tops (as such basis %)

	Dry matter	Crude protein	Ether extract	Crude fibre	NFE	Ash
Proximate analysis	24.8	1.9	0.5	7.7	14.5	2.2
Digestibility	58.0	55.0	50.0	60.0	65.0	-
Cane silage	25.0	1.7	0.6	7.5	12.9	2.3

2. Bagasse

Sugar cane bagasse is the fibrous product resulting from the crushing of cane sugar in the extraction or direct transformation of alcohol. Bagasse is the fibrous matter that remains after sugarcane or sorghum stalks are crushed to extract their juice.

It is used as a biofuel and in the manufacture of pulp and building materials. For each 10 tonnes of sugarcane crushed, a sugar factory produces nearly 3 tonnes of wet bagasse, since bagasse is a by-product of the cane sugar industry. The high moisture content of bagasse, typically 40 to 50%, is detrimental to its use as a fuel.

In general, bagasse is stored prior to further processing. For electricity production, it is stored under moist conditions, and the mild exothermic reaction that results from the degradation of residual sugars dries the bagasse pile slightly. For paper and pulp production, it is normally stored wet in order to assist in removal of the short pith fibres, which impede the papermaking process, as well as to remove any remaining sugar (Costa *et al*, 2015)

Table 2: Chemical composition of bagasse (%)

Cellulose	45– 55%
Hemicellulose	20– 25%
Lignin	18-24%
Ash	1 –4%
Waxes	<1%

Bagasse is often used as a primary fuel source for sugar mills; when burned in quantity, it produces sufficient heat energy to supply all the needs of a typical sugar mill, with energy to spare. To this end, a secondary use for this waste product is in cogeneration, the use of a fuel source to provide both heat energy, which is used in the mill, and electricity, which is typically sold on to the consumer electricity grid.

The resulting CO_2 emissions are less to the amount of CO_2 that the sugarcane plant absorbed from the atmosphere during its growing phase, which makes the process of cogeneration greenhouse gas-neutral.

The cellulose-rich bagasse is being widely investigated for its potential for producing commercial quantities of cellulosic ethanol. Bagasse is commonly used as a substitute for wood in many tropical and subtropical countries for the production of pulp, paper and board.

Table 2: Analysis of whole bagasse, bagasse fibre and bagasse pith (%) Ranjhan, 1990

	Whole bagasse	Bagasse fibre	Bagasse pith
Moisture	6.62	4.11	4.55
Ash	1.65	1.22	2.73
Water soluble			
Cold	5.91	4.16	7.02
Hot	7.85	6.35	9.84
Alkali soluble			
1 % NaOH	20.82	17.44	24.67
10 % NaOH	44.37	34.14	46.23
Ether Extract	1.62	0.81	2.88
Alcohol benzene extract	3.84	2.09	2.37
Lignin	20.03	18.98	21.26
Pentosans	23.86	24.01	24.82
Cellulose	53.78	58.28	50.22
Nitrogen	0.21	0.14	0.33
Protein (N* 6.25)	1.31	0.88	2.06

Sugar cane bagasse

Dry bagasse consists essentially of fibre and pith. The fibre content is 65% ash of dry weight. The pith including soluble matter constitutes remaining 35%. The optimum pressure for treatment of bagasse was 7 kg per cm^2 for 30 minutes and fortification with 1 percent urea and molasses.

3. Sugarcane juice

It contains about 15-16% soluble carbohydrate, 1-2% ash and 80-85% water. Sugar cane juice is usually cheaper than sugar by-products. It may be used to replace molasses for enhancing fermentation in ensiling. Sugar cane juice is easily available with the farmers and they can save time required for obtaining molasses.

Dehydrated sugar cane juices (or) gur contain 95-96%, 3-8% ash, 1% crude protein and 90-91% soluble sugars. Dehydrated sugar cane juice (rab and gur) is used for the supply of energy in the diets of calves, pigs, poultry and lactating animals.

4. Molasses

Molasses is another important by-product of sugar industry. It contains about 35 to 50% sugar depending on the quality. Uses include production of oxalic acid, lactic acid, butanol, acetone, glycerol, yeast, dextran, monosodium glutamate. It is cheap source of energy in the ration of the farm animals. Molasses has been mainly used in animal feeding 5 to 10% level.

Table 4: Composition of the molasses as follows (%) on DMB

Sugar	40-45
crude protein	3.2
soluble gums	2.2
Ash	8.2
combined and free acids	5.0.
NFE	84-88
Energy (ME) Mcal/kg.	2.5-2.6

These levels were mostly used as:

1. Carrier for urea impregnation of poor quality roughages like wheat bhusa, paddy straw and grasses.
2. Binder for commercially pelleted feeds for convenient and economic. Sweetener for increasing the voluntary intake of compounded feeds. In the past, it was assumed that the use of high level of molasses in the ration would decrease the feeding value of molasses and also cause diarrhoea in animals imposing a serious health problem.
3. Molasses is a sticky dark by-product of processing sugar cane or sugar beets into sugar.
4. Molasses can be a source of quick energy and an excellent source of minerals for farm animals and even humans.
5. Molasses can also be a key ingredient for cost effective management of feeds and pastures. The calcium content of sugar cane molasses is high (up to one percent), whereas the phosphorus content is low. Cane molasses is also high in sodium, potassium, magnesium and sulphur. Beet

molasses is higher in potassium and sodium but lower in calcium.

6. Molasses also contains significant quantities of trace minerals such as copper, zinc, iron and manganese. Supplementing poor quality hay with molasses will increase feed intake and improve palatability. Microbes in the rumen break down the sugars in molasses rapidly, which extensively causes a rapid release of energy that makes molasses very useful for balancing other feeds in the dairy diet all year round.
7. Feeding molasses to farm animals will improve digestion of pastures/hay; increase milk production, help maintain body condition and appetite and result in less feed waste. Cane sugar, which has similar benefits to molasses, is an inexpensive alternative to use. It is used in the production of urea-molasses-liquid feed. This is prepared by dissolving fertilizer grade urea in 2.5 litres of water, 2% mineral mixture,1% salt and 1 million IU of vitamin A per 100 kg in 92.0kg of molasses.
8. Molasses has also been used as binding agent with urea. Uromol compound was prepared by heating urea and molasses in the ration of 1:9 (w/w) at 110°C. In vitro studies showed slower release of ammonia by uromol than by urea.
9. At NDRI, Karnal, molasses bound urea was used as a protein replacer for the lactating animals. Urea- molasses mineral block lick can be used as a good supplement to straw-based diets.

It consists of

Molasses	-	46-50%
Urea	-	10-15%
MgO	-	4-6%
CaO	-	2-3%
Bentonite	-	5-6%
Salt	-	1%
Mineral mixture	-	1%
Bone meal	-	2%
Rice bran	-	20-25%

The lick could be prepared by hot process where molasses heated up to 80°C for 20 minutes and cold process where binding material is either cement or calcium oxide in place of MgO.

Benefits in animals

1. Molasses can reduce the dusty powdery nature of some finely ground feeds. In this role, it makes a feed mixture more palatable and edible to livestock.
2. Molasses can be added to replace missing sugar and trace minerals and help with fermentation in cases of low quality forages especially with low sugar levels.

Cow and Buffalo

- Provides sugar during early pasture growth
- Promotes animal health
- Increases milk solid production
- Increases diet density when intake is reduced before calving
- Improves milk let-down
- Improves digestion of fiber
- Helps reduce heat-related stress
- Helps growth and development of young stock
- Assists pregnancy rates (condenses calving patterns)

Horses

- Combines to reduce the dust in feed

- Increases palatability
- Reduces the ability of picky horses to sort through feed

Sheep and Goats

- Prevents pregnancy toxemia

5. Filter pressmud/scum

It is produced both in gur as well as sugar making. In the process of gur making it is largely removed with the help of large ladles and it does not contain calcium (or) sulphur compounds used in sugar industries.

Moisture	5.60%
Crude protein	10-15%
Ether extract	7-11%
Crude fibre	12-20%
NFE	40-43%
Ash	15-25%
SiO_2	25.54%
Al_2O_3	31.52
CaO	25.12
MgO	0.15
P_2O_5	2.18
Sulphur	2.31
Undetermined(by difference)	15.49

Fresh and dried filter press mud may be used to supply about 20-30% of the dry matter requirement of livestock by feeding in the concentrate mixture of fodders.

6. Distillary residue/ Yeast sludge

Large amount of diluted molasses is fermented for manufacture of alcoholic drinks and power alcohol. The residue contains high level of yeast cells and nutrients. Fresh, condensed and dried residues are used for the feeding of livestock (Amata, 2014). In poultry ration it was incorporated as an ingredient to a basal cereal- free ration at 20% level with good result.

Crude protein	15-30%
Crude fibre	Traces
NFE	40-50%
EE	0.5 -2%
Ash	20-30%

It is dried at higher temperature (130-135˚C) is more digestible than fresh and condensed products.

SUGAR CANE INDUSTRY PROCESSING

Physical and Chemical Treatment of Sugarcane Waste for Use in Animal Diet

1. Physical treatment

- Grinding
- Heat treatment
- Chopping and grinding has no beneficial effect on feeding value.
- Significantly reduced feed residues in the manger.
- It increases the rate of passage of ingesta and reduces the ruminal retention time and digestibility.
- Heat Treatment
- It increases digestibility and thus the utilization of nutrients.
- In pelleting, heat and pressure treatment are used, which are expected to improve the utilization of nutrients.

2. Chemical treatment

- Biomass Treated With Urea
- Biomass Treated With Ammonia
- Treated Biomass With Calcium Oxide

- Treated Biomass With Sodium Hydroxide
- Biomass Treated With Sulphur Compounds

Biomass treated with urea

- Increase of urea, the content of cellulose and lignin decreased. This was possibly by dissolving part of lignin and the disruption of the intermolecular ester bonds between uronic acid hemicellulose and cellulose, during ammoniation (Van Soest, 1994).
- Significant mean increases were observed in vitro digestibility (IVDMD)
- Improving the nutritional value of Brachiaria brizantha after treatment with urea.

Biomass treated with ammonia

- Souza *et al* cited by Pires (2004) treated waste from sugarcane with 4% anhydrous ammonia based on dry matter.
- The waste silos that received treatment with anhydrous ammonia showed no visible signs of fungus. For silos that received treatment with NH_3, observed increased CP and IVDMD, reducing NDF and hemicellulose and minor variations in the values of ADF and cellulose.
- There was considerable increase in crude protein content and IVDMD of hay.
- Souza *et al* cited by Rosa (1998) evaluated the effects of ammoniated hay Brachiaria decumbens adding 2 and 3% anhydrous ammonia based on dry matter. Reduction was observed in the contents of NDF and hemicellulose, due to ammoniation.
- The treatment did not cause significant changes in the contents of ADF, lignin and cellulose. Ammoniation increased the in vitro digestibility of dry matter.

Biomass treated with calcium oxide

- Souza *et al* cited by Geron (2010), used the quicklime calcium oxide at a dosage of 0.5% for each 100 kg of waste from sugarcane in natural (BIN) diluted in four liters of water to provide the formation of calcium hydroxide.
- The dry matter (DM), organic matter (OM), NDF, ADF and mineral matter (MM) showed no changes between the BIN and waste-sugar hydrolyzed (BHI) kept for 28 days.
- Processing of sugarcane waste-sugar fresh with lime (CaO) for making pulp from sugarcane hydrolyzed does not change the contents of dry matter, organic matter, neutral detergent fiber and acid detergent fiber and mineral matter in relation to the original raw material (waste sugarcane in natural) during the retention period of 28 days.
- So the crushed sugarcane can be kept fresh for 28 days without the use of lime.
- The crude protein and lignin from sugarcane waste were not affected by the addition of calcium oxide.
- According to Souza *et al* cited by Balieiro Neto (2007) the use of CaO as an additive promoted reduction in fiber content and increased digestibility.
- Reduce cell wall constituents, maintaining NDF and hemicellulose after opening the silo and promote

greater stability of the chemical composition and better silage quality.

Biomass treated with sodium hydroxide

- Souza *et al* cited by Sarmento (1999) have reported that increasing IVDMD material is treated with alkaline result of the partial solubilization of hemicellulose and cellulose of the expansion, which facilitates the attack the cell wall of microorganisms such material.
- Hydrolytic alkaline reagents such as sodium hydroxide cause partial solubilization of hemicellulose and lignin, by acting on the links that connect them, and covalent ester type, which are found in grasses (Van Soest, 1994).
- Significant effect of doses of NaOH, while a reduction in NDF, ADF, cellulose, lignin and hemicellulose. Also was no effect of increasing doses of NaOH on IVDMD. Souza *et al* cited by Manzano (2000) treated waste from sugarcane with 9% H2O2 and 7% NaOH based on dry matter.
- The treatment of sugarcane waste increased the in vitro digestibility of DM and OM.
- The NDF content decreased with treatment.

Biomass treated with sulphur compounds

- Souza *et al* cited by Pires (2004) used 2.5% Na2S and 2.5% Na2S + 4% NH3 to treat waste. There was a large loss of material treated only with sulfide. However, the mold material had been removed, providing only that they are no signs of visible fungi.
- The addition of Na2S did not change the composition or IVDMD. However, for the treatments in which we used NH3, reveals increased CP and IVDMD, reducing NDF and hemicellulose and minor variations in the values of ADF and cellulose.

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Effect of Mycotoxins on Poultry Health

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Abstract

In India, poultry production has registered a magnificent growth in the last two decades. Modern poultry operations are under enormous pressure to produce chicken meat and day old chicks for the lowest possible cost. Feed conversion, growth rate, mortality, fertility and hatchability are all monitored carefully and strategies are constantly reviewed to maximize efficiencies. As production is fine-tuned, there is increased attention placed on the effect of mycotoxins on the bird. Consumption of even low levels of mycotoxins can lead to decreased feed consumption, poor growth rate and increased susceptibility to disease challenge. Higher levels lead to clinical signs such as beak and intestinal lesions, fatty liver, kidney disorders and mortality.

Key words: *Poultry, Mycotoxins,Toxicity*

1.Introduction

Mycotoxins are secondary metabolites of fungi that have the capacity to impair animal health and productivity**.** A mycotoxicosis is a disease caused by a natural toxinwhich produced by a fungus. In poultry, this usually results when toxin-producing fungi grow in grain and feed. Hundreds of mycotoxins have been identified, and many are pathogenic. The significance of mycotoxin problems in poultry is probably considerable but yet insidious. The impact on poultry production may be best measured indirectly by the improvements in weight gain, feed efficiency, pigmentation, egg production, and reproductive performance thoseaccompanied byeffective control programs for mycotoxins.

2. Common Mycotoxicosis

2.1. *Aflatoxicosis:*

The aflatoxins are toxic and carcinogenic metabolites of *Aspergillusflavus*, *A.parasiticus*, and others. Aflatoxicosis in poultry primarily affects the liver but also impair'stheimmunologic, digestive and hematopoietic function. Aflatoxin can adversely affect weight gain, feed intake, feed conversion efficiency, pigmentation, processing yield, egg production, male and female fertility and hatchability.At necropsy finding's are mainly in liver and it can be reddened due to necrosis and congestion, yellowish discolouration due to lipid accumulation. Hemorrhages may occur in liver and other tissues. The

aflatoxins are carcinogenic, but tumor formation is rare with the natural disease, probably because the birds do not live long enough for this to occur.

2.2. *Fusariotoxicosis:*

The genus *Fusarium* produces many mycotoxinswhichinjurious to poultry. The trichothecenemycotoxins produce T-2 toxin, Diacetoxyscirpenol (DAS), Deoxynivalenol (vomitoxin, DON) and Zearalenone are common mycotoxins that are relatively nontoxic for poultry. Fusariotoxicosis in poultry caused by the trichothecenes results in feed refusal and acute digestive disease. Lesions include necrosis and ulceration of the oral mucosa, reddening of the GI mucosa, mottling of the liver, atrophy of the spleen and other lymphoid organs and visceral organs hemorrhages. Other *Fusarium* mycotoxins cause defective growth of long bones.

Table 1: Different type of Mycotoxins

Mycotoxins	**Moulds**
Aspergilus toxins	
Aflatoxins B1,B2	*A. flavus and A. parasiticus*
Cyclopiazonic acid	*A. flavus*
Ochratoxins	*A. ochraceus*
Sterigmatocystin	*A. vericolor*
Penicillium toxins	
Ochratoxins	*Penicilliumviridicatum*
Citrinin	*Penicilliumcitrinum*
Fusarium toxins	
T-2 toxin,	*Fusariumtricinctum*
Diacetoxyscirpenol	*Fusariumgraminearum*
Deoxynivalenol	*Fusariumsolani*
Fumonisins B1,B2	*F. moniliforme*
Zearalenone	*F. graminearum, F. roseum*

The fumonisin mycotoxins produced by *F.verticillioides* (formerly *F.moniliform*) impair feed conversion without causing specific lesions. Moniliformin is also produced by *F.verticillioides* andwhich is cardiotoxic and nephrotoxic in poultry.

2.3. Ochratoxicosis:

Ochratoxins are quite toxic to poultry. These nephrotoxins are produced chiefly by *Penicilliumviridicatum* and *Aspergilluso chraceus* in grains and feed. Ochratoxicosis causes primarily renal disease but also affects the liver, immune system, and bone marrow. Severe intoxication maycauses reduced spontaneous activity, huddling, hypothermia, diarrhea, rapid weight lossand death. Moderate intoxication impairs weight gain, feed conversion, pigmentation, carcass yield, egg production, fertilityand hatchability.

2.4. Ergotism:

Toxic ergot alkaloids are produced by *Claviceps* spp, which are the fungi that infest thecereal grains. Rye is especially affected, but also wheat and other leading cereal grains. The mycotoxins which affect the nervous system, causing convulsive and sensory neurologic disorders; the vascular system, causing vasoconstriction and gangrene of the extremities. In chicks, the toes become discolored due to vasoconstriction and ischemia. In older birds, vasoconstriction affects the comb, wattles, face, and eyelids, which become atrophied and disfigured. In laying hens, feed consumption and egg production are reduced.

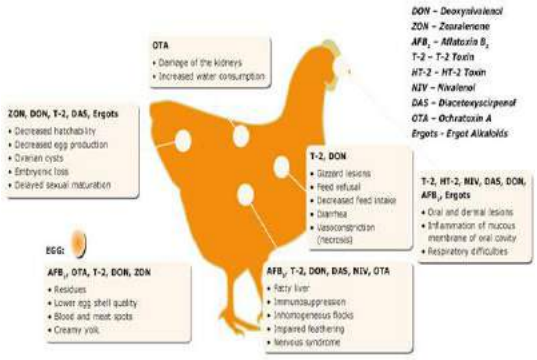
2.5. CitrininMycotoxicosis:

Citrininis produced by *Penicillium* and *Aspergillusspp* and whichis a natural contaminant of corn, rice, and other cereal grains. Citrinin causes a diuresis those results in watery fecal droppings and reductions in bodyweight gain.

2.6. OosporeinMycotoxicosis:

Oosporein is a mycotoxin produced by *Chaetomium* sppwhich causes gout and high mortality in poultry. *Chaetomium* are found on feeds and grains, including peanuts, rice, and corn. Oosporeinmycotoxicosis is seen as visceral and articular gout related to impaired renal function and increased plasma concentrations of uric acid.

Overall View of mycotoxins and its effect of poultry



2.7. **Cyclopiazonic Acid:**

Cyclopiazonic acid is a metabolite of *Aspergillusflavus*, which is the predominant producer of aflatoxin in feeds and grains. In chickens, cyclopiazonic acid causes impaired feed conversion, decreased weight gain and mortality.

2.8. **Sterigmatocystin:**

Sterigmatocystin, a biogenic precursor to aflatoxin, is hepatotoxic and hepatocarcinogenic but is less common than aflatoxin.

3. Diagnosis

Mycotoxicosis should be suspected when the history, signs and lesions are suggestive of feed intoxication, and especially when moldy ingredients or feed are evident. Feed and ingredient samples should be properly collected and promptly submitted for analysis. Mycotoxin formation can be localized in a batch of feed or grain. Multiple samples taken from different sites increase the likelihood of confirming a mycotoxin formation zone (hot spot).

4. Treatment

The toxic feed should be removed and replaced with unadulterated feed. Concurrent diseases should be treated to alleviate disease interactions and substandard management practices must be corrected. Some mycotoxins increase requirements for vitamins, trace minerals (especially selenium), protein, and lipids and can be compensated for by feed supplementation and water-based treatment.

5. Prevention

Prevention of mycotoxicosis should focus on using feed and ingredients free of mycotoxins and on management practices that prevent mold growth and mycotoxin formation during feed transport and storage. Regular inspection of feed storage and feeding systems can identify flow problems. Cleaning the feed mills and storage binsproperly. Provide sufficient ventilation in poultry houses to decreases the moisture available for fungal growth and toxin formation in the feed. Antifungal agents added to feeds to prevent fungal growth. Organic acids (propionic acid, 500–1,500 ppm [0.5–1.5 g/kg]) are effective inhibitors, but the effectiveness may be reduced by the particle size of feed ingredients. Sorbent compounds such as hydrated sodium calcium aluminosilicate (HSCAS) effectively bind and prevent absorption of aflatoxin.

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Indian agriculture:

Challenges and future prospects

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The nerves of our country still lie in its soil and agriculture is the backbone of our economy. In the next 25 years, India's population is expected to grow to 1.6 billion along with expansion and diversification of the consumption basket driven by economic growth. Agriculture will definitely play a pivotal role, in providing food security, employment generation and poverty reduction for sustainable and inclusive growth. With constraints like shrinking and degrading land and water resources, drought, flooding and global warming causing erratic weather patterns, there lies a big challenge for sustainable and profitable growth in agriculture in India. With opening of domestic market under WTO regime, challenges and prospects of Indian agriculture has increased manifold. Future road map for agriculture development should focus on integrated approach towards crop production and animal husbandry. Let us ponder over the areas which needs focused attention

INFORMATION COMMUNICATION TECHNOLOGY (ICT)

Investment in communication and technology development, their dissemination, basic infrastructure, improvement of technical capacity of producers and other players in the value chain, institutional support in core functions of production, logistics and marketing through concerted public sector support should be the key areas of activity. Coming years will be indeed an age of high edge technology. Forecasting Agricultural Output using Space, Agro-meteorology and Land-based observations will integrate land, space and weather data that will help in agro-climatic planning and will help the farmers to plan their activities and facilitate decision-making and planning at the local level. Information and Communication Technology in Agriculture will bring farmers, researchers, scientists and administrators together for the exchange of ideas and information. Electricity playing important and major roles in total development will be met by India's entry into nuclear energy programme and solar technology.

ACTIVE PUBLIC-PRIVATE PARTNERSHIPS

It is mandatory for provision of quality inputs, in particular seeds, strategies for land and water management, consolidation

and corporatization of land holdings through active and efficient public-private partnership model. It is required to lessen the already burdened public sector and bring in good agricultural practices, use of proven cost-reducing technology and finally agricultural research and extension services to fill in the gaps to tap the supply potential of India's agriculture resources.

ORGANIC FARMING

To make growing demand and supply constant there should be sustainable intensification and diversification of agriculture with organic farming given priority. Land degradation and environmental hazards could be minimized by practicing organic farming. Greater consumption of high value food and cash crops, organized food retail, bio-energy demand will create favorable business environment through enabling policies towards high value agriculture. Farmers can know chemical composition of land through lab testing to know fertility and what should they grow to make maximum profits.

BIOTECHNOLOGY IN AGRICULTURE

Application of modern biotechnological tools like DNA finger printing, tissue culture, terminator gene technology and genetic cloning will hold the key in raising the productivity. Easy access to storage facilities, better roads and speed transportation which make them tap the market efficiently, there will be rural urban continuum. Farmers will have entrepreneurial skills and productivity of labor will increase.

FARMER CENTRIC POLICIES

Government should create a favourable economic environment for increasing capital formation and farmer's own investments by removal of distortions. Improving the terms of trade with manufacturing sectors and bringing about external and domestic market reforms, backed by rationalization of domestic tax structure, easy availability of credit, insurance aginst crop-failures and other inputs, and infrastructure facilities for development ofagri-business industries and development of effective delivery systems and freeing movement of agro produce. And finally, our rich traditional knowledge and modern technology will make agriculture highly productive, improving overall socio-economic condition of farmers and attract youths in farming. Thus India would also be able to minimize agricultural imports and increase agricultural exports in the future. Therefore, with committed vision to better policy and enhanced infrastructure cascading benefits to Indian agriculture in the coming years can be brought, without compromising on the safety of environment and ecosystems.

Selection Strategies for Disease Resistance in Dairy Cattle and Buffalo

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Abstract

Selection for single trait of milk yield for many years have led to genetic erosion of genes responsible for disease resistance, adaptability etc. Therefore, selection strategies to include these neglected traits in the selection objectives of dairy animals are need of the hour for sustainable dairy in the unpredictable climatic condition.

Key words: Disease resistance, Selection for mastitis, Genetic improvement

The unique aspects of Indian dairy husbandry lies in its low to medium milk producing breeds, its scattered distribution, inadequate feeds and fodders, varying climate condition, suboptimal management and maze marketing. In this scenario, genetic improvement of dairy cattle and buffalo at farmers herd is feasible only through usual three 'tier' system. So far, selection objectives have been revolved around milk yield, and in some cases associated production traits such as fat per cent etc. Selection for disease resistance had not been in the selection objectives due to difficulty in trait measurements. As a result genetic erosion of genes responsible for disease resistance and adaptability is evident through increasing susceptibility to various diseases of high producing animals. Therefore to check the genetic death further, it is essential to induct disease resistance as one of the selection objectives in dairy animals not only to enhance the productivity but also to safeguard the welfare of the dairy animals.

Genetic basis of disease resistance

Susceptibility or resistance to disease is a threshold trait. Phenotypically, it behaves like a qualitative trait but genetically it follows quantitative traits. Measurement of trait *perse* is very difficult as there is no clear cut stage between the healthy animal and diseased animal. The term 'disease' refers to disorders caused by bacteria, viruses, parasites and feed-borne toxins, as well as to genetic disorders caused by inborn errors. These two types of diseases are described as 'disease from without' and 'disease from within', respectively

(Nicholas, 2005). In farm animal species, extensive genetic variability of disease incidence has been reported (Morris *et al*. 1998). This genetic variation prevailed in term of breed differences in disease resistance or susceptibility, and also within-breed inherited differences in disease resistance or susceptibility.

Disease from without

It can be approached via therapeutic, prophylactic and genetic resistance. The advantage of genetic resistance over other two approaches is it is permanent and cumulative. It should also be noted that resistance to disease from without is often multifactorial (polygenic) and therefore resistance is unlikely to be absolute. Thus selection for genetic resistance is likely to be used in conjunction with other disease control measures to reduce the size of challenge or to assist in treating existing infections or infestations.

Disease from within

Here, reducing the frequency of affected animals is the main commercial objective. In cases where the gene(s) controlling expression of an affected phenotype has been identified, then a gene test may be available, and selection can be applied to increase the frequency of the desirable phenotype in the population. If the genetic cause has not been identified, then removing or culling affected animals and their relatives may be effective, but the success depends on the disease incidence, and progress will become slower as the incidence falls. The bovine genome webpage http://omia.angis.org.au/ reveals that there are 367 'disorders / traits' in cattle. Of these, 65 are 'single-locus disorders or traits', with 34 being 'disorders and traits' for which the causative mutation has been identified at the DNA level.

Measurement of disease resistance:

Mastitis among other diseases has been a single, foremost condition in reducing dairy economy. Hence, augmentation of genetic resistance to mastitis in dairy animals would bring back the dairy economy in order. Recording of concerned trait is the first step in any successful breeding programme. Resistance traits (RT) are the traits by which the capacity of a cow to resist infection by udder pathogen can be measured. An ideal RT must be objectively, easily, cheaply and accurately measured, preferably early in life.

Selection for Mastitis resistance

Identification of mastitis resistance animals can be done directly based on occurrence of clinical mastitis or indirectly based on correlated traits such as somatic cell count (SCC), etc. or combination of both. Molecular markers and QTL are latest options available which could effectively be utilized in selection for disease resistance at younger age.

Direct Selection

The h^2 of clinical mastitis is 0.06-0.12. Response to selection (R) = h. i. σA. Progeny group size must be large to get more accuracy of selection (γ). For example, assume, h^2 of clinical mastitis is 0.03; Then the accuracy $\{\gamma=\sqrt{nh^2/4+(n-1)h^2}\}$ of

selection for different progeny groups will be 0.66, o.78 and 0.83 for 100, 200 and 300 progeny group size respectively. In addition σ_A of mastitis resistance is reasonably large. Thus, effective direct selection for mastitis resistance can be expected as long as proper recording and sufficiently large daughter groups are used for progeny test. There is 5% reduction in mastitis frequency among daughters of bulls with the best estimated BV for mastitis compared to daughters of bulls with the best estimated BV for milk yield.

Indirect Selection

It is based on SCC on monthly basis. The h^2 of SCC is 0.08-0.19. Efficiency of SCC as a proxy/an indicator of mastitis depends on the correlation between SCC and mastitis, progeny group size. The SCC covers sub-clinical mastitis mostly. Thus, selection on SCC alone appears to be less effective than selection directly on clinical mastitis.

Selection for other disease resistance

Apart from mastitis, there are enough evidences of heritability of different diseases which can effectively be included while formulating the selection strategies for disease resistance along with milk yield to improve the genetic architecture in totality for a given environment.

Conclusion

- Dairy population that have been genetically selected for high milk production seem to be more at risk for susceptibility to mastitis as these two traits are positively correlated.
- Data recording on health aspects of animals is an essential primary step to identify the resistant animals to diseases SCC is an excellent screening test for the presence of mastitis
- Detection of more and more QTLs for mastitis/disease resistance may make MAS a complementary or an efficient alternative selection strategy to improve the genetic disease resistance of dairy animals

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Table 1: Heritability of different traits of disease resistance

Diseases	Resistance traits	Heritability	References
Susceptibility to Nematode Parasites	Fecal egg count	0.04 (preweaning)	(Barlow and Piper, 1985)
		0.32 (post weaning)	(Morris *et al*., 2003)
	Antiparasite antibody concentration	0.3 (4-9 months) 0.22 (11-20 months) 0.3 (peripartum cow)	(Morris *et al*., 2003)
External parasitic infestation	Tick count	0.41 (Bos Taurus)	(Henshall, 2004)

Management of Cattle and Buffalo in Summer

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Heat stress occurs when the combination of environmental temperature, relative humidity, solar radiation and air movement cause the temperature of the cow and buffalo to exceed its zone of thermo-neutrality . The best way to deal with heat stress is to prevent it from occurring. High ambient temperature is also one of the major factors for reproductive failures and embryonic mortality, especially in buffaloes. Feed intake is reduced and milk yield is decreased. Buffaloes tend to be more uncomfortable because they have fewer sweat glands under the skin than cows.

Management of heat stress

Modification of the micro-environment/Use of cooling system: Cooling by reducing ambient air,temperature. Misters, high pressure foggers evaporative cooling pads and fans.

Misters: Misters the use of misters reduced water intake of the misted cows versus the unmisted controls. This was probably because the misted cows were not as hot and so transpired, sweated, less water than the unmisted controls. Misters under the shades in the loafing area should be used in conjunction with fans, at 8 to 9 feet above grade and oriented down at about 30 degrees, so that the water is blown onto the cows. The objective of misters is not to cool the air,

but to put water on the cows where its evaporation provides the cooling. Misters with fans have also been used successfully over the stall area. Cooling systems for cows in freestalls are less well understood than cooling systems for cows in drylots . However be sure that there is a roof peak opening and that the sides of the building can be opened to provide a free flow of air through the covered area.

Fans: The use of fans, particularly in areas of poor ventilation, is generally considered beneficial in preventing cows from becoming hot. One particularly critical housing area is the crowd pen prior to the milking parlor. Heat stress can occur here at relatively low environmental temperatures due to crowding. This area should have sufficient fans and misters to assure an air turnover of up to 1000 cubic feet per minute per cow. Their should also be an opening in the roof line peak to allow hot air to escape.

Sprinkling: Sprinkling (not misting) the cow with water to fully wet her body and using fans to evaporate the water cools the cow and encourages greater feed intake and milk production. Sprinklers and fans are usually placed next to the feed bunk so that the feeding area is the coolest place on the farm, helping to encourage greater feed intake.

ENHANCING THE COW'S NATURAL MECHANISM OF HEAT LOSS

Sprinkler and Fan Cooling Systems (Direct Evaporative Cooling) Sprayers in Parlor Exit Lanes Cooling Ponds/ Wallowing tanks Cooled Water Shades.

Cooled Water: It should be available as fresh and clean. Drinking water three-four times a day is a need in summer months in general farm practice. Showering/splashing of water on the body thrice a day (morning, noon and afternoon) decreases heat stress.

Wallowing : Wallowing is the cheapest and least laborious device to beat the heat in summer. Buffaloes are made to wallow in clusters in ponds, rivers, tanks or other water bodies for hours together, enables the animal to store body temperature. During wallowing rectal temperature decreases gradually but falls abruptly after the buffaloes leave the water. Wallowing becomes more effective if buffaloes remain in shade after getting out of water.

Shades: Shades The effect of shades on performance of dairy cows in dry lots demonstrate increased DMI and milk production from cows provided with shades in the loafing area. cows provided with shades in the loafing area are less hot (i.e., their body temperatures are lower) and that they feel less hot (i.e., their respiration rate is lower).

Natural Shade: Natural Shade Trees are an excellent source of shade and if given the choice cows will generally seek the protection of trees rather than man-made structures. They are not only effective blockers of solar radiation but the evaporation of moisture from leaf surfaces cools the surrounding air without appreciably interfering with air circulation.

Artificial Shade: Artificial Shade Portable shades These shades should be oriented in a North/South direction, provide about 40 to 45 square feet of shade per cow, and be about 12 to 13 feet high. There should be a raised dirt mound under the shades to prevent accumulation of moisture, and it should be groomed regularly. Mesh shadecloth is lightweight, available in numerous sizes, has reinforced grommets that make installation easy, and can be used in portable installations. When cows/buffaloes were provided with feed and water under shade that they were less hot (i.e., their body temperatures were lower) and that they felt less hot (i.e., their respiration rates were lower). Most studies on shade have used solid roof structures although shade cloths, such as

woven polypropylene, provide about 80 to 85% as much shading and are considerably less expensive.

Parlor Exit: Parlor Exit Cows should have access to fresh water at the parlor exit

even if water is available in the pens. In addition, cow activated showers as the cows walk back to their pens, have been effective at reducing heat stress. Only deliver enough water to wet the cow.

Cooling livestock buildings by integrated high pressure fogging system with air ventilation: Cooling livestock buildings by integrated high pressure fogging system with air ventilation. The main evaporative cooling methods used today are sprinkling, pad-and-fan, and fog. Sprinkling systems combine fans and spraying water from sprinklers onto the animals surface (mainly used in dairy cows housing), results in an increase of the free water surface area and consequently of the evaporation rate.

The pad-and-fan system: The pad-and-fan system is based on forcing outside air into the building through a wet pad , which humidifies and cools it only at the entrance, where the wet pad is situated. The disadvantages of the pad-and-fan system are: (a) the air must be forced through the pad, (b) significant temperature and humidity gradients, along the building, are created; (c) installation, operation and maintenance are expensive; (d) continuous operation and poor water quality cause progressive clogging of the pad, resulting in declining cooling performance.

The fogging system: The fogging system The fogging system is based on spraying the water as small droplets (in the fog range, 2-60 μ m in diameter) in order to increase the water surface in contact with the air. The free-fall velocity of the droplets is slow and they are easily carried by the air streams inside the building. These results in a high efficiency of water evaporation combined while keeping the animals and area dry.

Routine showering and keeping buffaloes in an open paddock during night are beneficial practices. Buffaloes in loose housing with a shade (may be shed) for feeding and shelter during the inclement weather and open area for night hours produce more milk than in conventional houses mismanaged and ill-equipped with cooling devices.

Immunopathology of Viral Diseases with Special Reference to Hypersensitivity Reactions

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When the balance between the protective and harmful effects of immunity shifts to immunity being the primary cause of tissue pathology and even death it is defined as *'Immunopathology'*. Immunopathology results from T-cell response, B-cell response; virus induced immune response to viral or host antigens (Autoimmunity)

Virus	Host	Site of persistent infection	Late disease
Aleutian disease virus	Mink	Macrophages	Hypergammaglobulinemia,arteritis, glomerulonephritis
Avian leukosis virus	Chickens	Widespread	Leukosis,leukemia
Equine infectious anemia virus	Horses	Macrophages	Anemia, vasculitis, glomerulonephritis
Feline immunodeficiency virus	Cats	Macrophages,lymphocytes	Opportunistic infections due to profound immunodeficiency
Human immunodeficiency virus	Humans	Macrophages,lymphocytes	Opportunistic infections due to profound immunodeficiency
Lymphocytic choriomeningitis virus	Mice	Widespread	Glomerulonephritis

Immunopathology due to:

CTLs (Cytotoxic T Lymphocytes):

(a) Lymphocytic choriomeningitis virus in mice

- Induces damage only in immunocompetent animals
- Release of proteins that recruit inflammatory cells

(b) Hepatitis B virus:

- CTL's attach to hepatocytes and induce apoptosis
- Cytokines released neutrophils and monocytes

(c) Coxsackievirus B infection: myocarditis in mice which is due to immune mediated tissue damage and virus induced cytopathology

CD4Th:

Eg: (a) Theiler's murine encephalitis virus (Picornavirus)

Immune mediated tissue damage and virus induced cytopathology and facilitate the inflammatory response. Activated macrophages and microglial cells release superoxide and nirtic oxide free radicals and lead to demyelination of the nervous system.

(b) Herpes simplex virus infection:

Herpes stromal keratitis is due to inflammatory reaction directed to uninfected cells in the stroma which is stimulated by secreted cytokines and produced by infected cells in the corneal epithelium.

Balance of Th1 & Th2 cells:

- Inappropriate Th1 or Th2 can have pathogenic effects. Vaccination which induces Th2 causes increased infiltration of eosionphils in lungs.

  Eg: Respiratory syncytial virus

B cells:

(a) Virus-antibody complexes:

- Antigen antibody complex are formed in sites inaccesible to immune system or continues in the presence of inadequate immune response and gets deposits in blood vessels,kidneys,brains.

- First described in mice (LCMV)

(b) Enhancement of viral infection:

- Eg:Dengue hemorrhagic fever-4 serotypes

Antibodies to 1 serotype do not protect against the other.Nonprotective Ab bind virus particles and facilitate uptake into nonsusceptible peripheral monocytes carrying Fc receptors. Infected monocytes produce proinflammatory cytokines ,stimulate T cells to produce more cytokines as a result there is plasma leakage and hemorrhage.

Result of virus infections:

Immunopathology can occur because of the following reasons i.e

(a)Viral infection causes polyclonal T and B cell activation.

(b)Viral infection evokes altered self or neo-antigens, the new determinants becoming targets of an immune response.

(c)Viral infection induces an anti-idiotypic response

(d) Viral infection of lymphoid cells causes aberrant unregulated expansion of immune effector cells.

Autoimmune damage caused by molecular mimicry: The presence of structural homologies between host and viral proteins is known as molecular mimicry. If viral and host determinants are similar enough to cross-react, yet different enough to break the normal immunologic tolerance, mimicry may induce autoimmune disease. Eg: (a) Coxsackie B4 virus crosses reacts with heart muscle. Onset of IDDM in children appears to be initiated by Coxsackie virus infection (b) In Sjogren's syndrome in humans, Epstein-Barr virus gene has been demonstrated in the salivary gland. (c) Neurologic disorders associated with maedi/visna and caprine arthritis-encephalitis virus infections.

Hypersensitivity reactions in viral infections:

Characteristics	Type I	Type II	Type III	Type IV
Time course				
Initiation	Minutes	Minutes	3-6 hrs	18-24 hrs
Persistence	Minutes	Dependent on antigen & antibody	Dependent on antigen & antibody	Weeks
Transfer with	IgE	IgM,IgG	IgG	T-lymphocytes
Complement required	Yes	No	Yes	No
Histamine dependent	Yes	No	Yes	No
Histology	Edema,congestion, eosinophils	Cell destruction, phagocytosis	Necrosis, neutrophils later plasma cells	Lymphocytes, macrophages, necrosis
Viral immunopathology	Minor,some erythema	Minor,some erythema	Major Acute:Fever Chronic:Immune complex disease	Major in brain, lung

Hypersensitivity Reactions:

The immune response in the form of either cell or humoral immune response sometimes elicited by antigens not associated with infectious agents may lead to severe and occasionally fatal results which are known as *hypersensitivity reactions*. Gell and Coombs in 1963 defined four types of hypersensitivity based on immunologic mechanism involved. The first three types I to III are immunoglobulin dependent reaction involving a humoral

B-lymphocyte mediated response. Type IV is a T-cell mediated hypersensitivity reaction. Over reaction to antigen is also described as 'allergy', the term originally coined by Von Pirquet. Allergic reactions occur when an already sensitized animal is re-exposed to the same allergen. In allergic conditions, foreign substances are non-harmful and thus the pathology induced is purely the result of immune response. Hypersensitivity reactions, thus demonstrate that immune response itself can cause significant pathology, as it does in some responses to infection.

The type of hypersensitivity depends on the nature of antigen and its immune response

Type 1 hypersensitivity:

Gell-Coombs implied that these disorders are driven predominantly by IgE which binds with high affinity to Fc receptors on the surface of mast cells and basophils bound . The IgE bound mast cells and basophils are referred to as sensitized. Subsequent exposure to the same allergen cross-links the membrane bound IgE on sensitized mast cells and basophils causing their degranulation and subsequent release of histamine causing an immediate reaction), leukotrienes (resulting in the more delayed symptoms) and other mediators, the classic symptoms of allergic airway disease result.

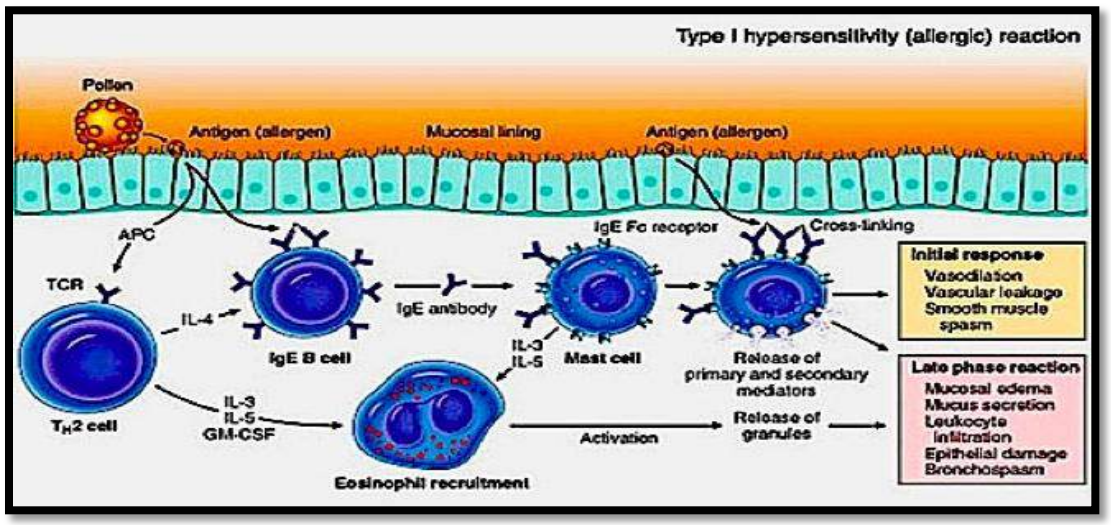


Fig: Mechanism of type 1 hypersensitivity

Primary mediators of anaphylaxis (preformed):

- Histamine – most important vasoactive amine of anaphylaxis. Enhanced vascular permeability, smooth muscle contraction, itching, increased glandular secretion.
- Serotonin – responsible for smooth muscle contractions.
- ECF – eosinophil chemotactic factor
- NCF – Neutral chemotactic factor
- Heparin – plays an important role in anaphylaxis in dogs.
- Other proteolytic enzymes

Secondary mediators-newly synthesized mediators:

- Slow reacting substances of anaphylaxis (SRS A) – Leukotrienes -*major mediator of smooth muscle constriction of bronchioles responsible for bronchial asthma.*
- Platelet activating factor (PAF) – causes broncho-constriction*, aggregation of platelets and release of vasoactive amines.*

Treatment:

- Anti histaminics such as – *Avil*
  - *Epinephrine*
  - *Corticosteroids (dexamethasone)*

Mostly seen in some adverse reactions to vaccine

Comparative features of anaphylaxis in animal and human:

Species	Shock organs	Major mediators
Avian	Respiratory tract	Serotonin, leukotrienes
Canine	Hepatic veins	Histamine, leukotrienes
Feline	Respiratorytract,intestinal tract	Histamine, leukotrienes
Swine	Respiratorytract,intestinal tract	Histamine
Equine	Respiratory tract, intestinal tract	Histamine, serotonin, kinins
Bovine	Respiratory tract	Serotonin. kinins, leukotrienes
Ovine	Respiratory tract	Histamine, serotonin, leukotrienes

Detection of Type I hypersensitivity:

(a) **Intradermal skin teasting**: Small amount of potential or suspected allergen is injected intradermally or by superficial scratching. If the animal is hypersensitive to the allergen, local mast cells will degranulate and the release of histamine will produce wheal and erythema within minutes.

(b) **Passive cutaneous anaphylaxis**: Used to detect IgE antibody in serum of the hypersensitive animals. In this test, several dilutions of serum from affected animals are injected into the skin of normal animal and the allergen is inoculated intravenously after 24-48 hrs. In positive reaction, acute inflammatory response develops at the injection sites.

(c) **Radio-allergosorbent test(RAST):** Allergen bound to beads are allowed to react with the test serum. The concentration of specific IgE bound to solid phase allergen is then measured by adding ^{125} I labeled anti-IgE and counting the bound radioactivity.

Type II hypersensitivity reaction:

Results when IgG or IgM binds to cell surface Ag and activate the complement system generating membrane attack complex and formation of pores in the foreign cells. Antibody by binding Fc receptors on Tc cells promoting ADCC(Antibody dependent cell cytotoxicity) mechanism. A number of cell including NKcells, macrophages, monocytes, neutrophils and basophils express receptors for Fc region of antibody on their surfaces. Cytotoxicity of these cells include release of lytic components, tumour necrosis factor, perforin, etc. Antibody bound to foreign red cell Ag acts as opsonin enabling phagocytic cells with Fc or C_3 receptor to bind and phagocytose the antibody coated cells. Eg: In EIA (Equine infectious anaemia), uninfected cells can be targets for type II reactions as when complement mediated lysis occurs after the virus binds to erythrocytes & contributes to the anemia seen in this disease

Fig : Replacement of bone marrow fat with dark red hemopoietic tissue

Type III hypersensitivity reaction(Immune complex-mediated cytotoxic reaction):

It is characterised by virus-Ab immune complex formation which causes inflammation and tissue damage by various mechanisms. If the reaction occurs in extravascular tissues, there is edema, inflammation and infiltration of polymorphonuclear leukocytes which is later replaced by mononuclear cells. This type of immune complex reaction constitute the classical Arthus response which is especially important in persistent viral infections. If antigen-antibody reactions occur in the blood, they produce circulating immune complexes which are found in most viral infections. The fate of the immune complexes depends on the ratio of antibody to antigen. If there is excess of antibody, each antigenic moiety becomes covered with antibody and is removed by macrophages that have receptors for the Fc component of the antibody molecule. If the amount f antibody and antigen are about equal, lattice structure develop into large aggregrates and are removed rapidly by reticuloendothelial system. Systemic immune complex activate enzymes of coagulation cascade leading to histamine release and increased vascular permeability. In such cases, fibrin gets deposited in the kidneys, lungs, adrenals and pituitary gland causing multiple thrombosis, infarcts and scattered hemorrhages. This phenomenon is known as disseminated intravascular coagulation.

Eg: Hemmorhagic fever (Arenaviruses,bunyaviruses,filoviruses,flaviviruses), kittens infected with feline infectious peritonitis virus, Fowl plaque,rabbit hemorrhagic disease ~ Arteritis due to deposits of Ab to hepatitis B virus or hepatitis B virus antigens, Equine viral Arteritis ~Rashes seen in measles and rubella is due to immune complex formation.

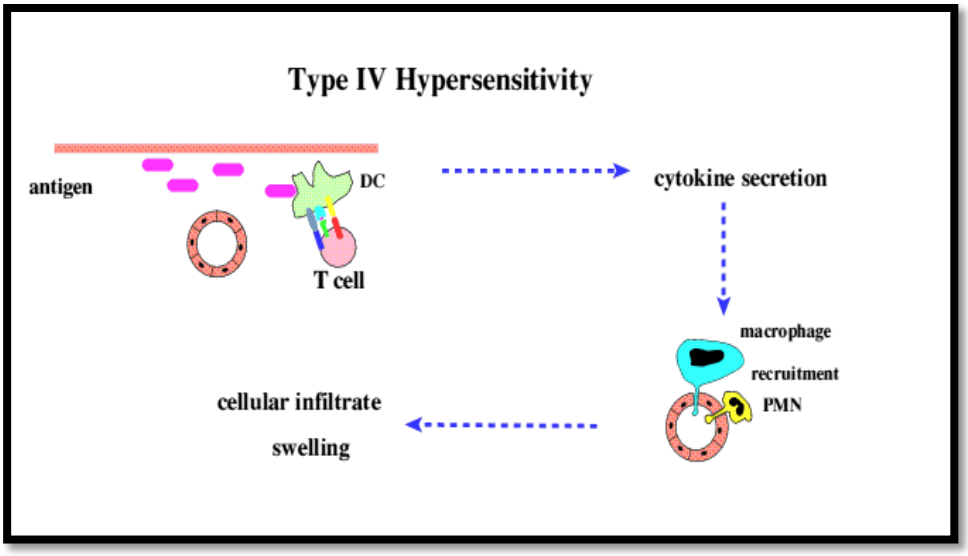
Uveitis in Canine Adenovirus

Type IV hypersensitivity reaction (Cell-mediated or Delayed hypersensitivity reaction)

- Mediated by sensitized T-lymphocytes and are manifested as inflammation, lymphocytic and macrophage infiltration.
  Eg: pathogenesis of Borna disease,influenza,parainfluenza
- Cutaneous basophil hypersensitivity(CBH):
  Eg: Chickens in response to intradermal Rous sarcoma virus

Mechanism of DTH reaction:

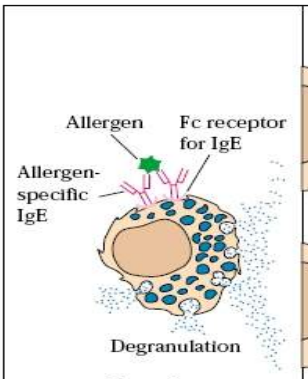
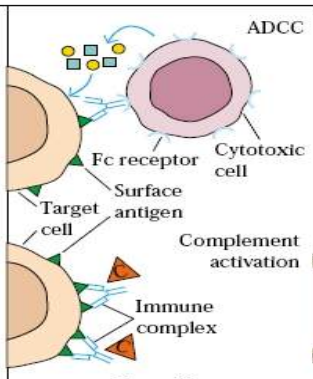
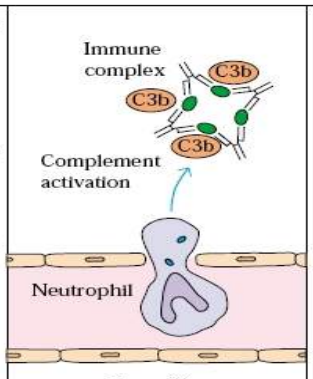
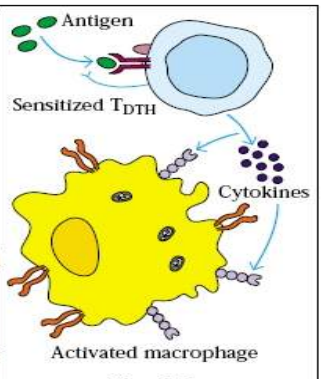
In the **sensitization phase**, the offending antigen causes activation of Th cells in 1-2 weeks.the antigen is processed by antigen presenting cells such as Langerhans cells and mcarophages and is presented to helper cell subpopulation of T cells in association with MHC class II molecules. The APC pick up the antigen from the site of infection or injection and bring them to the regional lymph nodes where T cells are activated and clonally expanded. The **effector phase** is induced by subsequent exposure with the similar antigen. Following intradermal injection, a part of the antigen is taken by the dendritic cells which migrate to the draining lymph node to activate sensitized T_{DTH} cells which in turn adhere to the endothelial cells and migrate to the antigen deposition site. Macrophage functions are the principal effector cells in DTH reaction. The cytokines secreted by the activated T_{DTH} cells cause increased expression of adhesion and MHC class II molecules. This results in the migration of blood monocytes in the surrounding tissues. They are differentiated into activated macrophages equipped with enhanced capability of phagocytosis and destruction of the microorganism. These activated macrophages also express increased levels of MHC class II and cell adhesion molecules so as to function more effectively as antigen processing cells.



Detection of type IV hypersensitivity reaction:

(a)**Lymphocyte proliferation assay**: It is carried out by incubating purified peripheral blood mononuclear cells with the antigen followed by measuring the uptake of incorporated thymidine by dividing lymphocytes.

(b)**IFN-ɤ assay**: Carried out by bioassay or sandwich ELISA using Mab (Monoclonal antibody)

 Type I	 Type II	 Type III	 Type IV
IgE-Mediated Hypersensitivity	IgG-Mediated Cytotoxic Hypersensitivity	Immune Complex-Mediated Hypersensitivity	Cell-Mediated Hypersensitivity
Ag induces crosslinking of IgE bound to mast cells and basophils with release of vasoactive mediators	Ab directed against cell surface antigens meditates cell destruction via complement activation or ADCC	Ag-Ab complexes deposited in various tissues induce complement activation and an ensuing inflammatory response mediated by massive infiltration of neutrophils	Sensitized T_H1 cells release cytokines that activate macrophages or T_C cells which mediate direct cellular damage
Typical manifestations include systemic anaphylaxis and localized anaphylaxis such as hay fever, asthma, hives, food allergies, and eczema	Typical manifestations include blood transfusion reactions, erythroblastosis fetalis, and autoimmune hemolytic anemia	Typical manifestations include localized Arthus reaction and generalized reactions such as serum sickness, necrotizing vasculitis, glomerulnephritis, rheumatoid arthritis, and systemic lupus erythematosus	Typical manifestations include contact dermatitis, tubercular lesions and graft rejection

Enterotoxemia- A Overeating Disease in Sheep

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Enterotoxemia, also known as overeating or pulpy kidney disease, is a condition caused by the absorption of a large amount of toxins from the intestines. Clostridium perfringens types C and D are bacteria normally found in the soil and as part of the normal microflora in the gastrointestinal tract of a healthy sheep. Under certain conditions, these bacteria can rapidly reproduce in the animals, producing large quantities of toxins. The toxins cause entero-colitis (inflammation of the intestine), increase the permeability of the blood vessels, and become absorbed in the blood. They circulate in the bloodstream, promoting swelling in the lungs and kidneys, giving the condition the name pulpy kidney disease. Young animals are most susceptible. Sudden and high mortality rates are concentrated in lambs and kids. Although adult animals are also susceptible to enterotoxemia, they develop immunity due to frequent exposure to these toxins. Clostridium perfringens types C & D are bacteria normally found in the soil. C. perfringens type B causes disease in several species and is especially harmful to newborns. It causes lamb dysentery and hemorrhagic enteritis in neonatal calves and foals. The two toxins associated with type B may be additive or synergistic (beta and epsilon toxin). Lamb dysentery is an acute disease of lambs

younger than 3 weeks. Many may die before signs are seen, but some newborns refuse to nurse and become listless. They often have blood tinged diarrhea and die within a few days. Mortality rates are 95%. In calves, there is acute diarrhea, abdominal pain, convulsions, and opisthotonos. Death will often occur within a few hours. In less severe cases animals survive for a few days and recovery is possible. Foals experience acute dysentery, toxemia, and rapid death. Iraq produced 90 gallons of C. perfringens (type unknown to us) as part of their biological weapons program. In 1945, Japan used a shrapnel bomb containing C. perfringens bacteria on ten Chinese victims who were tied to posts. The wounds inflicted infection and they all died a slow death from gas gangrene. Experiments such as these were carried out in Japan from 1937 to 1945 as part of Japan's ambitious biological warfare program directed by Japanese General Ishii. Historical information about Epsilon

Toxin of C. perfringens in biological warfare or bioterrorism is lacking.

Factors Associated with Enterotoxemia Outbreaks

Sheep and goats are more likely to produce too much Clostridium perfringens types C & D in the gut, and suffer from enterotoxemia, during conditions such as when kids and lambs excessively consume milk or feed with high quantities of grain while recovering from an illness or distress, natural immunity is compromised ,consequence of heavy infestations of gastrointestinal parasites, such as nematodes (worms) and coccidian ,animals have a diet rich in grains and low in dry matter (hay or green grass) ,animals have any condition or disease that slows the peristalsis (motility of the gastrointestinal tract)

AGENT

Clostridium perfringens Gram-positive bacteria, anaerobic rod found in soil, decaying matter and intestinal tract of mammals. Five types (A-E) Types B and D produce the epsilon toxin. Epsilon toxin Produced as an inactive protoxin increases intestinal permeability. Increases vascular permeability,Vascular damage and edema in brain, heart, lung and kidneys. C. perfringens D in Cattle and sheep produce neural manifestations. In Goats diarrhea is common. Mortality highest in lambs, Calves and goats non fatal subacute and chronic disease. The epsilon toxin is produced as an inactive protoxin. It becomes activated when trypsin removes a 13-residue N-terminal peptide. The toxin is known to increase intestinal permeability, and can also cause liver damage, elevate blood pressure and cause an increase in vascular permeability. This can lead to vascular damage and edema in many organs including brain, heart, lung and kidneys. The mode of action of the epsilon toxin is presently unknown, but recent work has indicated that the toxin remains on the outside of the cell, acting from the cell surface and causing an efflux of intracellular K+. This damages the cell membrane and eventually leads to cell death.

TRANSMISSION IN ANIMALS

C. perfringens is a normal intestinal inhabitant. Transmission can occur by fecal-oral route, or by ingestion of a large quantity of the bacteria through contaminated soil, water or feed. Another mode of transmission occurs when an animal's normal intestinal flora is disrupted and Clostridium are allowed to proliferate.

COMMON SIGNS OF ENTEROTOXEMIA

Most frequent in young animals, the peracute form is characterized by sudden death that occurs 12 hours after the first signs of the disease appear. Sudden death occurs only minutes after a lamb or a kid shows signs of central nervous system alteration. These signs are excitement and convulsion, followed by death, loss of appetite, abdominal discomfort, shown by kicking at the belly and arching the back,

Profuse diarrhoea (watery consistence with or without blood)

Diagnosis, Treatment, and Prevention

Diagnosis

Diagnosis is based on clinical signs, and history of sudden death that can be confirmed by necropsy. Diagnosis can be confirmed by positive identification of enterocolitis (inflammation of the intestine), Clostridium perfringens types C & D from the feaces, and gut content and kidneys cultured and isolated from the affected animals. The presence of glucosuria (high levels of sugar in the urine) can indicate enterotoxemia. Post-mortem data are important for the diagnosis of enterotoxemia. Therefore, dead animals should be submitted to necropsy and their tissues sent to a diagnostic laboratory for confirmation of the clinical diagnosis. A post-mortem examination of the large and small intestines can identify a high collection of watery blood and fibrinous clots, and numerous ulcers on the mucosa (internal layer of the intestines).

The brain and kidney tissues may show softening.

TREATMENT

Recommended treatments include Administering C & D Antitoxin according to the manufacturer's recommendation. Kids are normally treated with 5 mL of C & D Antitoxin subcutaneously. and administering penicillin. Orally administering an antacid. Administering anti-bloating medication. Administering thiamin (vitamin B1) intramuscularly. Replacing fluids intravenously or subcutaneously, and using corticosteroids. Using probiotics after treatment with antibiotics to encourage repopulation of the microflora in the rumen and guts.

PREVENTION

All animals in a herd should be vaccinated against enterotoxemia. Vaccination will reduce the chances that animals will contract enterotoxemia.

Recumbent lamb with acute type D enterotoxemia. Observe froathing by mouth. (Image; by F.A. Uzal, California)

VACCINATION PROTOCOL

Vaccinate pregnant animals with C/D & T vaccines during the fourth month of pregnancy. This procedure will enrich the colostrum (first milky secretion produced between one and three days after birth) with antibodies (specific proteins) that will protect the newborn against enterotoxemia. All young animals should be vaccinated at four weeks of age and then 30 days later. Administer another booster dose at the time of weaning. Vaccinate bucks and all adult animals once a year.

VACCINATION PROCEDURE

Do not vaccinate sheep within 21 days before slaughter. Vaccinate animals that appear healthy, avoid vaccinating animals sick with fever or worms. These animals may not respond well to the vaccine

because their immune systems may be weakened. Keep good records of the herd. Use a new needle for each animal. Disinfect the injection site with alcohol. Avoid causing stress to the animals during vaccination. Handle the herd in a peaceful manner; avoid accidents and noise when working with goats. In case of anaphylaxis caused by a vaccine, use epinephrine or its equivalent.