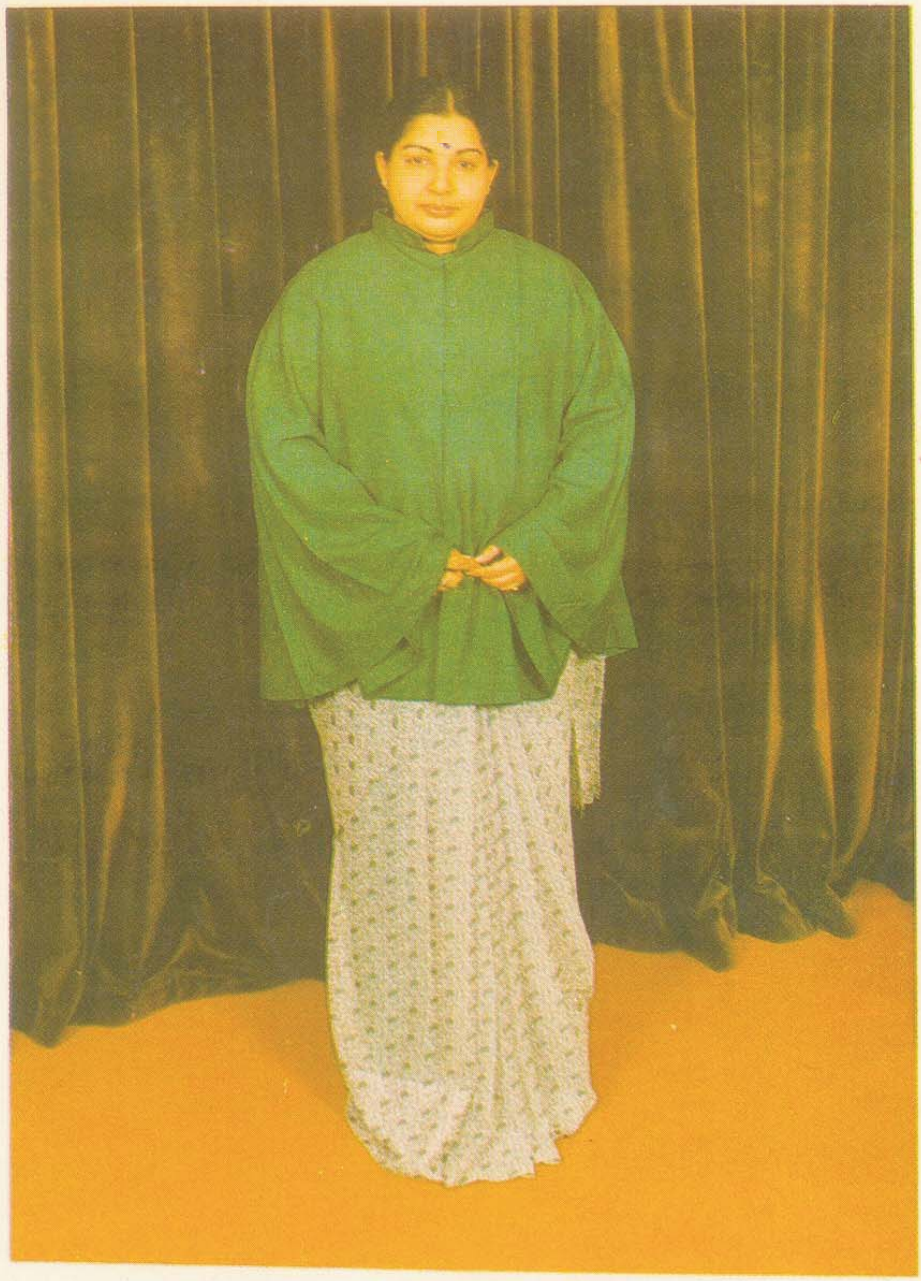
HON'BLE CHIEF MINISTER

PURATCHI THALAIVI **Dr. J. JAYALALITHA**

ENDOWMENT LECTURE



ORGANISED BY THE DIRECTORATE OF EXTENSION EDUCATION

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TAMIL NADU VETERINARY AND ANIMAL SCIENCES UNIVERSITY

First annual endowment lecture in the name of

PURATCHI THALAIVI Dr. J. JAYALALITHA

HON'BLE CHIEF MINISTER OF TAMIL NADU

By

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Organised by

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Thirumathi Periakkal Paramasivam, Tamil Pannai, Kuzhumani Post, Tiruchirapalli District, Tamil Nadu has kindly instituted Rupees One lakh to Tamil Nadu Veterinary and Animal Sciences University for an annual endowment lecture to be delivered by an eminent scientist in the honour of **PURATCHI THALAIVI Dr. J. JAYALALITHA** HON'BLE CHIEF MINISTER OF TAMIL NADU.

RECENT ADVANCES IN BIOTECHNOLOGY WITH REFERENCE TO VETERINARY SCIENCES

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I feel greatly honoured to be chosen by your University for delivering a lecture on biotechnological advances in veterinary sciences. At the out set, I wish to thank the Vice-Chancellor Dr. V. Gnanaprakasam and the organizers for giving me this opportunity. I am not a specialist in veterinary sciences, nor do I have any direct experience of handling this sector of the activity except that I had been deeply involved in the development of pharmaceutical industry and subsequently in the biotechnology industry in the country as a Science and Technology Manager for more than two decades. My involvement in these assignments had been project formulation, scrutiny, watch on implementation, generation of basic demand and consumption data and estimation of growth pattern in various sub-sectors besides identification of areas for investment opportunities. In today's meeting I have looked at the subject matter from my background and would like to discuss with you my thinking in this important area.

Our association with animals is as old as our civilization. Human developments in the initial phase of our civilization were largely dependent upon supports received from animals in terms of food, draught power and security. With the progress in industrialization. the developed countries have moved away from animals in terms of deriving advantage of draught power and security, but the developing countries including India are still largely dependent upon livestock for receiving major life support needs. In India, as per the latest estimate (1), the GNP at Current Prices during 1991 – 92 was Rupees 541888 crores (Rs. 212316 crores at 1980-81 prices) of which the contribution from agriculture and fishing accounted for nearly 30%. This clearly indicates the bias of development of Indian economy towards agriculture related activities. The *per capita* national income at current prices rose from Rs. 4934 in 1990-91 to Rs. 5529 in 1991-92 although in real terms at 1980-81 prices, the same shrunk to

some extent. The contribution of outputs from the livestock sector during 1989-90 (2) was Rs. 38178 crores at current prices (Rs, 16808 crores at 1980-81 prices) and is thus considered substantial. Every developmental support at National and State level needs therefore to be provided and strengthened to develop this sector of economic activity. To highlight the significance of this sector further, the value of outputs of major products during 1989-90 is indicated below (Table 1).

Table 1 Value of live Livestock : 1989 - 90

Particulars	Value Rs. Crores, at current prices
Milk and Milk group	24805
Meat group	5920
Eggs	1189
Wool and Hair	134
Dung and Others	6130
Total	38178

The value of draught power which is in addition to the above, is estimated to be over Rs. 5000 crores annually.

The objectives of the country towards livestock are therefore to be centred towards improving the productivity of the animals and increasing the animal power to support higher food production. These would include selection and genetic improvement of animals with higher performance; provide health-care support in terms of diagnosis of diseases, vaccination and therapy; plan for development of sufficient and cheaper nutrients; provide management expertise for scientifically managing the livestock; and develop expertise in scientifically processing the produce for longer shelf life and quality. In this discussion of today, I have concentrated upon **cattle** and **buffalo**, and the areas touched upon are current status as well as the future scenario on reproduction for genetic improvement including transgenics, animal health, and finally some aspects of food and feed improvement related to the above.

It has been estimated that the country possesses 23% of the world bovine population and yet the total milk production was 42.3 million tonnes in 1986 and 51.5 million tonnes in 1990, which was only 8%

of the world milk production (3, 4). According to FAO, 1985 (5) there were 182 million cattle, 64.5 million buffalo, 81.5 million goat, 41.3 million sheep and 8.8 million chicken in India. In 1986, the number of milching cows were only 56 million out of around 184 million total cattle population, with an average production of 187 kgs. of milk per cattle per annum (3). In order to improve the indigenous breeds, the local cattle had been bred with the semen of well-defined sires, either imported or procured locally after stringent selection. In the earlier years, semen of imported sires were used and the cross bred animals raised upto 1986 was estimated to be 10 million of which 3.8 million were milching cows producing about 1600 kgs of milk per animal per annum. The population of buffalo during the period was 62 million of which 34 million were milching buffaloes and the average production was 1000 kgs of milk per annum (3).

It has been estimated that 18% of the total stock of cattle and buffaloes belong to well-defined breeds while the remaining animals are non-descript ones.

The programmes launched in the country during the last four to five decades for improving the milk production had made some success in that the *per capita* milk consumption rose to 154 gms. in 1984, although this is much lower than the recommended level of 280 gms. as estimated by the Nutritional Advisory Committee (NAC) of the Government of India (3).

There had been marked improvement in the development of support facilities as well as processing infrastructure for milk since independence. Upto 1986, the country had 140 cattle breeding farms, 40 exotic animal farms and 48 frozen semen banks in operation. The processing capacity for milk in 1983-84 rose to 4,77,000 tonnes annually (3).

There is however, concern that there is a significant gap in the requirement of nutrients to the livestock to the tune of 44%. It has been argued that if the existing livestock could be given adequate nutrients, the milk production could be increased by over 40–50% straightway. The high milching animals would no doubt contribute significantly in such situations (3).

Taking into consideration the FAO figures of 1985 on the production of milk, meat etc. as the base and assuming a modest 5% annual increase in quantitative production and about 3% increase in *per capita* consumption, an estimate has been made about the production of livestock derived food and related products for 2000 AD as in Table 2.

Table 2 : Estimated requirements of outputs from livestock

Particulars	Production in 1985 (5) (Million Metric Tonnes)	Estimated production 2000 AD (Million Metric Tonnes)
Milk	41	85
Meat (excluding poultry)	1	2
Grease	38	79
Wool	25.4	53

These production levels are smaller when compared with *per capita* consumption of developed countries and even lower than that of many developing countries. The milk production from all sources by 2000 AD has been estimated by the Govt. at 80 million metric tonnes with the wishful thinking that many livestock improvement programmes taken up by the Govt. would show significant positive contribution. Indeed the milk production by 2000 AD should have been 102 million metric tonnes on an average daily *per capita* consumption of 280 gms. recommended by the NAC. This would not however be possible to attain.

Even the above estimated production levels would not materialise automatically, and Herculean tasks with painstaking methodical programmes would have to be chalked out to implement and materialise these. Let us therefore discuss what are our strengths in moving towards attainment of these targets.

GENETIC IMPROVEMENT

Natural Breeding :

Bulk of the milk, meat, wool and animal grease for 2000 AD would be obtained from the already existing livestock improvement programmes. These would however be further intensified. The country has made genetic selection experiments for the last 60 years and the results indicate the process of improvement of native breeds is very slow (6) Earlier, the selection of sires was on the basis of pedigree and not on the testing of progeny. Another study indicated that it would take around 90 years to double the milk production of the herd with an average lactation yield of 800 kgs. (7) Improvement through selection has not been rewarding mainly

due to the low level of milk production of Indian breeds. Therefore cross-breeding was considered the most suited alternative to introduce genes of desired traits in hybrids.

Cross breeding :

During 1961 to 1978, pure lines of Jersey, Holstein Friesian, Red Dane, Brown Swiss, Guernsey and Ayrshire to the extent of 7500 numbers were imported and distributed to the State Govts.; in addition the Central Govt. established several farms for multiplying the exotic breeds. During the 4th Plan preservation of well-defined breeds of local origin had started; in addition progeny-testing programmes were also initiated. By 1990, the country had established 330 cattle and buffalo farms. Breeding bulls were also produced in some of these farms The sires are currently being ranked on the basis of their relative breeding value, and semens are collected from the selected elite sires for breeding programmes. Currently there are 5500 artificial insemination centres (AI) distributed all over India. There are also 48 semen freezing stations in the country (3).

The major centres where considerable cross-breeding work was carried out include the Military Farms, National Agricultural Institute, Allahabad; National Dairy Research Institute, Karnal; and the All India Coordinated Research project on Cattle with 5 Centres with their Coordination Unit at Indian Veterinary Research Institute (IVRI), Izatnagar (7).

The cross-breeding experiments clearly demonstrated that Holstein crosses had outyielded all other breeds and breed crosses. The Friesians as well as the Jersey crosses had also performed well, though these were of lower performance than the Holstein crosses. The conclusions that were that it would be logical to maximise the breeds with Holstein crosses Of the Indian breeds, the best ones found were Sahiwal, Hariana, Gir, Red-Sindhi and Tharparkar.

Artificial insemination, semen freezing and semen dilution techniques were standardised and these made significant impact on the genetic improvement of livestock during the last eight decades through cross breeding programmes. With these techniques, improvement was possible only through the male path. These techniques would continue to dominate the scene during the next one decade both on cattle and buffalo. Although the main hurdle of these techniques is the low rate of female reproduction, the gains of success outweigh the natural insemination and natural selection processes. According to a recent estimate (7), out of the total production of milk from the cows, the contribution from cross bred cattle had risen from 18.5% in 1982 to 26.9% in 1990 and is expected to be 46% in 2000 A. D. Similar more contributions are anticipated from buffaloes also.

Multiple Ovulation and Embryo Transfer (MOET) :

Cross-breeding through artificial insemination (which is the MALE PATH) has its limitations in the speed of exotic breeding. Quantum jump in numbers is possible by standardising the FEMALE PATH of superovulation and coupling this technique with the MALE PATH, and further standardising the techniques of embryo transfer to foster mothers in "heat". Multiple Ovulation and Embryo Transfer (MOET) includes super-ovulation, recovery, transfer, culture and cryo-preservation of embryos.

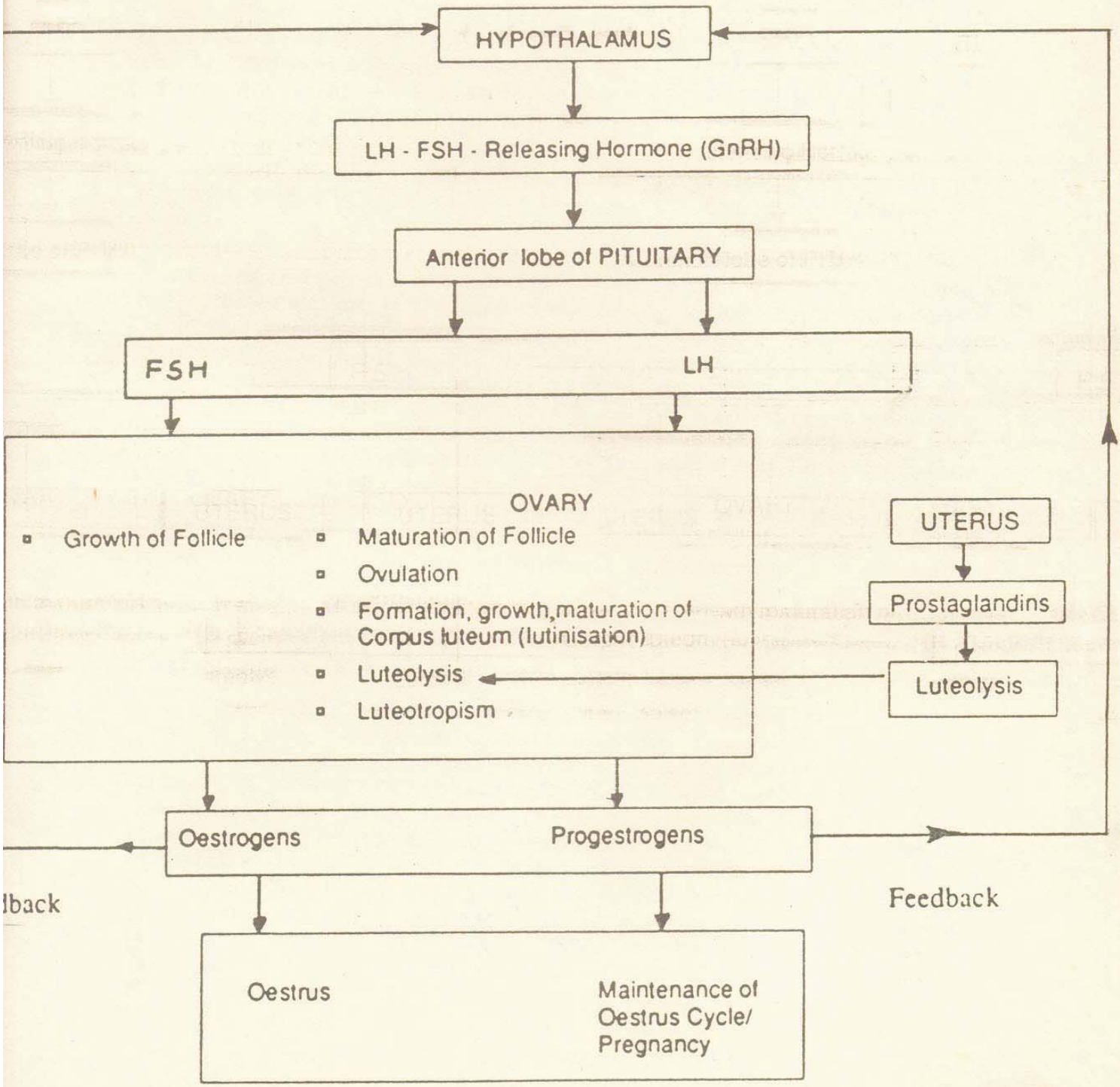
In order to discuss MOET, let us recapitulate the natural endocrinological events of reproduction (Fig. I).

Schally and Guillemin (8, 9) had independently discovered in 1971 the gonadotropin releasing hormone (GnRH), a decapeptide from porcine and ovine hypothalami respectively, and with their discovery the mechanism of regulation of reproduction of vertebrates was considerably understood. GnRH regulates the release of gonadotropins namely Follicle Stimulating Hormone (FSH) and Luteinising Hormone (LH) from the pituitary into the circulation which in turn causes the final gonadal maturation and ovulation in females Though the function of ovary are substantially controlled by the pituitary hormones, part of the function of the ovary of luteolysis is controlled by prostaglandins which are synthesised in the uterus and which find their way into the ovary through the ovarian artery or vein and induces the luteolytic process. During normal reproduction, a female produces only one ovule. The reproductive system can be stimulated either by activating the hypothalamus or the pituitary or by incorporating the gonadotropic hormones as well as the prosta-glandins in the right proportion on a right synchronization schedule. The reproductive endocrinology can be stimulated by injecting GnRH or biologically active analogues, and these would stimulate the anterior lobe of the pituitary which inturn would release additional quantities of FSH and LH, These gonadotropic hormones would inturn activate the ovary for super ovulation. FSH and LH responses could also be brought about by Pregnant Mare's Serum Gonadotropin (PMSG). The super-ovulation response is variable from animal to animal. The schedule that has been standardised in the country on cow and buffalo is to superovulate the donor using various combinations of FSH, PMSG, GnRH and Prostaglandin F_2 alpha. After superovulation, the animals are bred using semen of progeny tested sires. The embryos formed are collected by using normal saline, evaluated and then either transferred to foster mothers or stored in refrigerated conditions.

The Department of Biotechnology (DBT) took up the programme for cattle Herd Improvement through MOET in 1986-87. The programme was meant to improve the livestock faster and replace the poor quality of animal population with superior germplasm. The project

Figure - I

NATURAL SEQUENCE OF ENDOCRINOLOGICAL EVENTS OF REPRODUCTION (SCHEMATIC)



had been implemented with NDDB, Anand as the lead agency collaborating with NII, New Delhi; NDRI, Karnal; IVRI, Izatnagar; CFSP&TI, Hessarghatta. There are 25 state centres throughout the country which would provide the technology to the farmers. The total outlay of the project was Rs. 1684.69 lakhs and the project came to an end in March 1992.

Through the project, the basic MOET and cryopreservation were standardised in cattle and buffaloes. Viable embryo recovery rates achieved were 4 – 5 embryos per flush in cows and 2 - 4 in buffaloes. Techniques for the production of PMSG and FSH were standardised which would reduce the cost of imported chemicals to a considerable extent. An EIA kit for the detection of pregnancy in buffaloes was developed. An embryo sexing method was also developed (10).

Based on the techniques standardised, an estimate of the cost of ET cows and ET buffaloes have been worked out as in Table–3.

It is seen from the table that much further developmental work is to be carried out to make the cost of ET animals lower for enabling faster dissemination of the technology to the villages. The cost could be further reduced if methods for sexing would be very correctly determined and methods for breeding the desired sex of calves could be standardised. In this connection a news item appeared on Jan. 28, 1993 (11) described that a company in Eastern England in collaboration with Britain Institute of Animal Physiology and Genetic Research and the US Deptt. of Agriculture (USDA) had been successful in producing world's first calves of predetermined sex. The process described involves staining the sperms with a fluorescent dye and passing these through a laser beam that enables the scientists to separate the sperms bearing X chromosomes from the sperms bearing Y chromosomes. The separated sperms could then be used to breed the desired animals of predetermined sex. Such scientific work needs also to be initiated in the country.

Embryo splitting and cloning:

The MOET programmes would be very successful if embryos could be splitted to obtain cloned offsprings from selected embryos. The first successful experiments in this direction were the successful transfer of nucleus from blastomeres to enucleated zygotes in mice (12) and subsequently in the oocytes in sheep (13). The techniques have thereafter been perfected to a considerable extent. It is now possible to transfer cells of a multiple extent. It is now possible to transfer cells of a multiple cell embryo into the cytoplasm of enucleated oocytes. The oocytes could be collected even from the slaughter house from the sacrificed animals. Such oocytes have the ability of supporting the development of transplanted donor nucleus cells towards the formation of a new embryo (14).

Table 3 : Estimated cost of Embryo Transferred (ET) cow/buffalo (1992–93)

	Particulars	Cow (Rs)	Buffalo (Rs.)
1.	One flushing of superior embryos* ...	2500—2600	2500—2600
2.	Freezing cost of one flush ...	120— 150	120— 150
3.	Cost of one embryo transfer including transportation and incidentals ...	300— 350	300— 350
4.	Cost of successful single pregnancy assuming**		
	a) Two viable embryos per cow per flush ...	4075—4250	—
	b) Four viable empryos per cow per flush	2450 - 2500	—
	c) One and a half viable embryos per buffalo per flush ...	—	5860 - 6230
	d) Three viable embryos per buffalo per flush	--	3340—3630
5.	Raising cost of calves ...	3800—4000	4800- 5000
6.	Total cost of ET animal at lactation/maturity		
	a) Cattle i) Lower range ...	6250—6500	—
	ii) Upper range ...	7850—8250	
	b) Buffalo i) Lower range ...	—	8140—8630
	ii) Upper range ...		10660—11230
7.	Market price of		
	a) Cross bred cow/buffalo at lactation	Rs. 7500 - 10000	
	b) Cattle bull at maturity	Rs. 7000 - 7500	
	c) Buffalo bull at maturity	Rs. 7000 — 7500	

*The cost increases by about 20% if imported semen is used.

**Success rate of pregnancy in cows and buffaloes assumed as 40 and 35 percent respectively.

The above technique has the following 5 main steps :

- ★ Collection of Recipient Oocyte from the ovaries of say, staughter house and maturation in a medium in carbon dioxide incubator.
- ★ Sixteen and Thirty Two Cell Donor Embryo Collection from super cows after breeding and isolation of blastomeres with micromanipulators.
- ★ Micromanipulation of enucleated oocytes and donor blastomere cells. In this step, the first polar body along with a portion of the membrane bound cytoplasm of the oocyte opposite to polar body is inserted in an enucleation pipette and thereafter a donor blastomere is placed into the perivitelline space against the enucleated oocyte.
- ★ Fusion aud activation of oocyte-blastomere pairs in a cell fusion media in an electric field.
- ★ Culture of fused and activated oocyte-blastomere pairs in oviducts of intermediate hosts like sheep and rabbit.

The oviducts after maturation for about a week are removed and the embryos are isolated, graded and transformed to recipients to produce identical calves.

It has been estimated that from one original embryo nearly 30,000 or more identical clones could be produced. The embryo cloning steps are depicted schematically (14) in Fig. 2. The technique is being worked upon and it would take some time for its being fully perfected for routine use for breeding pnrposes.

Transgenic Animals :

In animal kingdom only the zygote formed by the fusion of an egg (ova) with a sperm cell can develop into a fully formed animal. All the blastomeres of 16 or 32 or even 64 or later stage embryos are identical in genetic make-up and and could also be manipulated to evolve into a full animal. The foreign genes inserted into the embryos or sperm or ova are expected to be inherited by the full grown animal and the foreign genes if expressed, are expected to elicit the gene products or the gene behaviour.

Foreign genes could be inserted into the chromosomes of an animal by various methods such as by using retroviruses as the vectors or by directly inserting copies of DNA pieces into the cells

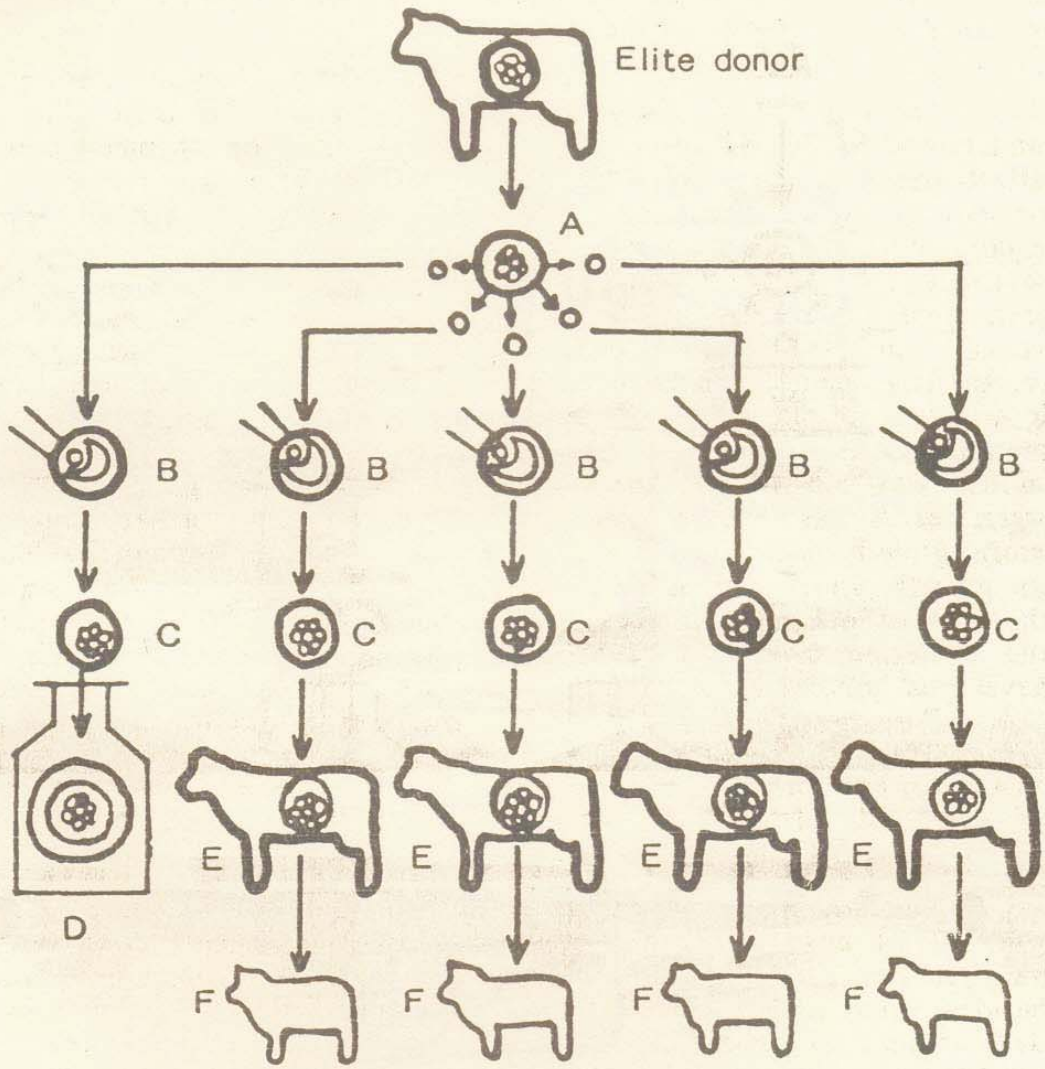
using a micropipette in the eggs before or just after fertilization. Methods have also been published wherein sperms have been treated (soaked) with foreign DNA fragments and the treated sperms have been used to fertilize the eggs. In the retrovirus method, the chosen vector is modified at certain regions of its genome by replacing the harmful genes and inserting the foreign genes to be incorporated in to the animal. The reconstructed vector is then used to infect the animal cells either at its embryonic stage or at some selected tissues to transfer the foreign gene (Figure-3).

The control of foreign genes into animal cells is not yet well understood. The insertion of foreign gene into the chromosomes can affect the expression in either way-it can block certain natural action or even turn on unwanted oncogenes producing tumors. However, experiments are being carried out all over the world to understand precisely the insertion and regulation mechanism of foreign genes into animal cells. Several unrelated foreign genes have been inserted into animals since 1981. Of the earlier successful and significant gene transfers in animals, mention may be made of rabbit globin gene expressed in a mouse, and the insertion of a growth hormone gene fused to a metallothionin gene again into a mouse. In the later experiment, the mouse was pushed to express the growth hormone gene in situations whenever it was exposed to certain heavy metals. This caused synthesis of more growth hormones in its body, thereby increasing and accelerating its growth under the influence of growth hormone to more than twice the size of the normal mice. A news appeared on 28-1-93 (11) informing the evolution of a transgenic sheep (named as Tracy), which was developed jointly by an Edinburgh based company and an institute in U.K. The sheep has now produced a second generation transgenic lamb (Tracy Two) which continues to secrete like its mother, alpha-1 antitrypsin (AAT) in its milk in quantities of 30 gms per litre of milk. The value of each litre of this milk is about 4000 pound sterling.

The beginning in this area has thus been well made. The future will tell us how would the outcome be. No transgenic cow or buffalo has yet been reported. Interest has however been directed to produce besides value added chemicals (to be secreted through the milk mode), development of genetically engineered animals which would be more productive and resistant to stress conditions for example disease resistance. Much work has been done on the use of bovine growth hormones (bGH), the genes of which have been cloned in unnatural hosts (microbes) and large quantities of recombinant material have been produced. Use of bGH on animals have shown faster, leaner and larger growth and the lactating animals have produced larger quantities of, milk (15). However the use of bGH in animals have produced several deficiencies in animals like udder inflammation (mastitis) increased subsequent sterility rates. marked rise in diabetic symptoms etc. Moreover

Figure - 2

Schematic diagram of embryo splitting and cloning



A = Donor Embryo

B = Transfer of donor cell to enucleated oocyte

C = Cloned embryos

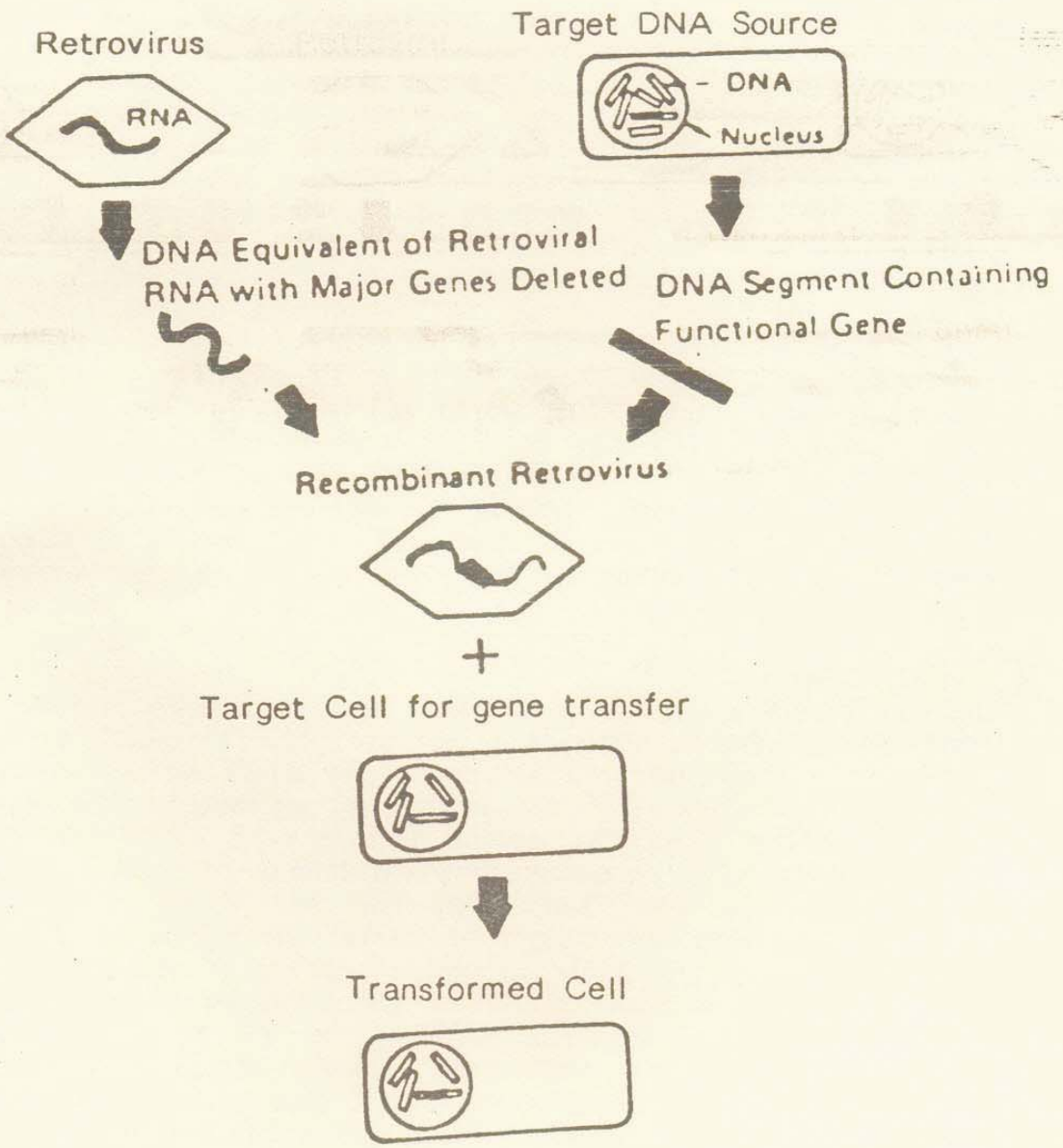
D = Frozen embryos

E = Cloned embryos transferred to surrogate mother cows.

F = Resulting identical calves.

Figure - 3

Schematic diagram of retrovirus mediated gene transfer



the milk produced has contained several other protein bodies like insulin like growth factor (IGF1–) etc. the effects on long term consumption of which on human health are not yet known. The work on the development of transgenic cattle or buffalo containing bGH gene would therefore have to take into consideration the untoward effects of bGH before economically valuable but socially acceptable animals could be produced. We have thus to go a long way before the transgenic animals of real value would be produced for our use.

Animal Health :

Increased livestock production with improved breed has to have strong infrastructure and material support for the maintenance of good health of animals. Animal health in India is affected by the non-availability of abundant doses of cheap and effective vaccines, chemotherapeutics, lack of diagnostic facilities, illiteracy and general lack of apathy for disease reporting, the depressed status of the small farmers owning the major portion of the livestock, low productivity of majority of the animals, and finally the absence of adequate organised marketing facilites of the livestock products which hinders the enthusiasm of the owner for increased attention. The number of animals in individual dairy farms usually vary between 3 to 25 with large numbers of them having only 5 to 10 animals. Very few organised farms exist with animals beyond 100 to 300 numbers. The animal health care system is thus to cater primarily to the needs of the small farms. With increased practice of cross breeding, the value of crossbred livestock with higher performance have gone up significantly; however, these animals are more susceptible to diseases also. The value of animal health care specially in the crossbred sector has thus gone up significantly during the last two decades.

Animal husbandry is a State subject in this country and this veterinary doctors (VETS) are the focal points for animal improvement programmes. The VETS are responsible for health care work; besides they are also responsible for animal breeding and advisory work. Usually the VETS have a small number of support staff for executing the tasks related to animal husbandry practices. The existing available supports for VETS are inadequate and need to be substantially augmented to enable the farmer to receive more personalised attention on their animals by the professionals. Moreover due to the nature of veterinary culture developed in the country, usually the livestock attendant who is not a qualified veterinary doctor does the main execution work in the villages like visiting the cattle shed and the diseased animals etc., and develop more human link with the farmers. In this background, the animal health support needs to be looked into.

Vaccines are by far easiest and cheapest modes of prevention and therefore as far as possible animals are required to be protected by active vaccination with as many animal vaccines as are available.

The major livestock diseases in India affecting cattle and buffaloes are Rinderpest, Animal Rabies, Foot and Mouth Disease (FMD), Anthrax, Black Quarter, Pneumonia, Haemorrhagic Septicemia, Lung worm, Liver flukes, Theileriasis, Tuberculosis, Trypnosomiasis, Dermatitis, etc.

Good progress has been made in producing vaccines agains major diseases like rinderpest, FMD, anthrax, black quarter, rabies and Haemorrhagic septicemia. Modern plants have been set up in several of these areas and in this connection the indigenous development at the Indian Immunologicals, Hyderabad producing BHK–21 tissue culture based rabies vaccine is significant. This unit along with other manufacturers are also making tissue culture based FMD and rinderpest vaccines, and are capable of meeting the full requirement of the country. Vaccine production centres have been established in 17 States in the country and these units produce several million doses of vaccines annually. Research work is also continuing on evolving vaccines against haemoprotista diseases like Theileriasis and Anaplasmosis. Scientists of National Dairy Development Board (NDDB) have already commercially introduced a tissue culture based vaccine against Theileriasis through Indian Immunologicals, Hyderabad. Another group in Punjab is also active in developing an efficient vaccine against Theileriasis. Research work on the production of a genetically engineered vaccinia virus incorporating the genes of rabies antigen has also been initiated in Delhi.

In addition, the roles of vectors in the transmission of diseases are being increasingly understood. Methods for the control of the ectoparasites like ticks, mites, lice and flies which cause large damage to livestock resulting severe economic loss to the owner are being known and products like synthetic pyrethroids are being used for controlling them. Similarly several chemotherapeutics are available for treating fascioliasis (like liver flukes) and worm infections. In addition, a part of the vast spectra of chemotherapeutics including antibiotics, synthetics, glandular and plant products available as therapeutics for human beings are also being used in veterinary practices in situations whenever these are warranted.

As regards the animal disease diagnostic devices, this area is currently receiving increased attention. The present level of development is low and the infrastructure is not yet adequate. There are only few diagnostic methods developed locally and these are not yet easily accessible. Imported diagnostic kits are available for detection of

certain diseases but these are expensive. There is a pressing need for having kits for the quick and reliable detection of bovine tuberculosis, brucellosis, enterotoxeamia, rhinotracheitist, heileriasis, rinderpest, rabies and anthrax. In addition, there is a need to detect chorionic gonadotropin in urine/blood and to quantify the hormonal levels in blood specially the LH surge, progesterone levels etc., as we would be intensifying practicing of MOET for raising elite livestocks. Several techniques like RIA, EIA, ELISA. latex/particle agglutination, and colour/spot development/disappearance modes are used for the detection of infective agents and usually blood/sera are used as the field condition kits or do it-yourself kits are not yet available, research projects are being centrally funded to develop kits suiting the local needs. Within a couple of years, this area is expected to develop significantly.

Animal Nutrition :

Animals with high genetic potential for increased production and performance characteristics as well as the capabilities for adaptation to the tropical climate would be generated through cross-breeding or through MOET. These animals would have to be provided with adequate nutritional inputs to make them perform their best on a sustained basis. Indigenous livestock with higher productivities would also require more feed with high nutritional value. The cost of livestock feed forms the major part of the total cost of production of milk as well as meat. It is therefore important for the country as well as for the individual farmer to optimise the production of forage and the intake of nutrients by the animals. In this direction, the absorption of nutrients from the digestive tract into the body of the animals along with feed conversion efficiency would also play an important role towards increasing the output and performance. It has been concluded that on perusal of the demand and supply situation of feed and fodder, there is a deficiency of 36% in green fodder, 40% in dry fodder and 44% in concentrates (16). Therefore, to provide high quality feed at optimum level to the livestock in the existing situation, the correct approach would be not only to improve the quality and utilisation of the available fodder, feed, agricultural residues and utilizable industrial by-products but study would also have to be made to manipulate the metabolic pathways of animals and to use bio-regulators to provide them maximum nutrition.

Although forages provide substantial part of the livestock feed, most forages naturally available have one or the other anti-nutritional factors. These include protease inhibitors, higher concentration of oxalates, cyanogens, tannins, phyto-haemoagglutinins etc. These substances may induce chronic intoxication and reduce feed efficiency. It is anticipated

that the recombinant DNA technology would be useful to modify the genome of plants to enable reduction or removal of the antinutritional compounds in future. The genetic variations found in the conventional plant breeding offers relatively narrow possibilities for the development of hybrids with desired characteristics. The understanding of the mechanisms of regulation of genes is gradually promoting the plant breeders to use the gene transfer techniques for the modification and selection of plants with increased desirable properties. Plants selected from the natural sources through the breeder selections as well as the new trans genics developed through genetic manipulations may have the desired qualities in the forages with minimum contents of antinutritional factors. It may also be possible to introduce other desirable traits such as higher protein content, better aminoacid profile, improved stress tolerance characteristics etc. through genetic manipulation. The new techniques that would be used for such manipulation would include besides the gene transfer techniques, cell and tissue culture or plants, clonal multiplication and use of somaclonal variation techniques. All over the world considerable work is being done to generate transgenic plants and several laboratories have claimed to have produced transgenics. However, as of date only one genetically engineerd species namely tomato plant has been permitted by the US government for use by the society. Besides other desirable properties, the plants produce tomatoes with delayed ripening characteristics. Considerable safety data are required to be generated before transgenic plants are permitted to be extensively used. Besides, transgenic-plants often land up with undesirable characteristics like sterility, and considerable variation in performance. It is thus evident that many years of sustained research would have to be carried out before quality improvement by genetic engineering becomes a reality. Improvement in the quality of the fodder plants by biotechnological methods may get delayed as priorities for any society are for improvement of food grains and vegetables.

The conventional ensiling process practiced in the country often reduce the quality of the forage. Controlled fermentation in the forage during storage is undoubtedly a very important area. This is related with efficient nutrient conservation and utilisation. It has been observed that the daily dry matter losses are between 1.5 – 3% for every 10 degree rise in silage temperature above ambient level (17). Therefore, efforts are to be made to control losses through uncontrolled fermentation by the external incorporation of microbial and other additives. Additives containing cultures of lactic acid bacteria have shown increased acceptance, as they are not only safe to handle but they can also accelerate the fermentation in the forages to research a low and stable pH quicker than would occur with spontaneous fermentation (18). It has been suggested that mixed cultures of lacto-bacillus are preferred over a single genus for such conversions (17). The properties which are imparted to the silage quality by use of mixed cultures, include rapid acid

formation, dominance of the cultures over epiphytic flora and their ability to utilise starch. The culture is active aerobically as well as anaerobically, and is resitant to bacterial phages. These do not have any proteolytic activity and have no action upon organic acids.

Nutrition in the Rumen :

The rumen microbial population plays a major role in the digestion of feeds by the cattle and buffaloes. During the current years, interests have been focussed on modifying the rumen microbial system by recombinant DNA technology with the objective of enhancing efficiency of feed conversion in the ruminants. The genetic modification of rumen microflora is carried out with the objectives of enhancing cellulolytic activity, reducing the deaminase and proteolytic activities, increasing the biuretase activity, enhancing microbial production of desired amino acids and imparting capabilities for nitrogen fixation. The molecular genetics of rumen cellulase system are very complex (19). The cloning experiments of genes with several rumen bacteria expressed in *E. Coli* have shown that the expression of the sequenced foreign genes are in agreement with the control mechanism of the host organism. Further, the data available from these experiments indicated that the sequences of enzymes which are involved in the degradation of structural polysaccharides of plant cells are many and at least 17 different enzymes are involved directly in the degradation of cellulose and lignocelluloses. These clearly reflect the great complexity of the structural rigidity of the lignocellulosic cell wall substrates. Lignin degradation is still more complex. Microorganisms which are capable of degrading lignin related structures have also been isolated from the rumen. These microorganisms have been fused with other related species to produce hybrid organisms which have been found to have ability to degrade both lignin and polysaccharides in controlled experiments (19, 20).

The above studies of the genetic systems of the microflora of the rumen are primarily limited to bacteria; however more knowledge is accumulating gradually. Very little work has until now been done on rumen fungi and anaerobic protozoa. The mechanism of action of the rumen microflora is complex. The bacteria, fungi, protozoa and other lifeforms generate a series of complex activities. Our wisdom lies in understanding these activities individually and in orchestrating these towards the benefits of the animals, eventually to benefit us. More basic information is therefore needed to be generated on the biology, biochemistry and physiology of all the lifeforms of rumen before it would be possible to control the rumen behaviour substantially.

Several feed additives such as cultures of yeast, preparation of probiotics and various enzyme concentrates have been used to improve

the feed of livestock. The use of these additives has usually improved the quantity as well as the quality of milk production, although currently these additives are expensive.

Several non-conventional agricultural-products available in India which are not usually used as animal feed, could also be processed for supplementing the animal feed needs. The suitability of several important agricultural products in these class were exhaustively examined at the Indian Veterinary Research Institute, Izatnagar. These include mango seed kernel, babul pods, panewar seeds, tamarind seeds, rain tree fruit, date stone, water washed neem seed cake, deoiled sal seed cake etc (7). Industrial byproducts like molasses from sugar industry mixed with 2% urea have also been used and this mixture has been found to hold potential for substantially supplementing the animal feeds. All these indigenous resources are valuable and need to be used to supplement our animal feed needs. Wherever possible, the qualities of these products should be improved by appropriate biotechnological methods.

India has large numbers of sugar mills scattered all over the country. All these industries, especially the modern ones have adopted energy conservation methods and thereby they have been able to generate surplus bagasse to the extent of 2 to 5% of their gross production of bagasse. Bagasse production (50% moisrure) constitute nearly 30% of the sugarcane crushed in a mill. The mills have crushing capacities ranging between 2000 to 6000 tonnes per day and the average duration of operation is about 150 to 200 days annually. Assuming a crushing capacity of 4000 tonnes of sugarcane per day, with 200 days annual working, a mill can generate nearly 12000 tonnes of surplus bagasse annually. If this could be modified into an effective animal feed by biotechnological methods using a series of microbes/enzymes, this would not only add to the animal feed production of the country, but would also ensure better profitability to the sugar mills.

CONCLUSIONS

The advances made in the field of livestock improvement through biotechnological interventions during the last three decades had been phenomenal. A large proportion of our cattle are being raised through cross breeding. Methods for MOET have also been largely standardised in cattle and buffalo and these hold substantial potential for enabling us to have a quantum jump in increasing the number of high yielding livcstocks, and if this strategy is successful, this would enable us to reduce our numbers of unproductive livestock to a considerable extent. The current costs of MOET raised animals are high. However, the prospects of increased turnover of of viable embryos are bright. Moreover, the success in pregnancy rates is also expected to rise. In addition, capabilities are being building up in the local production of hormones and biologicals required for MOET. All these factors are expected to reduce the cost

of MOET animals. The improvements already achieved in different spheres of livestock breeding have partly been transferred to the farmers and these are reflected in increased supplies of milk and meat to our population. There has also been better understanding of animal health and animal nutrition, and these understandings are manifested in the increased usage of vaccines, diagnostics and feed additives. It has also been recognised that as there is a shortage of animal nutrients, we need to utilise non-conventional agricultural residues to the maximum extent by improving their quality through biotechnological and other methods.

In vitro cultivation of oocytes into embryos is being perfected Once this technique becomes economically beneficial, this could be used to retrieve ovaries from a large number of highly productive cows and buffaloes which are currently being slaughtered in the slaughter houses of metropolitan cities. These animals are often moved to the urban cities from the villages after they start lactating. After the milking phase, these animals are usually not serviced for the next lactation but are sacrificed as the short-sighted urban animal owners do not consider the process of rearing the animals to the next lactation phase to be economically benefitting.

All said and done, in the ultimate analysis it emerges that currently our animals are very large in number and low in overall productivity. Our feed and fodder are in short supply and are not rationally managed. The scientific advances are yet to percolate fully into the rural massess for maximising the production and making the best use of our resources. On the top of it the burden of uncontrolled growth of population is exerting enormous pressure on the limited resources we have to our command. In this down - to - earth situation, we are indeed left with few options. Biotechnological interventions in all directly and indirectly related areas are expected to make noticeable changes to our benefits very fast. I therefore conclude by saying that this is our time for the use of biotechnology for our benefits. Let us embrace the opportunity and make full use of it intelligently by orchestrating breed improvement, feed management, animal health and higher productivity performances of our animals for raising, human food and for further securing humanwelfare through the latset scientific advances.

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