

Buffal Newsletter



Number 8 - December 1997

EUROPE - NEAR EAST

INFORMATION BULLETIN OF THE FAO INTER-REGIONAL COOPERATIVE RESEARCH NETWORK ON BUFFALO

FIFTH WORLD BUFFALO CONGRESS

The abstracts of all papers presented at the FIFTH WORLD BUFFALO CONGRESS - plenary sessions, short communications and poster sessions, can be found in:
<http://www.inea.it/isa/newnet.htm>

The Fifth World Buffalo Congress was held in Caserta (Italy) from 13-17th October 1997. Host organizations were the International Buffalo Federation, the Animal Production Research Institute of Rome, the University of Naples, FAO, the Italian Buffalo Breeders Association, the Buffalo Breeders Association of Caserta, the Animal Health and Pathology Institute of Naples, the Buffalo Project Consortium. The sponsorship of the City of Caserta and the Ministry for Arts and Culture let the Conference to be held inside the Royal Palace of Caserta, built in 1751 as the summer residence of the Borbon kings, and is a unique historical and artistic place.

Over 300 participant from more than 25 countries gathered for the Fifth Buffalo Congress. The scientific programme was opened by the Honorary President W.R.Cockrill and by G. De Francis, President of the International Buffalo Federation.

There were 5 plenary sessions. During the first, chaired by J. Boyazoghlu (EAAP) and A. Bordi (Italy), aspects of "Buffalo Production in different environments" were approached in three presentations: F. Addeo (Italy) "Tradition and innovation in the manufacture of mozzarella" described the production steps for the processing of this cheese stating the need to prevent the introduction of new technologies which might

modify the quality. N.C. Ganguli (India) presented "Milk production and quality in India" concluding by listing 12 action points consisting of several kinds of hygienic measures to be followed in order to obtain high quality milk. G. de Almeida (Brazil) described "Meat production in grazing conditions" concluding that buffalo surpass cattle more as weather conditions worsen, soil quality lowers and pastures become poorer. As buffalo behave very well under confinement conditions this system can be used for finishing the fattening and getting high profit out of the market season.

In the second plenary session, chaired by D. Matassino (Italy) and H.G. Wagner (FAO, Rome) "The Genetic Improvement of Buffalo" was considered. T.H.E. Meuwissen (Holland) talked of the "Impact of New Breeding Schemes on Genetic Progress and its Risk" concluding that the highest improvement is achieved with nucleus herds which may use progeny test of young bulls, and that these herds must endeavour to follow the same management as commercial herds in order to deal with genotype*environment interaction. The "Gene Mapping of Mediterranean Buffalo" was approached by L. Iannuzzi (Italy) who reported that total cattle syntenic groups assigned to the river buffalo genome are 18/31, bringing to 155 the total of expressed genes assigned to river buffalo chromosomes. All river buffalo biallelic chromosome pairs have now at least one molecular marker assigned to each chromosome arm. P. Masina (Italy) and his group presented the use of "Molecular Genetics for Improvement of Buffalo Productions" concluding that the

most effective contribution of molecular genetics to genetic improvement of buffalo is the identification of major genes by using the candidate gene approach. D. Di Bernardino (Italy) presented the "Chromosome Microdissection and Cloning in River Buffalo". The technique described consists of microdissecting GTG-banded chromosome and was performed with an inverted microscope equipped with a micromanipulator. Microdissection was performed by using microneedles obtained from a Nashridge apparatus and chromosome fragments were

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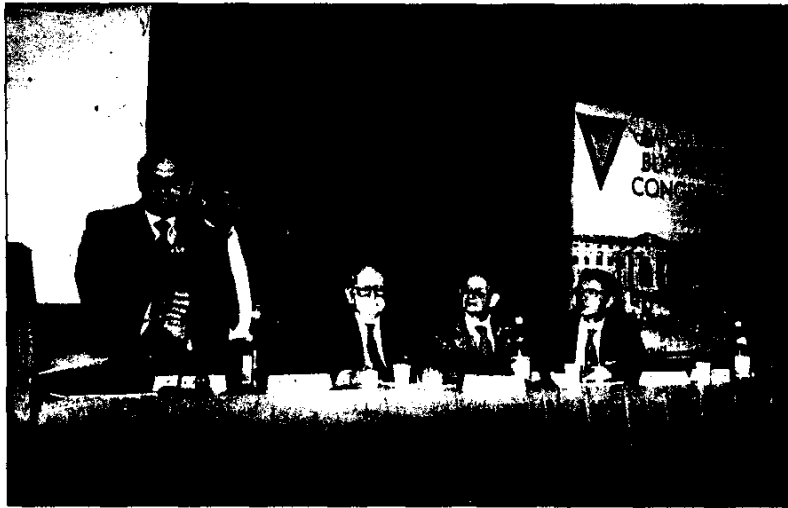
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collected in Eppendorf tubes. After collection, chromosome isolation can then be used for producing whole chromosome specific DNA libraries and DNA probes for gene mapping.

Plenary session 3 was dedicated to Reproduction and was chaired by A. Borghese (Italy) and S. El Nahas (Egypt). W.G. Vale (Brazil) gave a speech on "News on Reproduction Biotechnology in Males" concluding with the following recommendations: fertility is the only appropriate measure for determining the quality of frozen semen; check correct temperature of artificial vagina, water bath and diluents; ejaculate must be kept in its own plasma for 10-15 minutes; keep constant temperature during cooling time; best freezing rate seems to be 20 minutes from 4 °C to -140 °C.; research on new diluters must be stimulated. L. Zicarelli (Italy) referred on "News on Buffalo Cow Reproduction" concluding that the poor use of reproductive technologies in buffalo have not allowed for a substantial increase in productivity. Good results might be obtained by using not only the Embryo Transfer and In Vitro Fertilization but also the Juvenile Ovum Pick Up technique.

Plenary session 4, "Social and Economic Aspects of Buffalo Breeding" was chaired by F. El Keraby (Egypt) and F. Martillotti (Italy). During this session G.V. Pelagalli (Italy) presented his book



OPENING OF THE CONGRESS

"Morphostructural and Infrastructural Characteristics of Buffalo Stomach"; afterwards L. Cruz (The Philippines), J. Reggeti (Venezuela) and F. De Stefano (Italy) indicated the "Prospect of Buffalo Production" respectively in Asia, South America and Europe. Finally, F. Grasso (Italy) referred on the "Economics and Quality of Buffalo Tanned Hides" pointing out the possibility of increasing the added value of the whole leather production process: in Italy half of the whole supply is lost at present.

Plenary session 5 "Management,

processing and marketing problems" was chaired by G. De Almeida (Brazil) and F.P. Roberto (Italy). There were three presentations: in the first "Marketing Aspects of Mozzarella Cheese in Italy", F. Consalvo emphasized how the big social changes of the last years have influenced food consumption, opening new marketing prospects to fresh cheeses like mozzarella. O. Sekerden (Turkey) described the "Management and Marketing of Buffalo Products in Small Holdings" stating that the number of buffaloes in Turkey has decreased tremendously in the past 20 years due to the lack of marketing system. Buffalo meat is 10% lower paid than beef and the buffalo milk price is only slightly higher than that of cow milk.

The Scientific programme was enriched by the following Short Communication and Poster sessions: Milk Production and Processing; Nutrition Requirements and Forage Resources; Meat Production and Draught Buffalo; Genetic Improvement; Reproduction in Female Buffalo; Reproduction in Male Buffalo; Pathology; Management and Welfare, Haematological Profiles. Altogether 189 short communications were presented.

A special session was dedicated to the activity of the FAO research



G. ROSSI AND T. ALI: PRESENTATION OF THE BUFFALO NETWORK

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Network for Europe and the Near East and was chaired by T. Ali (FAO-RNE, Cairo). The coordinator of the Network (G. Rossi, Italy) summarized the purpose of the network, first as a tool for the exchange of information and material among researchers of the two regions. The Network has been active for 4 years and this goal has been achieved. The following countries have joined the Buffalo Conference emphasizing their will of cooperating: Azerbaijan (a scientist from this country took part for the first time in an International congress giving a presentation on buffalo farming), Bulgaria, Egypt, Greece, Iran, Iraq, Italy, Syria, Romania, Turkey. The presentation of G. Rossi was prepared together with the National coordinators of

the mentioned countries and pointed out the importance of buffalo research in the member countries as well as the future research goals. For the majority of the countries these are the genetic evaluation of buffaloes and the establishment of breeding schemes for improving milk yield and growth. Care as to the diffusion of AI and to the characterization of buffaloes, including studies on the genome will be also tackled in many countries. The member countries have decided to carry out a common project on the exchange of buffalo semen among European and Near Eastern countries, confrontation of genetic evaluation methods and standardization of rules and procedures.

The Congress was concluded with

the announcement of the new President of the International Buffalo Federation, Pablo Moser from Venezuela. The Sixth World Buffalo Congress will then be held in Venezuela in the year 2000.

On October 17th a full day field trip was attended by most participants in the congress. We first visited the historical botanic garden of Ninfa (Latina). Then we were welcomed by two buffalo farmers: Mr. Benedetti Panici and Mr. G. Jemma (Latina) where we not only visited the herds and modern equipment - milking parlours and fattening units - but we were offered a tasteful meal based on buffalo products.

STRATEGIC PLAN FOR TECHNOLOGICAL DEVELOPMENT OF CATTLE SYSTEMS IN COLOMBIA

M.V.E. Gonzalo Abad Arango

CORPOICA, Calle 11, Carreras 15 y 16, La Dorada, Caldas, Colombia

The Colombian Agricultural Research Agency (CORPOICA) is a basic mission the purpose of which is to generate scientific knowledge and technological processes of products to make agricultural production more efficient. The project here described was set up by CORPOICA and is going to be soon implemented.

CHARACTERIZATION OF THE BUFFALO PRODUCTION SYSTEM AS A MULTIPURPOSE SPECIES FOR BAD-DRAINAGED ECOSYSTEMS OF THE CARIBBEAN, CASANARE FOOTHILLS AND INTER-ANDEAN VALLEYS.

In Colombia there are three agroecological bad-drained or inundant areas:

1) the W zones, which are formed by alluvial and inundant flat, plain or plain and hollow lands, located in the Andean and Caribbean regions,

with an extension of 2292450 hectares;
2) the Kf zones which are formed by alluvial and inundant flat lands of the Orinoquia region, with an extension of 1089500 hectares;
3) the Kc zones composed of alluvial valleys with periodic inundation, located in the Orinoquia, Amazonia and Pacific-Andean regions, with an extension of 7327250 hectares.

The above areas are located at a warm thermic ground, in humid and perhumid soil. Asian buffalo, known as water buffalo due to its adaptation to tropics, is regarded as the alternative multipurpose animal to take advantage of the livestock production potential in the above areas. The estimated population is 10000 heads, but no data registration of their precise number and production performance has ever been done. With the actual buffalo population, the introduction of



MILKING A BUFFALO IN COLOMBIA

monitoring techniques, the beginning of epidemiological, ecophysiological and nutritional studies in cooperation with producers and universities, will be the starting point for the promotion of buffalo production. A network of producers, farmers associations and scientists will be created in order to define research goals and to establish a gradual and priority process of research, extension service and technology transfer. Epidemiology aspects will be the first to be tackled, because they have the major implication in buffalo husbandry.

Workshop on "ANIMAL RECORDING FOR SMALLHOLDERS IN DEVELOPING COUNTRIES"

K. R. Trivedi

National Dairy Development Board, Anand 388 001, India

The International Committee for Animal Recording (ICAR) with the support of FAO and of the National Dairy Development Board (NDDB) organized a successful Workshop on "Animal Recording for Smallholders in Developing Countries" at NDDB, Anand. The Workshop was attended by 45 delegates from 25 countries. A summary of the discussed items during three discussions Rounds, and recommendations is given below.

SUMMARY OF ROUND I:

Questions:

1. How to form the recording system on goals that provide the maximum management benefits, both short and long term to farmers?
2. Should service provision be integrated across recording, breeding, veterinary service and extension service?

Discussion:

1. An animal recording system should be for:
 - Monitoring of performance
 - Genetic improvement of animals
 - Understanding constraints
 - Providing decision support
 - Making culling decisions
2. A recording system must provide direct benefit to farmers.
3. A recording system should increase the value of recorded animals.
4. It is necessary to define needs of farmers, planners and policy makers in designing an animal

recording programme.

5. A recording system should be simple in operation.
6. A recording system must define the traits that are to be considered for improvement.
7. It is desirable to integrate extension, breeding, feeding and animal health care into a single recording system.
8. It would be better if a single organisation provides all recording, extension, breeding, feeding and animal health care services.
9. The recording system should identify the animal characteristics that are important to the farmer as the productivity of their animals increases. These traits should then be added to the recording system.

SUMMARY OF ROUND II:

Questions:

1. How to overcome the difficulties of establishing and sustaining a recording scheme

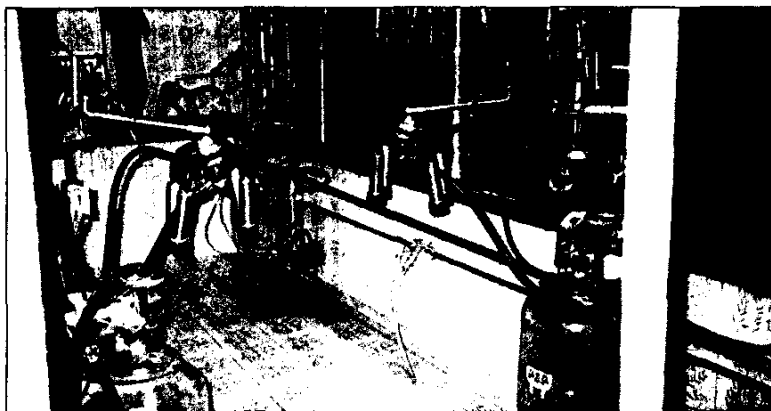
(Dairy, beef, sheep, goats)?

2. How to identify animals in recording schemes and how to do performance evaluations in small herds?

Discussion:

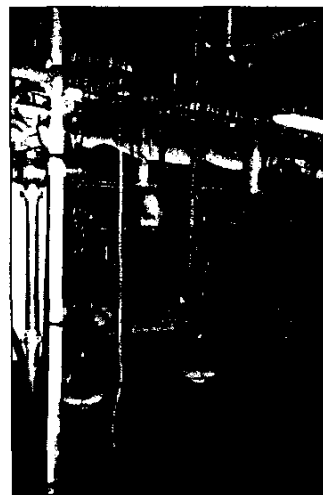
1. Overcoming the difficulties of establishing:
 1. 1. Obtaining support of the farmers through meetings and possible incentives.
 1. 2. Initiating a small pilot project.
 1. 3. Effective use of extension services.
 1. 4. Minimising initial direct costs to the farmer.
 1. 5. Identification of local institution to coordinate a programme is necessary for its success.
 1. 6. Networks of other local recording schemes may evolve later.
 1. 7. Obtaining adequate funds from the government or external sources is necessary to initiate

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MILKING PARLOUR (FARM "G. IEMMÀ", LATINA ITALY)

MILKING PARLOUR (FARM "BENEDETTI PANICI", LATINA ITALY)



the recording system and possibly to provide a part of the costs of operation for an agreed period. Subsequently, the running cost could be paid proportionately by the beneficiaries which could probably include the farmer.

1. 8. Overcoming the difficulties of sustaining:

- Ensure long term funding by obtaining firm financial commitment from all beneficiaries.
- Farmers' organisations should be responsible for obtaining payment by participating farmers.

2. Identification of animals:

A number of acceptable methods of animal identification were mentioned including:

- ear tags
- ear notching
- neck chains
- hot branding
- cold branding

It was agreed that each animal's number must be unique within the recorded population.

However, it was agreed that in very small herds where the individuals have names, the animals may not be marked with their identification numbers, but their recording will include both names and their unique number.

SUMMARY OF ROUND III:

Questions:

1. What proportion of costs should be paid by the government and by the farmers at the start and subsequently?
2. How to encourage farmers participation in recording schemes by providing information? What information should be provided to the small farmers?

Discussion:

The government and/or external sources should pay a very high proportion of the costs of establishing the infrastructure of the scheme and of its initiation. The use of extension personnel in recording is often desirable and may be cost effective, but the percentage of their time allotted to this purpose must be the subject of a long term agreement. Beneficiaries of recording

programmes may include: farmers; planners and policy makers; livestock product industries, and consumables. The cost of recording schemes should be borne proportionately by the beneficiaries once the scheme is established.

Those who contribute data to a recording scheme should receive information for decision making and planning from the scheme in a timely manner and in a form that is useful to the recipient. Assistance in the interpretation will probably be an essential part of the duties of recorders and extension workers who should receive training for this duty. The analysed and interpreted results provided to the farmer must permit and assist in improving productivity by appropriate changes in management recommended by the action lists and analysed results.

Providing farmers with comparative information on performance of their animals relative to others in the area may be a powerful tool to motivate farmers to adopt changes in management practices recommended by analysed and interpreted feedback from the recording scheme.

Recommendations:

1. FAO/ICAR should initiate pilot projects to demonstrate the economic benefits of animal recording.
2. FAO/ICAR should promote establishment of regional networks to exchange ideas, methods and experiences on animal recording and to assist in governmental arrangements.
3. The ICAR member organisations should coordinate recording systems nationally.
4. ICAR should provide guidelines for standardisation of data collection, communication and evaluation of animals of all species and products. These guidelines should address:
 - Quality of data
 - Low and medium input production systems
5. FAO in conjunction with ICAR should develop guidelines for initiating and structuring

national animal recording systems. The following principals should be included in these guidelines:

- Farmers should participate in conception, design and maintenance of the scheme.
- Local management of the recording programme should be encouraged through promotion of farmers' groups and cooperatives.
- The recording systems should be designed to meet the needs of farmers, planners, policy makers, breeding schemes and consumers.
- Government or other organisational funding is necessary to initiate and possibly to sustain recording. However, farmers should pay at least some cost of recording either directly or through their organisations.
- Cost of the programme should be borne proportionally by the beneficiaries.
- Recording should preferably be a part of an integrated local service / extension package.
- Recording activities (animal health recording, performance recording, animal resource characterisation, etc.) should be integrated among appropriate agencies and organisations.
- The recording system should include all measures necessary for economic evaluation of animals.
- The schemes should be no more complex than necessary to achieve the programme goals.
- 6. FAO in conjunction with ICAR should promote training activities in animal recording including training of field level extension staff, country coordinators of recording and extension, and university and ministry of agricultural personnel in small farm recording for management advise and record interpretation.
- 7. FAO should encourage government to use appropriate animal recording procedures to better characterise, utilise and monitor local animal genetic resources.
- 8. FAO / ICAR should promote the use of animal recording to government as a general mechanism for animal production and agricultural system development.

GENE MAPPING OF RIVER BUFFALO

Soheir M. El Nahas

Department of Cell Biology, National Research Center, El-Tahir St., Cairo, Egypt

SUMMARY: 1. GENERAL OVERVIEW • 2. DEVELOPMENT OF THE PHYSICAL GENE MAP OF RIVER BUFFALO • 3. USE OF SOMATIC CELL HYBRIDIZATION IN BUFFALO GENE MAPPING • 4. USE OF CHROMOSOMAL IN SITU HYBRIDIZATION (DIRECT ASSIGNMENT) IN BUFFALO GENE MAPPING • 5. THE PRESENT STATUS OF THE RIVER BUFFALO GENE MAP • The first four chapters are presented in this issue. Chapter 5 will be published in the next issue of the Buffalo Newsletter (n°9).

GENERAL OVERVIEW

The water buffalo (*Bubalus bubalis* L.) has for centuries been an important part of animal husbandry in many countries in Africa, Asia and in some countries in Europe. Water buffalo can be classified into: 1- the swamp buffalo ($2n=48$) of the eastern half of Asia, 2- the river buffalo ($2n=60$) of the western half of Asia, northern Africa and Europe. The difference in karyotypes between the two types results from a tandem fusion between chromosomes 4 and 9 of the river buffalo to make chromosome 1 in swamp where the centromere of number 9 fuses with the short arm of number 4 buffalo (Di Berardino and Iannuzzi, 1981). The Mediterranean buffalo is of the river type but has been isolated for so long that it has developed some unique characteristics. In Egypt the river buffalo (also called Egyptian buffalo), a representative of the Mediterranean buffalo, is of key importance to the country economy. The estimated herd number exceeds 3 million heads. In Egypt, buffalo herds vary in productivity and reproductivity. This variation may be attributed to different factors such as management, feeding and genetics. Genetic improvement of river buffalo, especially in reproductive performance and quantity of meat and milk production, ranks high among Egypt agricultural research needs. Buffalo selection for breeding is based mainly on the phenotype. Not much attention was given to modern biotechnical means for their genetic improvement as compared to cattle. In 1990 efforts were directed at the National Research Center, Egypt, towards the understanding of river buffalo

genetics by first establishing a physical gene map based on the chromosomal localization of markers. The development of a buffalo gene map is a tool for developing more efficient breeding strategies, in an attempt to raise the production and reproduction and to increase the disease resistance in buffaloes. The majority of traits in farm animals are polygenic in nature. They are controlled by the so-called Quantitative Trait Loci (QTL) which are difficult to improve by traditional genetic selection. Finding the locations of important gene on the chromosomes is a crucial step in the analysis of buffalo genome. Once precisely mapped within the genome, genes can be cloned, identified and characterized at the molecular level in order to understand their function and the way in which they are regulated.

A saturated map of markers applied to families segregating economically important traits will produce linkage associations of traits to markers, resulting initially in the use of markers for selective breeding. Marker-trait associations (mapping disease-associated genes to microsatellite marker) in cattle (Georges et al., 1993) is being developed, which will permit marker assisted selection (MAS). The development of buffalo gene map is expected to take shorter time, since, the development of gene maps in a variety of species has revealed genome conservation among distantly related species (Morizot, 1983), where large chromosomal regions have been conserved in mammalian evolution (Womack and Moll, 1986). This allows for the extrapolation of chromosomal localization of genes, in highly conserved regions, from one species to the other. Members of family *bovidae* were reported to show chromosomal conservation

(Gallagher and Womack 1992). Extensive chromosomal homology between cattle and buffalo chromosomes (Iannuzzi et al., 1990 and Report of the Committee for the Standardization of Banded Karyotypes of the River Buffalo, 1994) encourages the use of comparative mapping in the development of buffalo gene map. Thus the cattle map is used as a template for selecting gene loci to screen for in river buffalo. Comparative mapping will facilitate the assignment of genes, which were previously mapped in cattle, to buffalo chromosomes.

DEVELOPMENT OF THE PHYSICAL GENE MAP OF RIVER BUFFALO

The development of a buffalo physical map can be accomplished by the use of somatic cell hybrids or by direct assignment of genes to chromosomes. Somatic cell hybrids enable the study of synteny between genes by studying the syntenic relationships derived from isoenzyme, Southern and PCR analysis. Based on the genetic conservation expected between cattle and buffalo, use was made of the molecular markers (DNA probes or PCR primers) developed for cattle gene map. Assignment of syntenic groups to chromosomes can be accomplished either by comparative mapping, based on the extensive chromosome arm homology between cattle and river buffalo, or by cytological analysis of the hybrid. Direct assignment of certain genes is achieved by the molecular hybridization of specific DNA probes to fixed metaphase chromosomes. This method is

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called in situ hybridization. Signal of hybridization can be detected by fluorescence in situ hybridization technique (FISH) or by counting the number of grains if the probe used was radioactively labeled.

USE OF SOMATIC CELL HYBRIDIZATION IN BUFFALO GENE MAPPING

Production of Buffalo/Hamster Somatic Cell Hybrids

In general, somatic cell hybrids are obtained by fusion of nongonosomal cells of different animal species. During culture, these cells divide and lose part of their chromosomes, especially during early divisions. From each fusion experiment, several hybrids can be produced. Hybrid cells are isolated to produce a panel of independent hybrid clones with different chromosomal constitution. The first study on gene mapping in river buffalo was reported by de Hondt et al., (1991). It was based on the analysis of buffalo/hamster somatic cell hybrid panel produced through the collaboration between our laboratory and Dr. A.A. Bosma's laboratory at the Department of Functional Morphology, Utrecht University, The Netherlands. Buffalo/hamster somatic cell hybrids were produced through polyethylene glycol mediated fusion of river buffalo lymphocytes and Chinese hamster cells line wg3h deficient in hypoxanthine phosphoribosyl transferase (HPRT). The resulting hybrid clones were then selected in hypoxanthine/aminopterin/thymidine (HAT) medium. The fused buffalo/hamster cells contain the hamster and buffalo genome. With subsequent division of these cells, the buffalo chromosomes are partly lost whereas the chromosomes of the hamster remain. Different clones are then isolated and were cryopreserved in liquid nitrogen. Each clone in the established hybrid panel contains different set of buffalo chromosomes from the others.

Determination of Syntenic Relationship

River buffalo-hamster hybrid clones are analyzed for gene loci representing the cattle autosomal syntenic groups and the X chromosome using biochemical and molecular characterization. Analysis included electrophoretic separation of selected isozyme markers and testing DNA extract for marker loci using Southern and PCR analysis. The presence or absence of gene products, hybridized probes, or PCR amplification products in the different hybrids are then recorded and the cosegregation of the genes are determined statistically by calculating the % concordance and the correlation coefficient.

Biochemical and Molecular Characterization

For both biochemical and molecular characterization of the hybrid clones, cryopreserved hybrid cells are thawed and propagated in culture medium supplemented with foetal calf serum and antibiotics. Cells are incubated in CO₂ incubator at 37°C. When cells reach confluency, they are collected and processed. For biochemical analysis the enzymes were tested in lysed hybrid clones and parental cells (Chinese hamster cells and buffalo cells or tissues), using starch gel electrophoresis or cellulose acetate membranes, following the protocols of Harris and Hopkinson (1976). Comparison between the bands developed in hybrids with those developed in parental cells determines the presence or absence of studied enzymes in the hybrids. In molecular characterization, genomic DNA is extracted from buffalo, Chinese hamster cell line and hybrid cell lines following the established protocols (Blin and Stafford, 1976). The extracted DNA was used for testing marker loci using PCR or Southern analysis. The investigated PCR primers are designed from published bovine

sequences. PCR conditions have been optimized by establishing the highest annealing temperature that allows amplification of bovine-specific fragments. Total genomic DNA, extracted from hybrid clones were tested for the presence of PCR amplified product of bovine-specific fragments. This was done by electrophoresis of the PCR product on an agarose gel and the results were recorded. In Southern (1976) analysis, genomic DNAs are digested with restriction endonucleases, then subjected to agarose gel electrophoresis, and blotted onto nylon membranes. The membrane was hybridized with ³²P labeled probe DNAs developed for cattle to reveal river buffalo-specific bands. The choice of restriction endonuclease used with each probe was determined by test blot analysis of parental DNAs to select the enzyme that best allows discrimination between river buffalo-specific restriction fragments relative to that of hamster.

Statistical Analysis

The cosegregation of the studied genes in the hybrid panel elaborates on the syntenic relationship between these genes. After studying the segregation profile of the studied loci, synteny between markers are tested by calculating the % concordance and the correlation coefficient (φ). Loci are considered syntenic if they exceed a 90% concordance and a certain (value depending on the number of hybrids tested (Chevalet and Corpet, 1986).

Assignment of Syntenic Groups to Chromosomes

The assignment of certain genes or syntenic groups to chromosomes can be achieved by examining the clones cytogenetically or by comparative mapping. In cytogenetic analysis buffalo chromosomes are identified in each hybrid. A gene can be assigned to a specific

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chromosome when the gene product and the buffalo chromosome are simultaneously present or absent in the hybrid. To identify the buffalo chromosomes in the somatic cell hybrids, first we need to differentiate between the buffalo and the hamster chromosomes of the hybrids. This is done using fluorescent in situ hybridization (FISH) technique. This is accomplished by labelling river buffalo total genomic DNA with biotin-14-dATP using nick translation. In situ hybridization and fluorescein detection is then carried out. Fluorescein signal, marking the buffalo chromosome against Hoechst counter stained hamster chromosomes, can thus be photographed using high quality research microscope equipped for epifluorescence. To the same slide post hybridization QFH-banding or any other banding technique is applied. By coupling FISH of river buffalo total genomic DNA with banding, chromosomal characterization of the hybrids is simplified since buffalo chromosomes will be banded and easily discriminated from rodent chromosomes. The FISH technique is also a useful tool in revealing translocations between bovine and rodent chromosomes. Assignment of specific gene can be accomplished by comparative mapping. Sets of genes known to be syntenic in two species can be assigned to homologous chromosomes.

USE OF CHROMOSOMAL IN SITU HYBRIDIZATION (DIRECT ASSIGNMENT) IN BUFFALO GENE MAPPING

In this method, DNA labeled probes are hybridized to fixed metaphase chromosome which are G-, R-, or Q- banded, prior or post hybridization. Signal detection allows the localization of the probe to specific band on the chromosome.

Metaphase Preparation

Buffalo metaphases are harvested from blood or

fibroblast cultures according to standard techniques. Use of early or late BrdU incorporation is needed to perform Q or R banding, respectively.

Probe Labelling

DNA probes are labelled by nick translation using tritium (³H) for autoradiographic detection (Hassanane et al 1994) and biotin-11-dATP for fluorescein detection (Iannuzzi et al., 1993).

In Situ Hybridization and Signal Detection

Metaphase preparations (pretreated with RNase) and probe are denatured in 70% formamide in 2 X SSC at 70° C. The probe is then added to the slide, covered with a coverslip and sealed with rubber cement and kept in a moist chamber at 37° C for 14-16 hours. Slides are washed in 80% formamide / 2 X SSC, 2 X SSC and 0.1 X SSC.

For autoradiography, the slides are dipped in diluted nuclear track emulsion at 48° C, dried and placed in a light-tight box in 4° C for up to 25 days, and developed using Kodak D-19 developer, rinsed and fixed. Air dried slides are then stained with Giemsa. Metaphases with silver grains on the chromosomes are later identified using G or R-banding techniques. For fluorescence in situ hybridization (FISH), the slides are incubated at room temperature with fluorescein isothiocyanate (FITC) conjugated avidin. The intensity of biotin-linked fluorescence is amplified by adding a layer of biotinylated goat anti-avidin antibody followed, after washing, by another application of FITC conjugated avidin. Chromosome identification can simultaneously be achieved by QFH-banding using Hoechst 33258 in the antifade mountant. QFH-banded metaphase chromosomes are photographed, counterstained with propidium iodide and rephotographed using filter set for FITC excitation. R-banding

and G-banding can be also applied for chromosome identification.

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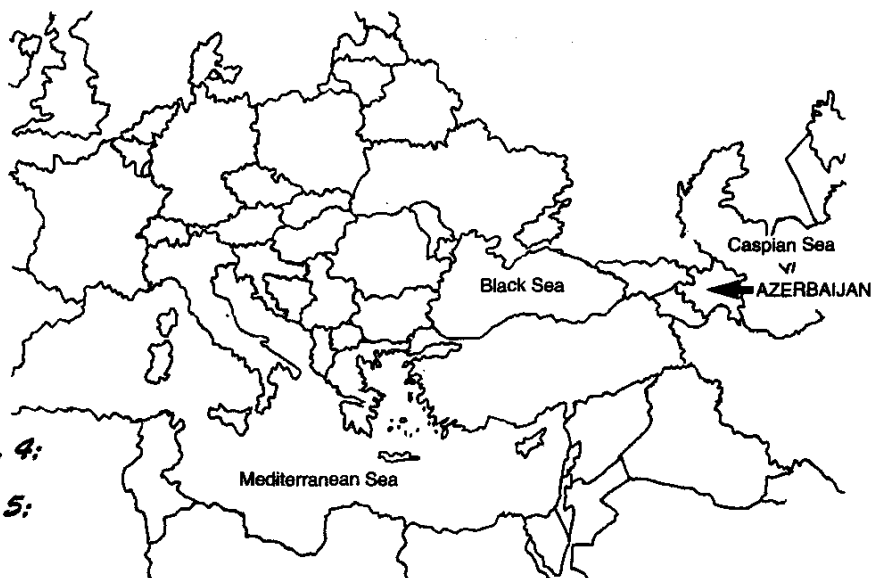
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In each issue of the Buffalo Newsletter a description of Buffalo farming in every member country is presented. Egypt and Syria appeared in n. 3; Bulgaria and Albania in n. 4; Turkey and Romania in n. 5; Iraq in n. 6; Italy in n. 7.



BUFFALO POPULATION AND PRODUCTION IN AZERBAIJAN

1 - ORIGIN, TYPE, CROSSING:

Asian buffalo, Azerbaijan breed, some crossing with Murrah breed.

2 - GEOGRAPHICAL AREAS:

Subtropical area of Azerbaijan.

3 - NUMBER IN 1996:

adult females 100,000
 adult males 5,000
 young stock 200,000
 TOTAL 305,000
 the number is slightly decreasing.

4 - RESEARCH HERDS:

4 (2 in Sheki-Zafatala area, 1 in Shirzan area, 1 in Gandja area).

5 - DESCRIPTION:

(see table 1)

6 - PRODUCTIVITY:

N. DAYS LACTATION/YEAR:
305.
 LACTATION MILK YIELD (KG):
kg 900 to 1000.
 AGE AT FIRST CALVING (MONTHS):
36 to 38.
 AVERAGE LACTATION NUMBER:
6-9.
 AGE AT SLAUGHTER:



male 18/24 months;
 female 96/120 months.

WEIGHT AT SLAUGHTER:
 male 420 kg;
 female 400 kg.

- IS THE CALF SUCKLING?

Yes for one month in all herds.

- ARE COWS MILKED ONCE A DAY?
 No.

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Table 1.

	ADULT MALE	ADULT FEMALE
- HEIGHT AT WITHERS	137 cm	131 cm
- WEIGHT	750/800 kg	455/550 kg
- COLOR	grey to black	dark grey to black
- HORNS	short	short

follows from page 9 / BUFFALO POPULATION AND PRODUCTION IN AZERBAIJAN

- ARE COWS MILKED TWICE A DAY?
Yes, at the farm.
 - ARE COWS MILKED BY HAND?
Yes.
 - ARE COWS MACHINE MILKED?
Yes, only in one farm.
 - TYPE OF MACHINES:
UDB - 100 F.

7 - FERTILITY:
 N. CALVES/YEAR: **1.**
 SEASON OF CALVING:
spring/autumn.

8 - HOUSING:
 Tied 100% .

9 - ARE BUFFALOES USED FOR DRAUGHT? No.

10 - SOURCE OF FEEDING:
 Maize silage, hay straw, haylage, oilcake concentrates; no grazing

11 - TOTAL ANNUAL PRODUCTION BY SPECIES.
 (see table 2).

OTHER PRODUCTS FROM BUFFALO:
No.

12 - MILK RECORDING:
 Yes in all research herds and pedigree farms.

13 - REPRODUCTION:
 HAS EACH FARM ITS OWN BULL?
Yes.
 ARTIFICIAL INSEMINATION?
Yes, for progeny testing
5,000 buffaloes inseminated every year.

14 - DISEASES AND PARASITES:
 Paratyphus in calves, fleas.

15 - SOCIAL POSITION OF BUFFALO FARMERS.
 Medium.

16 - PERSPECTIVES OF BUFFALO PRODUCTION:
 Effords are made to increase individual milk production. ●

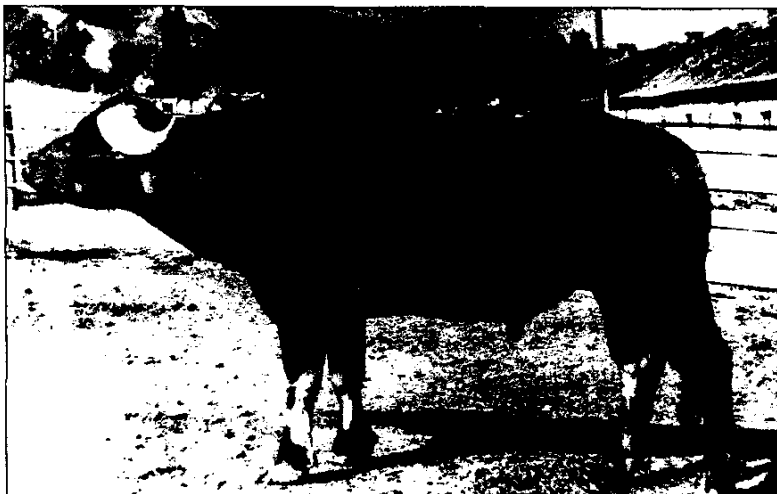


Table 2. Total animal production by species (metric tons) year 1986.

	N. ADULT FEMALES	TOTAL MILK	CONSUMED FRESH	CHEESE	BUTTER	MEAT
BUFFALO	100,000	100,000	40,000	-	10,000	750,000
DAIRY COW	700,000	886,000	-	-	-	-
EWES	2,500,000	-	-	-	-	-
GOAT	90,000	-	-	-	-	-

BUFFALO PRODUCTION IN VIETNAM - AN OVERVIEW

Nguyen van Thu

Department of Animal Husbandry, Faculty of Agriculture, Cantho University, Cantho city, Vietnam

ABSTRACT

Buffalo production is traditional and very important in the rice culture for draught and meat production of Vietnam. Swamp type buffaloes constitute the majority of the population in Vietnam. In 1995 the total buffalo population was 2,963,400 head (Lich, 1996). Murrah buffaloes have also been imported from India and China, however, the number are small for several reasons. Buffaloes in the Mekong delta (MD) are large in size as compared to those of other regions in Vietnam. (Thu, 1987). Average live weight of adult swamp buffalo cows of Vietnam is about 420kg and about 480kg for the adult female and male. Adult live weight of Murrah buffaloes raised in Vietnam was about 460kg and 595kg for the females and males, respectively (Xuong, 1981). Average dressing percentage of swamp buffaloes is about 45% and age at first calving is from 3.5 to 4.5 years. The pregnancy duration varies from 330 to 345 days and the buffalo cows normally give 2 calves per 3 years. In northern Vietnam buffaloes work singly, while in the south they normally work in pairs, and are able to plough from 0.3 to 0.4 ha/day. Feeding is based on extensive grazing with rice straw and other crop residues also used during the dry season. Some of the important diseases of buffaloes

are Pasteurellosis, Foot and Mouth disease, liver fluke and bloat. There is a lack of studies on resources to improve the swamp buffalo performance.

KEY WORDS: Swamp buffaloes, Murrah buffaloes, live weight, performance, feeding and diseases

INTRODUCTION

The swamp buffalo is an important animal in Vietnam for the preparation and cultivation of the rice fields. In addition they provide meat for the people and manure as important fertilizer source. From 1990 to 1996, although the total population has increased by 0.73% annually, there has been a significant reduction of their numbers in both the Mekong delta (MD) and the Hong river delta. In 1995 the buffalo population in Vietnam declined slightly because of an increasing use of tractors and increasing area under cultivation, which has resulted in a reduction in grazing land. Despite this, a participatory rural appraisal in 1994 in MD showed that many farmers still continue to raise buffaloes for land preparation, to ensure timeliness of seeding, and for profit from the sale of calves and meat (Ogle, 1994).

Murrah buffaloes have been imported and raised at several state farms of the country in order to evaluate their potential for milk production. However, their development has been limited due to lack of research resources and the fact that results from swamp buffalo have showed promise. Farmers in Vietnam have traditional experience in buffalo raising but techniques are primitive and are not widely applied in the rural areas to exploit their considerable potential for supplying draught power, beef, milk and manure. Therefore, their potential should be studied and suggestions for future research are necessary to help farmers to improve productivity

Table 2. Buffalo population distribution (%) in Vietnam

REGION	PERCENTAGE (%)
Mountainous and up-land areas in the North	51.8
Hong river delta	8.60
Central areas	28.2
South-eastern provinces	6.38
Mekong river Delta	4.96

Source: Lich (1966)

in sustainable farming systems.

POPULATION AND PHYSICAL CHARACTERISTICS

Recently, the rate of increase in the population of native buffalo in Vietnam has slowed and in 1995 there was a small reduction in the population due to a decrease in the grazing land as a result of an increase in the area under cultivation (table 1), in particular in the MD and Hong river delta provinces where the buffaloes are grazed extensively. However, there has been an increase in numbers in the mountainous and high-land areas, where grazing lands are still available (table 2).

Swamp buffaloes in Vietnam are similar in appearance to those in China, Thailand, Cambodia, etc. Adult swamp buffaloes are smaller than Murrah buffaloes, particularly the males. Buffaloes in the Mekong delta tend to be larger size than in other regions (table 3). Thu (1987) also reported that swamp buffaloes in Dong Thap province of the Mekong delta were the biggest in Vietnam with an average live weight of 593 kg for adult females and 706 kg for males.

Table 1 - Recent buffalo and cattle population changes in Vietnam (thousand head)

YEAR	BUFFALO	CATTLE
1990	2,854.1	3,120.8
1991	2,885.6	3,151.0
1992	2,883.4	3,193.8
1993	2,960.8	3,353.0
1994	2,971.1	3,466.7
1995	2,963.1	3,638.7

Source: Lich (1966)

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Table 3. Basic measurements and live weights of adult Swamp and Murrah buffaloes in Vietnam

REGION CRITERIA	MEKONG DELTA ⁽¹⁾	SOUTH-EASTERN PROVINCES ⁽²⁾	CENTRAL AREA ⁽³⁾	NORTHERN PROVINCES ⁽⁴⁾	MURRAH BUFFALOES ⁽⁵⁾
Females					
Height at whiter (cm)	127	126	121	118	131
Length of body (cm)	141	132	128	127	143
Heart girth (cm)	192	193	191	180	195
Live weight (kg)	458	434	417	368	457
Males					
Height at whiter (cm)	131	129	129	121	138
Length of body (cm)	143	135	136	133	154
Heart girth (cm)	198	197	198	190	207
Live weight (kg)	494	464	494	428	595

Source: ⁽¹⁾Thu (1967), ⁽²⁾Hoang (1978), ⁽³⁾Thao (1985), ⁽⁴⁾Xuong (1981).

PHYSIOLOGICAL CHARACTERISTICS AND REPRODUCTIVITY

Physiological criteria measured early in the morning are shown in table 4. Body temperature and pulse of Swamp buffaloes were lower than those of the Murrah, while respiration rate was higher in the Swamp buffaloes. Results are similar to those reported by Hoang (1978). Blood plasma protein of the Swamp buffalo was lower than in the Murrah, while erythrocyte and leukocyte counts and hematocrit were similar.

In the Swamp buffaloes, age at first calving is from 3.5 - 4.5 years and pregnancy duration from 330 - 345 days. The majority of swamp cows gives two calves per three years. The

total life time number of calves produced is variable, ranging from 5 - 10 due to variations in work load and nutrition. The estrus cycle was found to be 28 days. In Murrah buffaloes raised at the Buffalo Research Center in Song Be province, pregnancy duration was found to be 306±4.48 days (mean±std), calving interval 494±68.2 days and the estrus cycle 23.1±2.29 days. Average live weight at birth was 27.7±3.47 kg for females and 29.7 ±4.09 kg for males (Xuong, 1981).

MEAT, MILK AND WORK PERFORMANCE

The mean carcass weight of adult swamp buffaloes reported from the North of Vietnam was

187 kg with the live weight at slaughter of 428 kg (Hoang, 1978). It can be seen in table 5, that the carcass weight and live weight were higher at slaughter in the Mekong delta. Recently, Thu (1997) reported that the average daily milk yield of swamp buffaloes from the first to the fifth month of lactation in Mekong delta was 1.50kg, and the average milk fat 11.9% on a *sacciolepis indica* diet, and 8.68% on a diet of *eleocharis dulcis* and rice straw diet under village conditions. In the South of Vietnam swamp buffaloes usually work in pairs, while they normally work singly in the North. The ploughing capacity was found to be 630m²/half day/animal (Thao, 1979). For Murrah buffaloes the carcass percentage and milk yield were found to be higher than in the Swamp buffaloes, although milk fat percentage of the Murrah was lower than that of the Swamp buffaloes.

MANAGEMENT PRACTICES

Rural farmers in Vietnam have a long tradition and experience of buffalo raising, but techniques are mostly primitive. Management practices are based on extensive systems and buffaloes are freely grazed on natural grass lands, forests, roadsides, canal banks, rice fields after harvesting and dikes, etc. They are also fed with rice straw or other crop residues in the dry season and working season. They are occasionally supplemented with rice bran, broken rice soup, raw sugar or honey when farmers feel that their animals are in poor condition for working. Farmers normally do not fatten buffaloes, but middle men occasionally buy and fatten them by good quality grass lands for sale. They are mainly slaughtered when they are not able to work due to old age or as a result of accidents. Housing for buffaloes is simple and made of local materials such as wood or bamboo, with a palm leaves roof, and they are also tethered in the gardens or fields by nose strings.

Selection of draught buffaloes has been made according to the farmers experience, usually

Table 4. Average physiological characteristics (mean + std) of swamp and Murrah buffaloes over four years of age in Vietnam

CRITERIA	BREED	SWAMP (BOTH SEXES)	MURRAH (FEMALE)
Body temperature (°C)		37.4 - 37.8	37.8±0.27
Pulse (beats/min)		38.6 - 44.7	46.2±2.95
Respiration (beats/min)		25.6 - 29.7	15.2±1.77
Erythrocyte count (million/ml)		6.0 ± 0.72 ⁽²⁾	5.83 ⁽¹⁾
Leukocyte count (thousand/ml)		12.1 ± 2.24 ⁽²⁾	12.8 ⁽¹⁾
Hematocrit (%)		28.2 ± 1.60 ⁽²⁾	28.6 ⁽¹⁾
Blood plasma protein (g%)		6.53 ± 0.90 ⁽²⁾	7.34±0.46 ⁽¹⁾

Source: ⁽¹⁾Hoang (1978), ⁽²⁾Bay and Dong (1991), Xuong (1981 and 1987) and ⁽³⁾Thao (1985)

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Table 5. Meat and milk output and working performance of buffaloes in Vietnam

CRITERIA	BREED	SWAMP	MURRAH
Meat production			
Live weight (kg)		592 ⁽¹⁾	374 ⁽²⁾
Carcass (kg)		277 ⁽¹⁾	201-251 ⁽³⁾
Meat (kg)		215 ⁽¹⁾	-
Milk production			
Lactation period (day)		210-360 ⁽²⁾	236±57.3 ⁽⁴⁾
Milk yield (kg/day)		1.20-3.45 ⁽²⁾	5.55% ⁽⁴⁾
Milk fat (%)		8.31-15.0 ⁽²⁾	7.36% ⁽⁶⁾
Working capacity			
Ploughing (ha/pair/day)		0.30 ⁽¹⁾	-
Harrowing (ha/pair/day)		0.73 ⁽¹⁾	-

Source: ⁽¹⁾Thu (1987), ⁽²⁾Thac (1985), ⁽³⁾Ognjanovic et al (1970), ⁽⁴⁾Xuong (1981) and ⁽⁵⁾Thu (1979)

taking into account physical characters such as large size, high fore quarters as compared to hind quarters, tameness, appetite, wet nose, glossy hair, long and curved horns pointing forward (wind-presenting horns). Farmers also pay attention to hair swirls, usually prefer animals that have 4-5 swirls, two on the shoulder, two on the buttocks and one on the face. Hair swirls considered to be unlucky are Lachrymal, joke-resisting, inguinal and vulval. Breeding however has been neglected, except that farmers mate cows to good bulls, regardless of the risk of inbreeding, and usually a field-master bull mates all cows in a given area (Thu, 1987).

The important diseases of buffaloes in Vietnam are Pasteurellosis, Foot and Mouth disease, bloat, diarrhea. (Tam, 1985). Hung (1993) reported that liver fluke was a serious problem in some areas in the Mekong delta, while blood parasitic diseases have also been reported, including Anaplasmosis, Babesiosis and Trypanosomiasis. Murrah buffaloes were also reported to be more heavily infected with parasites than Swamp buffaloes (Hung, 1985). Recently, a veterinary network in rural areas has been more developed with aim of vaccinating buffaloes twice a year before the rainy seasons and dry seasons mainly against Pasteurellosis.

Development of buffalo production in Vietnam has slowed in recent years, while the requirements for milk, meat, draught power, manure and energy has increased, due to a growing human population and higher incomes. Buffalo nutrition and feeds, crossbreeding strategies, disease prevention and treatment and development of sustainable farming systems need to be studied in order to improve the multipurpose uses of both Swamp and Murrah buffalo such as meat, draft, milk, fuel (biogas) and fertilizer. Increased local-international research co-operation is required to increase production from buffalo, particularly the Swamp buffalo, which is declining in population in Asian countries.

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16:40-17:30 Role of Water Buffaloes in Crop-Livestock Farming System. Dr. Patricio S. Faylon (Philippines)

17:30-18:00 Use of Water Buffalo for Environmental Conservation of Waterland. Prof. A. Georgoudis (Greece)

News from Guatemala

In Guatemala there are very few and small herds of Buffalo (20 herds of 10 or less) with the exception of the herd of Oscar Molina (Ganaderia Rio Seco, Guatemala City) which presently has 800 buffaloes. They started the herd from Trinidad and Tobago 15 years ago by importing 12 females and a male. They used them mainly for draft, have palm oil plantations and use the males to get the fruit bunches to the extraction plant. About 2 years ago they started milking. They currently have 400 females in production; have a very active AI program since they purchased semen from other countries to better the genetics of the herd.

Buffalo recently introduced in Holland

Mr. Ijsbrand is a dairy farmer in Holland. He is now milking 10

buffaloes among black and white heifers. He also makes cheese. For the cheese he uses 25% Buffalo milk and 75 % normal cow milk. He makes this on the Gouda cheese way and is satisfied of the results. He will get the buffaloes pregnant by synchronisation and insemination with semen imported from Europe. His e-mail adress is: Spoor.cattle@tref.nl

Buffalo Network (Europe-Near East)

An updated list of all researchers

working on buffalo in Europe and the Near East is ready. It contains addresses, telephone numbers, fax and e-mail of 153 researchers. If interested to receiving it, contact the editor of the Buffalo newsletter.

The list of recent publications on buffalo in the European and Near Eastern countries is also ready. Contact the editor of the Buffalo newsletter for receiving it.

THE BUFFALO NETWORK AT THE FIELD TRIP



"Buffalo Newsletter"

Istituto Sperimentale per la Zootecnia
via Salaria, 31
00016 Monterotondo, Italy