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CYTOGENETIC AND GENOME STUDIES IN PAKISTANI BUFFALO (*BUBALUS BUBALIS*) - A REVIEW

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ABSTRACT

Pakistani river buffalo breeds (Azi-kheli, Kundi, Nili, Nili-Ravi and Ravi) are the least investigated ones for cytogenetic and genome studies. Although the first chromosomal study in buffalo was undertaken in 1960 however, there has been a long lapse until 2001. With the latest advancements in molecular biology tools and techniques including genome sequencing, PCR amplification and identification of SNPs of interest in different genes and high throughput microarray technology rapid progress in molecular determination of buffalo genome is expected in the near future. Similarly establishment of standard chromosome nomenclature of buffalo and clinical Cytogenetics has also been undertaken in Pakistan. Following paragraphs present the first ever review of chromosomal and genome studies at Cytogenetic and molecular genetic level providing a first comprehensive reference document for scientific use.

Key words: River buffalo, Cytogenetics, Chromosome, Genome

INTRODUCTION

River buffalo is among the most gentle of the domesticated dairy species in Indo-Pak subcontinent. However, the word "buffalo" evokes a different response in North America, a large section of Europe except Italy, and in many other parts of the world where buffalo have never been considered a domesticated species rather considered as a wild beast. Water buffalo is responsible for more than ten percent of world milk production but its contribution has seldom been recognized (FAO, 2007-08).

River buffalo *(Bubalus bubalis)* is economically important specie, especially in eastern, southern American and Mediterranean countries. Total world buffalo population is reported to be 161 millions of which 95 + 28 millions are inhabited in India and Pakistan alone (FAO, 2007-08). Under Pakistani dairy set up Nili-Ravi, Kundi and Azakheli breeds constitute an enormous dairy genetic resource for the country. Especially Nili-Ravi is the most docile and promising dairy breed which offers unlimited dairy potential because its milk and milk products are most favoured by Pakistani consumers and is considered as a symbol of pride in the rural set up of the country. It phenotypic beauty as a dairy animal is considered matchless and well fed buffaloes are able to produce up to 15 – 20 litres of milk per day with average 8% fat and 16 percent protein content. Buffalo milk contribution at present stands at 65% to the total national milk yield. Nili-Ravi buffalo are considered the best dairy animal in the tropics. Their home tract is in canal irrigated areas of central Punjab (Shah, 1991), where they are fed abundant green fodder. Owing to its potential, it produces far less milk in its home tract, which may be attributed to (1) little attention being paid in the past for improvement through selection and progeny testing (2) Late age at maturity (3) Silent heat and (4) long calving interval (Hussian et al., 2006). However, issues of quality over quantity are still being discussed and emphasis on improving buffalo milk producing merit through better feeding, management and enhancing its genetic worth is increasing.

For this purpose a large number of genetic and environmental evaluations for milk traits in buffalo have been undertaken (Khan, 1986) so far. Estimation of the magnitude of different genetic environmental parameters such as heritability, repeatability and genetic correlations for different dairy, growth and reproductive traits are available in plenty. However, work on the physical and molecular nomenclature of buffalo genome is extremely limited. In order to make the dairy buffalo of Pakistan, a promising commercial dairy animal a huge bulk of cytogenetic and molecular mapping of genes of economic importance as well as identification of molecular markers for selecting superior stock is the dire need of time. Here, we presents the review of the limited work that has so far been undertaken in Pakistan with some recommendations to gear up the efforts to determine the whole genome and desirable SNPs for milk and meat production, and disease resistance.

Buffalo Chromosomal Studies in Pakistan: Cytogenetic diversity within different livestock species is critical to delineate evolutionary genetic relationships and provide a basis to localize economically important genes on different chromosomes in the absence of genetic maps (Iannuzzi, 2007). Specific studies on physical genetic make up of river buffalo *(Bubalus bubalis)* is now taking some shape with its G-band standard nomenclature being published (CKSBB, 1994).

Some preliminary cytogenetic information is available only for Nili-Ravi breed. The earliest study in Pakistan, to determine the actual chromosome number dates back to 1960 (Khan, personal communication). Which revealed that buffalo has 60 chromosomes like cattle however no detailed information was provided about the morphology of chromosomes for the first time in Pakistan. Ali *et al*. (2001) karyotyped in Nili-Ravi buffalo and reported 2n = 50, XX and XY for female and male riverine buffalo respectively. The morphology of chromosomal complement in buffalo was described to be having five pairs of metacentric/submetacentric chromosomes whereas the rest of the autosomes were classified as acrocentric ones. Similarly X and Y chromosomes were also classified as acrocentric.

More recently the Cytogenetic lab of the department of Livestock Production, University of Veterinary and Animal Sciences, has furthered the work on buffalo chromosomes by producing a G-band karyotype of Nili-Ravi buffalo (under publication) for the first time in Pakistan. This helped comparing and identifying individual chromosomes of Nili-Ravi buffalo with already published standard G-band nomenclature of river buffalo (Iannuzzi, 2004). The results of the study clearly highlighted complete G-band homology with the standard buffalo karyotype as published by International Standard Chromosome Nomenclature of Domestic Bovids (CKSBB, 1994). The future plans entail screening of large populations of Nili-Ravi buffaloes to determine the incidence of chromosomal abnormalities in this species. For this purpose Pakistan Science Foundation has already approved funding of a meagre Rs. 1.7 million which will help initiating some pioneering work in the direction of clinical Cytogenetics.

Genome Studies: Like other livestock species, the majority of important economic traits such as milk production, growth rate and age at maturity in buffalo are polygenic making it impossible to isolate and identify them independently. Buffalo improvement by the genomic approach has been targeted by establishing physical and linkage maps to develop efficient breeding programs. Physical mapping mostly makes use of In-situ hybridization or some times somatic cell hybridization for syntenic determination of different loci and to assign them to specific regions of respective chromosomes. Similarly with the advancements in buffalo genome sequencing by the collaboration of Indian research institutes like Karnal-based National Bureau of Animal Genetic Resources (NBAGR) and the Hisar-based Central Institute for Research on Buffaloes (CIRB) would work on a network mode for the project, said the director-general of Indian Council of Agricultural Research (ICAR), Mangla Rai.

Similarly some major work on the molecular determination of gene of economic interest in river buffalo initiated at the University of Veterinary and Animal Sciences, Lahore , Pakistan would be a giant leap to produce a large body of knowledge about buffalo genome in next few years which will greatly help to improve this docile dairy animal to make it fit for new age intensive dairy production operations by genetically enhancing its milk production and reproductive ability on the similar lines as Italy has accomplished it through developing an improved strain of river buffalo called *'Bufala Mediterranea Italiana*'. This could be achieved through combination of selective breeding and marker assisted selection (MAS) for rapid improvement of *Bubalus bubalis* followed by evolving a crossbred dairy strain by mixing prominent dairy buffalo such as Nili-Ravi, Kundi and Murrah breeds.

Studies on physical gene mapping in Nili-Ravi buffalo and molecular determination of different genes of economic interest in buffalo are in an early stage with only a couple of reports available so far. Babar *et al.* (2008) identified DNA markers to study PRKAG3 gene in different Pakistani buffalo and cattle breeds. Main aim of the study was to develop molecular markers for identification and labelling of market beef. Similarly a trail of different studies on milk genes and other members of PRKA genes are in progress. Another report on random amplified polymorphic DNA (RAPD) analysis to assess genetic divergence/relationship among five buffalo breeds (Azi-kheli, Kundi, Nili, Nili-Ravi and Ravi) of Pakistan is under publication. The genetic similarity among these breeds ranged between 66.2 - 84.4 %. Work on physical mapping of loci in buffalo genome is also in the initiation phase at the University of Veterinary and Animal Sciences, Lahore under a two year research project funded by the Higher Education Commission of Pakistan.

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