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| |  |  | | --- | --- | | **ORIGINAL ARTICLE** |  | |  | | | **Year** : 2003  |  **Volume** : 57  |  **Issue** : 1  |  **Page** : 1--6 | |   Dystrophin gene deletions in South Indian Duchenne muscular dystrophy patients.  **GN Mallikarjuna Rao, T Hussain, N Geetha Devi, S Jain, GR Chandak, MP Ananda Raj**   Centre for DNA Finger Printing and Diagnosis Hyderabad,  **Correspondence Address**: G N Mallikarjuna Rao Centre for DNA Finger Printing and Diagnosis Hyderabad   |  | | --- | |  |   Abstract  66 unrelated patients from Southern India with Duchenne Muscular Dystrophy (DMD) were studied for intragenic deletion in 18 exons and Pm region of the DMD gene using multiplex PCR. Of these 41 (62.1%) showed intragenic deletions. 78% of the deletions were located at the distal hotspot region (44-55 exons) and 22% of the deletions were located at the proximal region (exon 2-19). Exon 50 is most frequently deleted. Deletions in isolated cases were significantly more compared to familial cases. The lower incidence reported from South India compared to North India, is suggestive of variations in the Southern and Northern population.   |  | | --- | | **How to cite this article:** Mallikarjuna Rao G N, Hussain T, Geetha Devi N, Jain S, Chandak G R, Ananda Raj M P. Dystrophin gene deletions in South Indian Duchenne muscular dystrophy patients. Indian J Med Sci 2003;57:1-6 |  |  | | --- | | **How to cite this URL:** Mallikarjuna Rao G N, Hussain T, Geetha Devi N, Jain S, Chandak G R, Ananda Raj M P. Dystrophin gene deletions in South Indian Duchenne muscular dystrophy patients. Indian J Med Sci [serial online] 2003 [cited 2015 Oct 1 ];57:1-6  **Available from:**http://www.indianjmedsci.org/text.asp?2003/57/1/1/11882 |   **Full Text**  Duchenne muscular Dystrophy (DMD) i; one of the most progressive X linked disorders with a frequency of about 1 it 3,500 live male births.[1] The dystrophin (gene spans 2500 Kb of DNA containing 79 exons.[2],[3] Gene deletion is the mos common mutation in DMD (approxi mately 65°x). Deletions are amenable fo analysis by Southern blot hybridization and PCR based methodology. Dystrophin deletions are spread non-ran domly through out the gene clusterin (around two hot spots, a major region o 44-52 exons and a minor one at the 5 end of the gene (exons 2-19).[6],[7]  The frequency of intragenic deletions varies in different populations. In American studies mutant alleles with gene deletions are reported in 55-70% of all DMD/BMD cases.[8],[9] Whereas in Asian and European studies much lower frequency of deletions were observed. In Europe, several regions of abrupt changes in gene frequencies have been identified, most coincident with linguistic boundaries.[10]  With a wide range of ethnic and geographical variations in Indian subcontinent, there is need to analyze systematically the mutational changes in this gene. 73% of deletions in DMD patients was observed in North Indian population.[11] Banerjee & IC Verma[12] observed deletion frequency of 64.4% in Indian population. The present study reports the= prop.;riion r na paiterip ci c, et onai nm.utations in DMD from south India, high lighting possible ethnic/geographical variations.  **MATERIALS AND METHODS**  Patients studied were primarily from Andhra Pradesh and its bordering states in South India. Detailed physical and clinical examinations were carried out including collection of information regarding age, parental consanguinity, birth order, pedigree, and reproductive histories of mother in a special proforma.  In DMD patients age of on set was seer between 1.5 and 5 years, who had calf muscle hypertrophy, positive Gower's sign and were confined to wheel chair by the age of 13 years. The cases were confirmed by EMG, muscle biopsy analy sis and by estimation of serum CK. For all clinically confirmed cases DNA analy sis was done.  Genomic DNA was isolated from lymphocytes from peripheral blood using phenol-chloroform extraction method.[13] Screening for deletions were carried out essentially as described by Chamberlain et a1[14] and Beggs et al[15] amplifying 18 exons and Pm region.  **RESULTS**  DNA samples from 66 patients were analyzed for deletions using multiplex PCR. 41 cases (62.1%) showed intragenic deletions, the localization of these deletions are depicted in the [Figure:1]. Majority of the deletions (78%) were located at distal hot spot region which encompasses exons 44-55 and 22% of the deletions were located at the proximal jot spot region (exons 2-19).  Total of 236 exons were deleted in 41 patients and the advantage number of exons deleted per patients was 5.73. Single exon was deleted in 14 patients (34.1 %) and in 27 patients (65.1 %) multiple exonic deletions were observed.  Most frequent single exon deleted were 50 & 51, the most common exons involved in multiple deletions were 49 (14.4%), 48 (13.6%) and 47 (11.2%). The most frequent exon deletion in overall deletions were 50 (14.38) followed by 48 (13.6%), 49 (12%) and 47 (10%). Intron with most 5' deletion break point is 44 (21 %) followed by 49 (9.7%), 43 (7.3%) and 46 (7.3%). Maximum 3' deleted break points were observed in intron 50 (29.2%) followed by 51 (19.5%).  We divided the families of DMD into familial and isolated cases [Table:1] and interestingly more deletions are found (78.1 %) in isolated cases than in the familial cases (21.9%). There is a significant reduction of identified deletions in familial cases (p<0.05).  Multiplex PCR analysis of deletions in dystrophin gene is a sensitive, rapid, reliable method in establishing the deletion frequency. About 98% of all dystrophin gene deletions can be identified by this method.[14],[15]  **DISCUSSION**  The proportions of deletions in different populations of Asia are compared in [Table:2] which shows a wide variation in the distribution of deletions. This variation may depend on the number of exons being examined or the number of samples being analyzed. Here we have compared those studies in which similar methods were applied and a minimum of 18 exons studied, covering major and minor hot spot regions. Highest deletion frequency of 86% was observed in Arab population, 8% in the proximal, 50% in the distal region of the gene and 42% covering both. Lowest deletion frequency of 37% was reported from Israel which is the lowest deletion frequency reported so far. No particular reasons have been persuasively attributed to explain population - based differences in mutations of the dystrophin gene, may be due to accumulation of differences in intronic sequences and their distribution over a time period as a consequence of genetic drift. Such sequences unique to the population may favour intragenic deletions due to mismatch at the locus.[16]  Most of the Asian population and the North Indian population have break point at intron 44. Our results are also in agreement with this. This may be due to conserved sequences at intron 44, which predisposes the higher preferential break point. However, break point distribution is different in European population viz., 44, 45' or 50.[10]  Familial cases were differentiated from the isolated cases by the presence of two or moe affected famil members. In 78% of the isolated cases deletions were detected, whereas in familial cases it was 21%. The processes operating in causing new mutations may be reflected by this preference.  In our study, deletion analysis was done for 18 exons covering major hot spot region (distal part of the gene) and minor hot spot region (exon 2-19). The break point deletion of 61.2% was found to be less compared to North Indian population (72.7%). A very high incidence of 720 x 10-[6] was observed in immigrants Indians of west midland region of Britain, who came from Northern and western parts of India. This is 2 to 3 folds high compared to the locals and other migrant Asians.[17] The frequency of deletions observed among North Indians is commensurate with the high prevalence observed.[11] The prevalence of DMD in south Indian population (Andhra Pradesh) was 47.8 x 10-[6] which is almost equal to studies reported from different countries (65.4 x 10-[6]).[1],[18] The prevalence of DMD in south Indian population is less compared to North Indian population. Thus the observed lower frequency of deletions in DMD in south Indian population is in keeping with the reported incidence frequency from the south, apparently underlying the differences in Northern and Southern population.  Detection of deletion of DMD gene is essential in the risk estimation, carrier detection and prenatal diagnosis. Screening of DMD patients for deletion using multiplex PCR is very accurate and reproducible compared to time consuming Southern blot technique. This study has helped greatly in establishing the deletion frequency in South Indian population and thus evolve a strategy in the carrier screening and prenatal diagnosis.  **References**   |  |  | | --- | --- | | 1 | Emery. "Duchenne Muscular Dystrophy" 2nd Ed. New York: Oxford University Press; 1988. | | 2 | Bebchuk KG, Bulman DE, D'Souza VN, Worton RG, Ray DN. Genomic organization of exon 22 to 25 of the dystrophin gene. Hum Mol Genet 1993;2:593-4. | | 3 | Roberts RG, Coffey AJ, Bobrow M, Bentley DR. 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