about 98% of all large deletions in the dystrophin gene (Beggs et al., 1990). If we want to detect startpoints and endpoints of deletions in some group of D/BMD patients, we have to use cDNA probes or PCR primers for more exons of the dystrophin gene. In the present study we had to use PCR primers for a total of 47 exon areas of the dystrophin gene for detection of all startpoints and endpoints of deletions in 50 unrelated D/BMD patients. Compared to other studies (Vitiello et al., 1992; Todorova et al., 1996; Banerjee and Verma, 1997; Herczegfalvi et al., 1999; Önengüt et al., 2000) we have examined by PCR the largest number of exons of the dystrophin gene. A large number of exons (more than 18) was analysed in similar studies by Todorova et al. (1996) from Bulgaria (57 patients with deletions) and by Herczegfalvi et al. (1999) from Hungary (116 patients with deletions). These authors used PCR primers for 38 exon areas, but they did not detect some deletion breakpoints of their patients. Further, Önengüt et al. (2000) did not manage to detect some of the deletion breakpoints in 146 patients from Turkey by using primers for 33 exon areas of the dystrophin gene. Similarly, Banerjee and Verma (1997) used primers for 27 exon areas for 103 D/BMD patients from India without detection of all deletion breakpoints. To detect all deletion breakpoints in selected groups of D/BMD patients it is necessary to use a larger number of primers, which will (particularly in large groups) probably be close to the number of exons in the examined area. An alternative way of detecting all deletion breakpoints, although more complicated, is the diagnosis by cDNA probes mentioned above, or the diagnosis by protein truncation test (PTT), which is being introduced by some laboratories mainly for identification of point mutations (Roest et al., 1993; Gardner et al., 1995).

The frequency of gross deletions found out in our D/BMD patients is 43.5%, which is considerably less than the expected 60–65%. It is also the lowest frequency of gross deletions of all the compared populations (Italy = 56.5% deletions, Turkey = 60%, India = 64.4%, Bulgaria = 67.8%, Hungary = 73%). On the other hand, lower frequencies of gross deletions were recorded e.g. in D/BMD patients from Russia (41% deletions) (Baranov et al., 1993), or in patients from Israel (37% deletions) (Shomrat et al., 1994). The differences in the frequency of gross deletions between different populations may be caused and theoretically accounted for by intragenous differences, similarly as the differences in the distribution of breakpoints. On the other hand, it is necessary to evaluate the differences in the frequencies of gross deletions obtained so far from different populations very carefully, as the non-deletion patients were probably diagnosed mainly on the basis of clinical symptoms. In order to prove the differences in the frequency of gross deletions in D/BMD patients from different populations, more detailed studies aimed at the detection of all mutations (that means also point mutations) in D/BMD patients from different populations are needed. Therefore, the part of our work dealing with possible interpopulational differences was mainly aimed at statistical comparison of breakpoints in deletion D/BMD patients from different populations.

The comparison of the distribution of breakpoints in individual introns or in segments of more introns (each segment = 20 introns) is one of the most widely used methods in population studies of D/BMD patients (Danieli et al., 1993; Todorova et al., 1996; Herczegfalvi et al., 1999; Önengüt et al., 2000). There are also studies which compared the distributions and frequencies of deletions (Banerjee and Verma, 1997), the frequencies of specific deletions (Önengüt et al., 2000) and the proportion of deletions encompassing 1–5 exons (Danieli et al., 1993; Önengüt et al., 2000). The use of statistical methods for comparisons of populations showed differences in the distribution of breakpoints in D/BMD patients from different populations (Danieli et al., 1993; Todorova et al., 1996; Herczegfalvi et al., 1999; Önengüt et al., 2000). However, Banerjee and Verma (1997) pointed out that the differences among populations with respect to deletions in the dystrophin gene could be doubtful if small groups are compared or if small numbers of exons are analysed. According to Önengüt et al. (2000) the data used provide precise information about the deletions in the major deletion hot spot (exons 43–52). We agree with this idea as well and we think that for population studies it is convenient to use the statistical comparison of distributions of breakpoints in the main hot spot of the dystrophin gene. The breakpoints in this area are the most frequent and mostly all introns contain breakpoints. That is why it is possible and suitable to apply the χ^2 test in a contingency table. In order to detect all breakpoints in this area it is sufficient to add only primers for exons 42, 46 and 53 to the commonly used primers according to Chamberlain et al. (1990) and Beggs et al. (1990). In case of comparing larger groups (in our opinion a few hundreds of patients with deletions) from different populations it would be suitable to extend the analysis to the intron area of the second hot spot (exons 1–20) or to the area between both hot spots of the dystrophin gene as well. According to our study, the 6 compared populations (Czech, Hungarian, Bulgarian, Italian, Turkish, Indian) exhibit statistically significant non-homogeneity in the breakpoint distribution within the main hot spot (introns 43–52). In order to detect the cause of this non-homogeneity, we performed pair comparisons of the populations in question. These pair comparisons revealed statistically significant differences between the populations from Bulgaria and Hungary, Bulgaria and Turkey, Hungary and Italy. From the point of view of breakpoint distributions in the area of the main hot spot of the dystrophin gene, the Bulgarian and Hungarian populations differ the most from the other studied populations because only

Bulgarian and Hungarian populations are significantly different from the two other studied populations. Danieli et al. (1993) and Herczegfalvi et al. (1999) had also previously noted the differences in distributions of deletion breakpoints among groups of D/BMD patients from different countries. On the other hand, Önengüt (2000) did not find any statistically significant differences in the distribution of breakpoints (divided into 3 intron segments, each segment included 20 introns) between the groups of D/BMD patients from India (from all over the country), North India, Turkey and Europe. As for the pair comparison of the groups of D/BMD patients from India and Turkey, we have not found any significant differences in the distributions of deletions breakpoints in introns 43–52, either. However, we have not compared (as did Önengüt et al. (2000)) the data from individual countries with a multi-population European group of D/BMD patients (reported by Danieli at al. (1993)), because we wanted to find out especially the differences among individual populations. We have not compared the proportion of deletions encompassing 1–5 exons like Önengüt et al. (2000) either, because in our opinion it does not necessarily express the differences in intron sequences in the dystrophin gene.

The results of our study support the theory suggested by other authors (Danieli et al., 1993; Herczegfalvi et al., 1999; Önengüt et al., 2000) that specific differences in intron sequences of the dystrophin gene can exist in different populations. As was first suggested by Danieli et al. (1993), these differences can be a result of a genetic drift.

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