CYP2A6 is a polymorphic enzyme, and the genotype of CYP2A6 has been shown to be associated with smoking habits and lung cancer. We investigated CYP2A6 polymorphism in Japanese from four different geographic areas of Japan, and in Ovambo and Turk populations. Two polymerase chain reaction (PCR)-restriction fragment length polymorphisms (RFLP) were used to identify the functionally important variants- CYP2A6*1A, *1B, *1F, *1G, *4A and *4D. In the Japanese population, the highest frequencies of CYP2A6*1A allele were observed in subjects from Fukuoka (Kyushu island) and Ehime (Shikoku island) prefectures, while subjects in Shimane and Tottori (both located on the Japan Sea side of Honshu island) showed the highest frequencies of the CYP2A6*1B allele. No subject homozygous for the CYP2A6*4A allele, a whole gene deletion type and prevalent among Orientals, was found in the Tottori and Shimane groups. In the Ovambo and Turk populations, CYP2A6*1A alleles were predominant. Furthermore, two alleles undetected in the Japanese were observed in these latter two ethnic groups: CYP2A6 variants *1G was solely in the Ovambos, and *1F solely in the Turks. The present study is the first to show inter-prefecture differences in CYP2A6*1A, *1B, *1F, *1G, *4A and *4D polymorphism of Japanese in relatively close but distinct geographic areas, and to evaluate the variations among these Japanese, Ovambo and Turk populations. The distribution results of these alleles could help to define the true significance of CYP2A6 polymorphism as a genetic susceptibility marker in the worldwide populations.