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# Polymorphism of the Microsatellite DXS8377 in a Population of Northern Thai Males

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## Abstract

**Objective:** This research investigated the polymorphism of the X-chromosomal short tandem repeats (X-STR) DXS8377 in Northern Thai males.

**Methods:** Genomic DNA was extracted from blood or buccal cells of 200 unrelated healthy individuals using Nucleospin® Blood or Nucleospin® Tissue (Macherey-Nagel), respectively. The DXS8377 locus was amplified by polymerase chain reaction (PCR) and the PCR product was analyzed by agarose gel electrophoresis. The allelic ladder was constructed and the size of all PCR products was analyzed by polyacrylamide gel electrophoresis using allelic ladder as a marker. The nucleotide sequences of the different-sized PCR products were analyzed to identify the DXS8377 alleles. Lastly, the Polymorphic information content *(PIC)* and Power of discrimination (PD) of DXS8377 DNA marker were calculated and the analysis of population relationships was performed using chi-square test of homogeneity.

**Results:** Eleven DXS8377 alleles were observed in the study population, the allele 49 was the most frequent with the allele frequency of 0.4400. According to previous studies, allele 49 was found in all of the populations studied and it was common among various populations. The polymorphism and the probability of DXS8377 to discriminate two unrelated individuals were moderate with a PIC and PD of 0.6973 and 0.7305, respectively. Most interestingly, the DXS8377 DNA marker was able to differentiate Mongoloid from Caucasoid and Negroid and it was also able to differentiate Northern Thai males from other Asian populations, including Korean, Japanese and Chinese.

**Conclusion:** Even though DXS8377 possessed moderate PIC and PD, this DNA marker was able to differentiate Mongoloid from Caucasoid and Negroid and it was also able to differentiate Northern Thai males from Korean, Japanese and Chinese.

**Keywords:** X-STR; DXS8377; Northern Thai males; Allele frequency; Polymorphic information content; Power of discrimination; Population relationships

## Introduction

Forensic science has been extensively developed especially in the study of population genetics. Because events that are harmful to persons and things happen all the time, forensic evidence that is able to link individual offenders to these events is very important. DNA evidence is reliable and its use recognized internationally. Population genetics investigates gene or allele frequency, both of which represent the genetic composition of a population. In this study, the population genetics of a population of Northern Thai males was evaluated using short tandem repeats (STRs).

Short tandem repeats (STRs) are microsatellites that are abundant throughout the human genome, especially on non-coding regions. STRs are advantageous as genetic markers because they are highly polymorphic and have a wide range of applications in forensic science and casework, including individual identification and the study of relationship between populations. Forensic STR analysis can be performed with biological evidence, such as blood cells, buccal cells and bones [1,2].

STRs can be found on both autosomal and sex chromosomes, including STRs on the Y-chromosome (Y-STRs) and X-chromosome (X-STRs). However, the recombination and mutation rates of STRs on autosome are higher than STRs on sex-chromosome. Therefore, STRs on sex-chromosome are highly useful for individual identification, paternity and kinship testing. Population genetics studies using Y-STRs as genetic markers are numerous, thus providing considerable reference information, whereas, studies of X-STRs are relatively new, although demand for them is increasing [3].

Children inherit X-STRs through both father (paternal inheritance) and mother (maternal inheritance). Male individuals inherit their one X-chromosome from their mothers, while female individuals receive two X-chromosomes — one each from the mother and father. Therefore, X-STRs are useful genetic markers for studying population genetics [4-6].

Athanasiadou et al. [4] developed a quadruplex PCR to analyze four X-STRs, including DXS101, DXS8377, HPRTB and STRX-1 (DXS981) in German population. The results showed that, among the four X-STRs used in their study, DXS8377 was the most polymorphic and the alleles 45 and 48 were found to be the most frequent. In 2005, Tabbada et al. [5] analyzed four X-STRs, including DXS101 HPRTB DXS8377 and DXS981(STRX-1), in Chinese, Japanese and Thais. The results revealed that, similar to the findings of Athanasiadou et al. [4], DXS8377 was the most polymorphic. In 2006, Gomes et al. [7] analyzed four X-STRs, including DXS7423, DXS101, DXS8377and HPRTB in Galician resided in Northern Spain and it was revealed that DXS8377 was the most polymorphic. In 2011, Aşicioğlu et al. [8] analyzed five X-STRs, including, DXS8377, DXS101, DXS6789, STRX-1 and HUMHPRTB, in Turkish and DXS8377 was found to be highly polymorphic and possessed high power of discrimination (PD). To summarize the analysis of DXS8377 in various populations, this DNA marker was highly polymorphic and, thus, can be applied in the individual identification and forensic caseworks.

The population relationships were normally employed to analyze the similarities between two different populations [9]. Katsuyama et al. [10] analyzed the genetic relationships of various populations, including Japanese, Northern Han, Hui, Uygur, Kazakh, Greek, Saudi Arabian, and Italian, by using four variable number of tandem repeats (VNTRs; D1S80, D4S43, COL2A1, D17S5) and one STR (ACTBP2) loci. By analyzing the allele frequencies of five genetic markers, it was revealed that these genetic markers can differentiate Asian and European populations. When considering a dendrogram constructed based on the allele frequencies, two clusters were clearly formed, in which five Asian populations (Japanese, Northern Han, Hui, Uygur, and Kazakh) were presented in one cluster, while two Europeans and one West Asian (Italian, Greek, and Saudi Arabian) were presented in another. Gonzalez-Perez et al. [11] analyzed the population relationships in the Mediterranean populations by using the combination of genetic markers; 18 Alu insertion polymorphisms (polymorphisms based on the presence and absence of an Alu element at a specific location of chromosome) and 3 STR loci. The results showed that, when considering the Alu/STR haplotype frequencies, the differentiation between Northern and Southern Mediterraneans was significantly noticeable.

The aim of this study was to investigate the polymorphism of X-STR DXS8377 in the population of Northern Thai males. The obtained data can be applied in the analysis of forensic casework, such as deficiency paternity testing and complicated kinship testing. Moreover, as Northern Thailand shares borders with Myanmar and Laos, and immigration for employment is common, the obtained data on DXS8377 in a population of Northern Thai males might be essential should a major incidence occur, like an earthquake that requires individual identification.

## Experimental Methods

### Population sampling and DNA extraction

DNA was collected from 200 unrelated healthy males, aged between 18-60 years old, and are resided in the North of Thailand (Figure 1). All participants signed informed consent forms. The Ethics Committee of the Faculty of Medicine, Chiang Mai University, Chiang Mai, Thailand approved this study (study code No. NONE-2557-02446 (2015). Genomic DNA was extracted from either blood cells or buccal cells using Nucleospin® Blood or Nucleospin® Tissue (Macherey-Nagel, Germany), respectively, according to the manufacturer's recommendation. The integrity of DNA was analyzed by 1.2% agarose gel electrophoresis.

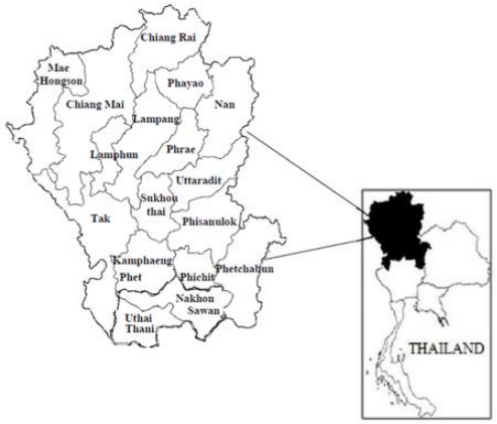


**Figure 1:** Geographical location of the populations used in this study. DNA samples were collected from 200 unrelated healthy individuals residing in the North of Thailand.

### DXS8377 amplification

PCR amplification of the DXS8377 STR marker was performed in a 50 µl reaction containing 50 ng to 100 ng of DNA template, 1x PCR buffer, 1.5 mM $MgSO_4$, 0.2 mM of each dNTPs, 0.5 µM of each primer (H-8377-F and H-8377-R primers; nucleotide sequences shown in Table 1) [4] and 1.0 µl of KOD-Plus-Neo DNA polymerase (Toyobo, Japan) using a Mastercycler® Personal thermal cycler (Eppendorf, USA). The STR amplification was cycled through the following PCR program: initial denaturation at 98°C for 10 min, 30 cycles of denaturation at 98°C for 10 sec, primer annealing at 60°C for 10 sec and product extension at 68°C for 20 sec, followed by final extension at 68°C for 10 min. The expected PCR products (225 bp to 264 bp) were examined by 2.5% ethidium bromide stained agarose gel electrophoresis using Low range DNA marker-A (25 bp to 500 bp) (Bio Basic Inc., Canada) as the standard DNA marker.

### Construction of allelic ladder

The different-sized PCR products were analyzed by 8.5% polyacrylamide gel electrophoresis (PAGE) using Low range DNA marker-A (25 bp to 500 bp) (Bio Basic Inc., Canada) as the standard DNA marker. Briefly, 1 µl of PCR product was mixed with 1 µl of 6x loading dye (50% v/v Glycerol, 200 mM EDTA pH 8.0, 0.125% w/v Bromophenol blue) and applied to the 8.5% PAGE. After electrophoresis at 120 volts for 2 h, the fragments were separated and detected by silver staining. Briefly, 100 ml of 1% (v/v) nitric acid was added to the gel and agitated for approximately 10 min. Nitric acid (1%) was subsequently discarded and the gel was rinsed with deionized water. Silver nitrate solution (100 ml of 0.012 M) was added to the gel and agitated for approximately 30 min. After 30 min, 0.012 M silver nitrate solution was discarded and the gel was rinsed with deionized water. Sodium carbonate (50 ml of 0.28 M) and 0.019% (v/v) formalin solution were added to the gel; the solution was discarded when it turned from colourless to brown. Sodium carbonate (50 ml of 0.28 M)

was repeatedly added to the gel until the DNA fragments were noticeable. Following the appearance of DNA fragments on the gel, 100 ml of 10% (v/v) glacial acetic acid was added to stop the reaction. After approximately 5 min, 10% glacial acetic acid was discarded and the gel was rinsed with deionized water until the acetic acid odour was undetectable.

PCR products of different sizes were selected and the DXS8377 loci of these samples were re-amplified from genomic DNA by PCR using different pairs of primers, H-8377-CF and H-8377-R (nucleotide sequences shown in Table 1). The obtained PCR products were purified from 2.5% ethidium bromide stained agarose gel using NucleoSpin® Gel and PCR clean-up (Macherey-Nagel, Germany). The purified PCR products were used as DNA templates for a second PCR amplification using H-8377-F and H-8377-R primers. Nucleotide sequences of the amplicon obtained from the second PCR amplification were subsequently analyzed by First BASE Laboratories SdnBhd (Malaysia) using a BigDye® Terminator v3.1 cycle sequencing kit. The designation of the DXS8377 allele was based on its complex composition: $(AGA)_x$-$(GGA\text{-}AGA)_y$-$(AGA)_2$-GGA-$(AGA)_6$. The total number of repeat units (x+2y+9) was calculated to identify DXS8377 alleles [12].

| Primers | Nucleotide sequences (5'->3') |
|---|---|
| H-8377-CF | CCCTTCTGCAACAGTTCTGCC |
| H-8377-F | ACCACTTCATGGCTTACCACAG |
| H-8377-R | TATGGACCTTTGGAAAGCTAG |

**Table 1:** Nucleotide sequences of primer.

## Identification of DXS8377 alleles

The X-STR DXS8377 alleles of all samples were identified by comparing the size of the PCR products to the allelic ladder constructed on 8.5% PAGE.

## Data analysis

Nucleotide sequences were analyzed and the allele frequencies ($P_i$) were determined using the equation $P_i$=(number of allele i)/(total alleles). The polymorphic information content *(PIC)* was determined using the equation $PIC=1-\Sigma P_i^2-(\Sigma P_i^2)^2+(\Sigma P_i^2)^4$, in which $P_i$ is the allele frequency of i. The power of discrimination (PD) was also determined using the equation $PD=1-\Sigma(P_i)^2$, in which $P_i$ is the frequency of genotype i [13].

The analysis of population relationships were also performed using chi-square test of homogeneity. This test was applied to analyze the similarities between two different populations using the equation $E_{ij}=[(R_{Ti})(C_{Tj})]/N$, in which $E_{ij}$ is the frequencies expected for population i at level j, $R_{Ti}$ is the total observations from population i, $C_{Tj}$ is the total observations at treatment level j and N is the total number of sample.

The chi-square test was determined using the equation $X^2=\Sigma[(O_{ij}-E_{ij})^2/E_{ij}]$, in which $O_{ij}$ is the frequencies observed in population i for level j of the categorical variable and $E_{ij}$ is the frequencies expected in population i for level j of the categorical variable. The degree of freedom (df) was determined using the equation df=(r-1)(c-1), in which r is the total number of populations and c is the total number of levels for the categorical variable [9].

| Allele | Allele frequency | Amplicon length (bp) | Nucleotide sequences |
|---|---|---|---|
| **42** | 0.005 | 225 | $P_F$-$N_{31}$-$(AGA)_{23}$-$(GGA\text{-}AGA)_5$-$(AGA)_2$-GGA-$(AGA)_6$-$N_{25}$-$P_R$ |
| **43** | 0.02 | 228 | $P_F$-$N_{31}$-$(AGA)_{24}$-$(GGA\text{-}AGA)_5$-$(AGA)_2$-GGA-$(AGA)_6$-$N_{25}$-$P_R$ |
| **45** | 0.01 | 234 | $P_F$-$N_{31}$-$(AGA)_{24}$-$(GGA\text{-}AGA)_6$-$(AGA)_2$-GGA-$(AGA)_6$-$N_{25}$-$P_R$ |
| **46** | 0.005 | 237 | $P_F$-$N_{31}$-$(AGA)_{29}$-$(GGA\text{-}AGA)_4$-$(AGA)_2$-GGA-$(AGA)_6$-$N_{25}$-$P_R$ |
| **47** | 0.005 | 240 | $P_F$-$N_{31}$-$(AGA)_{26}$-$(GGA\text{-}AGA)_6$-$(AGA)_2$-GGA-$(AGA)_6$-$N_{25}$-$P_R$ |
| **48** | 0.155 | 243 | $P_F$-$N_{31}$-$(AGA)_{29}$-$(GGA\text{-}AGA)_5$-$(AGA)_2$-GGA-$(AGA)_6$-$N_{25}$-$P_R$ |
| **49** | 0.44 | 245 | $P_F$-$N_{31}$-$(AGA)_{28}$-$(GGA\text{-}AGA)_6$-$(AGA)_2$-GGA-$(AGA)_6$-$N_{25}$-$P_R$ |
| **50** | 0.145 | 248 | $P_F$-$N_{31}$-$(AGA)_{31}$-$(GGA\text{-}AGA)_5$-$(AGA)_2$-GGA-$(AGA)_6$-$N_{25}$-$P_R$ |
| **51** | 0.17 | 251 | $P_F$-$N_{31}$-$(AGA)_{30}$-$(GGA\text{-}AGA)_6$-$(AGA)_2$-GGA-$(AGA)_6$-$N_{25}$-$P_R$ |
| **54** | 0.035 | 260 | $P_F$-$N_{31}$-$(AGA)_{29}$-$(GGA\text{-}AGA)_8$-$(AGA)_2$-GGA-$(AGA)_6$-$N_{25}$-$P_R$ |
| **55** | 0.01 | 264 | $P_F$-$N_{31}$-$(AGA)_{34}$-$(GGA\text{-}AGA)_6$-$(AGA)_2$-GGA-$(AGA)_6$-$N_{25}$-$P_R$ |

$P_F$: sequence of primer H-8377-F (22 bp)

$P_R$: complementary sequence of primer H-8377-R (21 bp)

$N_{31}$: CAGCAGAAGAAGAAGAAGCAGCAGCAGCAGC

$N_{25}$: CAGGGAACGAAGGAACAAAAATACA

**Table 2:** Allele nomenclature, allele frequency, amplicon length and nucleotide sequences of DXS8377 observed in Northern Thai males.

## Results and Discussions

### Designation of DXS8377 allele

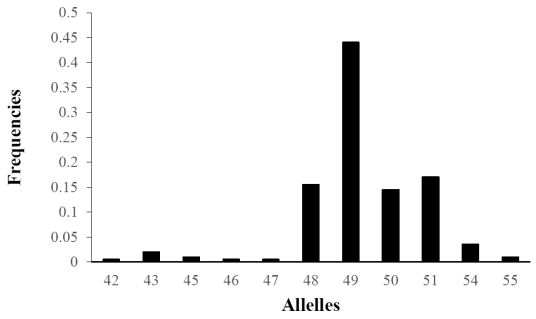


**Figure 2:** The DXS8377 allele distribution in Northern Thai males.

Eleven DXS8377 alleles were identified in this study: allele 42 (PCR fragment of 225 bp), allele 43 (228 bp), allele 45 (234 bp), allele 46 (237 bp), allele 47 (240 bp), allele 48 (243 bp), allele 49 (246 bp), allele 50 (249 bp), allele 51 (252 bp), allele 54 (261 bp) and allele 55 (264 bp) as shown in Table 2. According to this study, allele 49 was the most frequent with the allele frequency of 0.4400 and three alleles, including allele 42, 46 and 47, were the least frequent with the allele frequency of 0.0050 (Table 2). The DXS8377 allele distribution in Northern Thai males was presented in Figure 2.

When comparing the results of this research to previous studies that analyzed X-STR DXS8377 in other populations, allele 49 was found in all of the populations studied (Table 3) [4,5,15-21].

Interestingly, allele 49 was common among various populations as it was also the most frequent in Japanese, Xinjiang Uigur and Inner-Mongolia Mongol (China), Korean, German, Italian and Galician (Spain) populations [5,7,15-19] (Table 3 and Figure 3).

| Populations | Sample size | DXS8377 alleles (number of allele) | Highest allele frequency | PIC | PD |
|---|---|---|---|---|---|
| Northern Thai males (this study) | 200 | 42-43,45-51, 54-55 (11) | 49=0.4400 | 0.6973 | 0.7305 |
| German [4] | 120 | 37-56 (19) | 45,48=0.1390 | N/D | N/D |
| Thai [5] | 157 | 40,44-56,58 (16) | 47=0.1515 | N/D | M=0.9005/F=0.9817 |
| Japanese [5] | 95 | 42-55,58 (15) | 49=0.1684 | N/D | M=0.8971/F=0.9805 |
| Galician (Spain) [7] | 68 | 40-57 (17) | 46,48,49,50,53=0.0960 | N/D | N/D |
| Japanese [15] | 401 | 42-58 (17) | 49=0.1710 | 0.889 | M=0.8870/F=0.9820 |
| Japanese [16] | 546 | 41-58 (18) | 49=0.1520 | 0.9 | M=0.9000/F=0.9780 |
| Philippines [5] | 155 | 37,43-55, 57-58 (16) | 48=0.1329 | N/D | M=0.9092/F=0.9846 |
| Chinese [5] | 90 | 40,42-56 (16) | 47=0.1800 | N/D | M=0.8876/F=0.9771 |
| Xinjiang Uigur (China) [17] | 235 | 41-58 (18) | 49=0.1398 | 0.9101 | M=0.9020/F=0.9846 |
| Inner-Mongolia Mongol (China) [17] | 209 | 42-55 (14) | 49=0.1608 | 0.8987 | M=0.8853/F=0.9814 |
| Korean [18] | 300 | 41-59 (19) | 49=0.1360 | 0.897 | 0.983 |
| Italy (Umbrian, Perugia, Terni) [19] | 200 | 40-56 (17) | 48,49=0.1184 | 0.9 | 0.97 |
| Slovak [20] | 116 | 40-56,58 (18) | 48=0.1550 | N/D | M=0.9100 |
| Belarusian [20] | 180 | 42-57,60 (17) | 48=0.1390 | N/D | M=0.9100 |
| German [5] | 105 | 40-56 (17) | 49 = 0.1500 | N/D | M=0.9148/F=0.9865 |

PIC: Polymorphic information content
PD: Power of discrimination
N/D: Not determined

**Table 3:** Allele frequency, PIC and PD of DXS8377 observed in various populations.

In this study, the subordinate allele was allele 51, with an allele frequency of 0.1475 (Table 2 and Figure 2). Previous studies also found allele 51 to be subordinate, including the genetic analysis in populations of Han Chinese, Galicians (Spain) and Slovakians, as shown in Table 3 [7,17,20]. Allele 48 was the third frequent in this study with the allele frequency of 0.1450, as shown in Table 2 and Figure 2. This result is similar to the analysis of DXS8377 in populations of Chinese, Japanese and Inner-Mongolia Mongol (China), in which allele 48 was also the third frequent (Table 3) [5,16,17]. Allele 50 was the fourth frequent in this study with the allele frequency of 0.1200, as shown in Table 2 and Figure 2. This result is similar to the analysis of DXS8377 in Chinese, Japanese and Inner-

Mongolia Mongol (China), in which allele 50 was also the fourth frequent (Table 3) [5,15-17]. Interestingly, Tabbada et al. [5] found allele 47 was the most frequent of DXS8377 observed in Thai populations, while in this study, allele 47 was among the least frequent alleles observed in Northern Thai males. However, Tabbada et al. [5] did not mention the regions of Thailand in which they collected the DNA samples.

## Statistical analysis of DXS8377

The forensic efficiencies of X-STR DXS8377 in the population of Northern Thai males studied here are presented in Table 3. It was revealed that the polymorphism of this DNA marker is moderate, with a PIC of 0.6973 or 69.73%. The probability of DXS8377 to discriminate two unrelated individuals is moderate, with a PD of 0.7305 (73.05%). Most interestingly, according to the analysis of population relationships, the DXS8377 DNA marker was able to differentiate Mongoloid from Caucasoid and Negroid and it was also able to differentiate Northern Thai males from other Asian populations, including Korean, Japanese and Chinese.

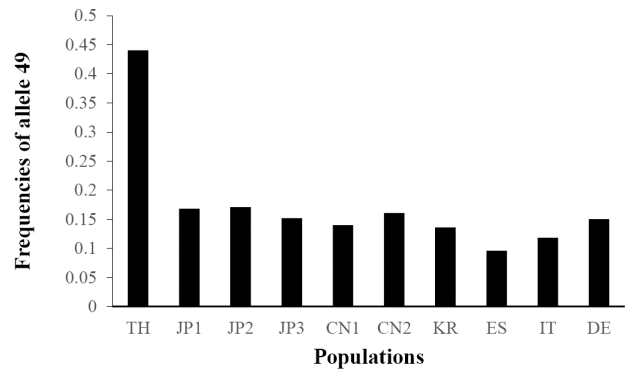


**Figure 3:** Various populations in which allele 49 of DXS8377 was the most frequent.
TH: Northern Thai males (this study) CN2: Inner-Mongolia Mongol (China) [17]
JP1: Japanese [5] KR: Korean [18]
JP2: Japanese [15] ES: Galician (Spain) [7]
JP3: Japanese [16] IT: Umbrian, Perugia, Terni (Italy) [19]
CN1: Xinjiang Uigur (China) [17] DE: German [5]

## Polymorphic information content *(PIC)*

The polymorphic information content *(PIC)* obtained from this study was 0.6973, which was moderate compared to previous studies in Japanese, Xinjiang Uigurs, Inner-Mongolia Mongol, Korean and Italian populations[15-19] (Table 3). According to previous studies, the overall PIC of DXS8377 is high, with the PIC of 0.8890-0.9101, meaning this DNA marker is highly polymorphic. However, the fact that most of the DNA samples used in this study were obtained from Thai male volunteers who resided in Chiang Mai might explain the moderate PIC observed.

## Power of discrimination (PD)

The power of discrimination obtained from this study was 0.7305 (Table 3), which was moderate when compared to the analysis of DXS8377 in other populations from previous studies, including Thai, Japanese, Philippines, Chinese, Xinjiang Uigur, Inner-Mongolia Mongol, German, Slovakian, Korean and Italian populations [5,15-20] (Table 3). According to previous studies, the overall PD of DXS8377 is high, with the PD of 0.8853-0.9865, meaning this DNA marker possesses a high probability of discriminating between two unrelated individuals. However, the limited number of alleles identified in the population of Northern Thai males from this study might explain the moderate PD observed.

## Population relationships

The analysis of population relationships to examine the similarity between Mongoloid, Caucasoid and Negroid populations using DXS8377 DNA marker revealed that these populations were significantly different at chi-square ($X^2$) of 226.57, ($p<0.0001$, df=40). The DXS8377 allele distribution in Mongoloid, Caucasoid and Negroid (Figure 4) [19,21].

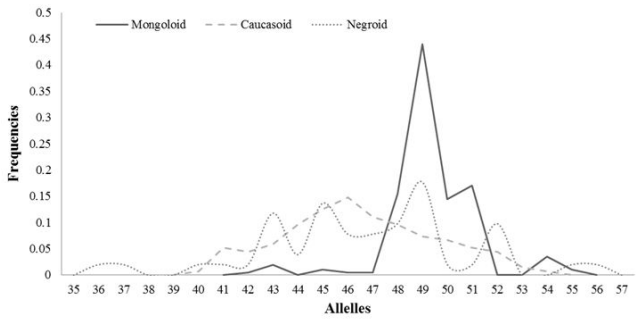


**Figure 4:** The DXS8377 allele distribution in Mongoloid, Caucasoid and Negroid.

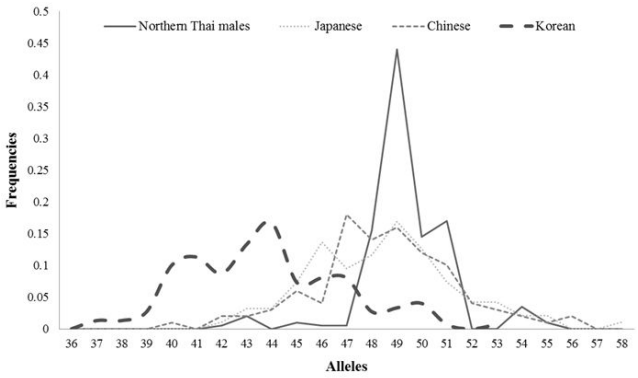


**Figure 5:** The DXS8377 allele distribution in Northern Thai males, Japanese, Chinese and Korean.

Furthermore, the analysis of population relationships to examine the similarity between Northern Thai males (this study) and other Asian populations, including Korean, Japanese and Chinese, suggested that Northern Thai males and Korean, Northern Thai males and Japanese and Northern Thai males and Chinese were significantly

different at $X^2$ of 111.27 ($p<0.0001$, df=16), $X^2$ of 92.976 ($p<0.0001$, df=14) and $X^2$ of 87.039 ($p<0.0001$, df=14), respectively. The DXS8377 allele distribution in Northern Thai males, Japanese, Chinese and Korean (Figure 5) [5,18]. These results suggested that the X-STR DXS8377 marker was able to differentiate Mongoloid from Caucasoid and Negroid populations. Most interestingly, DXS8377 was able to differentiate Northern Thai males from other Asian populations, including Korean, Japanese and Chinese.

To summarize, the analysis of DXS8377 in a population of Northern Thai males in this study revealed that the polymorphism of this DNA marker is moderate, with a PIC of 0.6973, or 69.73%. The probability of DXS8377 to discriminate two unrelated individuals is moderate, with a PD of 0.7305 (73.05%). The fact that most of the DNA samples used in this study were obtained from Northern Thai males who were resided in Chiang Mai, Thailand and the limited number of alleles identified in this study might explain the moderate PIC and PD observed [14]. Interestingly, according to the analysis of population relationships, DXS8377 alleles observed in Mongoloid were significantly different from Caucasoid and Negroid. Moreover, the DXS8377 alleles observed in Northern Thai males were significantly different from other Asian populations, including Korean, Japanese and Chinese.

## Conclusion

This study analyzed the polymorphism of DXS8377 in 200 unrelated healthy individuals of Northern Thai males. Eleven DXS8377 alleles were observed in the population examined, of which the PCR products ranged from 225 bp to 264 bp. The PIC and PD of DXS8377 in the population of Northern Thai males were 0.6973 and 0.7305, respectively. DXS8377 alleles observed in Mongoloid were significantly different from Caucasoid and Negroid and the DXS8377 alleles observed in Northern Thai males were significantly different from other Asian populations, including Korean, Japanese and Chinese. Therefore, the DXS8377 DNA marker was able to differentiate Mongoloid from Caucasoid and Negroid and it was also able to differentiate Northern Thai males from Korean, Japanese and Chinese.

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## References

1. Butler JM (2010) Fundamentals of Forensic DNA Typing. Academic Press, Amsterdam, Netherlands.
2. Hudson R, Thompson M, Maughan N, Hudden D, Brideson A, et al. (2004) Forensic sampling and DNA Databases in Criminal Investigations. Melbourne Government Printer, Melbourne, Australia.
3. Schaffner SF (2004) The X chromosome in population genetics. Nat Rev Genet 5: 43-51.
4. Athanasiadou D, Stradmann-Bellinghausen B, Rittner C, Alt KW, Schneider PM (2003) Development of a quadruplex PCR system for the genetic analysis of X-chromosomal STR loci. Int Congr Ser 1239: 311-314.
5. Tabbada KA, Ungria MCA, Faustino LP, Athanasiadou D, Stradmann-Bellinghausen B, et al (2005) Development of a pentaplex X-chromosomal short tandem repeat typing system and population genetic studies. Forensic Sci Int 154: 173-180.
6. Edelmann J, Szibor R (2001) DXS101: A highly polymorphic X-linked STR. Int J Legal Med 114: 301-304.
7. Gomes I, Carracedo A, Amorim A, Gusmao L (2006) A multiplex PCR design for simultaneous genotyping of X chromosome short tandem repeat markers. Int Congr Ser 1288: 313-315.
8. Aşicioğlu F, Akyüz F, Ozdilli K, Oguz F, Çarin M (2011) X-Chromosomal short tandem repeat loci in the Turkish population. Afr J Biotecnol 10: 1334-4338.
9. Bolboaca SD, Jantschi L, Sestras AF, Sestras RE, Pamfil DC (2011) Pearson-Fisher Chi-Square Statistic Revisited. J Inf Sci 2: 528-545.
10. Katsuyama Y, Inoko H, Imanishi T, Mizuki N, Gojobori T, et al. (1998) Genetic relationships among Japanese, northern Han, Hui, Uygur, Kazakh, Greek, Saudi Arabian, and Italian populations based on allelic frequencies at four VNTR (D1S80, D4S43, COL2A1, D17S5) and one STR (ACTBP2) loci. Hum Hered 48: 126-137.
11. Gonzalez-Perez E, Esteban E, Via M, Gaya-Vidal M, Athanasiadis G, et al. (2010) Population Relationships in the Mediterranean Revealed by Autosomal Genetic Data (Alu and Alu/STR Compound Systems). Am J Phys Anthropol 141: 430-439.
12. Edelmann J, Deichsel D, Hering S, Plate I, Szibor R (2002) Sequence variation and allele nomenclature for the X-linked STRs DXS9895, DXS8378, DXS7132, DXS6800, DXS7133, GATA172D05, DXS7423 and DXS8377. Forensic Sci Int 129: 99-103.
13. Butler JM (2012) X-Chromosome Analysis. Academic Press, Burlington, USA.
14. Bar W, Brinkmann B, Budowle B, Carracedo A, Gill P, et al. (1997) DNA recommendations. Further report of the DNA Commission of the ISFH regarding the use of short tandem repeat systems. International Society for Forensic Haemogenetics. Int J Legal Med 110: 175-176.
15. Asamura H, Sakai H, Ota M, Fukushima H (2006) Japanese population data for eight X-STR loci using two new quadruplex systems. Int J Legal Med 120: 303-309.
16. Nakamura Y, Samejima M, Tamaki K, Minaguchi K (2013) Multiplex PCR for 18 X-chromosomal STRs in Japanese population. Leg Med (Tokyo) 15: 164-170.
17. Liu QL, Lu DJ, Li XG, Zhao H, Zhang JM, et al. (2011) Development of the nine X-STR loci typing system and genetic analysis in three nationality populations from China. Int J Leg Med 125: 51-58.
18. Shin KJ, Kwon BK, Lee SS, Yoo JE, Park MJ, et al. (2004) Five highly informative X-chromosomal STRs in Koreans. Int J Leg Med 118: 37-40.
19. Coletti A, Lottanti L, Lancia M, Margiotta G, Carnevali E, et al. (2006) Allele distribution of 6 X-chromosome STR loci in an Italian population sample. Int Congr Ser 1288: 292-294.
20. Cybulska L, Wysocka J, Re K, Kapin E, Mikulich AI, et al. (2008) Polymorphism of four X-chromosomal STR loci in Belarusians and Slovaks. Forensic Sci Int 1: 145-146.
21. Gomes I, Alves C, Maxzud K, Pereira R, Prata MJ, et al. (2007) Analysis of 10 X-STRs in three African populations. Forensic Sci Int 1: 208-211.