

## 조직별 및 나이에 따른 미토콘드리아 DNA 결손 ( $\Delta$ mtDNA<sup>4977</sup>)의 축적

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2 . 3 . 2 . 2 . 1 . 1  
1 . 1 . 1 . 1,2

### Accumulation of mtDNA Deletion ( $\Delta$ mtDNA<sup>4977</sup>) showing Tissue-Specific and Age-Related Variation

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**Objectives:** Controversial arguments exist on both the case for and against on the accumulation of mitochondrial DNA (mtDNA) deletion in association to tissue and age. The debate continues as to whether this mutation is a major contributor to the phenotypic expression of aging and common degenerative diseases or simply a clinical insignificant epiphenomenon. The objective of this study was to determine whether the accumulation of mtDNA deletion is correlated with age-related and tissue-specific variation.

**Materials and Methods:** One hundred and fifty-seven tissues from blood, ovary, uterine muscle, and abdominal muscle were obtained from patients ranging in age from 31~60 years. After reviewing the clinical reports, patients with mitochondrial disorder were excluded from this study. The tissues were obtained at gynecological surgeries with the consent of the patient. Total DNA isolated from blood, ovary, uterine muscle, and abdominal muscle was amplified by two rounds of PCR using two pairs of primers corresponding to positions 8225-8247 (sense), 13551-13574 (antisense) for the area around deleted mtDNA and 8421-8440 (sense), 13520-13501 (antisense) for nested PCR product. A statistical analysis was performed by  $\chi^2$ -test.

**Results:** About 0% of blood, 94.8% of ovary, 71.4% of uterine muscle, and 86.1% abdominal muscle harbored mtDNA deletion. When we examined the proportion of deleted mtDNA according to age deletion rate was 90% of ovary, 63.6% of uterine muscle, 77.7% of abdominal muscle in thirties and 100% of all tissue in fifties.

**Conclusion:** The findings of this study suggest that the mtDNA deletion is varied in tissue-specific

pattern and increases with aging.

**Key Words:** Mitochondrial DNA, Common deletion, PCR

DNA 16.5-kb 가 가  
 DNA , , DNA  
 가 가 .  
 가 2.  
 2~10 copy DNA (mtDNA) 1) DNA  
 가 .<sup>1</sup> DNA  
 homoplasmy .<sup>2</sup> , , , 55  
 DNA , 가 10 mM Tris-HCl, 1 mM EDTA (pH 8.0), 0.1 mg/ml  
 . DNA proteinase K (Invitrogen, CA, USA), 0.5% SDS  
 heteroplasmy 가 . DNA phenol : chloro-  
 form : isoamylalcohol (25 : 24 : 1)  
 chloroform : iso-  
 amylalcohol (24 : 1) . DNA  
 가 0.3 M NaCl 2 가 -80  
 1 , 4 12,000 rpm  
 , 70% alcohol .  
 DNA 가 DNA 200~400 µl 10 mM Tris-HCl, 1 mM EDTA  
 (pH 8.0) -20 .  
 2) (PCR)  
 PCR Perkin Elmer GeneAmp 2400 Thermal  
 cycler , PCR 2 U Taq polymerase  
 (Promega, WI, USA), 1×PCR buffer, 2.5 mmol/L MgCl<sub>2</sub>,  
 1 mM dNTPs, 0.2 µmol/L primer, 100 ng DNA  
 50 µl . , MT-1 (5'-AT-  
 TCCCCTAAAAATCTTTGAAATG-3') MT-3 (5'-AG-  
 AGTAATAGATAGGGATCAGGGG-3') primer  
 first PCR , MQ-1 (5'-AGAGTAATAG-  
 ATAGGGATCAGGGG-3') MQ-3 (5'-CCTAGGATT-  
 GTGGGGGC-3') primer second PCR  
 . PCR 94 5 , 94 30 ,  
 1. 60 30 , 72 30 30 cycles  
 72 10 . PCR  
 157 가 2% agarose gel EtBr , UV

3) 100% (Table 2).

*t*-test Chi-square ( $\chi^2$ -test)  
5%

DNA 가  
90%

가

0%, ROS (Reactive  
Oxygen Species) ROS 가

94.8%, 71.4%,  
86.1% 가 가  
DNA ROS DNA  
DNA  
가

(Table 1).

(p=0.0067)

가 (Table 1).

30, 40, 50  
, 30  
가  
50

3-6 가  
 $\Delta$ mtDNA<sup>4977</sup> (common deletion) Common  
deletion

DNA  
가

**Table 1.** mtDNA deletion rate of various tissues

Tissue	mtDNA deletion (%)
Ovary	37* / 39† (94.8)
Abdominal muscle	31 / 36 (86.1)
Uterine muscle	32 / 35 (71.4)‡
Lymphocyte	0 / 47 (0)

\*: number of mtDNA deletion, †: total number of sample,  
‡: inter-tissue variation of ovary and uterine muscle,  
p=0.0067 (p<0.05)

**Table 2.** mtDNA deletion rate according to age

Age	Ovary	Uterine muscle	Abdominal muscle
30~39	10* / 11† (90%)	7* / 11† (63.6%)	7* / 9† (77.7%)
40~49	20 / 21 (95%)	14 / 20 (70%)	17 / 20 (77.7%)
50~59	7 / 7 (100%)	4 / 4 (100%)	7 / 7 (100%)

\*: number of mtDNA deletions, †: total number of samples

DNA direct repeat DR1 (Direct repeat 1, nt 13447-13459) DR2 (Direct repeat 2, nt 8470-8482)가  
 heavy strand DR1 light strand DR2  
 가  
 가  
 DNA DR1  
 가<sup>13</sup> DNA  
 가  
 ROS  
 가<sup>14</sup>  
 DNA  
 가  
 가

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