Alteration of SF3B1 and SRSF2 Genes in Myelodysplastic Syndromes Patients in Upper Northern Thailand

Phuttirak Yimpak¹, Adisak Tantiworawit², Thanawat Rattanathammethee², Sirinda Angsuchawan¹, Sikrai Laowatthanapong¹, Witoon Tasuya¹, Kanokkan Bumroongkit¹*  

Abstract

**Background:** The frequency and pattern of mutation in SF3B1 and SRSF2 RNA splicing machinery genes were found to vary among myelodysplastic syndrome (MDS) patients in different populations. There have been less reports of incidence of these gene mutations in Thailand especially in upper northern Thailand. This study therefore had aims to investigate the frequency and pattern of mutation in mutational hotspot of SF3B1 and SRSF2 genes among MDS patients in upper northern Thailand and to investigate the clinical features associated with the mutations. **Methods:** Fifty-five MDS patients who underwent treatment at Maharaj Nakorn Chiang Mai Hospital participated in this study. The detection of SF3B1 and SRSF2 hotspot mutations was carried out using polymerase chain reaction followed by Sanger sequencing. In addition, clinical features of individual patients with these gene mutations were also investigated. **Results:** SF3B1 mutations (SF3B1mut) were found in 9 patients (16.4%) including E622D (1/9), R625C (1/9), H662Q (1/9), K700E (5/9), and Q699H co-mutation with K700E (1/9). SRSF2 mutations (SRSF2mut) were found in 4 patients (7.3%) which included P95H (3/4) and P95L (1/4). The SF3B1mut was associated with lower hemoglobin levels (p = 0.023) and higher platelet counts (p = 0.047) when compared with MDS patients without SF3B1mut, while SRSF2mut tended to occur in patients with a higher percentage of bone marrow blasts (p = 0.074). **Conclusion:** The findings confirmed the difference in frequency of SF3B1 and SRSF2 mutations among different populations. Specifically, we found a co-mutation of Q699H and K700E that has not been previously reported in MDS patients in the COSMIC database. It was also found that SF3B1mut was strongly associated with low hemoglobin level, and high platelet counts whereas SRSF2mut was mostly clustered in MDS with excess blasts subsequently increasing the probability of progression to acute myeloid leukemia.  

Keywords: Myelodysplastic syndrome- SF3B1 gene mutation- SRSF2 gene mutation- splicing machinery gene

**Introduction**

Myelodysplastic syndromes (MDS) has been defined as a group of clonal bone marrow (BM) disorders characterized by ineffective hematopoiesis which contributes to morphologic dysplasia in hematopoietic cells and peripheral blood cytopenia(s) (Arber et al., 2016). Currently, the mutations in RNA splicing machinery genes have been reported as being concretely proportional in MDS patients. The RNA splicing is a key to the regulation of gene expression. Intact and accurate RNA splicing is essential for the accuracy of final protein products. Any alterations in these pathways contribute to the dysfunction of the final protein products and subsequently are the cause of disease. SF3B1, one of the RNA splicing machinery genes, is located on chromosome 2q33.1. This gene encodes the subunit 1 of the splicing factor 3b complex which is an essential component of the U2snRNP complex and is important for the recognition of the 3' splice site between intron and exon in normal RNA splicing (Padgett, 2012; Cazzola et al., 2013). SF3B1mut is frequently found in MDS patients and is associated with the presence of ring sideroblasts (RS) which are erythroid precursors showing iron deposition in the mitochondria cover around the nuclear circumference (Papaemmanuil et al., 2011; Cazzola et al., 2013). Currently, in the World Health Organization (WHO) 2016 guidelines, SF3B1mut is used as a biomarker for the classification of MDS (Arber et al., 2016). Several studies have reported that MDS with SF3B1mut are associated with a favorable prognosis (Malcovati et al., 2011; Papaemmanuil et al., 2011; Cui et al., 2012). In addition to SF3B1, SRSF2mut are commonly detected in MDS patients. SRSF2 is located on chromosome 17q25.1 and encodes for the serine/arginine

¹Department of Anatomy, ²Division of Hematology, Department of Internal Medicine, Faculty of Medicine, Chiang Mai University, Chiang Mai, Thailand. *For Correspondence: kanokkan.bumr@cmu.ac.th
rich splicing factor 2 (SRSF2) which is associated with the regulation of constitutive and alternative pre-mRNA splicing (Long and Caceres, 2009; Wu et al., 2012). SRSF2mut has an unfavorable impact on MDS patients and its presence predicts shorter overall survival when compared with MDS patients without SRSF2mut (Thol et al., 2012). As the regulation of RNA splicing is essential for normal functioning of the cell, the alteration in SF3B1 and SRSF2 splicing machinery genes may certainly be involved in the pathogenesis of MDS. There have been few studies regarding SF3B1 and SRSF2 gene mutations and clinical features of MDS patients in Thailand especially in upper northern Thailand. Therefore, this study aimed to investigate the frequency and patterns of the mutations along with the clinical features including RS in SF3B1 and SRSF2 gene mutations among MDS patients in upper northern Thailand.

Materials and Methods

Patients

From 2017 – 2018, a total of 55 BM samples and 37 dried BM smear slides of MDS patients with ≥18 years old from the Division of Hematology, Department of Internal Medicine, Maharaj Nakorn Chiang Mai Hospital (Chiang Mai, Thailand) were recruited onto the study. All of patients were diagnosed MDS confirmed by a hematologist according to WHO 2016 classification criteria. Clinical features and hematological data were collected from electronic medical records. This study was approved by the Ethics and Research Committee of the Faculty of Medicine, Chiang Mai University [Certificate No. 327 /2017 Study code ANA-2560-04844].

Prussian blue staining for detecting ring sideroblasts (RS)

The protocol was modified from Jouihan (2012) (Jouihan, 2012). Dried BM smear slides were immersed in a freshly mixed solution of 2% potassium ferrocyanide and 2% HCl in equal proportions and counterstained with nuclear fast red. One hundred erythroid cells were counted for each specimen (Mufti et al., 2008). RS was defined by the presence of at least five siderotic granules extending over at least one third of the nucleus circumference in a stained BM smeared slide (Lee et al., 2008). The presence of ≥15% RS from 100 erythroid cells was defined as positive for RS.

Detection of SF3B1 exon 14, 15 and SRSF2 exon 1 mutation

Genomics DNA was extracted using inorganic salting out method modified from Seielstad et al., (1999)(Seielstad et al., 1999) and the QIAamp® DNA Mini Kit (QIAGEN, Hilden, Germany) following the manufacturer’s instructions. The genomic DNA was amplified by using polymerase chain reaction (PCR) targeting the mutational hotspot of SF3B1 (exon 14 and 15) and SRSF2 (exon 1). The sequences of each primer are shown in Table 1. The PCR process was as follows: initial denaturation step at 95°C for 1 minute, followed by 35 cycles of denaturation at 95°C for 15 seconds, annealing at 60°C for 15 seconds, and extension at 72°C for 10 seconds on a thermocycler (Eppendorf Mastercycler, USA). PCR products were purified using NucleoSpin® Gel and PCR Clean-up (Macherey-Nagel, Germany) and sequenced bidirectionally using the BigDye® Terminator Version 3.1 Cycle Sequencing Kit (Applied Biosystems, Foster City, California, USA). Sequencing reactions were purified using the Ethanol/EDTA/Sodium acetate precipitation method and run on the ABI Prism 3130® DNA Analyzer (Applied Biosystems, Foster City, California, USA). The electropherograms were analyzed using the Seqscape program V2.5 (Applied Biosystems, Foster City, California, USA) to detect the mutations by comparing the resultant products with reference DNA sequences from Genbank (NG_032903.2 for SF3B1 gene and NG_032905.1 for SRSF2 gene). In all cases of mutational detection, PCR and sequencing were repeated to confirm the results.

Statistical analysis

The frequencies of gene mutation were counted and percentages calculated. The Mann-Whitney U test or Student’s t-test was used for the comparison of numerical variables (such as age, BM blast, white blood cell count (WBC), absolute neutrophil count (ANC), platelet count, and hemoglobin between groups. Fisher’s exact test or the Chi-square test was performed to analyze the significance of the association between gene mutations and categorical variable parameters, such as sex, WHO classification, RS, cytogenetic categories and IPSS-R categories. A p-value less than 0.05 was considered to be statistically significant.

Results

Clinical data and laboratory feature of all 55 MDS patients

The characteristics of 55 upper northern Thai MDS patients are shown in Table 2. Out of the 55 patients, 37 patients had BM smeared slides for RS detection. RS was positive in 4 patients (10.8%). The morphology of RS is shown in Figure 1. The clinical data of patients with relation to SF3B1mut
SF3B1 and SRSF2 Gene Mutations and Clinical Impact in MDS Patients

For PCR and sequencing Exon 14
Forward GTGGACAAACCTGACCTAACC
Reverse CCTCACCCCGTTTACCT

Table 1. The Primers for PCR Amplification and Sequencing of SF3B1 and SRSF2 Genes

SF3B1

| For PCR | Exon 14-15 | Forward | TAGAGTGGAAGGCCGAGAGA | (Kang et al., 2015) |
|---------|------------|---------|-----------------------|-------------------|
|         | Reverse    | TTCAAGAAAGCAGCCAAACC | (Kang et al., 2015) |

For Sequencing Exon 14
Forward TTAGTGGAAGGCCGAGAGA
Reverse CAACCTACATGTCAAAGACCTC

Exon 15
Forward GTTGATATATTGAGAGAATC
Reverse TTCAAGAAAGCAGCCAAACC

SRSF2

For PCR
Exon 14-15 Forward TAGAGTGGAAGGCCGAGAGA | (Kang et al., 2015) |
Reverse TTCAAGAAAGCAGCCAAACC | (Kang et al., 2015) |

For Sequencing Exon 14 Forward TAGAGTGGAAGGCCGAGAGA | (Kang et al., 2015) |
Reverse CAACCTACATGTCAAAGACCTC | (Kang et al., 2015) |

Exon 15 Forward GTTGATATATTGAGAGAATC | (Kang et al., 2015) |
Reverse TTCAAGAAAGCAGCCAAACC | (Kang et al., 2015) |

Figure 2. Electropherograms by Sanger sequencing show mutations in splicing machinery genes: (a), (b), and (c), SF3B1mut in exon 14; (d) and (e), SF3B1mut in exon 15; (f) and (g), SRSF2mut in exon 1; Red arrow, point of the location of base substitution

Figure 3. World Health Organization Classification of MDS Patients with SF3B1 and SRSF2 Mutation

and SRSF2mut status are shown in Table 2. The SF3B1mut were found in 9 patients (16.4%), one of the 9 patients having 2 affected codons. The SRSF2mut were found in 4 patients (7.3%). There was a statistically significant difference in the World Health Organization classification between patients with SF3B1mut and without SF3B1mut (p = 0.042). On analysis of the clinical data (blood count), patients with transformation to AML were excluded.
Table 2. Clinical Characteristics of MDS Patients According to the Alteration of SF3B1 and SRSF2 Status

| Characteristics | All patients (n = 55) | SF3B1 wild type (n = 46, 83.6%) | SF3B1 mutation (n = 9, 16.4%) | P | SRSF2 wild type (n = 51, 92.7%) | SRSF2 mutation (n = 4, 7.3%) | P |
|----------------|-----------------------|---------------------------------|--------------------------------|----|---------------------------------|--------------------------------|----|
| Median Age (years), (range) | 65 (31-93) | 65 (31-93) | 62 (42-76) | 0.358 | 65 (31-91) | 67 (56-93) | 0.484 |
| Sex | | | | | | | |
| Male, n (%) | 29 (52.7) | 26 (56.5) | 3 (33.3) | | 26 (51.0) | 3 (75.0) | |
| Female, n (%) | 26 (47.3) | 20 (43.5) | 6 (66.7) | | 25 (49.0) | 1 (25.0) | |
| WHO classification, n (%) | | | | | | | |
| MDS-SLD | 11 (20.0) | 9 (19.6) | 2 (22.2) | 0.042* | | | 0.372 |
| MDS-MLD | 14 (25.5) | 13 (28.3) | 1 (11.1) | | 14 (27.5) | | |
| MDS-RS | 2 (3.6) | - | 2 (22.2) | | 2 (3.9) | | |
| MDS with isolate del(5q) | 1 (1.8) | - | 1 (11.1) | | 1 (2.0) | | |
| MDS-EB-1 | 2 (3.6) | 2 (4.3) | - | | 2 (3.9) | | |
| MDS-EB-2 | 10 (18.2) | 8 (17.4) | 2 (22.2) | | 8 (15.7) | 2 (50.0) | |
| MDS-U | 10 (18.2) | 9 (19.6) | 1 (11.1) | | 9 (17.6) | 1 (25.0) | |
| Transformation to AML | 5 (9.1) | 5 (10.8) | - | | 4 (7.8) | 1 (25.0) | |
| Blood counts, median (range) (n=50) | | | | | | | |
| BM blasts (%) | 1.0 (1.0 - 19.0) | 1.0 (0.0 - 19.0) | 1.0 (0.0 - 12.0) | 0.467 | 1.0 (0.0 - 19.0) | 13.0 (1.0 - 19.0) | 0.074 |
| WBC (>10^9/L) | 4.4 (0.7 - 47.7) | 4.4 (0.7 - 47.7) | 5.3 (2.3 - 13.0) | 0.92 | 4.4 (0.7 - 18.2) | 5.5 (4.0 - 47.7) | 0.213 |
| ANC (>10^9/L) | 4.2 (0.9 - 9.1) | 4.3 (0.9 - 9.1) | 2.7 (2.2 - 5.0) | 0.116 | 4.2 (0.9 - 9.1) | 4.0 (4.0 - 5.7) | 0.61 |
| Hemoglobin (g/dL) | 8.5 (4.2 - 13.4) | 8.7 (6.2 - 13.4) | 6.7 (4.2 - 10.1) | 0.023* | | 8.5 (4.2 - 13.4) | 8.7 (6.2 - 8.9) | 0.591 |
| Platelets (>10^9/L) | 66.0 (5.0 - 485.0) | 58.0 (5.0 - 485.0) | 129.0 (36.0-290.0) | 0.047* | 62.0 (5.0 - 485.0) | 139.0 (77.0 - 307.0) | 0.111 |
| Cytogenetic categories, n (%) | | | | | | | |
| Very good | 3 (5.5) | 3 (6.5) | - | | 3 (5.9) | - | |
| Good | 39 (79.1) | 32 (69.6) | 7 (77.8) | | 37 (72.5) | 2 (50.0) | |
| Intermediate | 6 (11.0) | 5 (10.9) | 1 (11.1) | | 5 (9.8) | 1 (25.0) | |
| Poor | 2 (3.6) | 2 (4.3) | - | | 2 (3.9) | - | |
| Very poor | 4 (7.3) | 3 (6.5) | 1 (11.1) | | 4 (7.8) | - | |
| N/A | 1 (1.8) | 1 (2.2) | - | | - | 1 (25.0) | |
| IPSS-R categories, n (%) | | | | | | | |
| Very low | 7 (12.7) | 6 (13.0) | 1 (11.1) | | 7 (13.7) | - | |
| Low | 25 (45.5) | 20 (43.5) | 5 (55.6) | | 24 (47.1) | 1 (25.0) | |
| Intermediate | 5 (9.1) | 4 (8.7) | 1 (11.1) | | 5 (9.8) | - | |
| High | 7 (12.7) | 6 (13.0) | 1 (11.1) | | 6 (11.8) | 1 (25.0) | |
| Very high | 10 (18.2) | 9 (19.6) | 1 (11.1) | | 9 (17.6) | 1 (25.0) | |
| N/A | 1 (1.8) | 1 (2.2) | - | | - | 1 (25.0) | |
| RS, (n=37) | | | | | | | |
| Present (n) | 4 (10.8) | 2 (6.1) | 2 (50.0) | 0.05 | 3 (8.6) | 1 (50.0) | |
| Not present (n) | 33 (99.2) | 31 (93.9) | 2 (50.0) | | 32 (91.4) | 1 (50.0) | |

* Patients with SF3B1mut had lower hemoglobin levels (p = 0.023) and higher platelet counts (p = 0.047) than those patients without SF3B1mut. There was no obvious correlation in age, sex, bone marrow blasts, white blood cell counts, absolute neutrophil counts, cytogenetic category, and IPSS-R category between patients with SF3B1mut and without SF3B1mut. In the patients with SF3B1mut there seemed to be an association with ring sideroblasts, however, there was no significant difference (p = 0.050). As regards SRSF2mut, there was no significant difference in age, sex, World Health Organization classification, bone marrow blasts, white blood cell count, absolute neutrophil count, hemoglobin, platelet counts, ring sideroblasts, cytogenetic category, and IPSS-R category between patients with and without SRSF2mut (Table 2).

Frequency and pattern of SF3B1mut and SRSF2mut in MDS patients

Mutations in the SF3B1 and SRSF2 splicing machinery genes were detected in 13 out of 55 MDS patients (23.6%). We found SF3B1mut in 9 patients (16.4%), one of the 9
patients had 2 affected codons. SRSF2mut were found in 4 patients (7.3%) (Table 2). These MDS patients displayed either an SF3B1 or SRSF2 gene mutation. None of patients carried both gene mutations. All the mutations of these genes were heterozygous missense mutations (Table 3 and Figure 2). Patients with SF3B1mut were 2 MDS-SLD (22.2 %), 1 MDS-MLD (11.1 %), 2 MDS-RS (22.2 %), 1 MDS with isolated del(5q) (11.1 %), 2 MDS-EB-2 (22.2 %), and 1 MDS-U (11.1%) (Table 2 and Figure 3). We found 10 affected codons in 9 patients including E622, R625, H662, Q699, and K700 (Table 3, Figure 2 and 4). The K700 mutation was the most common SF3B1mut observed in this study which was positive in 6 patients. Additionally, one out of these patients displayed a co-mutation with Q699. Regarding SRSF2mut, the mutations were found in MDS-EB-2, MDS-U subtype and transformation to AML (tAML) for 2 (50.0%), 1 (25.0%) and 1 (25.0%) patients respectively (Table 2 and Figure 3). All of these showed heterozygous missense mutations in codon P95 (Figure 2 and 4).

**Discussion**

The mutations in RNA splicing machinery genes have been reported in a concrete proportion of adult MDS patients with varying frequencies between the different populations (Yoshida et al., 2011; Damm et al., 2012; Thol et al., 2012; Kang et al., 2015). The incidence of these gene mutations have been less frequently reported in Thailand especially in the upper northern population. We identified the alteration of SF3B1 and SRSF2 genes at the mutational hotspot exon by the PCR technique followed by Sanger sequencing. The results showed the frequency of these gene mutations were different to those in other populations and the mutations were associated with some clinical data.

SF3B1mut at exon 14 – 15 was detected in 9 out of 55 patients (16.4%). When considering the frequency of these exons mutation in other populations, SF3B1mut were found in 6.6% (6/91) of Brazilian patients (Donaires et al., 2016) which is lower than this study. In Asian populations the frequency of SF3B1mut was different from this report, including a 52.9% (55/104) incidence in Chinese patients (Cui et al., 2012); 10.0% (48/479) in Taiwanese patients (Lin et al., 2014a), and 7.0% (9/129) in Korean patients (Kang et al., 2015). However, the frequency of these gene mutations in our study was similar to that reported in a study by Rujirachaivej (2018) which cited 13.9% (10/72) in Thai MDS patients who were undergoing treatment at Ramathibodi Hospital located in central Thailand (Rujirachaivej et al., 2018). The various in frequencies and pattern of SF3B1 gene mutations among different population could depend on many factors such as diversity of genetic background, heterogeneity of disease, environment, individual lifestyle or the number of samples. Individual laboratory methods may also contribute to these differences. The people in upper northern Thailand experience different environment factors and come from a different genetic pool to those in other parts of Thailand. The information regarding SF3B1mut in MDS in the upper northern Thai population might be of further use for diagnosis and prognosis in this patient group.

This study detected five different SF3B1 missense mutations in 9 patients (Table 3). K700E is the most frequent mutation pattern of SF3B1 in this study, a similar finding to prior studies (Papaemmanuil et al., 2011; Cui et al., 2012; Patnaik et al., 2012; Seo et al., 2014; Donaires et al., 2016). Codon K700 is located on exon 15 of the SF3B1 gene, and the K700E missense mutation leads to
a lysine to glutamic acid substitution. The other affected pattern on exon 15 is Q699H, resulting in a glutamine to histidine substitution and this affected codon co-occurring with K700E. A co-mutation of Q699H and K700E that has not been previously reported in MDS patients in the COSMIC database, however, this affected codon has been reported in cases of pancreatic cancer (Catalogue of Somatic Mutations in Cancer). In addition, we found a mutation on exon 14, which is similar to previous studies (Papaemmanuil et al., 2011; Cui et al., 2012; Patnaik et al., 2012; Seo et al., 2014; Donaires et al., 2016). Codon E622D was found in 1 patient leading to a change in amino acid from glutamic acid to aspartic acid, and another mutation found in another patient was codon R625C, resulting in a change from arginine to cysteine. The final mutation found on exon 14 was codon H662Q, leading to a change in amino acid from histidine to glutamine. Mutational hotspots of the SFSB1 gene were found clustered in the fourth, fifth and sixth of the C-terminal HEAT domains (Papaemmanuil et al., 2011; Darman et al., 2015). In missense mutations, the amino acid substitutions may change the size and/or polarity of the side chains of protein structure and can lead to disease. Papaemmanuil (2011) reported that the mutation in the SFSB1 gene is involved in the pathogenesis of MDS (Papaemmanuil et al., 2011).

To our knowledge, there is no report about the SRSF2 gene mutation in the Thai population. We found the frequency of mutation in upper northern Thai patients was 7.3%. This frequency is lower than those in some previous reports. In western populations frequencies differ, for example SRSF2mut were found in 12.4% (24/193) of German MDS patients (Thol et al., 2012). In an Asian population, Wu et al., (2012) found the mutation in 14.6% (34/233) of Taiwanese MDS patients (Wu et al., 2012). In contrast, the frequency found by the current study was higher than those found among Chinese MDS patients (4.6% (5/108)) (Lin et al., 2014b). Our study detected the following 2 patterns of missense mutations in 4 patients: P95H (3/4) and P95L (1/4). P95 was the most frequent codon mutation which is consistent with the results of previous studies (Thol et al., 2012; Wu et al., 2012; Kang et al., 2015). The P95H and P95L mutations resulted in proline to histidine and leucine substitutions, respectively. Since SRSF2 protein needs P95 in order to bind to the target RNA, a mutation of P95 affected the function of this protein which may result in the reduction of the RNA binding affinity of the SRSF2 protein (Wu et al., 2012). Previous studies have demonstrated that not only missense mutations but also frameshift deletions have been found in MDS patients. Wu et al., (2012) found that 26.5% (9/34) of SRSF2mut in Taiwanese MDS patients were deletion mutations (Wu et al., 2012). Our study found only missense mutations in the Thai population, which may be due to the genetic background and also the limit in number of the test population (n = 55). Further studies with a larger sample size might provide more evidence of the pattern of mutation in the SRSF2 gene in MDS patients.

This study additionally analyzed the correlation between the clinical data and SF3B1mut and SRSF2mut in MDS patients. Similar to prior studies, patients who carried the SF3B1mut were found to have significantly lower hemoglobin level (p = 0.023) and a higher platelet counts (p = 0.047) (Cui et al., 2012; Damm et al., 2012; Rujirachaivej et al., 2018). As was found in some studies which demonstrated that SF3B1mut showed a direct correlation with RS (Malcovati et al., 2011; Papaemmanuil et al., 2011; Damm et al., 2012), this study also found that SF3B1mut mostly occurred in patients with RS, however, statistically it was a borderline significant difference (p = 0.050). Several previous studies demonstrated that patients with SF3B1mut had a favorable prognosis (Malcovati et al., 2011; Papaemmanuil et al., 2011; Cui et al., 2012), but there was a study which reported that SF3B1mut had no relevance as regards a favorable prognosis (Damm et al., 2012). The conflicting results possibly indicate the heterogeneity of the disease, the selection of analytical variables or co-occurrence with mutations in other genes. There are many current studies concerning the mutational co-occurrence of splicing machinery genes and genes involved in epigenetic regulation of transcription (ASXL1, EZH2, and DNMT3A) (Damm et al., 2012; Thol et al., 2012; Martin et al., 2017). There was a statistically significant difference in shorter overall survival and higher risk of AML progression in patients with co-occurrence of SF3B1 and DNMT3A mutation than those with SF3B1mut but DNMT3A wild type (Martin et al., 2017). An investigation focusing on Thai MDS patients needs to be carried out regarding this correlation in further studies.

The correlation of the clinical impact with SRSF2mut was also evaluated in this study. Wu et al., (2012) demonstrated that patients with SRSF2mut showed a strong association with male sex (Wu et al., 2012). However, this correlation was not observed in our study, although, 75% of the SRSF2mut patients were male. In addition, the patients with SRSF2mut tended to have a higher percentage of blasts than those without mutation (p = 0.074). Two of the patients with SRSF2mut were found in the MDS-EB-2 subtype and one in the transformation to AML group. The high percentage of BM blasts is an unfavorable factor, giving patients an increased probability of progression to AML. Previous studies indicated that SRSF2mut had a strong unfavorable impact in MDS patients (Thol et al., 2012; Wu et al., 2012; Lin et al., 2014b). The SRSF2 gene encodes for the serine/arginine rich splicing factor 2 (SRSF2), which involves the splicing E/A complex in the early stage of spliceosome assembly and is involved in the regulation of genomic stability (Xiao et al., 2007). Therefore, a mutation in the SRSF2 gene might contribute to an adverse prognosis in MDS (Thol et al., 2012).

In summary, this study showed that the frequencies of SF3B1mut and SRSF2mut were different from other populations previously studies. We found a co-mutation of Q699H and K700E which has not been previously reported as occurring in MDS patients in the COSMIC database. Patients with the SF3B1mut were associated with lower hemoglobin levels, and a higher platelet count. SRSF2mut patients appear to have a higher percentage of bone marrow blasts which increases the probability of progression to AML. The information pertinent to SF3B1mut and SRSF2mut and their possible correlation...
with clinical features in MDS in an upper northern Thai population might be of further use in diagnosis and prognosis in this patient group. Further studies with a larger sample size might provide more information of gene mutation in MDS patients.

Conflict of Interest
The authors declare no conflicts of interest.”

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