Molecular Profiling of Follicular Variant of Papillary Thyroid Cancer Reveals Low-Risk Noninvasive Follicular Thyroid Neoplasm with Papillary-Like Nuclear Features: A Paradigm Shift to Reduce Aggressive Treatment of Indolent Tumors

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Abstract

Introduction: Encapsulated follicular variant of papillary thyroid carcinoma (EFVPTC) has been reclassified into noninvasive follicular thyroid neoplasm with papillary-like nuclear features (NIFTP) and invasive EFVPTC. NIFTP is considered a low-risk neoplasm. Therefore, follicular variant of papillary thyroid cancer (FVPTC) presently has two distinct histopathological subtypes – invasive EFVPTC and infiltrative/diffuse FVPTC. Molecular characteristics of these groups remain unclear. **Methodology:** Thirty FVPTCs (10 NIFTPs, 12 invasive EFVPTCs, and 8 infiltrative/diffuse variants) were reviewed and screened for BRAF and RAS mutations by restriction fragment length morphism-polymerase chain reaction (PCR) and Sanger sequencing. The mRNA expression levels of iodine-metabolizing genes were analyzed using real-time PCR. The mutations status and mRNA expression levels were correlated with clinicopathological features. **Results:** All 10 NIFTPs had predominant follicular pattern. One case showed NRAS mutation, whereas none showed BRAF mutation. All invasive EFVPTC had capsular and/or lymphovascular invasion and 4/12 showed lymph node metastasis. BRAF and NRAS were seen in three cases each of invasive FVPTC. All eight infiltrating/diffuse FVPTCs showed infiltration into adjacent thyroid parenchyma and lymph node metastasis. **Conclusion:** BRAF mutation was observed in 62.5% of cases; however, no NRAS mutation was found. Sodium iodide symporter (NIS) expressions in NIFTP were similar to that of normal thyroid tissue, whereas it was downregulated in invasive and infiltrative/diffuse FVPTC. Our study supports the argument that NIFTP can be considered as low-risk follicular thyroid neoplasm. Those tumors that harbor BRAF mutations may be offered a complete thyroidectomy because they show decreased expression of NIS gene which confers a tendency to lose radioactive iodine avidity and further recurrence of the tumor.

Keywords: BRAF mutations, infiltrative/diffusing follicular variant of papillary thyroid cancer, invasive encapsulated follicular variant of papillary thyroid carcinoma, iodine-metabolizing genes, noninvasive follicular thyroid neoplasm with papillary-like nuclear features, RAS mutations

INTRODUCTION

Papillary thyroid cancer (PTC) accounts for 80%–90% of well-differentiated thyroid cancers.[1,2] However, different histological variants of PTC are well recognized which often display variable rates of aggressive behavior in the spectrum between well-differentiated conventional PTC (cPTC) and solid variant of PTC. The degree of aggressiveness is measured in terms of clinical presentation, local invasion, recurrence, and distant metastasis as well as their ability to take up radioiodine. Among the histological variants of PTC, follicular variant of PTC (FVPTC) is the most frequent variant of occurrence after classical PTC.[3-6] Previously, FVPTCs were histopathologically classified into two subtypes:

How to cite this article: George N, Agarwal A, Kumari N, Agarwal S, Krisnani N, Gupta SK. Molecular profiling of follicular variant of papillary thyroid cancer reveals low-risk noninvasive follicular thyroid neoplasm with papillary-like nuclear features: A paradigm shift to reduce aggressive treatment of indolent tumors. Indian J Endocr Metab 2018;22:339-46.

Access this article online

Quick Response Code:

Website:
www.ijem.in

DOI:
10.4103/ijem.IJEM_86_18

encapsulated (encapsulated follicular variant of papillary thyroid carcinoma [EFVPTC]) and infiltrative subtypes.[7,8] However, recently, EFVPTCs were histologically divided into two subtypes as noninvasive and invasive.[9-11] A study by Nikiforov *et al.* reclassified the EFVPTCs into two histological subtypes: noninvasive follicular thyroid neoplasm with papillary-like nuclear features (NIFTP) and invasive EFVPTC.[10] NIFTP is considered as neoplasm of low risk of malignant potential. The histological criteria for NIFTPs include the following major features comprising encapsulated or clear demarcation, follicular growth pattern, nuclear features of PTC, and minor features comprising dark colloid, irregularly shaped follicles, intratumoral fibrosis, and multinucleated giant cells within follicles. Some features that are considered to be absent in NIFTP include papillae >1%, psammoma bodies, infiltrative border, tumor necrosis, and mitosis >3/10 HPF. Invasive EFVPTC subtype has similar histological features; however, unlike NIFTP, it shows vascular and/or capsular invasion. Infiltrative FVPTC shows infiltration in the adjacent thyroid parenchyma.[10,12,13]

Molecular and clinicopathological characteristics of NIFTP and invasive EFVPTC have been shown to be similar to follicular thyroid cancer (FTC; low-nodal metastasis, less frequency of BRAF, and higher RAS mutations) and the infiltrative FVPTC is like cPTC (high-nodal metastasis and BRAF mutations).[8,10] Surgery followed by radioiodine treatment is the common method of treatment for the invasive encapsulated and infiltrative subtypes of FVPTC.[14,15] Response to radioiodine is an important factor in thyroid cancers, and iodine-metabolizing genes play a major role in this mechanism.[16] Hence, we decided to check the levels of iodine-metabolizing genes in our set of patients. And also, this is the first attempt to study the mutational profile of two subtypes of FVPTC (invasive and infiltrative) in comparison with NIFTP in patients.

METHODOLOGY

Sample selection and histopathological analysis

Forty FVPTC patients undergoing surgery from an endemic goiter region of North India over a period of 10 years from 2004 to 2014 were screened for the study. Of these, only 30 cases had formalin-fixed paraffin-embedded (FFPE) tissue available for review and were selected for the further study. All cases with papillary nuclear features and prominent follicular pattern with true papillae <1% were taken as FVPTC. The selected slides were reviewed and reclassified into NIFTP, invasive EFVPTC, and infiltrative FVPTC, according to the established criteria.[10,12,13]

The tumor nodules were not entirely submitted, and one section/cm of the tumor was taken for the study. An average of 1–8 sections was sampled per case depending on the size of the tumor. NIFTP was classified when the tumor showed encapsulation or clear demarcation, follicular growth pattern, nuclear features of PTC, and minor features comprising dark colloid, irregularly shaped follicles, intratumoral fibrosis, and multinucleated giant cells within follicles. Features such as papillae >1%, psammoma bodies, infiltrative border, tumor necrosis, and mitosis >3/10 HPF was absent. Invasive FVPTC was classified when an encapsulated tumor showed capsular and/or lymphovascular invasion. Infiltrative FVPTCs were categorized when there was infiltration in the adjacent thyroid parenchyma [Figure 1]. The pathologists were blinded from all clinical and mutational outcomes of the studies. The study was approved by the institutional ethical committee (IEC code: 2012-172-EMP-66).

Clinical parameters

Hospital information system was used to collect the patient details such as age, sex, and radioiodine treatment. It was classified on the basis of lymph node metastasis, size of the tumor,
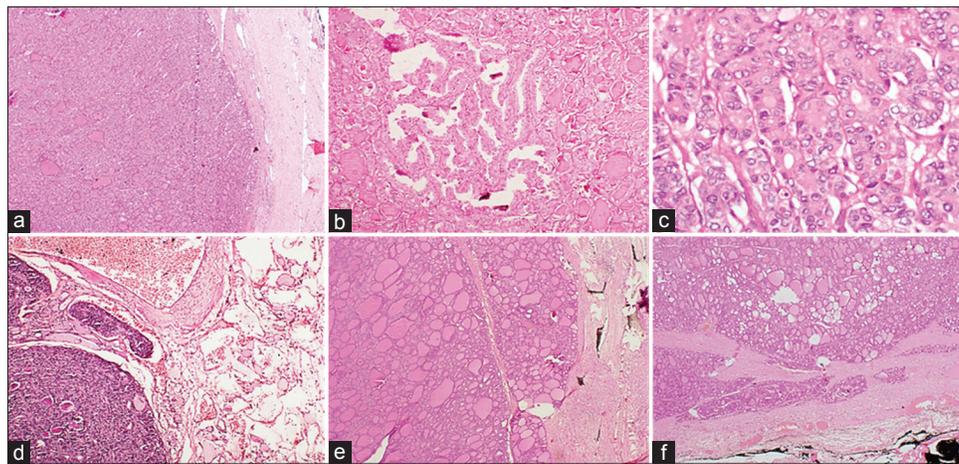

Figure 1: (a) Low-power photomicrograph showing encapsulated tumor (H and E, ×4). (b) Small focus of true vascular papilla (<1%) in a tumor with predominant follicular pattern (H and E, ×20). (c) High-power photomicrograph showing papillary nuclear features with optically clear nuclei, nuclear grooving, overlapping, and pseudoinclusions (H and E, ×40). (d) Invasive tumor showing lymphovascular invasion with tumor embolus present in a vascular channel (H and E, ×10). (e) Low-power photomicrograph showing capsular invasion with the presence of tumor nodule outside the capsule (H and E, ×10). (f) Infiltrative follicular variant of papillary carcinoma showing multiple nodules of tumor (H and E, ×4)

extrathyroidal extensions, and distant metastasis. The clinical parameters were further correlated with the mutational status.

Screening for BRAF V600E, RAS mutations

The tumor areas were confirmed and marked from the slides stained by hematoxylin and eosin stain. Four sections of 10 μ from each FFPE tissue block were subjected to DNA extraction using QIAamp FFPE Tissue Kit (Qiagen, Germany). The quality and quantity of the DNA were measured by using the NanoDrop 2000c (Thermo Fisher Scientific, USA).

Polymerase chain reaction-restriction fragment length polymorphism and sequencing

The most common BRAF V600E mutation reported in thyroid carcinomas is confined to exon 15. We therefore amplified BRAF exon 15 by polymerase chain reaction (PCR) using the following primers: forward '5GCTTGCTCTGATAGGAAAATGAG3'; reverse '5GATACTCAGCAGCATCTCAGG3'. The denatured PCR products were electrophoresed [Figure 1a], and digestion of the 237-base pair (bp) PCR fragment with restriction endonuclease TspRI showed three major bands of 117, 87, and 33 bp for the wild-type allele [Figure 1b]. The T1799A mutation abolished the restriction sites, which resulted in a prominent band of 237 bp from the mutant allele and residual bands from the normal allele.[17] Randomly selected three BRAF-positive samples were sequenced [Figure 1c] using Applied Biosystems 3500 Genetic Analyzer and reconfirmed the presence of BRAF mutations.

RAS mutation screening

Point mutations in codons 12/13 of the H-RAS and K-RAS genes were analyzed by sequencing method. The protocol used to analyze the tissue for point mutations in codon 12/13 of *K*-RAS has been described in a previous study. DNA isolated from patient sample was used for PCR using specific primers F5′-GGCCTGCTGAAAATGACTGA-3′ and R5′-TAGCTGTATCGTCAAGGCAC-3′. The resulting PCR products were sequenced for one of the six possible activating point mutations in codon 12/13. F5'-AATTGATGGAGAAACCTGTCTCTT-3'and R5'-TCCTCATGTACTGGTCCCTCATT-3' were used for the codon 12/13 of the KRAS for targeting other mutations.[18] Primers for codon 12/13 of HRAS F5'-TGA GGA GCG ATG ACG GAA-3'and R5'-GCG CTA GGC TCA CCT CTA T-3' were used for PCR and sequencing.[19] Codon 61 of NRAS was amplified using specific primers F5'-CCCAGGATTCTTACAGAAAACA-3' and R5'-TAATATCCGCAAATGACTTGC-3' and sequenced.[20]

Iodine-metabolizing genes expression levels

Total RNA was isolated from the five normal thyroid tissues and 30 FFPE FVPTC tissues using RecoverAll Total Nucleic Acid Isolation Kit for FFPE (Thermo Fisher Scientific, USA). Quantity and quality were measured by NanoDrop 2000c (Thermo Fisher Scientific, USA). Yield and quality of RNA were not affected by the procedure and storage. Further cDNA was synthesized using Revert Aid First

strand cDNA Synthesis Kit (Thermo Fisher Scientific, USA). Real-time PCR was performed for the iodine-metabolizing enzymes; thyroglobulin (Tg), thyroid peroxidase (TPO), sodium iodide symporter (NIS), and thyroid-stimulating hormone receptor (TSHR) along with internal control glyceraldehyde 3-phosphate dehydrogenase (GAPDH) using Applied Biosystems 7500 Fast Real-Time PCR System. The amplified products were checked on agarose gel electrophoresis [Figure 2]. The PCR reactions were performed in triplicate, and the Ct was obtained using Applied Biosystems software and averaged (standard deviation <1.0). The expression levels of iodine-metabolizing genes were normalized by GAPDH and the fold changes in the expression levels of iodine-metabolizing genes were calculated using Δct. All the primers were taken from a previous study.[21]

Statistical analysis

Categorical variables were analyzed using the Fisher's exact probability test. Kruskal–Wallis H-test was performed for analyzing iodine-metabolizing gene expression levels. $P < 0.05$ was considered statistically significant.

RESULTS

Clinicopathological features

A total of 30 cases were included in the study. Among 30 FVPTC samples, 10 patients were NIFTP, 12 patients were invasive EFVPTC, and eight samples were classified as infiltrative/diffusing FVPTC. Clinical features of NIFTP, invasive EFVPTC, and infiltrative subtypes were studied, and there was no significant change in the age, gender, distant metastasis, radioiodine treatment, staging, risk group, tumor size, and surgical protocol [Table 1]. Only lymph node metastasis was found to have significant difference between the three categories. The rate of lymph node metastasis was significantly higher in patients who had infiltrative/diffusing FVPTCs (100%) compared with patients who had EFVPTCs (4 [33.33%] of 12 patients) and patients with NIFTP (no lymph node metastasis; $P = 0.0004$). Tumor size was
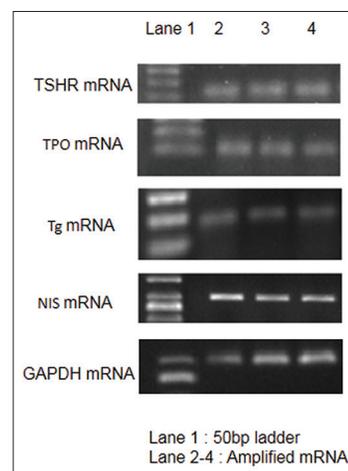

Figure 2: Representative agarose gel images of amplified iodine-metabolizing genes

Table 1: Clinico-pathological features of FVPTC

Characteristics	No. of patients (%)			
	NIFTP (*n*=10)	Invasive EFVPTC (*n*=12)	Infiltrative/diffusing follicular variant (*n*=8)	*P*ᵃ
Age				
≤45	9 (90%)	8 (66.67%)	5 (62.5%)	NS
>45	1 (10%)	4 (33.33%)	3 (37.5%)	
Gender				
Female	5 (50%)	10 (83.33%)	4 (50%)	NS
Male	5 (50%)	2 (16.67%)	4 (50%)	
Tumor size Median				
≤4	10 (100%)	6 (50%)	1 (12.5%)	NS
>4	0	6 (50%)	7 (87.5%)	
Lymph Node Metastasis				
Present	0	4 (33.33%)	8 (100%)	0.0004
Absent	10 (100%)	8 (66.67%)	0 (%)	
Distant Metastasis				
Present	0	2 (16.67%)	3 (37.5%)	NS
Absent	10 (100%)	10 (83.33%)	5 (62.5%)	
Thyroid surgery				
Lobectomy	0	1 (8.33%)	0	NS
Thyroidectomy	10 (100%)	11 (91.67%)	8 (100%)	
Radioactive Iodine Therapy				
Yes	8 (80%)	12 (100%)	8 (100%)	NS
No	2 (20%)	0	0	
Stage				
1&2	NA	10 (83.33%)	4 (50%)	NS
3&4	NA	2 (16.67%)	4 (50%)	
Recurrence				
Yes	0	3 (25%)	1 (12.5%)	NS
No	10 (100%)	9 (75%)	7 (87.5%)	
Risk group				
Low risk	10 (100%)	5 (41.7%)	0	NS
Intermediate risk	0	5 (41.7%)	4 (50%)	
High risk	0	2 (16.6%)	4 (50%)	

ᵃFisher's exact probability test, *P*<0.05 were considered significant. The comparison was performed between invasive EFVPTC and Infiltrative/diffusive FVPTC

Table 2: Histological features in NIFTP, Invasive EFVPTC and infiltrating/diffusing FVPTC

Features	NIFTP (*n*=10)	Invasive EFVPTC (*n*=12)	Infiltrating/ diffusing FVPTC (*n*=8)
Encapsulation/clear demarcation	10	12	0
Follicular growth pattern	10	12	8
Papillary architecture			
Absent	7	6	3
<1%	3	6	5
Psamomma bodies	0	3	4
Capsular invasion	0	11	1
Lymphovascular invasion	0	4	2
Tumor necrosis	0	0	0
Infiltrative growth pattern	0	0	8
Solid/trabecular/growth pattern	0	0	0
Multinucleated giant cells	0	0	1
Dark colloid	4	3	2

found to be >4 cm in 87.5% of the infiltrative/diffusing FVPTCs and 50% of the EFVPTCs. Size of all NIFTPs was ≤4 cm.

All the FVPTC cases underwent thyroidectomy except for one EFVPTC patient who had undergone hemithyroidectomy. Numbers of patients with age group above 45 years were higher in infiltrative/diffusing FVPTC (37.5%), followed by invasive EFVPTC (33.33%) and NIFTP (10%) patients. Interestingly, 16.67% of the FVPTC patients had distant metastasis. Among distantly metastasized patients, three of them were in the infiltrative/diffusing subtypes and two patients were in the invasive EFVPTC subtype group. When patients were classified on the basis of stages of papillary thyroid carcinoma as per the American Thyroid Association (ATA) guidelines, the occurrence of higher stages of PTCs was found to be higher in infiltrative/diffusing FVPTCs in comparison with invasive EFVPTC subtype patients (50% vs. 16.67%). The subtypes were further classified on the basis of risk factors of recurrence (ATA low risk, ATA intermediate risk, and ATA
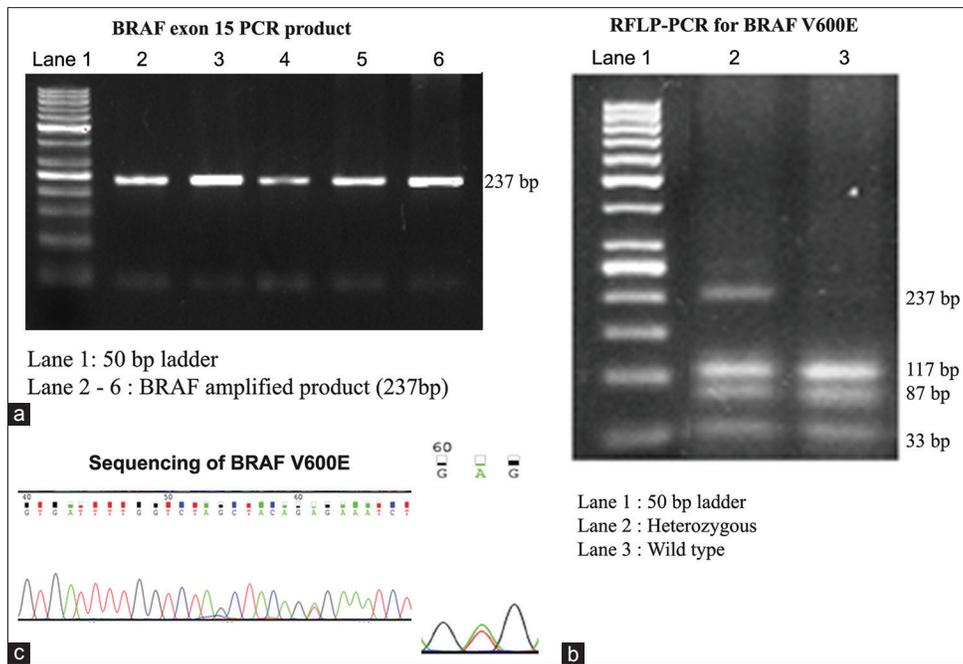
Figure 3: Polymerase chain reaction-restriction fragment length polymorphism. Analysis and sequence of codon 15 of the BRAF gene: (a) Representative agarose gel electrophoresis image of amplified Polymerase chain reaction product of BRAF gene. (b) Representative agarose gel electrophoresis image of Polymerase chain reaction-restriction fragment length polymorphism of the wild-type and mutant heterozygous BRAF gene. (c) Sequence of the V600E BRAF gene. The sequencing result show a mixture of T and A at the 1799 site
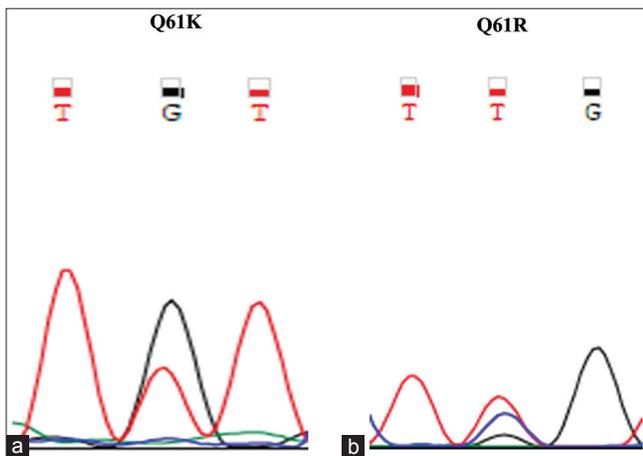

Figure 4: Results of RAS sequencing: (a) NRAS Q61K mutation, (b) NRAS Q61R mutation

high risk). Every infiltrative/diffusing subtype patient showed high-risk and intermediate-risk features and all the NIFTP patients were at low-risk category. Most of the invasive EFVPTC subtypes were in the intermediate-risk (36.36%) and low-risk category (45.45%). The major features of aggressiveness such as lymph node metastasis, distant metastasis, higher stages, and risk factors were observed at higher rates in infiltrative FVPTC than the other two groups of FVPTC.

The histological features of NIFTP, invasive EFVPTC, and infiltrative/diffuse FVPTC are shown in Table 2. The nuclear features of papillary carcinoma including ground-glass or optically clear nuclei, nuclear overlapping, grooving, and

pseudoinclusion were present in all cases on the basis, of which they diagnosed as papillary tumor of the thyroid. No mitotic activity was seen in either NIFTP or invasive EFVPTC, whereas very occasional mitoses were seen in infiltrative FVPTC.

Genotyping
BRAF mutations
Eight BRAF V600E mutations (26.67%) were identified in 30 FVPTCs studied. Infiltrative/diffusing FVPTC alone had 62.5% (5/8) occurrence of BRAF V600E mutations, while invasive EFVPTC had 25% (3/12) BRAF mutations [Table 3 and Figure 3]. NIFTP lacked the presence of BRAF V600E mutations. On presentation, three patients of infiltrative and two patients of invasive EFVPTC had distant metastasis and four of them showed BRAF V600E mutation. Further, BRAF V600E was analyzed for the correlation with all the clinicopathological features and none of them found to be significantly associated.

RAS mutations
Four RAS mutations (13.3%) were observed in a total of 30 samples. All of these genetic alterations were NRAS mutations. Two each Q61R and Q61K NRAS mutations (4/30) were observed on sequencing [Figure 4a and b]. Invasive EFVPTC subtypes harbored a single Q61K mutation and two Q61R (3/12) mutations. NIFTP subtypes were screened with one Q61K mutation (1/10). There were no RAS mutations in infiltrative/diffusing subtype [Table 3]. No mutations were detected on HRAS and KRAS screening.

Table 3: Genetic alterations in FVPTC

Genotype	No of patients (%)			
	NIFTP (*n*=10)	Invasive EFVPTC (*n*=12)	Infiltrative/diffusing follicular variant (*n*=8)	*P*a
BRAF mutations	0/10	3/12 (25%)	5/8 (62.5%)	0.0238
RAS mutations	1/10 (10%)	3/12 (25%)	0/8	NS
Total	1/10	6/12	5/8	

aFisher's exact probability test was performed, *P*<0.05 was considered significant
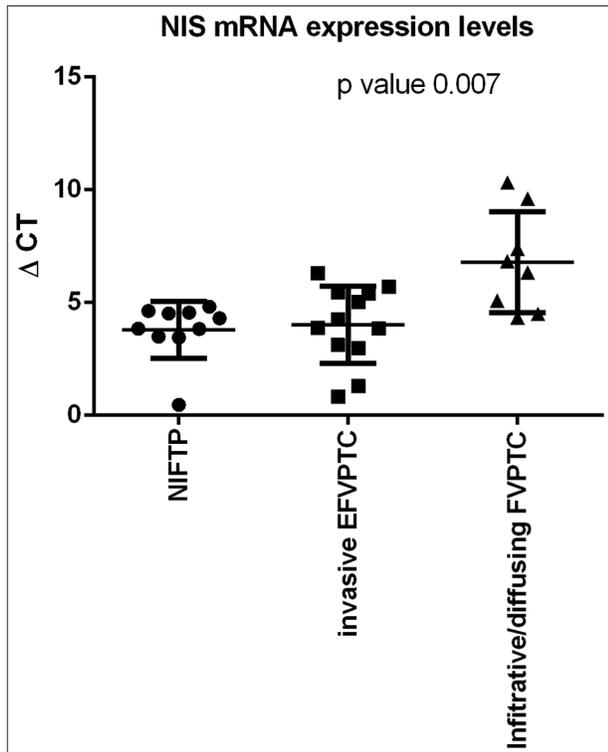

Figure 5: BRAF mutational status and relative expression level of sodium iodide symporter in different groups. Follicular variant of papillary thyroid cancer samples determined by semiquantitative polymerase chain reaction (*P* = 0.007). Kruskal–Wallis H-test was performed for analyzing iodine-metabolizing gene expression levels. *P* < 0.05 were considered statistically significant

Iodine-metabolizing genes expression levels

A reduction in expression levels of all iodine-metabolizing genes (NIS, Tg, TSHR, and TPO) was observed in comparison with iodine-metabolizing gene expressions in normal thyroid tissue, but it was not found to be significant. When iodine-metabolizing genes were analyzed in three subtypes of FVPTC, NIS mRNA (expressed as Δct) expression was found to significantly reduced in infiltrative and EFVPTC samples on comparison with NIFTP samples (*P* = 0.007) [Figure 5].

DISCUSSION

Clinicopathological findings of the previous studies have suggested that FVPTC has hybrid metastatic capacity with lymph node metastasis through the lymphatic system similar to cPTC and distant metastasis through the heterogeneous route

similar to FTC.[8,22] This heterogeneity can be explained by the fact that there are two distinct histologic "subtypes" which have distinct genetic mutations. Previous studies have shown that encapsulated or well-circumscribed form of FVPTC very rarely metastasizes, while the infiltrative form shows significant lymph node metastasis. Rivera *et al.* were the first to perform a comprehensive survey on oncogenic mutations in FVPTC according to its histologic subtype which helped in classifying these tumors into clinically relevant entities.[7,8,23,24] The authors reported 26% of BRAF mutations in infiltrative subtype while none was reported in encapsulated variants; conversely, RAS mutations were present in 36% of encapsulated subtypes and only 10% in infiltrative variants. The authors concluded that the infiltrative subtype has a molecular profile close to cPTC, while encapsulated types have a molecular profile similar to follicular adenoma or FTC.[8] However, recently, Nikiforov *et al.* reclassified EFVPTC into NIFTP and invasive EFVPTC. The major difference in invasive EFVPTC to NIFTP was the presence of vascular and/or capsular invasion of tumor. NIFTP was considered as follicular thyroid neoplasm.[10] Therefore, we included three groups in our studies; NIFTP, invasive EFVPTC, and infiltrative/diffusing FVPTC. Clinicopathological features and molecular profile of invasive EFVPTC and NIFTP were analyzed in different series of patients, but none of those studies included the infiltrative/diffusing FVPTC.[1,10,11,25-27]

On clinicopathological correlation analysis, we found the rate of lymph node metastasis in infiltrative subtype of FVPTC is close to the rates of cPTCs which was highly significant in comparison with invasive EFVPTC and NIFTP (*P* = 0.0004) [Table 1]. Li *et al.* have done a study in 359 FVPTC patients and found high frequency of distant metastasis (9.2%) in FVPTC cases.[22] Our study showed an increased incidence of distant metastasis (37.5%) in infiltrative subtype, and 16.67% (2/12) of patients had distant metastasis in invasive EFVPTCs, while none from the NIFTP group exhibited distant metastasis. The major reason of higher incidence of distant metastasis in FVPTCs in this particular set of study cohort may be due to the delay in presenting patients to surgeons with minimum of 2 years of neck swelling. Furthermore, this is the first study from this endemic goiter region of North India. More lymph node metastasis, higher incidence of distant metastasis, high stages of cancer, and higher degree of risk were noted in infiltrative variant.

In addition to clinicopathological data, infiltrative subtypes showed 62.5% (5/8) of RAF mutations in our set of patients which was similar to the rates of CPTCs from the previous studies.[11,27,28] Surprisingly, invasive EFVPTC had 25%

of BRAF V600E mutations, and one of the recent studies conducted by Kwon *et al*. also observed similar rate of BRAF mutations (31.25%) in invasive EFVPTCs of our study,[29] while none of NIFTP had BRAF mutations. Invasive EFVPTC subtypes showed 25% (3/12) RAS mutations while NIFTP had 10% (1/10) RAS mutations. Invasive EFVPTC and NIFTP groups showed similar range of RAS mutations occurring in FTCs.[10,13,30] From the above mutational profile and clinicopathological features, NIFTP can be accounted as low-risk neoplasm, but the other two groups require more attention. More lymph node metastasis, distant metastasis, risk factors, and higher incidence of BRAF V600E mutations in infiltrative subtypes make this group more aggressive.

In this set of FVPTC patients, no correlation was found between BRAF mutations and clinicopathological features. This suggests that a different molecular event might be associated with FVPTC. Although many authors demonstrated that BRAF is an important genetic event in PTC and FVPTC, discrepancies exist regarding the evidence of BRAF being responsible for more aggressive tumors in terms of advanced stage of presentation, lymph node metastasis, and local recurrence.[31-33] Oler and Cerutti suggested a possible association between BRAF mutational status and changes in the gene expressions of iodide-metabolizing genes. Thus, it is suggested that these BRAF mutation-positive FVPTC may not show aggressive clinicopathological features; however, at molecular level, they may have the tendency to lose radioiodine avidity and thus show a poor outcome.[28] This was also shown by Riesco-Eizaguirre *et al*., who demonstrated that the high risk of disease recurrence in tumors with BRAF mutations was associated with less differentiated tumors due to the loss of NIS-mediated I[131] uptake. In addition, it was demonstrated *in vitro* that BRAF mutations decreased the promoter activity of NIS, TSHR, and TPO.[34] However, the results of this association are controversial. While Riesco-Eizaguirre *et al*. and Durante *et al*. found an association between BRAF mutations and decrease in NIS expression,[34,35] Mian *et al*. found no such association.[36] Therefore, expression levels of iodine-metabolizing genes were studied in all samples.

When all iodine-metabolizing gene expression levels were analyzed, expression levels of NIS gene were found to be decreasing significantly in both invasive EFVPTC and infiltrative FVPTCs. Since the previous studies reveal BRAF V600E mutations partially influence the lowering of NIS mRNA expressions,[2,28,36,37] these phenomena may be due to the high rate of BRAF V600E mutations in invasive EFVPTC and infiltrative FVPTC subtypes. Thus, as a natural consequence, altering MAP kinase pathway by inhibiting BRAF may improve the expression levels of NIS gene and thus can improve the radioiodine uptake.[38-40]

CONCLUSION

Our study provides molecular evidence to support the argument that NIFTP can be considered as low-risk follicular thyroid neoplasm, which requires only close observation as treatment. Furthermore, FVPTC can be separated into two subtypes; infiltrative/diffusing subtype which has a molecular profile close to cPTC and invasive EFVPTC subtype which has a molecular profile in between cPTC and FTC (occurrence of BRAF V600E and RAS mutations). This study suggests that the patients undergoing hemithyroidectomy for indeterminate cytology and where the final pathology is FVPTC should undergo mutation analysis for BRAF and RAS genes. Those patients who harbor BRAF mutations may be offered completion thyroidectomy and radioactive iodine (RAI) treatment because they show decreased expression of NIS gene which confers a tendency to lose RAI avidity. As the significant loss of NIS expression is only in patients who had a total thyroidectomy, it will be beneficial to know BRAF mutations status before giving RAI. However, those harboring only RAS mutations may benefit from close observation instead of completion thyroidectomy and radioiodine treatment.

Conflicts of interest

There are no conflicts of interest.

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