H-Ras alterations in papillary thyroid cancer: a pilot clinicopathological study

Smith RA1, Salajegheh A1, Lam AKY1,2
1School of Medicine, Griffith University, Gold Coast Campus
2Queensland Health Pathology Services

Introduction:
Thyroid cancer is the most common cancer of the endocrine glands in Australia and in other parts of the world. Thyroid cancer occurs primarily in young and middle aged adults, with approximately 122,000 new cases per year worldwide. However, compared with other cancers, little in-depth research has been performed in thyroid cancer in Australia. The main problem that hinders care of thyroid cancer patients is the lack of proper classification of thyroid cancer. This is because thyroid cancer is composed of different histological entities with variable biological behaviour.

More than 95% of thyroid cancer is carcinoma, with papillary thyroid carcinoma accounting for approximately three quarters of thyroid carcinomas. Other common types of thyroid carcinomas include follicular carcinoma, poorly-differentiated carcinoma and undifferentiated carcinoma. Many variants of papillary thyroid carcinoma have been described and some are known to have prognostic significance. Some patients with papillary thyroid carcinoma can also progress to poorly-differentiated carcinoma or even to undifferentiated carcinoma of the thyroid. The development pathway taken by a thyroid carcinoma may be influenced by the acquisition of mutations in particular genes (see Figure 1).

As with the majority of cancers, the development of thyroid cancer is influenced by changes to the structure and behaviour of certain genes. There are genetic alterations in three genes that are commonly reported in papillary thyroid carcinoma, namely the BRAF, Ras and RET/PTC oncogenes. Ras is part of the major growth control pathway in cells. Ras is a family of genes rather than a single gene, all of which are monomeric membrane localised GTPases of roughly 21 kDa each. Ras genes function as signal inducers, taking information directly from receptors and passing that information on to numerous downstream transducers and effectors (see Figure 2). Being a crucial step in the major cellular growth pathway, Ras mutations can have a profound effect on carcinogenesis, affecting cellular growth, differentiation and even cellular mobility and invasion. Many human cancers show Ras mutations. Since the structure of the Ras family is highly conserved, these mutations have similar effects in most Ras family members, tending to fall into the 12, 13 and 61 codons. These mutations eliminate the need for cofactors and lock the Ras genes in an active state. Ras mutations have been known to coexist with other major transformation mutations at fairly high rates in some cancer types, though this has been found not to be the why this should not be the case in thyroid cancer is not yet fully clear.

Objective:
The aim of the present pilot study is to investigate the clinicopathological roles of mutations in codons 12/13 and 61 of the H-Ras gene in thyroid cancer.

Materials and methods:
The formalin fixed paraffin embedded tissues were collected from 10 patients (2 men, 17 women) with a diagnosis of papillary thyroid carcinoma from the Gold Coast Hospital with full ethical approval. The age, gender, clinical presentation, and other clinicopathological data was collected in a computerized database. Four of these tissues also had associated lymph node metastases available. DNA was extracted from these tissues using QIAGEN DNA extraction kits. Mutations of the H-Ras genes in codons 12/13 and 61 were determined by polymerase chain reaction, followed by restriction enzyme digestion and visualised on an agarose gel. PCR experiments are summarised in Table 1 and Figures 3 and 4.

Results:
Chi-square analysis proved negative for all clinicopathological characteristics, with the exception of the H-Ras mutation in codon 12/13, which was more often in patients with advanced (stage 3) lesions (p=0.023). Analysis of lymph node metastases was not viable due to low numbers of samples, though it was noted that in these metastases, mutational status was the same, with the exception of one sample which had acquired an additional mutant version of codon 61. Chi-square results and population genotypes are shown in Tables 2 and 3.