Successful Treatment of Anaplastic Thyroid Carcinoma with a Combination of Oral Valproic Acid, Chemotherapy, Radiation and Surgery

HITOSHI NOGUCHI, HIROTO YAMASHITA, TSUKASA MURAKAMI, KEISUKE HIRAI, YASUSHI NOGUCHI, JUNKO MARUTA, TADAO YOKOI AND SHIRO NOGUCHI

Noguchi Thyroid Clinic and Hospital Foundation

Abstract. Anaplastic thyroid carcinoma (ATC) is the most aggressive of thyroid cancers whose treatment is not yet established and mortality is extremely high. Recent in vitro studies have shown that valproic acid (VA), a newly identified histone deacetylase (HDAC) inhibitor, induces apoptosis, modulates differentiation gene expression of thyroid tumors and enhances the sensitivity of anaplastic cancer cell lines to doxorubicin. We report a case of successful treatment of anaplastic thyroid carcinoma with a combination of oral valproic acid, chemotherapy consisting of cisplatin and doxorubicin, external and intra-operative radiation and surgery. Tumor volume decreased by 50.7% under CT measurement and 44.6% under sonogram measurement over the course of the treatment. No significant rebound of tumor size was observed between each cycle of chemotherapy. Serial cytology performed via fine needle aspiration (FNA) presented a rapidly changing profile of cell types, starting with anaplastic and proceeding through increasingly well differentiated presentations. Only microscopic remnants of ATC cells were found in the histological examination of the resected thyroid. Ga scintigraphy and whole body PET scan six months after surgery revealed no evidence of recurrence or metastasis. As of Nov. 22, 2008, the patient is alive and disease free two years after diagnosis.

Key words: Anaplastic carcinoma, Thyroid, HDAC, Valproic acid

Case Report

Case

A 51 year old male, in otherwise good health, presented with a rapidly growing nodule with mild tenderness on the right side of his throat. He noticed a vague discomfort in his throat on November, 15, 2006, but could not locate the tumor by palpation. By the time he visited his local ENT on Nov. 18, the tumor was not only palpable but also visible as a protruding mass of finger tip size. He was referred to Noguchi Thyroid Clinic on Nov. 22 by his ENT as a possible case of subacute thyroiditis. The thyroid mass was hard and immobile on palpation. Sonogram and rapid FNA revealed the tumor to be ATC. Computerized tomography on the same day showed a thyroid mass of 30 × 41 × 35 mm occupying most of the right lobe of the thyroid with involvement of the anterior neck muscle. The tumor presented with acute pain. There was no evidence of lymph node or pulmonary metastasis. White blood cell count was 8460/µl. The case gauged 1/4 in the Sugitani Prognostic Index. Tc-99m scintigraphy presented a defect in the right lobe of the thyroid compatible to sonogram and CT findings. The patient had no previous history of cancer or thyroid ailments.

Clinical Course

The patient was immediately admitted and started on 1200 mg of oral VA daily, the upper therapeutic dose for epilepsy, and pre-hydrated for chemotherapy
which consisted of 100 mg of cisplatin per square meter of body area and 50 mg of doxorubicin per square meter of body area. The patient underwent three cycles of the above regimen at 3–4 week intervals during which he received 40 Gy of external beam radiation to the affected area. Serum concentration of VA was 53.6 \( \mu g/ml \) in spot measurement. Estimated maximum concentration was 78.3 \( \mu g/ml \), estimated minimum was 52.8 \( \mu g/ml \) and estimated mean was 66.4 \( \mu g/ml \) (SRL, Japan).

Tumor size decreased noticeably immediately after start of therapy. Sonogram measurements suggested a volume decrease of 11.4% in the first week. By Dec.26, after the second cycle of chemotherapy, tumor volume decreased by 31.7% as measured by CT (28 × 35 × 30 mm). By Feb.8, 2007, tumor volume decreased by 50.7% by CT (23 × 33 × 28 mm). 40 Gy of external radiation was administered with chemo.

Surgery was performed on Feb. 14, 2007, 83 days after diagnosis, with 15 Gy of intra-operative radiation. The patient was then administered an additional 30 mg of doxorubicin per square meter of body area. One month after surgery no evidence of residual tumor could be found in sonogram, neck and chest CT or Ga scintigraphy. He was discharged on foot, March 17th. Oral VA was administered daily through the entire course of the treatment and continued after discharge. He underwent Ga scintigraphy and whole body PET scan about six months after surgery which revealed no evidence of recurrence or metastasis, after which he was tapered off of VA (Fig. 3).

**Surgery Observation**

Examination upon surgery revealed less extensive invasion to the surrounding tissues than anticipated. The tumor had invaded into the anterior neck muscle, mostly to the sternothyroid muscle but also to the sternohyoid muscle to a lesser extent. Portions of both muscles were resected with the adhesion to the tumor intact. There was no pathologic adhesion to the trachea or the major blood vessels. The right recurrent laryngeal nerve was away from the tumor and undamaged. The right lobe of the thyroid was resected with the adhered muscle portions. Right lateral lymph nodes of II, III and IV areas and the anterior neck lymph nodes (VI) were dissected. Macroscopic lymph node metastases were not observed. The left lobe of the thyroid and the upper right parathyroid gland were preserved (which is standard procedure in Japan for papillary thyroid carcinoma of this size) as were the laryngeal nerve and the blood vessels. 15 Gy of radiation was administered to the 5 cm square surrounding the entry point of recurrent laryngeal nerve to the larynx before the wound was closed. There was no voice hoarseness or hypocalcemia after surgery.

**Cytology and Pathology**

FNA was performed on Nov. 22, Dec. 18, Jan. 9 and Feb. 5 in 3 to 4 week intervals. FNA sample taken on Nov. 22 was air dried and stained with modified ultrafast Papanicolaou stain. All subsequent FNA samples were fixed in a 95% alcohol solution and stained with standard Papanicolaou stain.

The first FNA showed very low differentiated tumor cells with giant nuclei and coarse chromatin amid a thick background of necrotic debris and inflammatory cell debris. After the first round of chemo, various cell shapes appeared but with similar low-differentiated nuclei. After the second round of chemo and seven weeks of oral valproic acid, spindle shaped cells became dominant. Pseudo-inclusions could be seen in some of the nuclei. After the third round of chemo, FNA presentation became typical of papillary carcinoma and the patient was ready for surgery.

**Fig. 1.** (a) The first FNA sample showed typical characteristics of anaplastic carcinoma cells with large nuclei with coarse chromatin amid a background of inflammatory cells and necrotic debris. (b) Four weeks later, after the first round of chemo, various cell shapes appeared but with similar low-differentiated nuclei. (c) After the second round of chemo and seven weeks of oral valproic acid, spindle shaped cells became dominant. Pseudo-inclusions could be seen in some of the nuclei. (d) After the third round of chemo, FNA presentation became typical of papillary carcinoma and the patient was ready for surgery.
cells, which are typical findings for ATC (Fig. 1-A). The second FNA findings were still those of very low differentiated cells with inflated nuclei, coarse chromatin and pronounced nucleoli, but the cells themselves were in various shapes, such as elongated like fibrous tissue and clustered like adenocarcinoma, but all sharing the same low differentiated nuclei. There were substantially less necrotic debris and almost no inflammatory cells (Fig. 1-B). By the third FNA, the elongated cells became predominant. The nuclei still showed low uniformity in size, coarse chromatin and pronounced nucleoli, but the cell bodies became predominantly spindle shaped. Giant nuclei were difficult to find and some of the nuclei had pseudo-inclusions (Fig. 1-C). The fourth FNA presented drastically fewer spindle shaped cells and clusters with round, closely arranged nuclei with pseudo-inclusions typical to papillary carcinoma (Fig. 1-D).

Post operative pathology presented a tumor with a necrotic core surrounded by fibrous tissue divided by a thin ring of papillary carcinoma (Fig. 2-A). HE

1200mg oral VA (tapered after Aug. 2007)
40Gy External Beam Radiation

Clinical Course

![Clinical Course](image)

Fig. 3. Clinical Course: The patient was admitted on Nov. 22, 2006 and immediately started on 1200 mg of oral VA and given three courses of chemotherapy consisting of 50 mg of doxorubicin per 1 sq. meter of body surface and 100 mg of cisplatin per 1 sq. meter of body surface and 40 Gy of external beam radiation to the neck. Surgery was performed on Feb. 14, 2007, with 15 Gy of intra-operative radiation. An additional 30 mg of doxorubicin per 1 sq. meter of body surface was administered after surgery. Oral VA was tapered off after Aug. 2007.
stained slides revealed the encapsulating fibrous tissue containing microscopic pockets of very large nuclei cells that may have been remnants of ATC cells (Fig. 2-B). The papillary carcinoma segment was very well differentiated (Fig. 2-C). Microscopic lymph node metastases were found in 4 of the 26 lymph nodes dissected, all of which were papillary carcinoma. There were no low differentiated cells found in the lymph nodes.

Toxicity

Side effects of chemotherapy were not more severe than anticipated. The patient complained of nausea, taste disturbance, hair loss, dizziness and hypersensitivity to smell. Episodes of agranulocytosis, which followed each cycle of chemotherapy, were treated with G-CSF. Ultrasound cardiography showed a slight decrease in the ejection fraction of the left ventricle but within normal limits. 24-hour creatinine clearance was not compromised at discharge. Serum creatinine levels remained below 1.3 mg/dl throughout the treatment. Postoperative free T3 remained within normal bounds due to the partial resection of the thyroid, but 25 micrograms of levothyroxine was added two months later to correct sub-clinical hypothyroidism. Patient complained of dizziness and numbness while on therapy. One unexpected side effect was recurrent severe headache, although valparates are sometimes used in the treatment of headache. The symptom profile resembled that of cluster headache more than that of migraine and substantially improved after the patient was tapered off of VA. Serum ammonia levels, known to increase during VA use, were periodically monitored but no abnormalities were found. The patient is still suffering from a relatively mild case of post-traumatic stress.

Discussion

Anaplastic thyroid carcinoma is a highly lethal cancer with median survival of 4 to 12 months from the time of diagnosis [1]. It is known to be highly resistant to chemotherapy. Typically, the tumor is initially diminished in response to chemotherapy, only to rebound in size before the next round of chemotherapy could be administered. A combination of chemotherapy, radiation and surgery has been reported to be effective in very early and incidental cases, but there remains no standard method of treatment for this particular cancer [2].

Histone deacetylase (HDAC) inhibitors are a promising class of anti-neoplastic agents that induce differentiation and apoptosis. Moreover, they may enhance the cytotoxicity of drugs targeting DNA through acetylation of histones. It has been reported in recent years that VA acts as a HDAC inhibitor at therapeutic concentrations and induces apoptosis, promotes differentiation [3] and enhances sensitivity to chemotherapy of thyroid cancer cells [4].

Increased sensitivity to chemotherapy was demonstrated with the cell lines CAL-62 and ARO, both of which are cell lines derived from human ATC. In the report, Catalano et al describe the survival of the cancer cells cultured in a doxorubicin solution dropped significantly when they were pretreated with VA. The survival of CAL-62 fell from 48% to 15% and the survival of ARO fell from 43% to 20%.

Other HDAC inhibitors are known to have similar effects in laboratory experiments, but have yet to be tested for human use [5]. Valproic acid has long been known as a treatment for epilepsy and is also sometimes used to treat persistent migraine. Side effects are rare and the side effect profile is well documented. Its potential role as an HDAC inhibitor is still under evaluation. The substance is thyroid suppressing under long term use.

There are also some very good laboratory data showing that rosiglitazone also enhances the effect of chemotherapy agents on ATC cell lines in vitro. However, rosiglitazone is not yet clinically available in Japan [6]. (We subsequently used pioglitazone on another case with some encouraging results.) There is also data to suggest that all-trans retinoic acid (RTRA) and 13-cis retinoic acid also have helpful effects [7, 8]. The use of RTRA, however, is known to cause serious side effects whose mechanism is not clear. Supposedly, retinoic acid syndrome in patients of acute promyelocytic leukemia is caused by the rapid maturing of myeloid cells and therefore should not be a concern to patients of ATC. But in this case, the very existence of such a syndrome was enough to exclude retinoic acid as the first drug to be tested. We chose valproic acid because it was clinically available and had a well documented side effects profile.

At least some ATCs are believed to be secondary mutations of existing well-differentiated thyroid carci-
nomas, such as papillary carcinoma. Histologically, ATC tissues often coexist with papillary carcinoma in the same tumor [1]. The papillary carcinoma found in the postoperative pathology may be the remnant of the papillary element of a mixed papillary-anaplastic tumor after the ATC element had been purged by chemotherapy. Another possibility is that the differentiating effect of valproic acid converted the surviving ATC cells into papillary carcinoma. It could be argued that there was no ATC in the first place and that it was all papillary carcinoma from the very beginning. If that were the case, it would be difficult to explain the rapid growth of the tumor before treatment and the distinctive FNA presentation at the outset, as well as the responsiveness to chemotherapy. It could also be argued that this was not precisely ATC, but poorly differentiated thyroid carcinoma (PDTC) which has a slightly better prognosis than ATC. If that were the case, it still needs to be explained how well-differentiated papillary carcinoma became so dominant in the resected tumor. Through a composite clinical picture of rapid growth, FNA presentation, laboratory data, radiological and sonogram findings as well as response to chemotherapy, we diagnose this case as ATC and conclude that this case is consistent with the in vitro findings that valproic acid promotes re-differentiation and enhances sensitivity to chemotherapy and radiation.

It is difficult to say how much of the treatment effect can be attributed to the use of valproic acid and how much to chemotherapy and radiation. When compared with the prognosis of previous cases which employed similar treatments without valproic acid, this case has been exceptionally successful. As of Nov. 22, 2008, the patient has survived two years from the day of initial diagnosis and has lived without recurrence since discharge.

Clinical trials for the use of vaproic acid in the treatment of malignant tumors are already under way. We believe this case supports the importance of such trials.

References