Hum Genet (1995) 96:27–32 © Springer-Verlag 1995

ORIGINAL INVESTIGATION

Jan Zedenius · Göran Wallin · Ann Svensson
Lars Grimelius · Anders Höög · Göran Lundell
Martin Bäckdahl · Catharina Larsson

Allelotyping of follicular thyroid tumors

Received: 22 November 1994 / Revised: 5 January 1995

Abstract To elucidate further the genetic mechanisms for follicular thyroid tumor development and progression, we allelotyped follicular thyroid tumors and other thyroid lesions from 92 patients. In general, a low frequency of loss of heterozygosity (LOH) was found, the highest being for chromosomes 3q, 10q, 11p, 11q, 13q, and 22q (10%–15%). However, detailed study of LOH of these chromosome arms with regard to the different histopathological diagnoses indicates that a locus on chromosome 10q may be involved in follicular thyroid tumor progression. In addition, the majority of Hürthle cell adenomas showed LOH on either chromosome 3q or 18q, in contrast to the other tumor types. This discrepancy in genetic alterations may contribute to the divergent clinical features occurring in these tumors.

Introduction

Tumors of the thyroid gland are common among the general population, but only a minority are malignant (Hedinger et al. 1988). Thyroid carcinomas vary strongly in aggressiveness, ranging from the indolent occult papillary carcinomas to the wildly growing anaplastic carcinomas, the latter being one of the most lethal neoplasms in mankind. It has been debated whether a multistage tumorigenesis similar to that in colo-rectal and breast carcinomas exists in the thyroid (Fagin 1992; Wynford-Thomas 1993; Farid et al. 1994; Zedenius et al. 1992). It is also uncertain whether the anaplastic carcinoma emerges from a differentiated form, and whether a follicular thyroid adenoma turns malignant if left untreated.

The vast majority of thyroid tumors are follicular adenomas. Apart from the common (trabecular/solid, microfollicular, normofollicular, macrofollicular) adenomas, there are two subgroups: atypical and Hürthle (oxyphil) cell adenomas (Hedinger et al. 1988). The atypical adenoma shows high cellularity, less regular architectural and cytological patterns, with often the presence of mitotic activity, but no signs of capsular or vascular invasion. Follicular carcinomas are usually characterized as minimally or widely invasive (Hedinger et al. 1988). If the tumor does not exhibit general signs of metastasis or perithyroid growth, there is no accurate method of distinguishing preoperatively between follicular adenomas and carcinomas. Hence, the follicular tumor must be surgically removed in order to verify its histopathological diagnosis.

Apart from mutations in the *ras* oncogenes, considered as early events in follicular thyroid tumorigenesis (Lemoine et al. 1989; Suarez et al. 1990; Namba et al. 1990a), and involvement of the p53 gene in poorly or undifferentiated tumors (Ito et al. 1992; Fagin et al. 1993; Donghi et al. 1993), little is known about the genetic mechanisms behind follicular thyroid tumor development. Loss of heterozygosity (LOH) at chromosome 11q13, the region containing the candidate multiple endocrine neoplasia type 1 (MEN1) gene, has been described in a subset of follicular adenomas (Matsuo et al. 1991). We report here the results from allelotyping a large number of follicular neoplasms to identify further regions of chromosomal loss that might contain loci for tumor suppressor genes involved in thyroid tumor development and progression.

J. Zedenius (✉) · G. Wallin · A. Svensson · M. Bäckdahl
Department of Surgery, Karolinska Hospital,
S-171 76 Stockholm, Sweden

J. Zedenius · C. Larsson
Department of Molecular Medicine (Endocrine Tumor Unit),
Karolinska Hospital, S-171 76 Stockholm, Sweden

A. Höög
Department of Pathology, Karolinska Hospital,
S-171 76 Stockholm, Sweden

G. Lundell
Department of Oncology, Karolinska Hospital,
S-171 76 Stockholm, Sweden

L. Grimelius
Department of Pathology, Uppsala University Hospital,
S-751 85 Uppsala, Sweden

Table 1 Allelotypes of the follicular tumors and other thyroid lesions from the 92 studied patients

Chromosomal location	Locus	Marker[a]	Method[b]	No. of cases investigated	No. of informative[c] cases/no. of scored cases (%)	No. of LOH/ informative cases (%)
1p36	D1S243	AFM214yg7	CA	92	75/92 (82)	2/75 (3)
1p36.12–36.2	D1S96	p1–45	*Taq*I	90	38/77 (49)	2/38 (5)
1q32–44	D1S81	pTHH33	*Rsa*I	72	51/68 (75)	4/51 (8)
2p15–23	D2S6	pXG18	*Taq*I	90	27/77 (35)	2/27 (7)
2q14–22	D2S44	pYNH24	*Taq*I	67	60/66 (91)	4/60 (7)
3p21	D3F15S2	RIK	*Taq*I	62	31/62 (50)	1/31 (3)
3q13–22	D3S1267	116xh2	CA	92	74/91 (81)	7/74 (9)
4p16	D4S227	C5E	*Taq*I	85	36/72 (50)	2/36 (6)
4q28	FGA	pAF1	*Taq*I	90	26/74 (35)	2/26 (8)
5p15.3	D5S48	CRI-L123	*Rsa*I	72	29/51 (57)	–/29 (0)
5q21	D5S346	LNS-CA	CA	91	71/83 (86)	–/71 (0)
6p21.3–22	D6S10	pCH6	*Taq*I	86	46/76 (61)	1/46 (2)
6q27	D6S21	CRI-1065	*Rsa*I	72	50/72 (69)	1/50 (2)
7p13	IGFBP1	chBP1–1:2	*Taq*I	87	38/81 (47)	–/38 (0)
7q31	MET	pmetH	*Taq*I	73	33/72 (46)	2/33 (6)
8p22–23	D8S17	pYNM3	*Rsa*I	66	16/66 (24)	1/16 (6)
8q13–22	CA2	H25-3.8	*Taq*I	80	17/49 (35)	–/17 (0)
8q24	TG	phTg3	*Taq*I	69	7/46 (15)	1/7 (14)
9p21–24	D9S157	AFMO67xd3	CA	89	51/89 (57)	4/51 (8)
9q34	D9S7	pEFD126.3	*Rsa*I	72	41/72 (57)	2/41 (5)
10pter-13	D10S28	cTBQ7	*Rsa*I	72	50/55 (91)	3/50 (6)
10q26	D10S25	EFD75	*Taq*I	89	46/82 (56)	6/46 (13)
11p15.5	HRAS	pTBB-2	*Taq*I	88	47/71 (66)	4/47 (9)
11q13	D11S97	pMS51	*Taq*I	89	68/89 (76)	6/68 (9)
11q13	INT2	SS6	*Taq*I	36	13/28 (46)	2/13 (15)
11q23.3	D11S29	L7	*Taq*I	36	8/35 (23)	1/8 (12)
12p13	F8VWF	EMBL M25716	CA	92	62/91 (68)	1/62 (2)
12q23	IGF1	MFD1	CA	91	57/91 (63)	–/57 (0)
13q14	RB1	p68RS2.0	*Rsa*I	44	18/44 (41)	2/18 (11)
13q14	RB1	RB1.20	CA	91	71/91 (78)	9/71 (13)
14q32	D14S13	pMLJ14	*Rsa*I	56	40/51 (78)	2/40 (5)
15q11–12	GABRB3	EMBL X63670	CA	91	65/91 (71)	3/65 (5)
16p13.3	HBA1	pSstalpha2	*Rsa*I	69	39/69 (57)	3/39 (8)
16q24	APRT	Huap15	*Taq*I	79	29/74 (39)	1/29 (3)
17p13.3	D17S5	pYNZ22.1	*Rsa*I	55	38/53 (72)	–/38 (0)
17q22	D17S74	pCMM86	*Rsa*I	38	36/38 (95)	1/36 (3)
18p11.3	D18S71	AFM254yd5	CA	92	72/92 (78)	2/72 (3)
18q21.3	DCC	DCC	CA	92	71/92 (77)	5/71 (7)
19p13.3	D19S21	pMCOB5	*Rsa*I	66	34/59 (58)	2/34 (6)
19p13.3	D19S177	Mfd120	CA	91	74/90 (82)	5/74 (7)
19q13.3	D19S51		CA	92	80/90 (89)	2/80 (2)
19q13.4	D19S214	AFM163xf8	CA	92	66/92 (72)	2/66 (3)
20p11.2–12	D20S66	Mfd 136	CA	91	69/83 (83)	1/69 (1)
20q13.1	D20S120	AFM276xh1	CA	91	71/91 (78)	4/71 (6)
21q22.2	D21S167	NA-F	CA	92	56/89 (63)	1/56 (2)
22q11.1–11.2	D22S9	p22/34	*Taq*I	81	31/74 (42)	5/31 (16)
22q11.1–11.2	D22S10	22CI-8	*Taq*I	81	16/74 (22)	1/16 (6)
22q11.2-qter	MB	pHM27.B2.9	*Taq*I	68	27/66 (41)	4/27 (15)
22q13-qter	D22S94	KI-1105	*Taq*I	68	27/67 (40)	1/27 (4)
Y/Xq21.31	DXYS1X	pDP34	*Taq*I	84	36/64 (56)	1/36 (3)

[a] Marker names are taken from their original descriptions
[b] Markers used for Southern blot technique appear with their respective restriction enzyme. *CA* PCR-based microsatellite technique used
[c] Constitutionally heterozygous

Materials and methods

Tissue specimens

The study included tissue samples from different thyroid lesions in 93 patients: 59 common follicular adenomas, five atypical adenomas, 14 Hürthle cell adenomas, three atypical Hürthle cell adenomas, three follicular carcinomas, four nodular goitres, and five glands from patients with Graves' disease. Immediately after surgical removal, the thyroid tissue was frozen in liquid nitrogen and stored at –70° C. The tumors were histopathologically classified as suggested by the WHO committee (Hedinger et al. 1988), using routinely processed tissue preparations.

DNA was extracted from the fresh frozen tissue by phenol-chloroform extraction and ethanol precipitation as previously described (Larsson et al. 1990). To determine the representativeness of the tumor material, pieces were cut from all specimens for histopathological examination. Only tissue samples containing more than 60% tumor cells were included in the study. One adenoma did not fulfill these criteria. DNA extracted from blood samples obtained from the 92 remaining patients served as constitutional control DNA for the allelotyping.

Two different techniques were used for allelotyping: Southern blot hybridization using restriction fragment length polymorphism/variable number of tandem repeat markers, and the polymerase chain reaction (PCR)-based detection of microsatellite repeats. The 50 markers and their chromosomal locations are listed in Table 1.

Southern blot hybridizations

The extracted high-molecular-weight DNA was digested, separated on agarose gels, and transferred to Zeta-Probe nylon filters (Bio-Rad Lab) as described (Bergerheim et al. 1989). Restriction enzymes were *TaqI* and *RsaI*. Hybridization and autoradiography conditions were as described (Bergerheim et al. 1989). LOH was detected by the naked eye as either a total absence, or ≥ 50% reduced signal intensity, of one of the constitutional alleles in the thyroid tumor tissue DNA.

Dinucleotide repeat polymorphisms (CA-repeats)

Standard PCR reactions were carried out in a final volume of 10 μl, containing 40 ng high-molecular-weight DNA, 50 m*M* KCl, 10 m*M* TRIS-HCl (pH 8.3), 1.5 m*M* $MgCl_2$, 125 μ*M* each dNTP, 2 pmol each oligodeoxynucleotide primer (one of which was end-labeled with either [^{32}P] or [^{35}S]), and 0.2 U DNA polymerase (Dynazyme, Finnzyme Oy). The standard thermal cycling conditions were: incubation at 94° C for 4 min, 25 step cycles at 94° C for 30 s, at 55° C for 30 s, and at 72° C for 30 s, with a final extension for 4 min at 72° C. Alternatively, step cycles were at 94° C for 1 min, 60° C for 1 min, and at 72° C for 1 min, with a final extension for 7 min at 72° C. Aliquots of the PCR product were denatured with formamide, heated to 80° C and electrophoresed on standard denaturing 6% polyacrylamide DNA sequencing gels. Gels were then fixed, dried, and subjected to autoradiography for 12–24 h. LOH was detected as described above.

Table 2 LOH in follicular thyroid neoplasms with regard to the histopathological diagnoses. *Fo ad* common follicular adenoma, *At ad* atypical adenoma, *Hü* Hürthle cell adenoma, *At Hü* atypical Hürthle cell adenoma, *Fo ca* follicular carcinoma

Chromosome arm	Fo ad	At ad	Hü	At Hü	Fo ca	Total	(%)
1p	1/52[a]	1/5	–/11	–/3	2/3	4/74	(5)
1q	3/32	1/4	–/6	–/2	–/3	4/47	(9)
2p	2/16	–/2	–/2	–/1	–/2	2/23	(9)
2q	3/42	–/1	1/6	–/1	–/3	4/53	(8)
3p	1/21	–/–	–/2	–/2	–/3	1/28	(4)
3q	2/48	1/5	4/12	–/2	–/3	7/70	(10)
4p	2/22	–/2	–/5	–/2	–/–	2/31	(6)
4q	1/19	–/1	1/2	–/1	–/1	2/24	(8)
5p	–/18	–/2	–/2	–/1	–/2	–/25	(0)
5q	–/44	–/5	–/11	–/2	–/2	–/64	(0)
6p	1/30	–/1	–/6	–/1	–/2	1/40	(2)
6q	1/30	–/2	–/7	–/1	–/3	1/43	(2)
7p	–/27	–/1	–/4	–/1	–/2	–/35	(0)
7q	1/20	1/2	–/5	–/1	–/2	2/30	(7)
8p	–/9	1/1	–/1	–/2	–/2	1/15	(7)
8q	1/19	–/1	–/1	–/–	–/2	1/23	(4)
9p	3/31	1/3	–/7	–/2	–/2	4/45	(9)
9q	2/22	–/4	–/5	–/2	–/1	2/34	(6)
10p	–/30	3/3	–/7	–/3	–/3	3/46	(7)
10q	–/28	2/3	1/4	1/3	2/2	6/40	(15)
11p	2/33	1/3	–/4	–/–	1/2	4/42	(10)
11q	3/47	2/3	–/9	1/2	–/3	6/64	(9)
12p	–/38	–/4	–/7	1/3	–/1	1/53	(2)
12q	–/39	–/2	–/5	–/2	–/2	–/50	(0)
13q	4/48	2/4	1/10	1/3	1/3	9/68	(13)
14q	2/26	–/1	–/6	–/2	–/1	2/36	(6)
15q	2/43	–/2	–/11	–/3	1/1	3/60	(5)
16p	2/27	–/2	1/5	–/–	–/1	3/35	(9)
16q	1/18	–/1	–/4	–/3	–/2	1/28	(4)
17p	–/22	–/2	–/2	–/3	–/3	–/32	(0)
17q	–/19	–/1	–/6	–/3	1/2	1/31	(3)
18p	1/46	1/4	–/11	–/3	–/3	2/67	(3)
18q	–/42	1/5	4/12	–/2	–/3	5/64	(8)
19p	4/51	1/5	1/10	–/3	–/3	6/72	(8)
19q	1/53	1/5	–/13	–/3	–/3	2/77	(3)
20p	1/42	–/4	–/9	–/3	–/3	1/61	(2)
20q	1/44	2/5	1/10	–/3	–/2	4/64	(6)
21q	1/34	–/1	–/10	–/3	–/2	1/50	(2)
22q	5/40	1/4	–/5	1/3	–/3	7/55	(13)
X/Y	–/24	1/1	–/4	–/1	–/2	1/32	(3)
Total LOH (%)[b]	22/58 (38)	4/5 (80)	9/14 (64)	2/3 (66)	3/3 (100)	39/83 (47)	

[a] The figures refer to the number of tumors in each diagnostic group showing LOH, compared with the number of informative cases. If more than one marker has been used on one chromosome arm, the accumulated data are given

[b] The percentage in each histopathological group of tumors showing LOH at any locus

Results

In the present study, various thyroid lesions from 92 patients were analyzed for LOH on all chromosome arms. The majority of the lesions represented follicular tumors of various types. A comprehensive list of all markers used for this allelotyping is shown in Table 1, which further shows the percentage of informative cases for each probe, and the proportion of cases showing LOH. The LOH frequency per marker varied from zero to approximately 15%. No LOH was seen in nodular goitres or in thyroid tissue from patients with Graves' disease.

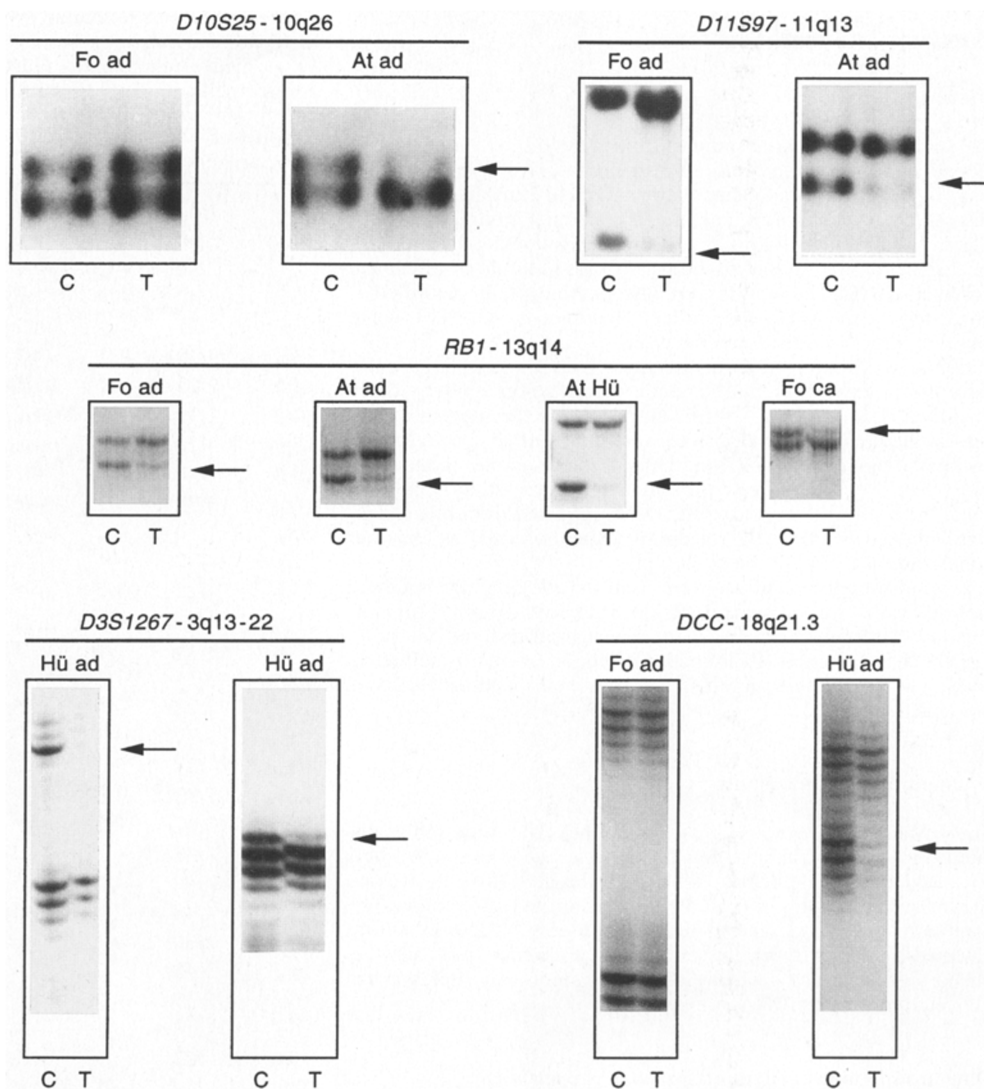

Fig. 1 Autoradiograms from Southern blot (D10S25 and D11S97), and microsatellite repeat (RB1, D3S1267, and DCC) analyses. Paired DNAs from follicular thyroid tumors *(T)* and corresponding constitutional tissue *(C)* were analyzed with the markers listed in Table 1. The *arrows* indicate loss of one of the constitutional alleles (LOH), whereas absence of the *arrow* corresponds to retained heterozygosity. *Fo ad* common follicular adenoma, *At ad* atypical adenoma, *Hü ad* Hürthle cell adenoma, *At Hü* atypical Hürthle cell adenoma, *Fo ca* follicular carcinoma

Table 2 shows the LOH frequency in the tumors with respect to the different histopathological diagnoses. Accumulated LOH frequency for each tumor type was highest for chromosomes 3q (10%), 10q (15%), 11q (9%), 13q (13%), and 22q (13%).

Detailed study of LOH with regard to the various histopathological types of follicular neoplasms revealed that five of eight informative tumors with atypical and malignant features showed LOH of chromosome 10q (Table 2, Fig. 1). LOH of 10q was not seen in the common adenomas, but in one of four Hürthle adenomas. LOH of chromosomes 3q and 18q was mainly restricted to Hürthle cell adenomas (Table 2, Fig. 1). Indeed, seven of ten Hürthle cell adenomas informative for both markers showed LOH at either 3q or 18q, one showing LOH on both chromosome arms; the other tumor types showed only single cases with LOH at these loci.

LOH of the MEN1 locus on chromosome 11q13 was found in about 10% of the tumors investigated. Of all tumors, 13% showed LOH at the Rb gene locus on chromosome 13q, and LOH was seen in all types of tumors (Table 2, Fig. 1).

The total number of tumors with LOH at any locus varied between the tumor types (Table 2, bottom line). No LOH was seen in the non-neoplastic lesions examined, i.e., the nodular goitres and glands from patients with Graves' disease. Of the common adenomas, 38% showed LOH, in comparison with the other tumor types, in which the vast majority showed LOH. For example, four of five atypical and all three carcinomas exhibited LOH for at least one locus.

Discussion

The underlying mechanisms of follicular thyroid tumorigenesis and tumor progression are yet not well known. However, some events are clearly discernible. Follicular neoplasms are usually clonal, arising from a single precursor cell (Namba et al. 1990b; Hicks et al. 1990). Mutations of the *ras* oncogenes have been reported, and are regarded as early events in tumor progression as they are found both in adenomas and in carcinomas (Lemoine et al. 1989; Suarez et al. 1990; Namba et al. 1990a). Thy-

rotropin is a stimulatory growth factor for the follicular thyroid cell. Mutations in the thyrotropin receptor cause hyperfunctioning adenomas (Parma et al. 1993). This is also true for mutations in the α-chain of the G_s protein involved in the cAMP-dependent signal transduction pathway (O'Sullivan et al. 1991; Suarez et al. 1991). Mutations in this gene (the *gsp* oncogene) have also been reported in other follicular thyroid tumors (Goretzki et al. 1992).

The only tumor suppressor gene established as being involved in thyroid tumorigenesis is the p53 gene, which mutates mainly in poorly or undifferentiated thyroid tumors (Ito et al. 1992; Fagin et al. 1993; Donghi et al. 1993). Few studies of LOH in follicular thyroid tumors have been reported. Matsuo et al. (1991) have found LOH of chromosome 11q13, the region for the MEN1 disease gene, in four of 27 follicular adenomas. The overall frequency of LOH is however low, as is the case in a study by Kubo et al. (1991). Herrmann et al. (1991) have shown LOH on chromosome 3p in all six follicular carcinomas investigated. A similar finding has also been reported in cytogenetic studies (Jenkins et al. 1990; Roque et al. 1993). Further cytogenetic findings include, e.g., trisomy of chromosomes 7 (the most frequent trisomy) (Belge et al. 1994), 12 and 22 (Taruscio et al. 1994; Antonini et al. 1993), and translocations of the long arm of chromosome 19 (Roque et al. 1992; Belge et al. 1992).

The overall LOH frequency in this report is low compared with that found in several other neoplasms, a finding that agrees with previous reports (Matsuo et al. 1991; Kubo et al. 1991). A frequency of 15% and below does not significantly exceed that expected as "background noise" when studying LOH in a large group of tumors. However, detailed study of LOH of some loci with regard to the various histopathological types of follicular neoplasms reveals some results of interest. The finding of LOH on chromosome 10q in five of eight informative tumors with atypical and malignant features indicates that a locus in this region may be involved in follicular thyroid tumor progression (Fig. 1). A previous report of a girl who was treated for an invasive follicular carcinoma by the early age of ten may support this hypothesis. She was found to have a constitutional ring 10 chromosome with the breakpoint at 10q26 (Sparkes et al. 1978; Tommerup and Lothe 1992). Cytogenetic studies of follicular thyroid adenomas have shown the loss of chromosome 10q genomic material. However, these reports did not distinguish between common and atypical variants of the adenomas investigated (Bartnitzke et al. 1989; Antonini et al. 1991).

The retinoblastoma gene plays a role in the development of several different tumor types. Interestingly, deletion of part of chromosome 13q has been seen cytogenetically in a follicular adenoma (Belge et al. 1991). In contrast, Farid et al. (1994) report alterations of the retinoblastoma gene in 55% of thyroid carcinomas, but none in benign tumors. In parathyroid tumors, LOH at the Rb gene locus is also restricted to the malignant variant (Cryns et al. 1994). In our study, 13% of the tumors show LOH at this locus, and all tumor types are involved.

No loss of chromosome 3p was seen in the studied carcinomas, in contrast to earlier reports (Herrmann et al. 1991; Jenkins et al. 1990; Roque et al. 1993). However, the number of carcinomas in this study is too small to allow any conclusion.

When comparing the different tumor types with regard to the proportion of tumors with LOH at any locus, the difference is striking (Table 2, bottom line). Although atypical adenomas and Hürthle cell adenomas behave mainly as benign tumors, the finding that these tumors show genetic instability in comparison with common adenomas suggests a progression of these tumors. In the light of our report, we propose that LOH at chromosome 10q in atypical follicular thyroid tumors, and LOH at 3q and 18q in Hürthle adenomas should be added to the previously proposed schemes of follicular thyroid tumor progression (Fagin 1992; Wynford-Thomas 1993; Farid et al. 1994).

Acknowledgements We thank Lisa Ånfalk for technical assistance with the thyroid tissues, and Ing-Mari Lund for help with collecting blood samples. This study was supported by the Swedish Medical Research Council (no. 102, 2330), the Cancer Society in Stockholm, the Swedish Cancer Foundation, the Swedish Society for Medical Research, the Marcus Borgström, Robert Lundberg, Magnus Bergwall and Lars Hierta Foundations, and the Karolinska Institute.

References

Antonini P. Vénuat AM, Linares G, Caillou B, Schlumberger M, Travagli JP, Berger R, Parmentier C (1991) Cytogenetic abnormalities in thyroid adenomas. Cancer Genet Cytogenet 52:157–164

Antonini P, Lévy N, Caillou B, Vénuat A-M, Schlumberger M, Parmentier C, Bernheim A (1993) Numerical aberrations, including trisomy 22 as the sole anomaly, are recurrent in follicular thyroid adenomas. Genes Chromosom Cancer 8:63–66

Bartnitzke S, Hermann ME, Lobeck H, Zuschneid W, Neuhaus P, Bullerdiek J (1989) Cytogenetic findings on eight follicular adenomas including one with a t(10;19). Cancer Genet Cytogenet 39:65–68

Belge G, Thode B, Bullerdiek J, Bartnitzke S (1991) Deletion of part of the long arm of chromosome 13 as the only karyotypic aberration in a follicular thyroid adenoma. Cancer Genet Cytogenet 56:277–280

Belge G, Thode B, Bullerdiek J, Bartnitzke S (1992) Aberrations of chromosome 19. Do they characterize a subtype of benign thyroid adenomas? Cancer Genet Cytogenet 60:23–26

Belge G, Thode B, Rippe V, Bartnitzke S, Bullerdiek J (1994) A characteristic sequence of trisomies starting with trisomy 7 in benign thyroid tumors. Hum Genet 94:198–202

Bergerheim U, Nordenskjöld M, Collins VP (1989) Deletion mapping in renal cell carcinoma. Cancer Res 49:1390–1396

Cryns VL, Thor A, Hong-Ji X, Shi-Xue H, Wierman ME, Vickery AL, Benedict WF, Arnold A (1994) Loss of the retinoblastoma tumor-suppressor gene in parathyroid carcinoma. N Engl J Med 330:757–761

Donghi R, Longoni A, Pilotti S, Michieli P, Della Porta G, Pierotti MA (1993) Gene p53 mutations are restricted to poorly differentiated and undifferentiated carcinomas of the thyroid gland. J Clin Invest 91:1753–1760

Fagin JA (1992) Molecular defects in thyroid gland neoplasia. J Clin Endocrinol Metab 75:1398–1400

Fagin JA, Matsuo K, Karmakar A, Chen DL, Tang S-H, Koeffler HP (1993) High prevalence of mutations of the p53 gene in poorly differentiated human thyroid carcinomas. J Clin Invest 91:179–184

Farid NR, Shi Y, Zou M (1994) Molecular basis of thyroid cancer. Endocrine Rev 15:202–232

Goretzki PE, Lyons J, Stacy-Phipps S, Rosenau W, Demeure M, Clark OH, McCormick F, Röher H-D, Bourne HR (1992) Mutational activation of RAS and GSP oncogenes in differentiated thyroid cancer and their biological implications. World J Surg 16:576–582

Hedinger CE, Williams ED, Sobin LH (1988) Histological typing of thyroid tumours. The WHOs international histological classification of tumours, 2nd rev edn. Springer, Berlin Heidelberg New York

Herrmann MA, Hay ID, Bartelt DH, Ritland SR, Dahl RJ, Grant CS, Jenkins RB (1991) Cytogenetic and molecular genetic studies of follicular and papillary thyroid cancers. J Clin Invest 88:1596–1604

Hicks DG, LiVolsi VA, Neidich JA, Puck JM, Kant JA (1990) Clonal analysis of solitary follicular nodules in the thyroid. Am J Pathol 137:553–562

Ito T, Seyama T, Mizuno T, Tsuyama N, Hayashi T, Hayashi Y, Dohi K, Nakamura N, Akiyama M (1992) Unique association of p53 mutations with undifferentiated but not with differentiated carcinomas of the thyroid gland. Cancer Res 52:1369–1371

Jenkins RB, Hay ID, Herath JF, Schultz CG, Spurbeck JL, Grant CS, Goellner JR, Dewald GW (1990) Frequent occurrence of cytogenetic abnormalities in sporadic nonmedullary thyroid carcinoma. Cancer 66:1213–1220

Kubo K, Yoshimoto K, Yokogoshi Y, Tsuyuguchi M, Saito S (1991) Loss of heterozygosity on chromosome 1p in thyroid adenoma and medullary carcinoma, but not in papillary carcinoma. Jpn J Cancer Res 82:1097–1103

Larsson C, Byström C, Skoog L, Rotstein S, Nordenskjöld M (1990) Genomic alterations in human breast carcinomas. Genes Chromosom Cancer 2:191–197

Lemoine NR, Mayall ES, Wyllie FS, Williams ED, Goyns M, Stringer B, Wynford-Thomas D (1989) High frequency of ras oncogene activation in all stages of human thyroid tumorigenesis. Oncogene 4:159–164

Matsuo K, Tang S-H, Fagin JA (1991) Allelotype of human thyroid tumors: loss of chromosome 11q13 sequences in follicular neoplasms. Mol Endocrinol 5:1873–1879

Namba H, Rubin SA, Fagin JA (1990a) Point mutations of ras oncogenes are an early event in thyroid tumorigenesis. Mol Endocrinol 4:1474–1479

Namba H, Matsuo K, Fagin JA (1990b) Clonal composition of benign and malignant human thyroid tumors. J Clin Invest 86:120–125

O'Sullivan C, Barton CM, Staddon SL, Brown CL, Lemoine NR (1991) Activating point mutations of the gsp oncogene in human thyroid adenomas. Mol Carcinog 4:345–349

Parma J, Duprez L, Sande J van, Cochaux P, Gervy C, Mockel J, Dumont J, Vassart G (1993) Somatic mutations in the thyrotropin receptor gene cause hyperfunctioning thyroid adenomas. Nature 365:649–651

Roque L, Castedo S, Clode A, Soares J (1992) Translocation t(5;19): a recurrent change in thyroid follicular adenoma. Genes Chromosom Cancer 4:346–347

Roque L, Castedo S, Clode A, Soares J (1993) Deletion of 3p25 →pter in a primary follicular thyroid carcinoma and its metastasis. Genes Chromosom Cancer 8:199–203

Sparkes RS, Ling SM, Muller H (1978) Ring 10 chromosome: 46, XX, r10(p15q26). Hum Genet 43:341–345

Suarez HG, Villard JA du, Severino M, Caillou B, Schlumberger M, Tubiana M, Parmentier C, Monier R (1990) Presence of mutations in all three ras genes in human thyroid tumors. Oncogene 5:565–570

Suarez HG, Villard JA du, Caillou B, Schlumberger M, Parmentier C, Monier R (1991) gsp mutations in human thyroid tumours. Oncogene 6:677–679

Taruscio D, Carcangiu ML, Ried T, Ward DC (1994) Numerical chromosomal aberrations in thyroid tumors detected by double fluorescence in situ hybridization. Genes Chromosom Cancer 9:180–185

Tommerup N, Lothe R (1992) Constitutional ring chromosomes and tumour suppressor genes. J Med Genet 29:879–882

Wynford-Thomas D (1993) Molecular basis of epithelial tumorigenesis: the thyroid model. Crit Rev Oncog 4:1–23

Zedenius J, Auer G, Bäckdahl M, Falkmer U, Grimelius L, Lundell G, Wallin G (1992) Follicular tumors of the thyroid gland; diagnosis, clinical aspects and nuclear DNA content. World J Surg 16:589–594