Original Article

Diagnosis of Adrenocortical Tumors by Reticulin Algorithm

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Abstract

Aims: To apply reticulin algorithm (RA) to the diagnosis of adrenocortical tumors on adrenalectomy specimens and compare its efficacy to the modified Weiss criteria or Lin–Weiss–Bisceglia (LWB) criteria for oncocytic variant. **Materials and Methods:** Adrenocortical tumors (ACTs) diagnosed on resected specimens including the variants during January 2010–June 2016 were retrieved from the pathology records. The demographic and clinical data were obtained from medical records. The functional status of the tumor was noted based on clinical and biochemical evaluation. The location, size, and gross appearance of the tumor were noted. The corresponding hematoxylin and eosin-stained slides were independently assessed by two pathologists applying modified Weiss criteria and LWB criteria for the oncocytic variant as applicable. Reticulin stain was performed on representative sections in all cases. All the tumors were classified according to RA, and the diagnoses made by each system were correlated. **Results:** There were 15 ACTs in the study period. There were two adenomas including one oncocytoma which showed Weiss score (WS) of 2 and intact reticulin framework. There were 13 adrenal cortical carcinomas including two oncocytic variants with WS ranging from 4 to 7. There was disruption of reticulin and thick, irregular reticulin fibers in all tumors, irrespective of the histology. It correlated with modified Weiss and LWB criteria. **Conclusions:** The RA was simple, easy to apply, and correlated well with modified Weiss criteria in the diagnosis of ACTs including the oncocytic variant.

Keywords: Adrenal cortical adenoma, adrenal cortical carcinoma, modified Weiss criteria, oncocytic variant, reticulin algorithm

Introduction

Adrenal cortical tumors (ACTs) include adrenal cortical adenoma (ADA) and adrenal cortical carcinoma (ACC). ACC is a rare and aggressive disease with an incidence of one case per million with two peaks of incidence in early childhood and adults.[1,2] The pathological diagnosis of ACC is challenging owing to its rarity, wide morphological spectrum, and lack of uniform criteria applicable to all its variants. Modified Weiss criteria are widely used but have limitations in differentiating ACC with low Weiss score (WS) from anterior cerebral artery (ACA), low diagnostic accuracy among general pathologists, and nonapplicability to the variants. Volante *et al.* proposed a simplified system called the "reticulin" algorithm (RA) for the diagnosis of ACC which was validated by Duregon *et al.*, which defines malignancy through an altered reticulin framework associated with one of the three following parameters: necrosis, high mitotic rate, and vascular invasion.[1-3] This RA showed a similar diagnostic performance as compared with the WS but was faster and easier to apply including the variants.[3]

We aim to apply RA to ACC diagnosed by modified Weiss criteria or criteria applicable to variants (Lin–Weiss–Bisceglia [LWB] criteria for oncocytic ACC) on resected specimens and compare the efficacy of RA to the existing criteria.

Materials and Methods

ACTs diagnosed on resected specimens including the variants during January 2010–June 2016 were retrieved from the pathology records. The demographic and clinical data were obtained from medical records. The functional status of the

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tumor was noted based on clinical and biochemical evaluation. The location, size, and gross appearance of the tumor were noted. The corresponding hematoxylin and eosin-stained slides were independently assessed by two pathologists applying modified Weiss and LWB criteria for the oncocytic variant as applicable. Tumors were confirmed to be of adrenal cortical origin by applying immunohistochemistry using alpha-inhibin and vimentin. Reticulin stain was performed on representative sections in all cases. Two sections from normal adrenal were included as controls. The sections were examined under both low and high magnification. Reticulin network as seen in the normal adrenal gland was defined as intact when reticulin fibrils of same thickness completely surround the adrenal cortical cells in nests and cords. Loss of reticulin fibrils or qualitative disruption as defined by reticulin fibrils having variable, irregular thickness with a frayed appearance surrounding single or small groups of cells was noted. All the tumors were classified according to RA, and the diagnoses made by each system were correlated.

Results

There were 15 ACTs in the study period including two ACAs and 13 ACCs.

Adrenal cortical adenoma ($n = 2$)

There were two males aged 49 and 57 years. Both were nonfunctional and presented with nonspecific abdominal pain. They were right-sided tumors, each measuring 3.5 cm and 4.5 cm, respectively. Grossly, the tumors were well circumscribed with gray-yellow areas. Microscopically, one was an oncocytoma and the other was composed of clear cells predominantly. There were no areas of necrosis, mitoses, capsular/vascular invasion [Table 1]. The WS was 2 in both ACAs. Reticulin stain showed intact reticulin framework composed of uniform reticulin fibers surrounding cords and nests of cells [Figure 1].

Adrenal cortical carcinoma ($n = 13$)

These included all females with age ranging from 18 to 67 years. Only one patient aged 18 years presented with virilism and the rest of the patients presented with mass per abdomen. There were two right-sided and 11 left-sided tumors with size varying from to 7 cm to 21 cm. Grossly, there were gray-white areas with hemorrhage, necrosis, and gray-yellow areas. Normal adrenal was identified in five tumors, and capsular irregularity and thickness were identified in six tumors. Microscopically, two were diagnosed as oncocytic carcinoma according to LWB criteria and 11 were classified as ACC according to modified Weiss criteria [Table 1]. The WS ranged from 4 to 7. Reticulin pattern was lost or disrupted in all tumors, irrespective of the histology. The loss of reticulin fibers was focal in wide areas of the tumor, whereas disruption of reticulin was appreciated in most areas [Figure 1].

Table 1: Demographic, clinical, gross, microscopic features and modified Weiss score and Lin–Weiss–Bisceglia score in adrenal cortical tumors (n=15)

Age (years)	Gender	Location	Functional status	Weight (g)	Size in greater dimension (cm)	Mitoses	Atypical mitoses	Necrosis	<25% clear cells	Capsular invasion	Vascular invasion	Diagnosis by modified Weiss/ LBW criteria	Weiss score	RA diagnosis
53	Female	Right	NF	750	14	-	-	1	2	1	1	ACC	4	ACC
55	Female	Right	NF	400	9	2	1	1	-	1	1	ACC	5	ACC
43	Female	Left	NF	2500	21	2	1	1	2	1	1	ACC	7	ACC
30	Female	Left	NF	1700	19	2	1	1	-	1	1	ACC	5	ACC
49	Female	Left	NF	750	11	2	1	1	2	1	1	ACC	7	ACC
67	Female	Left	NF	200	7.5	2	1	-	2	1	1	ACC	6	ACC
40	Female	Left	NF	100	7	2	1	-	2	-	-	ACC	5	ACC
54	Female	Left	NF	750	15	2	1	1	2	1	1	Oncocytic variant ACC	7	ACC
29	Female	Left	NF	600	15	2	1	1	2	1	1	ACC	7	ACC
35	Female	Right	NF	750	16	-	-	1	2	1	1	Oncocytic variant ACC	4	ACC
40	Female	Left	NF	500	18	2	1	-	2	1	1	ACC	5	ACC
35	Female	Left	NF	900	18	2	-	1	2	1	1	ACC	6	ACC
18	Female	Left	Functional	225	10.5	2	1	1	2	1	1	ACC	7	ACC
49	Male	Right	NF	150	4.5	-	-	-	2	-	-	ACA oncocytic	2	ACA
57	Male	Right	NF	25	3.5	-	-	-	2	-	-	ACA	2	ACA

LBW: Lin–Weiss–Bisceglia, RA: Reticulin algorithm, ACA: Adrenal cortical adenoma, ACC: Adrenal cortical carcinoma, NF: Nonfunctional

Reticulin algorithm

Applying RA, both the adenomas were classified as adenomas including oncocytic adenoma. All 13 ACCs including two oncocytic carcinomas were diagnosed as ACC. The RA was simple to apply and diagnose ACC including oncocytic variants and differentiate from ACA [Table 1].

Discussion

The pathologic diagnosis of ACC is based on the recognition of several morphologic parameters.[4] Several classification criteria have been proposed, but none have 100% sensitivity and specificity. The RA proposed by Duregon *et al*. to define malignancy in ACT was applied in the present study.[3]

Both the cases of adenoma including one oncocytic adenoma had a WS score of 2 and intact reticulin network similar to normal adrenal. There were two cases of ACC with a WS score of 4; however, reticulin was disrupted in both the cases facilitating the diagnosis of ACC. Oncocytic neoplasms are likely to be overdiagnosed as malignant using the modified Weiss criteria. In the present study, two of the oncocytic ACCs diagnosed on LWB criteria also showed reticulin disruption. Hence, the application of RA facilitated the diagnosis of ACC with low scores and differentiated it from ADA including the variants.

In the present study, reticulin disruption was observed in all cases of ACC with different growth patterns. These observations were similar to those made by Duregon *et al*.[3] Volante *et al*. in a study on 92 ACCs observed that on morphological analysis and appropriate histochemical and immunohistochemical reactions, all ACC had disruption of reticulin framework. In addition, they concluded that apart from diffuse growth pattern, even cord-like, nesting or other patterns affected by loss of the reticulin network favored a diagnosis of ACC. This new concept of growth pattern as a primary feature in ACC diagnosis corroborates with their recent findings on the expression by adrenal

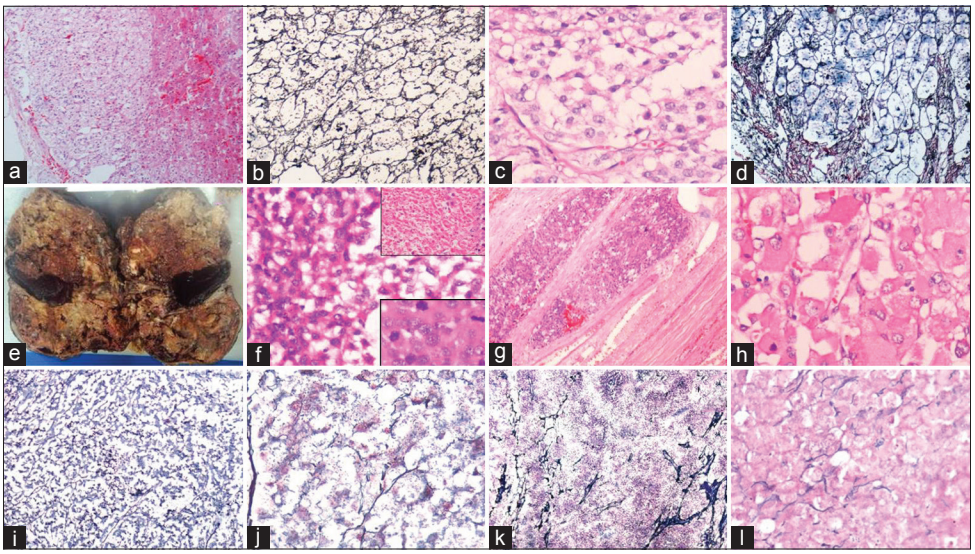

Figure 1: Normal adrenal cortex (a) H and E; (b) reticulin, ×100; adenoma (c) H and E; (d) reticulin with regular fibers of same thickness surrounding nests and cords; adrenal cortical carcinoma (e) gross specimen with thick capsule, areas of hemorrhage and necrosis; (f) sheets of polygonal cells with clear cytoplasm: insets showing necrosis and mitoses; (g) capsular and vascular invasion; H and E, ×100; (h) oncocytic variant H and E, ×400; (i) loss reticulin, ×100 (j) disruption (k) thickened and frayed fibers; (l) disrupted framework in oncocytic variant; reticulin, ×400

Table 2: Comparison of the present study with other studies

Clinicopathological features	Papotti *et al*.[1] (*n*=245)		Volante *et al*.[2] (*n*=139)		Present study (*n*=15)	
	ACA	ACC	ACA	ACC	ACA	ACC
Female/male ratio	1.9	1.45	3.2	1.5	2 males	13 females
Age (year)	54 (16-80)	48 (9-97)	53 (15-77)	46 (20-88)	54 (49-57)	40 (18-67)
Location (left/right)	28/3	92/79	23/24	48/45	0/2	10/3
NF	11	54	13	50	2	12
Oncocytic variant	7	37	4	12	-	2
WS						
0-2	61	-	47	-	2	-
3-5	-	43	-	38	-	6
6-9	-	141	-	54	-	7

WS: Weiss score, ACA: Adrenal cortical adenoma, ACC: Adrenal cortical carcinoma, NF: Nonfunctional

cancer cells of proteins involved in matrix digestion, such as matrix metalloproteinase type 2, 25 and on the deregulation of gene transcription of laminin isoforms in malignant compared with nonneoplastic adrenal tissue.[2] Table 2 gives the comparison of the present study with other large series using RA.

Duregon *et al*. in a multicentric validation study on 245 ACTs concluded that reticulin staining is a fast and cheap technique and an easy-to-interpret system, in which both quantitative and qualitative changes are considered in the evaluation of the reticulin framework disruption. They also observed that RA had high interobserver reproducibility and confirmed efficacy in the rare oncocytic variant.[3] The present study had similar observations. There was in concordance of diagnosis between both the pathologists, and the application of RA was particularly useful in equivocal cases.

The pathologic diagnosis of malignancy in ACTs plays a relevant role to stratify patients for further molecular studies and specific treatments. Scoring systems or algorithms for assessing malignancy need to be simple and reproducible to achieve the goal. The RA helps in achieving that goal.

Conclusions

This retrospective study, though small in numbers, confirms the diagnostic value of RA as compared to modified Weiss criteria. This is probably the first study of its kind evaluating and validating the RA in the Indian scenario.

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Conflicts of interest

There are no conflicts of interest.

References

1. Papotti M, Libè R, Duregon E, Volante M, Bertherat J, Tissier F. The Weiss score and beyond – Histopathology for adrenocortical carcinoma. Horm Cancer 2011;2:333-40.
2. Volante M, Bollito E, Sperone P, Tavaglione V, Daffara F, Porpiglia F, *et al.* Clinicopathological study of a series of 92 adrenocortical carcinomas: From a proposal of simplified diagnostic algorithm to prognostic stratification. Histopathology 2009;55:535-43.
3. Duregon E, Fassina A, Volante M, Nesi G, Santi R, Gatti G, *et al.* The reticulin algorithm for adrenocortical tumor diagnosis: A multicentric validation study on 245 unpublished cases. Am J Surg Pathol 2013;37:1433-40.
4. Papotti M, Duregon E, Volante M, McNicol AM. Pathology of the adrenal cortex: A reappraisal of the past 25 years focusing on adrenal cortical tumors. Endocr Pathol 2014;25:35-48.