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Current Perspective in the Diagnosis and Treatment of Adrenocortical Carcinoma

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Adrenocortical carcinomas are rare, highly malignant tumors that account for only 0.2% of deaths due to cancer. Their incidence has been estimated at two per million people per year. Approximately 50% of these tumors are functioning and produce hormonal and metabolic syndromes leading to their discovery. The other 50% are silent and discovered only when they attain large size and produce localized abdominal symptoms or metastases. Occasional children have been found to have adrenocortical carcinomas but most cases occur between ages 30 and 50 [1]. An exception to this age distribution occurs in southern Brazil, where the annual incidence of adrenocortical carcinoma in children is unusually high, ranging from 3.4 to 4.2/million children, compared with a worldwide incidence of 0.3/million children younger than 15 [2]. The etiology of adrenal cancer is unknown but molecular cytogenetic cloning studies in the past five years have described chromosomal abnormalities possibly associated with tumorigenesis [3]. These abnormalities consist of loss of heterozygosity for genes coding for tumor suppressor p53 (on 17p13.1) [4–7], p57 (on 11p15) and insulin growth factor II (on 11p15.5) [8]. Mutations in tumor suppressor genes are an important factor in hereditary cancer syndromes such as the Li Fraumeni, characterized by sarcoma, breast cancer, brain tumors, lung cancer, carcinoma of the larynx and leukemia [9]. Adrenocortical carcinoma occasionally develops in these families and patients with the Beckwith-Widenam syndrome that maps to 11p15 often develop adrenocortical cancer. In some cases, chromosomal gains have been described [2]. These genetic defects may predispose to adrenal cortical tumor formation. Environmental factors have been implicated in southern Brazil because the distribution of the tumors follows a regional rather than familial pattern.

Clinical Presentation

The early diagnosis of a functioning adrenocortical carcinoma is based on the physician's ability to recognize clinical manifestations of excessive steroid hormone production by the tumor. A definition of the biochemical abnormality present can be obtained with specific measurements of cortisol, aldosterone, androgen or estrogen levels. Computerized tomography or magnetic resonance imaging helps to localize the tumor and define the presence or absence of local or distant metastases. The following cases illustrate these clinical presentations.

Case 1

A 53-year-old woman was admitted with a two-year history of polyphagia and an 18 kg weight gain and a two-month history of generalized edema, weakness, fatigue, dyspnea and facial hirsutism. She had a past history of non-insulin-dependent diabetes mellitus treated with sulfonylureas. Physical examination revealed moon facies, trunkal obesity and facial lanugal hirsutism but no striae, acne or skin atrophy. Blood pressure was 150/96 mm/Hg and she had a trace of pretibial edema. Biochemical studies revealed urinary 17-hydroxycorticoids of 11.8 mg/d; urinary free cortisol of 399 μg/d and a cortisol secretion rate of 41.3 mg/d. Serum cortisol levels were 43.2, 43.7 and 46.4 at 0800, 1200 and 2200 hours respectively. Cortisol levels failed to suppress with dexamethasone. An adrenal venogram revealed a 5 cm right suprarenal mass; iodocholesterol scintigraphy showed this mass did not take-up the tracer. She underwent surgical resection of a well-circumscribed adrenocortical carcinoma. The patient required postoperative replacement with cortisol, 30 mg daily. She was begun on adjuvant therapy with mitotane, 3 gm daily and was maintained on a dose of 1 gm daily for the following seven years. She was last evaluated 18 years postoperatively and had no evidence of either hormonal or anatomical tumor recurrence.

Case 2

A 27-year-old woman was admitted with a two-year history of irregular menses. She was initially treated with oral contraceptives but developed hypertension and they were discontinued. Subsequently, she noted acne over the trunk and extremities, scalp hair loss, weight gain and depression. Physical examination showed a blood pressure of 122/90 mm/Hg and mild hirsutism. Biochemical studies revealed high serum total testosterone, 1.44 ng/ml (N: 0.2–0.8), free testosterone 28 pg/ml (N: 3–13) and DHEA-S 435 μg/dl (N: 80–300). Cortisol was normal, 13 μg/dl but ACTH was suppressed. Urinary 17-ketosteroids was 21.4 mg/d and remained elevated at 24.3 mg/d after dexamethasone. Urinary free cortisol, 79 μg/d, was at the upper limit of normal and did not suppress on dexamethasone. Imaging studies including an abdominal sonogram and a CT scan showed a 6.4 cm left adrenal mass. Iodocholesterol scintigraphy failure to visualize either gland was consistent with a functioning (cortisol and androgen-secreting) adrenocortical carcinoma. The patient underwent resection of a left adrenal carcinoma and was placed on adjuvant therapy with mitotane, 2 gm daily. Four years later, the patient showed no evidence of either hormonal or anatomical tumor recurrence.

Cushing's syndrome is the most common clinical presentation in the adult patient [10]. Characteristically, these patients describe rapid development (3–6 months) of the clinical manifestations of cortisol excess including weight gain, muscle weakness, easy bruising, irritability and insomnia. In addition, there commonly are manifestations of androgen excess, including hirsutism, acne and irregular menses or amenorrhea in women. The androgen excess may decrease the severity of the catabolic effect of hypercortisolemia such that skin and muscle atrophy may not be as readily apparent as in those with benign tumors. Patients with metastatic disease have anorexia and weight loss rather than weight gain. Adrenocortical carcinomas causing Cushing's syndrome are large tumors with an average weight of 800 gm, but the clinical manifestations of hormone excess lead to earlier diagnosis and the finding of smaller tumors.

Studies of patients with clinical manifestations of Cushing's syndrome include measurements of urinary free cortisol, serum cortisol and DHEA-S at baseline and during dexamethasone suppression. Those with cortisol-secreting adrenocortical carcinomas demonstrate elevated baseline cortisol and DHEA-S levels and failure to suppress with a high (8 mg) dose of dexamethasone. ACTH levels are usually suppressed. Occasionally, the steroid profile in serum or urine can help distinguish between benign and malignant adrenocortical tumors because of the presence of intermediary precursors in the steroid biosynthetic pathway or their metabolites in the serum of patients with malignant neoplasms [11].

Sex hormone producing carcinomas lead to virilization in women and manifestations of feminization in men. Women with virilizing adrenocortical carcinomas present with marked androgen type hirsutism, male pattern baldness, deepening voice, breast atrophy, clitoral hypertrophy, decreased libido and oligo or amenorrhea. Manifestations of androgen excess are less noticeable in men. In prepubertal boys, androgen excess will cause precocious puberty without concomitant testicular enlargement. Feminizing tumors in women may cause breast tenderness and dysfunctional uterine bleeding. Estrogen-secreting tumors in men are associated with gynecomastia, breast tenderness, testicular atrophy and decreased libido. In prepubertal girls, feminizing tumors cause early breast and uterine development and onset of menarche.

Patients with virilizing tumors demonstrate elevated serum levels of testosterone, androstenedione and DHEA-S, while patients with feminizing tumors have high serum estradiol levels. These levels are usually very high with total testosterone in women frequently greater than 2.0 ng/ml (N female, 0.3–0.6 ng/ml).

Aldosterone-producing adrenocortical carcinomas are rare [12,13]. They present with clinical manifestations of primary aldosteronism with hypertension and hypokalemia. Compared to patients with benign aldosterone-secreting adenomas, those with carcinoma have larger tumors, higher aldosterone levels and more severe hypokalemia. Evaluation should include measurement of serum electrolytes, aldosterone and plasma renin levels. The usual findings are severe hypokalemia with potassium levels below 2.5 mEq/l, hypernatremia and metabolic alkalosis. Serum aldosterone levels are high and plasma renin levels are suppressed.

The detection of non-functioning tumors is frequently incidental to the investigation of nonspecific abdominal complaints.

Role of Imaging in the Evaluation of Adrenal Cancer

A variety of imaging procedures can be used to localize and determine the possible benign or malignant character of an adrenal cortical mass. On computerized tomography (CT) malignant adrenal masses are usually larger than 5 cm and have an inhomogeneous pattern because of areas of necrosis within the tumor. The CT procedure helps determine the presence of involved lymph nodes and hepatic or pulmonary metastases. A definition of metastatic involvement is important in determining

treatment goals for a given patient. The tumors are frequently invasive of the upper pole of the adjoining kidney and of the inferior vena cava. By ultrasonography, large malignant lesions vary in echo texture with focal or scattered echopenic or echogenic zones representing areas of tumor necrosis, hemorrhage and/or, calcification [14]. By magnetic resonance imaging (MRI) tumors appear as hypointense masses compared to the liver on T-1 weighted images and hyperintense compared to the liver on T-2 weighted images. The MR also demonstrates displacement or invasion of adjacent organs as well as liver metastases. Superior blood vessel identification and the multiplanar capabilities of MRA makes it the imaging modality of choice in evaluating the extent of disease and the degree of vascular invasion and in planning surgical excision [15,16]. Scintigraphic imaging characteristics can help determine if an adrenal tumor is benign or malignant [17]. Benign tumors show uptake of ^{131}I-6-β-iodomethyl-19-norcholesterol (NP-59) on adrenal scintigraphy while malignant tumors fail to take-up this radiolabeled tracer. Since hypercortisolemia suppresses ACTH secretion and the function of the contralateral adrenal gland, patients with cortisol-producing adrenocortical carcinomas fail to show an image either at the site of the tumor or the contralateral gland. On the other hand, aldosterone, androgen or estrogen-secreting tumors usually appear as an area of decreased uptake on the side of the tumor mass. The decreased or absent tracer uptake by adrenocortical carcinomas are in contrast with the increased concentration of radionuclide by benign tumors. However, this distinction is not absolute. Patients with adrenocortical carcinoma may occasionally give positive nuclear scans. This is illustrated by Case 3.

Case 3

A 65-year-old woman presented with severe Cushing's syndrome. A CT scan of the abdomen revealed a large adrenal mass consistent with an adrenocortical tumor. Iodocholesterol scintigraphy revealed intense uptake on the side of the tumor concordant with the CT image and with suppression of uptake by the contralateral adrenal. Resection of the mass revealed it to be an adrenocortical carcinoma.

A positive scan can mislead the physician into thinking that the tumor is benign and choose a surgical approach to the tumor (posterior or laparoscopic) that is inappropiate for the resection of an adrenal carcinoma.

Pathological Definition of Malignancy

Various systems of diagnosis have been proposed but the most commonly used is the one described by Weiss [18,19]. Nine histologic findings have been described: (1) high nuclear grade, (2) mitotic grade greater than 5 per 50 high power fields (HPF), (3) atypical mitotic figures, (4) eosinophilic tumor cell cytoplasm, (5) diffuse architecture, (6) necrosis, (7) venous invasion, (8) sinusoidal invasion and (9) capsular invasion. Malignant tumors have four or more of these histologic criteria. The three most commonly found are a mitotic rate greater than 5 per 50 HPF, atypical mitotic figures and venous invasion. The mitotic rate is an important criterion not only for distinguishing malignant from benign tumors but also for predicting clinical virulence of adrenocortical carcinomas. Patients with carcinomas with a high mitotic rate (more than 20 mitoses per 40 HPF) have a shorter disease-free survival period as compared with those with low mitotic rates (less than 20 mitoses per 40 HPF).

Most adrenocortical carcinomas are of large size; however, size alone is not the only factor determining survival. Depending on the degree of cell differentiation, adrenal cortical carcinomas have been classified as well differentiated or anaplastic. While better-differentiated carcinomas may have a less aggressive course than the anaplastic tumors, cell differentiation does not predict survival independently of the mitotic rate.

Attempts have been made to study adrenocortical carcinomas by immunohistochemical analysis. These tumors are frequently positive for vimentin. In contrast to other malignant neoplasms, they do not consistently express keratin, α-1 antitrypsin, α-fetoprotein, β-2 microglobulin or lectins. Anti-alpha-inhibin, an antibody directed against the peptide inhibin, appears to reliably distinguish between adrenal and renal cell carcinoma [20].

The measurement of cellular DNA by flow cytometry has been used to distinguish benign from malignant adrenocortical tumors [21]. These measurements have been related to histopathologic criteria of malignancy and survival. While aneuploid stemlines are identified more often in carcinomas than in adenomas, the sensitivity and specificity of aneuploidy for predicting clinical outcomes is only 56 and 65%, respectively. Identification of specific gene mutations in adrenal cortical carcinomas may, in the future, be helpful in determining not only diagnosis but the malignant grade and prognosis of individual tumors.

Staging of Adrenocortical Carcinoma Based on Size and Extent of Tumor Involvement

Patients with clinical and imaging diagnosis of an adrenocortical carcinoma should undergo staging of

Table 1.

Stage	Size	L Nodes	Local invasion	Metastases	TNM
I	< 5 cm	−	−	−	$T_1N_0M_0$
II	> 5 cm	−	−	−	$T_2N_0M_0$
III	Any Size	+	+	−	$T_{1,2}N_1M_0$
IV	Any Size	+	+	+	$T_{1,2}N_1M_1$

their disease by appropriate imaging procedures. Staging not only determines prognosis but also the selection of treatment. The MacFarland classification [22] as modified by Sullivan [23] is described in Table 1.

Patients in stage I have tumors that measure less than 5 cm in size and have no evidence of lymph node involvement or metastases; patients in stage II have tumors larger than 5 cm but are also free of lymph node involvement or metastases. Patients in stage III exhibit tumors of any size with local lymph node invasion or have experienced local recurrence. Patients in stage IV have distant metastases. The sites of tumor spread in stage IV are summarized in Table 2. The most frequent sites for metastases are lung, liver, lymph nodes, and bone. The stage at which an adrenal cortical carcinoma is defined determines prognosis [24,25]. While 50% of patients in stages I, II or III are alive 40 months after diagnosis, only 10% of patients in stage IV are alive at that time.

Treatment Possibilities of Adrenal Cancer

Therapeutic interventions used to treat patients with adrenal cancer include surgery, radiation therapy, systemic chemotherapy and mitotane [10].

Surgical resection, even when incomplete, should be considered the initial step in therapy. Because most adrenal carcinomas are large, the surgical approach should be either transabdominal or thoracoabdominal with an incision sufficiently wide to allow adequate exposure for resection of contiguous organs and if necessary, to remove all visible tumor. The surgical goal should be the resection of the entire tumor mass when possible. When this is not possible because of local extension into other structures, tumor debulking to the maximum degree possible is indicated. It is frequently necessary to remove the adjoining kidney in block with the tumor because of invasion by the tumor of the upper pole. In cases of liver metastases, a partial lobectomy with resection of the involved portion of the liver has led to long-term remission [26]. Patients have been operated even when there was tumor invasion of the inferior vena cava and neoplastic thrombus extending from the right atrium to the bifurcation. In some cases, this thrombectomy required cardiopulmonary bypass and vascular reconstruction. Patients tolerated this extensive procedure without perioperative deaths and with disease-free intervals of 3–31 months [27]. These aggressive efforts to excise all visible tumor are justified because they may increase life expectancy. Several of the larger series indicate that surgical resection of the primary tumor and metastases results in extended survival in 56% of patients [28].

Table 2. Sites of metastasis in stage IV adrenocortical carcinoma

Organ	Percent ($n = 33$)
Lung	45
Liver	42
Lymph nodes	24
Bone	15
Pancreas	12
Diaphragm	12
Spleen	6
Miscellaneous	12

(Brain, peritoneum, skin, palate).

Adrenocortical carcinomas are generally resistant to radiation therapy that causes only transient reduction of local disease [29]. Conventional abdominal irradiation is associated with tumor response in 15% of the cases [28]. More recently, we have used 3-dimensional conformal radiotherapy directed to the suprarenal space after local tumor recurrence and repeat surgical excision. Patients have tolerated this form of radiotherapy without complications and without evidence of recurrence after one year of follow-up. The number of patients treated is small and the length of follow-up not long enough for a determination of the efficacy of this modality of treatment, but more extensive use of this approach will help determine the value of this form of radiotherapy in the treatment of adrenocortical carcinoma.

Mitotane (o,p′-DDD), is an adrenalytic drug with selective activity on the adrenal cortex which has been found to be effective in inducing a tumor response in 33% of patients treated [10]. The duration of response has varied between 1 and 204 months. Most of the experience with mitotane comes from its use on patients in advanced stages of the disease but its effectiveness under those circumstances has been disputed [30,31]. Decreases in elevated urinary steroid levels, measurable disease response and overall clinical response have been described. Mean survival however is short (8.4 months) when the drug is used after the appearance of metastatic disease. Isolated case reports have described impressive remissions and even cures of adrenocortical carcinoma following mitotane monotherapy. However, the drug has been used more commonly in combination with other systemic chemotherapy.

The initial dose is 1 g twice daily and gradually escalated to tolerance. The optimal dose should be

determined by measuring blood levels. These levels should be $> 14\,\mu g/dl$ [32]. By following blood levels it has been possible to decrease the dose and limit toxicity [33]. The therapeutic threshold is reached after 3–5 months of therapy. The drug should be administered with fat containing foods since its absorption and transport appears to be coupled to lipoproteins.

The cortisol response to mitotane therapy can be determined by measuring urinary free cortisol excretion. Serum cortisol levels are frequently elevated even when the circulating level of free cortisol is not, because mitotane increases the binding of cortisol to CBG [34]. Tumor response can be determined by measuring the size and number of metastatic lesions by CT or MRI.

Treatment with mitotane not only affects the tumor, but also the function and integrity of the contralateral uninvolved adrenal gland. Patients need to be replaced with hydrocortisone in doses of 25–35 mg daily. When given in low doses (2–4 gm daily), mitotane has less adrenalytic effects on the zona glomerulosa of the contralateral gland and is less likely to suppress aldosterone production. With larger doses, replacement with 9-alpha-flurocortisol may be necessary.

A limiting factor in the use of mitotane as a therapeutic agent is its toxicity. Adverse effects of mitotane therapy are found to be dose-dependent. The prominent early side effects of large doses of mitotane are anorexia and nausea. Side effects can be reversed if therapy is interrupted for several days and restarted at lower dose levels. Occasionally, maculopapullar exanthem and exfoliative dermatitis with mucosal involvement have been found in susceptible patients taking mitotane. Similarly, hepatotoxic effects requiring interruption of therapy have been observed in some patients.

It is likely that the tumor response depends on the ability of adrenal tumors to metabolize mitotane, a step required for its adrenalytic activity. Mitotane belongs to the class of drugs that requires metabolic transformation for therapeutic action. As a result of this transformation, active metabolites are produced that cause toxicity either through covalent binding to specific targets within the cells or oxygen activation with superoxide formation. The concept that mitotane requires metabolic transformation for activity stems from observations of its variable activity in different animal species. The dog adrenal, the most responsive to mitotane is also the most capable of metabolite formation and covalent binding. In contrast, the human adrenal is less capable of both transformation and binding and is less responsive [35].

This metabolic transformation is P-450 mediated and it generally occurs in normal adrenals where it causes suppression of function and necrosis. Adrenal carcinomas may not retain the ability to metabolize mitotane and become unresponsive to the drug. Studies showing an ability of mitotane to suppress the expression of a multidrug resistant (MDR) gene in adrenocortical cancer cells [36] suggest a possible use of mitotane as an enhancer of the concentration of other chemotherapeutic agents within the adrenocortical tumor.

We proposed a pathway of mitotane metabolism that follows the well-known process by which chloramphenicol causes toxicity. Mitotane is hydroxylated at the β-carbon and quickly transformed by dehydrochlorination into an acyl-chloride. The acyl-chloride either covalently binds to bionucleophiles in the target cells or, by losing water, is transformed to the acetic acid derivative (DDA) for renal excretion (Fig. 1). The initial hydroxylation step is carried out in the mitochondria and is catalyzed

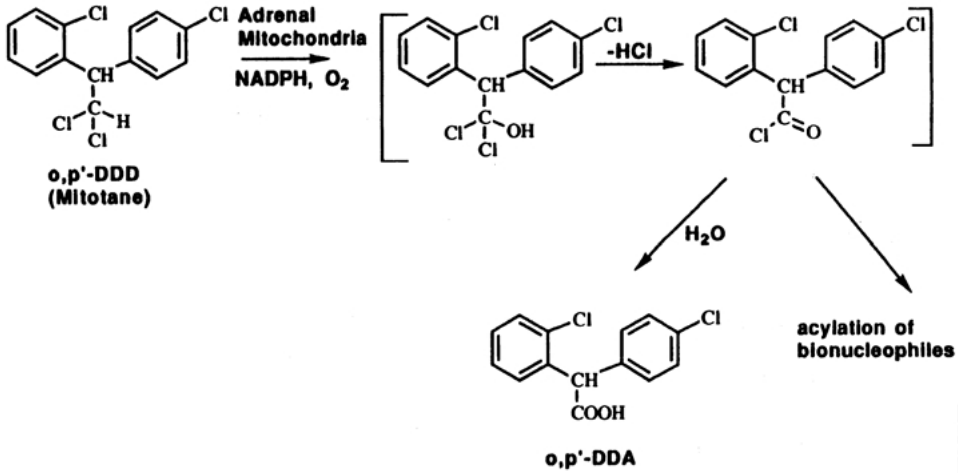

Fig. 1. *Proposed mechanism for the metabolism of mitotane. Mitotane is hydroxylated at the β-carbon and quickly transformed by dehydrochlorination into an acyl-chloride. The acyl-chloride either covalently binds to bionucleophiles in the target cells or, by losing water, is transformed to the acetic acid derivative (DDA) for renal excretion.*

through a P-450 enzyme. The importance of this metabolic transformation can be tested by the introduction of a methyl group at the β-carbon of o,p′-DDD, a procedure that blocks the metabolic transformation to the acyl-chloride. Whereas dogs treated with mitotane show prompt suppression of cortisol secretion and increases in serum ACTH levels, treatment with the methylated analog (mitometh) does lack this effect. Similarly, while mitotane causes necrosis of the adrenal cortex, mitometh has no significant effect [37].

The pathway of metabolic transformation of mitotane has been extensively studied in our laboratory [38]. Formation of hydroxylated derivatives can be shown by incubation of dog adrenal homogenates with radiolabeled o,p′-DDD. HPLC analysis of the homogenate after incubation shows the appearance of hydroxylated metabolites and the production of DDA. We have characterized the adrenal enzymes involved in mitotane metabolism. The metabolic reaction is dependent on O_2 and NADPH; and is inhibited by 68% with a mixture of O_2-CO [20 : 80]. The metabolic tranformation is inhibited by ketoconazol, but not by other specific enzyme inhibitors such as aminoglutethimide and metyrapone, or substrates such as cholesterol, 11-deoxycorticosterone, 11-deoxycortisol, corticosterone and androstenedione. It is possible that the mitotane-metabolizing enzyme is a novel non-steroidogenic P-450 present in the adrenal cortex and active in the metabolism of xenobiotics.

We have also investigated the cellular target to which mitotane metabolites are covalently bound. Incubation of normal adrenal and adrenal tumor homogenates with a radiolabeled analog of mitotane show that most of the radioactivity is associated with proteins with molecular weights of 49.5 and 11.5 kD. The sequence and structure of these proteins have not been worked out.

Another possible mechanism mediating the adrenalytic effect of mitotane is oxidative damage through production of free radicals. We have tested the effect of tocopherol acetate on the antiproliferative activity of mitotane and shown that this activity is reversed by the addition of this antioxidant to NCI-H295 adrenal cancer cell cultures.

Thus, metabolic transformation and free radical formation are mechanisms involved in the cytotoxicity of mitotane. Tumors vary in their ability to metabolize mitotane and the ability of tumors to transform mitotane may predict the clinical response to the drug. Preliminary data show a possible correlation between metabolic activity of neoplastic adrenocortical tissue and response to mitotane. We have developed a tritium release assay to test the ability of adrenal tumors to metabolize mitotane [39]. Tritiated mitotane is incubated with adrenal homogenates or cell suspensions. The unreacted substrate is removed and the amount of tritium released to the aqueous media is determined. The amount of tritium released correlates with the metabolic transformation into the acyl-chloride. Results from the tritium release assay correlate with data obtained using ^{14}C labeled compound. This assay has potential clinical application for selecting patients who are responsive to mitotane from those who are not likely to respond.

Combination of Surgery and Mitotane

Early treatment with mitotane following surgical resection is also associated with longer survival. This combined approach is illustrated by cases 1 and 2 described above and by the following case.

Case 4

A 20-year-old woman was first evaluated with a one-year history of irregular menses, six months of hirsutism and deepening of her voice and four months of amenorrhea. Physical examination revealed hirsutism, slight clitoromegaly and a palpable left upper quadrant mass. CT scan of the abdomen showed a large left suprarenal mass and large hepatic metastases. Urinary 17-ketosteroids obtained at an outside hospital were very high. She underwent resection of the left adrenal mass at another hospital. Surgery included splenectomy, distal pancreatectomy and left nephrectomy. The pathological report revealed a poorly differentiated adrenal carcinoma. The patient was started on chemotherapy with mitotane 10 gm and adriamycin. However, this treatment was poorly tolerated and discontinued. The patient subsequently underwent right hepatic trisegmentectomy with gross total removal of the tumor. Postoperatively, she was placed on mitotane 6 gm daily and hydrocortisone 35 mg daily. For the next three years the patient demonstrated low cortisol and androgen levels. However, repeat CT of her chest showed a 3×5.5 cm mass in the left lung base and a 1.5 cm nodule in the right lung apex. Both of these lesions were resected through thoracotomies. The patient continued treatment with mitotane in gradually decreasing doses from 3 to 1 g daily. Seven years after the initial diagnosis, she had no evidence of radiographic or biochemical tumor recurrence.

In a study of patients undergoing surgical resection of an adrenocortical carcinoma, 26 patients took mitotane from 10 months to 10 years; 13 patients started treatment immediately after surgery and 13 after a delay. Forty six percent of these patients had long survival compared to 28% who had surgery alone without mitotane [40].

In spite of these reports, the use of mitotane as adjuvant therapy in patients with stages I and II

adrenocortical carcinoma is controversial [41,42] because of lack of convincing data that the drug can prevent tumor recurrance and the significant toxicity associated with its administration. Prospective, large multicenter trials are necessary to determine the proper role of mitotane as adjuvant therapy.

Systemic chemotherapy has shown response in less than 10% of patients treated for stages III and IV. Several chemotherapeutic protocols have been employed with variable success. In a series of 14 patients with progressive metastatic disease and large tumor burden, treatment with a combination of 5-fluorouracil, cisplatin and doxorubicin had an overall response rate of 23% but complete remission lasting 42 months was achieved in only one patient. Significant toxicity was observed with these drugs [43]. A combination of cisplatin and etoposide was reported to have induced partial remission after only one cycle of therapy [44].

Combination of Systemic Chemotherapy and Mitotane

Combinations of adriamycin vincristin, cisplatin, etoposide and mitotane have been used in escalating doses with some success. Mitotane has been used in these instances because of its potential to suppress a multidrug resistance gene and facilite the action of the other drugs. Several trials have been reported. Twenty-eight patients with inoperable, advanced stage adrenocortical carcinoma were studied as part of an Italian multicenter phase II trial. The protocol consisted of etoposide, 100 mg/m^2, doxorubicin, 20 mg/m^2, cisplatin 40 mg/m^2 given between days 1 and 9 and mitotane, up to 4 g daily. A complete response was reported in two patients and a partial response in 13 for an overall response rate of 53% [45]. Not all of these trials have been successful. In another phase II trial, 45 patients were treated with cisplatin and etoposide, followed by mitotane at disease progression. Cycles were repeated every 21 days. Objective responses were 11–13% with median survival of 10 months [46]. The difficulty in assessing the effectiveness of published treatment protocols stems from the fact that most series are limited in the number of patients studied. There is great variability in the drugs used, the stage and extent of the tumor, and the malignancy grade. In addition, there is lack of a uniform definition of response, the duration of response is unclear and multiple treatments are given in variable sequence.

Several treatment strategies have resulted in temporary or partial tumor regression but very few cases have attained long survival [1].

Other anticancer drugs including taxol, gemcitabine, suramin and gossypol have been used with equivocal results [47]. Taxol [48] and paclitaxel [49] have been shown to have antiproliferative effects *in vitro* on the human adrenocortical carcinoma cell line NCI-H295, but this effect needs to be demonstrated *in vivo*. Suramin, an antiparasitic drug, had been shown to have adrenocorticolytic effects in primates. When given as a single agent to patients with metastatic disease, a partial to minor response was observed in some.

Prognosis and Response to Therapy

The results of therapy for adrenocortical carcinoma are uniformly poor. There are a significant number of patients on whom therapy can extend life expectancy with acceptable morbidity. However, recurrence can occur even after long periods of remission.

Several series of patients receiving treatment for adrenocortical carcinoma have been evaluated for long term response. In a comparison of 18 patients treated with mitotane alone and 15 patients treated with combined surgical resection and mitotane chemotherapy, those who underwent surgical treatment had a more favorable response, with 33% of patients living more than five years from the time of first recurrence [50]. In a study of 49 patients with adrenal carcinoma, surgical excision offered the best opportunity for prolonged survival. Forty three percent of patients with a completely resectable tumor were alive with no evidence of disease an average of 7.3 years postoperatively [51]. Comparing various types of therapy in 110 patients with adrenocortical carcinoma, it was noted that 56% of patients responded to surgery for localized and regional disease with a disease free survival time of at least two years. In contrast, abdominal radiation therapy was effective in 15%, systemic chemotherapy in 9% and mitotane in 29% [28]. In a review of 82 patients, it was noted that survival of patients with metastatic disease was poor and not improved by treatment with mitotane, cytotoxic chemotherapy or radiation therapy [52].

The pathological characteristics of the tumor may predict survival. Patients with primary tumors of less than 5 cm in diameter and without local or distal extension appear to have relatively good prognosis and long survival. In contrast, patients with larger tumors or local metastatic disease have short life expectancy [24]. The degree of ''histologic'' malignancy also correlates with survival. In a study of 21 patients with adrenocortical carcinoma, the median survival time of patients with anaplastic tumors was only five months, while median survival of those with differentiated tumors was 40 months. Three of five patients with differentiated

adrenal tumors had objective responses to mitotane [53]. The mitotic rate has a strong statistical association with patient outcome. In 42 cases of adrenocortical carcinoma, the 21 patients who had tumors with greater than 20 mitoses per 50 HPF had a median survival of 14 months. In contrast, the 21 patients with carcinomas with less than 20 mitoses per 50 HPF had a median survival of 58 months [24].

As indicated above, the use of mitotane as adjuvant therapy is controversial. An important limitation of its use under these circumstances is the toxicity associated with its long-term administration and lack of information on how long adjuvant treatment should be continued. Case 5 illustrates the possible advantage of post-surgical treatment with mitotane.

Case 5

A 45-year-old man had resection of a primary left adrenal cortical carcinoma. Several months later he developed a recurrent left suprarenal mass that was surgically resected and weighed 868 gm. He also had a left nephrectomy and splenectomy. Post op, the patient was started on mitotane therapy. Six months later he had developed adrenal insufficiency and was started on cortisol replacement therapy. He had no recurrence of this tumor. Three years later he developed an adenocarcinoma of the rectum and underwent AP resection and sigmoid colostomy. Two years later he was noted to have developed a large left retroperitoneal mass and underwent debulking resection of this mass. He went on to develop an obstructive uropathy secondary to recurrence of his rectal carcinoma, and underwent chemotherapy. He eventually died of complications of his rectal cancer.

Follow-up of Patients with Adrenal Cancer

A hormonal profile should be determined in every patient with an adrenal mass and especially patients who may have a primary adrenal cortical carcinoma. Cortisol, androgens and estrogens are the adrenal cortical steroids most commonly found elevated in these patients. A hormonal profile should also be obtained on patients with apparently non-functioning adrenal tumors. Some apparently non-functioning tumors may produce biosynthetic steroid pathway intermediates such as progesterone and 11-deoxycortisol. It is important to determine the levels of these steroids on patients with adrenocortical cancer prior to surgery since these hormones can be used as biochemical markers in the postoperative follow-up. In Case 6, the hormonal profile continued to be abnormal after surgery, indicating significant residual.

Case 6

A 39-year-old woman presented with dysmenorrhea and was treated with endometrial ablation. She subsequently complained of bloating, weight gain and hirsutism. She was investigated for possible thyroid disease. Six months later she was noted to have a large abdominal mass. A CT revealed a $14 \times 21 \times 22$ cm left suprarenal mass and hepatic and pulmonary metastases. A FNB of the mass revealed sheets of pleomorphic cells consistent with adrenal carcinoma. Biochemical studies showed: UFC 807 μg/d; U17KS 228 μg/d; serum cortisol 32–40 μg/dl without circadian variation; serum DHEA-S 993 μg/dl; serum testosterone, total 2.4 ng/ml; free 18.7 pg/ml (N: 0.3–2.2). The patient underwent debulking resection of the primary tumor that weighed 1800 gm and resection of the ipsilateral kidney. The mass had areas of necrosis. Histologically, this was an adrenocortical carcinoma with angiolymphatic invasion and invasion of the left adrenal vein. Repeat biochemical studies one week post-op showed the following values: UFC 158 μg/d; U17KS 75.5 μg/d; serum cortisol 18.3–21.1 μg/dl; serum DHEA-S 647–983 μg/dl; serum testosterone, total 1.0 ng/ml; free 10.8 pg/ml. The patient began chemotherapy but died eight months later with metastases.

Use of FNB in the Diagnosis of Adrenal Cancer

FNBs of adrenal masses may be helpful in the detection of metastatic disease to the adrenal but it is not recommended when there is a high probability of a primary adrenocortical carcinoma. In a series of 48 patients who had undergone FNB for incidentally discovered adrenal masses, 23 showed evidence of metastatic disease to the adrenal. Eighteen of the 23 had known malignancy [54]. The most common primaries (61%) were adrenal metastases from a primary lung carcinoma. Of importance is that the cytological characteristics of the metastases are similar to those of the primary tumor. Thus, FNB is useful in documenting metastatic disease. We do not advise the use of FNB on patients with a high probability of a primary adrenocortical carcinoma. Tracking of tumor cells along the path of the needle may result in transplantation of the tumor to the liver without metastasis. This is illustrated by the following case.

Case 7

A 33-year-old woman was diagnosed with Cushing's syndrome secondary to an adrenocortical carcinoma. She had presented with weight gain, night sweats and headaches. Because of concomitant abdominal pain, an

abdominal ultrasound was performed that revealed a large intra-abdominal mass compressing the inferior surface of the right lobe of the liver. The mass was initially thought to be hepatic and she underwent a fine needle aspiration biopsy of the mass prior to surgery. The tumor mass was resected and the Cushing syndrome remitted. She had periodic follow-up abdominal CT scans. Six months later she was noted to have a small, 2.5 cm hepatic lesion but because she became pregnant the lesion was not followed-up until after delivery. At that time the mass was large, 9 cm and she had re-developed clinical and biochemical manifestations of Cushing syndrome. She underwent a partial hepatectomy. The lesion, cylindrical in shape, was consistent with adrenocortical carcinoma, most likely implanted in the liver by the needle. She did not have further recurrance of her disease 30 months later and did not receive any adjuvant chemotherapy.

Metabolic Management of Patients with Functioning Adrenocortical Carcinoma

If the patient continues to have residual disease, the metabolic changes associated with excessive hormonal production may cause significant morbidity and shortened life expectancy. A variety of inhibitors of adrenal function have been used to suppress steroid hormone production and improve the clinical manifestations of the disease. The most commonly used inhibitors include ketoconazole and aminoglutethimide: ketoconazole is an imidazole derivative that inhibits the synthesis of cortisol by inhibiting mitochondrial cytochrome P-450 dependent enzymes such as cholesterol side chain cleavage and 11-beta-hydroxylase in rat and mouse adrenal preparations. It has been found to be an important inhibitor of gonadal and adrenal steroidogenesis *in vivo* when given in doses as low as 200–600 mg/day. Ketoconazole has been used to treat Cushing's syndrome caused by adrenal tumor [55]. Clinical improvement occurs frequently but regression of metastatic disease is rare [56].

When patients are treated with ketoconazole, adrenal insufficiency is avoided by decreasing the dose sufficiently to maintain normal cortisol levels. The most frequent adverse reactions with ketoconazole are nausea and vomiting, abdominal pain and pruritus in 1–3% of patients. Hepatotoxicity, primarily of the hepatocellular type, has been associated with its use. Aminoglutethimide inhibits cholesterol side chain cleavage and the conversion of cholesterol to delta-5-pregnenolone in the adrenal cortex. As a consequence, the synthesis of cortisol, aldosterone and androgens is suppressed. The drug has been used both in adults and children in doses of 0.5–2.0 gm per day. Cortisol levels fall gradually with regression of the clinical manifestations of Cushing's syndrome [57]. Eventually, patients may need glucocorticoid replacement. The effect of aminoglutethimide is promptly reversed by interruption of therapy. Aminoglutethimide causes gastrointestinal (anorexia, nausea, vomiting) and neurologic (lethargy, sedation, blurred vision) side effect and can cause hypothyroidism in 5% of patients. Skin rash is frequently observed during the first ten days of treatment; this usually subsides despite continuation of treatment. Headaches have also been observed with larger doses.

Future approaches to the treatment of adrenocortical carcinoma are likely to be based on blocking or reversing the biological mechanisms of tumorigenesis. For example, angiogenic and immunological factors may play a role in adrenal tumor growth. Inhibition of these factors may result in inhibition of tumor growth.

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