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A fly in the ointment: reassessing mitotane's role in the treatment of adrenocortical carcinoma

"In light of recent developments, we need to step away from using mitotane simply because it is the only approved treatment."

KEYWORDS: adrenocortical carcinoma ■ CYP3A4 ■ FIRM-ACT ■ mitotane

Mitotane (2-[o-chlorophenyl]-2-[p-chlorophenyl]-1,1-dichloroethane or o,p'-DDD), an isomer of the banned pesticide 1,1,1-trichloro-2,2-di(4-chlorophenyl)ethane (DDT), is the current standard of care for patients presenting with advanced and metastatic adrenocortical carcinoma (ACC) [1–3]. Mitotane or combinations containing mitotane have remained the standard of care for patients with ACC for over 30 years despite a lack of understanding regarding its mechanism of action or interactions with other agents and a suboptimal effectiveness. The results from the recently reported FIRM-ACT trial [4] and evidence from a study of urine steroid profiles of patients with ACC cast further doubt on the broad use of mitotane in treating ACC patients [5]. These studies highlight a need to identify markers of mitotane resistance and put significant effort into the development of new treatments, and possibly treatment paradigms, in order to improve outcomes for patients with ACC.

Why mitotane is the standard of care for the treatment of ACC

ACC is an aggressive cancer of the adrenal cortex. The outcome for patients with ACC has remained unchanged in the past 25 years, with the overall 5-year survival of patients undergoing surgical resection being approximately 40% [6]. The disease may have a variable course. Certain patients generally fare better, including those with stage II disease whose 5-year survival rates approach 60%, and improve to greater than 95% if their tumor does not recur within 16 months [7]. However, patients who present with large, locally invasive tumors, have involved surgical margins or present with metastatic disease fare considerably worse with 5-year survival rates of 10–20% [6].

The rationale for using mitotane as treatment for ACC stems from early observations of the adrenolytic effect of the pesticide DDT

on adrenal glands of dogs and the finding that patients treated with mitotane showed reduced metastasis and urinary steroid secretion [8,9]. Mitotane is used in an adjuvant setting for patients with resectable tumors due to high tumor-relapse rates [1–3,10]. However, this recommendation is controversial because of various study design issues [10]; it was a nonrandomized, retrospective study spanning 20 years and included patients in multiple centers receiving variable doses of mitotane [10].

A serum therapeutic concentration range of 14–20 mg/l of mitotane is sufficient to achieve positive responses, with an optimum range of 12–14 mg/l [1–3,11]. While monotherapy with mitotane has shown some positive responses in patients treated with ACC (response rates of up to 31% in patients with advanced ACC), the narrow therapeutic window can be difficult to achieve, and severe side effects are generally dose limiting during treatment [1–3,11]. Combination therapy with cytotoxic drugs was initially shown to be more effective than mitotane alone [12]. However, recent results from the FIRM-ACT trial suggest that this may not be the case [4]. FIRM-ACT was the first ever randomized Phase III clinical trial to compare chemotherapy regimens combined with mitotane. Etoposide, doxorubicin and cisplatin–mitotane was evaluated against streptozotocin–mitotane. The etoposide, doxorubicin and cisplatin–mitotane had better responses compared to the streptozotocin–mitotane as a first-line therapy (23.2 and 9.2%, respectively) and had better progression-free survival as a second-line therapy (5.6 and 2.2 months, respectively) [4]. However, despite these differences, overall survival was still bleak and did not improve significantly with treatment [4]. Elucidating the mechanism of mitotane action will be essential for the development of better treatment options for ACC.

Aditi A Bapat

Adrenocortical Carcinoma Research Program, Integrated Cancer Genomics Division, Translational Genomics Research Institute (TGen), Scottsdale, AZ, USA

Michael J Demeure

Adrenocortical Carcinoma Research Program, Integrated Cancer Genomics Division, Translational Genomics Research Institute (TGen), Scottsdale, AZ, USA and Virginia G Piper Cancer Center, Scottsdale HealthCare, Scottsdale, AZ, USA

Kimberly J Bussey

Author for correspondence: Adrenocortical Carcinoma Research Program, Integrated Cancer Genomics Division, Translational Genomics Research Institute (TGen), 13208 E Shea Boulevard, Suite 100, Scottsdale, AZ 85259 USA Tel.: +1 602 343 8817 kbussey@tgen.org

Mitotane's mechanism of action

For mitotane to be therapeutically active, it needs to be transformed into active metabolites by an as yet unidentified CYP450 enzyme. It has been postulated that mitotane first undergoes β -hydroxylation and is subsequently transformed into an acyl-chloride derivative by dehydrochlorination. This acyl chloride is considered to be the active metabolite of mitotane [1,13,14]. The metabolites are believed to contribute to the adrenolytic action of mitotane by destroying the adrenocortical mitochondria, leading to cell death and necrosis. Additionally, oxidative damage caused by reactive oxygen species and superoxide formation during the process of mitotane metabolism have also been speculated to contribute to adrenolysis [1,14]. Mitotane inhibits adrenal steroidogenesis at lower concentrations than required for adrenal cortex destruction [2,3,14]. It also results in increased levels of the cortisol-binding globulin protein, further reducing cortisol levels by binding to free circulating cortisol in the blood, therefore helping to combat adrenal hormone excess seen in ACC patients [13,15]. Thus, glucocorticoid replacement is usually required along with mitotane therapy to counteract the drastic reduction in adrenal hormone levels in ACC patients and to prevent adrenal insufficiency [1,3].

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Interaction with known mediators of drug resistance

Adrenal glands naturally express high levels of P-gp, encoded by the multidrug resistance gene (*ABCB1/MDR-1*). P-gp functions as an ATP-dependent drug exporter with broad affinity, the expression of which can be elevated in ACC. Mitotane inhibits P-gp, blocking the export of compounds such as doxorubicin and etoposide [16]. While this effect was observed *in vitro*, adequate inhibition was not seen in patients treated with doxorubicin, vincristine, etoposide and mitotane in a Phase II clinical trial [16].

Mitotane treatment has been shown to increase the activity of CYP3A4 in patients [13,17]. In flies, the orthologous gene to *CYP3A4*, *Cyp6a2*, mediates resistance to DDT, further supporting the role of this enzyme in mitotane response [18].

CYP3A4 metabolizes several clinically relevant drugs, including doxorubicin and etoposide, and CYP3A4 activity remains elevated long after cessation of mitotane administration due to mitotane's long half-life [13]. This observation has important implications in partly explaining the reduced efficacy of combination therapy with mitotane, as increased activity of CYP3A4 by mitotane may increase the metabolism of the chemotherapeutic agents and thus reduce efficacy [13,17]. This may partially explain the similar overall survival of the two groups in the FIRM-ACT trial, as doxorubicin and etoposide are substrates of CYP3A4, but cisplatin and streptozotocin are not. The investigators emphasized the importance of monitoring of CYP3A4 levels for this reason [4]. Mitotane may also increase the activity of other CYP450 enzymes leading to increased metabolic activation of mitotane and creating a positive feedback loop that leads to continued resistance to cytotoxic chemotherapeutic agents as well as some targeted agents such as erlotinib [13].

“...while mitotane may benefit some patients with adrenocortical carcinoma, the data overwhelmingly support a cautious application of mitotane in a broad context.”

Moving away from a mitotane-based approach to treating ACC

Given the recent results of the FIRM-ACT trial and the discovery that mitotane strongly induces the expression of CYP3A4, a protein that not only rapidly metabolizes mitotane, but several additional therapeutic agents, the use of mitotane as therapy for ACC is controversial especially without a thorough understanding of its mechanism of action. The strongest rationale for its continued use, the suppression of adrenal steroid synthesis, can be accomplished through other compounds such as ketoconazole with more favorable toxicity profiles and fewer potential interactions with potentially useful cancer therapeutics. Thus, while mitotane may benefit some patients with ACC, the data overwhelmingly support a cautious application of mitotane in a broad context. We should identify markers of resistance and predictors of responsiveness so that only those patients with a reasonable hope of responding are exposed to the risks mitotane therapy carries, both in terms of toxicity and impacting additional treatment options if a patient progresses in spite of mitotane treatment. One obvious candidate is assaying for the expression of CYP3A4 in the tumor. This is an

attractive prospect because it not only provides information relative to initial mitotane resistance, but resistance to other therapeutics as well [13].

Moving beyond mitotane will require a more detailed understanding of the pathobiology of ACC. The rarity of ACC suggests that there is a single genomic event that drives the disease, yet identification of this event remains elusive. Whole-genome sequencing of patient tumors could prove to be invaluable in finding this event. Several profiling studies have implicated genes involved in the gap-2/mitosis (G2/M) cell-cycle transition as well as the IGF2, β -catenin, and p53 pathways [19,20]. An understanding of which of these genes is driving tumorigenesis as opposed to promoting it will be essential in successfully applying the knowledge clinically. Expression-profiling studies have also highlighted the heterogeneity of ACC, both in expression profile and in survival, raising the possibility that there may not be a 'one-size-fits-all' treatment regimen for ACC. Successful treatment of ACC will require personalizing therapy based on the molecular characteristics of a given tumor at a given point in time. Technologies such as molecular

expression profiling of patient tumors and/or clinical genomic sequencing can help identify rational treatment alternatives to mitotane and provide a way to monitor tumor evolution as a consequence of therapy [19,20].

In light of recent developments, we need to step away from using mitotane simply because it is the only approved treatment. Several other rare tumor types have no approved first-line therapy. It may be time to put ACC back into that category and start to use the emerging clinical profiling technologies to elucidate a first-line therapy based on the genomics of each individual patient's tumor.

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