Development of mitoxantrone

Richard J. White and Frederick E. Durr
Infectious and Neoplastic Disease Research Section, Medical Research Division, Lederle Laboratories, American Cyanamid Company, Pearl River, NY, U.S.A.

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Summary

Mitoxantrone (Novantrone®; dihydroxyanthracenedione) belongs to a new structural class of antineoplastic agents, the anthracenediones. It was the outcome of a program in synthetic chemistry, at the Medical Research Division of the American Cyanamid Company, which started from a molecule with structural features predicted to favor intercalation with double stranded DNA.

The initial lead compound had immunomodulatory effects and was subsequently found also to possess significant activity against transplantable murine tumors. A large series of analogues was synthesized and mitoxantrone was selected for clinical trial on the basis of its potency and excellent antitumor activity in mice. It is a cytotoxic agent that will kill both proliferating and non-proliferating cells.

A variety of experiments conducted with both intact cells and cell-free systems have revealed mitoxantrone’s ability to bind to single stranded and double stranded RNA and DNA. The drug inhibits cellular RNA and DNA synthesis to about the same extent and causes chromosomal aberrations. In vivo experiments using murine models have demonstrated good activity for mitoxantrone against a variety of transplantable tumors including both leukemias and solid types, in many cases giving putative cures. Surprisingly, it is effective when given up to 30 days before tumor implantation. Combination studies with standard anticancer agents gave evidence of therapeutic synergy in a number of cases.

Preclinical studies in several animal models indicate that mitoxantrone does not have the cumulative cardiotoxic liability associated with anthracycline antibiotics such as doxorubicin.

Rationale leading to synthesis of mitoxantrone

Mitoxantrone (Novantrone®; dihydroxyanthracenedione) is a synthetic anticancer drug belonging to a new class of chemotherapeutic agents called the anthracenediones. It was discovered independently by groups working at American Cyanamid (1) and the Midwest Research Institute (2). The program leading to mitoxantrone at American Cyanamid had its origins in studies on a series of compounds considered to possess certain structural features favoring intercalative binding with DNA: they possessed planar tricyclic chromophores and had basic side chains attached. Such molecules had been shown to possess a wide range of pharmacological effects encompassing antiviral, antibacterial, antiprotozoal, anticancer, and immunomodulating activities. One of the structures investigated was initially found to have immunomodulating activity, and subsequently it was shown to possess modest but reproducible activity.

Address for reprints: Richard J. White, Infectious and Neoplastic Disease Research Section, Medical Research Laboratories, Lederle Laboratories, American Cyanamid Company, Pearl River, NY 10965, U.S.A.
in the P388 murine leukemia model and became the lead compound. It served as the starting point for a synthetic program aimed at developing analogues with increased activity and potency. Over 300 compounds in this series have been synthesized and evaluated against murine P388 leukemia and B16 melanoma. Structures of the initial lead and mitoxantrone, along with their effect on P388 leukemia are given in Fig. 1. Also presented are selected analogues from the synthetic program that illustrate the range of activity and potency achieved. Mitoxantrone was the most potent analogue synthesized and possessed excellent activity against a broad spectrum of animal tumors, furthermore it gave a significant number of putative cures, or long term survivors. On the basis of this potency and activity, mitoxantrone was rapidly progressed to clinical evaluation. The high potency of mitoxantrone is relevant to potential cosmetic side-effects as the aminoanthraquinones are intense blue dyes and their i.v. or i.p. administration in animals can result in a persistent blue coloration of the internal organs and skin. The lower doses of mitoxantrone required in comparison with other members of this series should lessen the potential for this undesirable side-effect in the clinic (2).

**Structure activity relationships**

Analysis of the results obtained for the different analogues in the P388 leukemia and B16 melanoma test systems permits some conclusions to be made concerning the relative importance of specific structural features of the molecule to *in vivo* activity (1, 2).

1. Ring hydroxylation at positions 5 and 8 enhances potency and activity whereas hydroxylation at positions 5 and 6 has little effect.

2. The presence of two basic side chains does not appear to be essential to activity. Certain “one-armed” derivatives, with a hydrogen or amino function replacing the side chain, retain significant activity.

<table>
<thead>
<tr>
<th>Compound</th>
<th>Optimal dose (mg/kg)</th>
<th>Activity P388~30 Days</th>
</tr>
</thead>
<tbody>
<tr>
<td>Initial lead (CL 55,343)</td>
<td>100</td>
<td>68</td>
</tr>
<tr>
<td>(CL 227,791)</td>
<td>50</td>
<td>&gt;228</td>
</tr>
<tr>
<td>Ametantrone (Cl232,129)</td>
<td>25</td>
<td>&gt;200</td>
</tr>
<tr>
<td>Mitoxantrone (Cl232,315)</td>
<td>1.6</td>
<td>&gt;186</td>
</tr>
</tbody>
</table>

* 1,5 and 9 day schedule

Fig. 1. Development of 1,4-bis(alkyl amino) anthraquinones.