Mitomycin C

- A molecule in the complex world of chemotherapeutic agents -

Mitomycin C (MMC) is a cytostatic antibiotic inhibiting cell division and thus tumor growth. That happens as a result of inhibiting the synthesis of the deoxyribonucleic acid (DNA). Also, the cellular ribonucleic acid (RNA) plus the protein synthesis can be suppressed in the presence of a high concentration of MMC.

The preliminary stage of MMC is isolated from a bacterium called Streptomyces caespitosus.

Physical and chemical properties

- CAS-Number: 50-07-7
- Form: Powder
- Color: blue-purple
- Odor: odorless
- pH-value (5g/l) at 20°C: 6-8
- Melting point / range: >360°C
- Boiling point / range: not defined
- Flammability: not flammable
- Solubility in water: soluble

Categorization of MMC as a dangerous substance

- T: toxic
- R25: toxic if swallowed
- Xn: harmful
- R40: limited evidence of a carcinogenic effect

Source: http://www.sigmaaldrich.com/medium/structureimages/09/mfcd00078109.png
Hazard pictograms

GHS06:  
Source: http://www.svfg.de/91-elemente/gefahrenzeichen/sicherheitszeichen-gif-jpg/ghs06.gif

GHS06:  
Source: http://www.svfg.de/91-elemente/gefahrenzeichen/sicherheitszeichen-gif-jpg/ghs08.gif

History of MMC

The drug Mitomycin was first discovered by Hata at the Kistasatio Institute in Japan in 1955. There are 17 different derivates of mitomycin, of which 16 show a biological activity. Most of them have an antibiotic and cytostatic effect.

The structure of mitomycin derivats

MMC was firstly separated by Wakaki at Kyowa Hakko Kogyo (now: Kyowa Hakko Kirin) as purple crystals.

The drug is very stable and is most effective, looking at antineoplastic activity (meaning the strongest effect against malignant tumors), of all derivates.

In Japan, the evaluation of its clinical efficacy started in 1957.

In 1963, the medicament was approved in Japan. Fifteen years later, the drug has been introduced in Germany. By and by, it has been approved all around the world, in more than 80 countries.

Source: http://www.beilstein-journals.org/bjoc/content/inline/1860-5397-5-33-i2.png?max-width=550&background=EEEEEE

1 mitomycin.net am 20.06.13
2 Kishi – Mitomycins A & C 1977, 99, 4835-4836
3 http://www.medac.de/data/downloads/urologie/mitoBasisdokumentation-web.pdf on 29.06.13
Chemical Synthesis

Practical total Synthesis of MMC via Isomitomycin A

The substances Isomitomycin A (2) and Albomitoycin A, which are in equilibrium with the wanted Mitomycin, were isolated from cultures of *Streptomyces caespitosus* in 1955. The equilibrium reaction was named "Mitomycin rearrangements" and includes a Retro-Michael- and a Michael-addition. In order to find a technical representation of Isomitomycin A from the chalcon (5) this synthesis was developed by Tohru Fukuyama and Lihu Yang in 1989. The shown chalcon can be synthesized from 2,6-dimethoxytoluene in thirteen steps, which will not be discussed any further.

![Scheme I. Diagram of the synthesis process](image)


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Step-by-step-synthesis from chalcon (5) to Isomitomycin A

<table>
<thead>
<tr>
<th>Reaction type</th>
<th>Educts</th>
<th>Products</th>
<th>Reactants and conditions</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>a</strong> [3+2] DA, endo</td>
<td>3 + 4</td>
<td>5</td>
<td>SnCl₄ (0.1 equiv), CH₂Cl₂, -78 °C; Py (1 equiv)</td>
</tr>
<tr>
<td><strong>b</strong> Intermolecular azide-olefin cycloaddition</td>
<td>5</td>
<td>6</td>
<td>Toluene, 110 °C 3 h.</td>
</tr>
<tr>
<td><strong>c</strong> Partial reduction</td>
<td>6</td>
<td>-</td>
<td>DIBAL, THF, -78 °C</td>
</tr>
<tr>
<td><strong>d</strong> Subsequent acetylation</td>
<td>-</td>
<td>7</td>
<td>Ac₂O, Py</td>
</tr>
<tr>
<td><strong>e</strong> Ozonolysis &amp; oxidation</td>
<td>7</td>
<td>8</td>
<td>RuO₂ (0.05 equiv), NaIO₄ (5 equiv), EtOAc/H₂O (1 : 1), 23 °C</td>
</tr>
<tr>
<td><strong>f</strong> Reduction</td>
<td>8</td>
<td>9</td>
<td>NaBH₄, MeOH</td>
</tr>
<tr>
<td><strong>g</strong> Ammonolysis</td>
<td>9</td>
<td>10</td>
<td>CCl₃CONCO, CH₂Cl₂, 23 °C</td>
</tr>
<tr>
<td><strong>h</strong> Reduction</td>
<td>10</td>
<td>(11, 12) 13</td>
<td>NH₃, MeOH, 23 °C; NaBH₄</td>
</tr>
<tr>
<td><strong>i</strong> Hydrogenolysis</td>
<td>13</td>
<td>(14) 15</td>
<td>CSA (0.3 equiv), MeOH, 23 °C</td>
</tr>
<tr>
<td><strong>j</strong> Oxidation</td>
<td>-</td>
<td>2</td>
<td>H₂ (1 atm), 10% Pd/C, EtOH</td>
</tr>
<tr>
<td><strong>k</strong> Nucleophil substitution</td>
<td>2</td>
<td>16</td>
<td>NH₃, MeOH, 23 °C, 5 h</td>
</tr>
</tbody>
</table>

Mitomycin Rearrangements in detail

The N-2-atom (green) attacks the C-7a-atom nucleophile, whereat the electron density is shifted to the C-3a-atom. (→ AlboMMC)

In the next step the N-7b-atom (black) splits from the C-7a-atom heterolytically and attacks the hydrogen-atom of the C-3a-atom. The electron density is shifted back to the C-7a-atom, thus the second double bond of the quinone between the C-7a-atom and the C-3a-atom is rebuild. (→ MMC)

The yield from MMC from IsoMMC is 85 %⁶, the overall yield from MMC from 2,6-dimethoxytoluene is 10 %⁷.

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MMC by E. Korotkova, L. Lopez Dolz, J. Menke and L. Noll
Mechanism of action

In general, MMC inhibits the DNA synthesis by an irreversible alkylation of the DNA strands. The pharmacological mechanism of MMC starts with being activated by enzymes, such as the NADPH-cytochrome P450 reductase (CPR), which is generally involved in drug metabolism. After the enzymatic activation to either a hydroquinone or a semiquinone, the heterocyclic nitrogen is transformed from an amido to an amino group. Accordingly, the electron rich amino group promotes the elimination of the cyclic methoxy group. Tautomerization of the iminium ion followed by an elimination of the carbamate group results in an electrophilic center, sensitive to the nucleophilic attack by a DNA base. This monofunctional binding to either adenine or guanine is promptly followed by a second nucleophilic attack on the opposite DNA strand. Both strands are thus alkylated (cross-linked) and cannot be separated by helicases during mitosis. MMC is most effective during the G1- and S-phase in the cell cycle, particularly when the DNA is duplicated.

Pharmacology

MMC is primarily broken down in the liver, but is also metabolized in all tissue cells. The brain cells are one exception, since the drug is unable to pass the blood-brain barrier.

The degree of damage is less severe in resistant cells than in sensitive cells, plus fast proliferating cells are more affected than those in biological dormancy (G0-Phase).

The administration of MMC is either i.v., by an intravesical (locally) or intraperitoneal installation (bladder cancer). The dose depends on the grade of carcinoma. Usually the patient is given 20mg/40mg MMC dissolved in 20ml NaCl either as a one-time treatment, or given several times a
lesser dose over 10 days. As for pharmacokinetics, \( c_{\text{max}} \) (50 ng/ml plasma) is reached after the first 60 min. after administration. An overdose causes additional breaking of DNA due to the formation of peroxide radicals. Approximately 10% are excreted unchanged in the urine via the kidneys.\(^\text{12}\)

**As a chemotherapeutic agent (bladder carcinoma)**

Bladder cancer can be classified into three damage levels (low risk < 3 cm diameter, intermediate risk and high risk), if non-muscle invasive. The follow-up of a post-operative transurethral resection is an initial treatment with a one-time (higher) dose of MMC within 24 h, commonly 40 mg, regardless of the severity of carcinoma. Then MMC is administered through an implanted catheter (intravascular installation).\(^\text{13}\) The resection of an intermediate-risk carcinoma requires up to three-year recurrence prevention, initial- and maintenance therapy. In the case of a high-risk tumor, the patient is additionally given BCG (Bazillus Calmette-Guerin), a vaccine developed from a mycobacterium strain\(^\text{7}\), to recover the immune system. MMC should be administered over a six-week period as an initial treatment, followed by three-times a week applications every three to six months.\(^\text{14}\)

Muscle invasive carcinoma (stage III, picture) implements a cystoscopy in which the men’s urine bladder and prostate, and the women’s uterus and ovaries are being removed. A chemotherapy with MMC comes next.
Generally, chemotherapy with MMC requires frequent medical checks, which consist of monitoring the leucocyte and thrombocyte numbers to prevent bone marrow suppression (reduction in the number of blood cells providing immunity).\textsuperscript{15}

MMC is also used in a combination therapy with hyperthermia.\textsuperscript{16} The thermotherapy is a type of cancer treatment where the affected tissue is exposed to higher temperature (40°C-44°C) via microwave probes to kill cancer cells.\textsuperscript{17} Another combination therapy includes an electro-magnetic field induction (EMDA – “electromotive drug administration”). This type of treatment enhances membrane permeability for MMC in tissues.

\begin{center}
\includegraphics[width=\textwidth]{passive_activation.png}
\end{center}

Simlar anticancer drugs

Comparing Dexorubicin and Epirubicin to MMC, the recurrence rate when administering MMC is lower.\textsuperscript{13}

\textsuperscript{15} http://www.diagnosia.com/at/medikament/mitomycin-c-kyowa-10-mg-trockenstechampullen on 17.06.13
\textsuperscript{16} http://www.medac.de/data/downloads/urologie/mitoBasisdokumentation-web.pdf on 01.06.13
\textsuperscript{17} http://www.cancer.gov/cancertopics/factsheet/Therapy/hyperthermia on 17.06.13
Side effects

Not recommended for patients with\textsuperscript{18}

MMC is not recommended for patients with a decreased amount of blood plates (thrombocytopenia), changes in the clotting, bleeding disorder, problems in the bunch system and sodium-poor diet (9,5 mg Na/mg MMC) because Mitomycin powder contains sodium. Patients on a sodium-poor diet are at a higher risk of a heart stroke.

Administration\textsuperscript{19}

The administration should always be dispensed under the vigilance of trained doctors. The speed of the filling is dependent from age. If the filling is not correctly administrated, it will cause local ulcerations and cellulitis, as to the spilled liquids.

The skin of the one who administrates the product should never be exposed with the powder or the liquid because MMC causes allures on the skin and eyes. Also, the doctor shouldn’t apply creams or other, which would facilitate the absorption of MMC.

Recommendations for patients\textsuperscript{20}

Patients with limited liver function should avoid the use of MMC. The patients` ability to operate crafts is impaired by MMC. The usage of MMC is not recommended to women during pregnancy or breastfeeding due to the lack of information in this field.

Adverse Effects\textsuperscript{21}

The main point of toxicity by MMC is the impairment of the medulla. This causes "thrombocytopenia" (decreasing of the number of blood plates), "leucopenia" (decreasing of the number of leucocytes) and refractory anemia (decreasing of the number of red blood cells).

The toxicity caused by MMC is accumulative, so patients should be controlled after every administration. If the side effects are too severe, the treatment is discontinued.

\textsuperscript{18}http://www.vademecum.es/medicamento-mitomycin-c_prospecto_68816 on 30.06.13
\textsuperscript{19}http://chemocare.com/chemotherapy/drug-info/MitomycinC.aspx on 01.06.13
\textsuperscript{20}http://www.vademecum.es/medicamento-mitomycin-c_prospecto_68816 on 05.06.2013
\textsuperscript{21}http://prospectos.org/prospectos/mitomycinc-40-mg-polvo-para-solucion-inyectable-1-vial/ on 17.06.13
**Secondary effects**

**Dysfunction of the immune system:**

*Frequently:* Fever, pruritus.

**Dysfunction of the skin and the subcutaneous tissue:**

*Frequently:* Necrosis, pain in the injection point.

**Vascular dysfunction:**

*Frequently:* Tromboflebitis.

**Dysfunction of the stomach-bowel system:**

*Very frequently:* Nausea, vomiting.

*Frequently:* Anorexia, canker (mouth ulcer).

**Dysfunction of the urine- and bunch systems:**

*Frequently:* Growth of the nitrogen content from urine.

**Dysfunction of the nervous system:**

*Frequently:* Somnolence feeling.

**Dysfunction of the breathing system:**

*Rarely:* Pneumonia, cough, breathing difficulties (Dyspnea).

**Dysfunction of the blood and lymphatic systems:**

*Very frequently:* refractory anemia, thrombocytopenia, Leucopenia.

*Rarely:* Anemia (Anaemia)

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22 [http://www.vademecum.es/medicamento-mitomycin-c_prospecto_68816 on 05.06.2013](http://www.vademecum.es/medicamento-mitomycin-c_prospecto_68816 on 05.06.2013)
Interaction with other drugs

*Dexorubicin*: Toxicity (cardiac arrhythmia) by some patients.

*Vinblastin*: Contraction of the bronchia (bronchospasm).

*Fluoruracil and Tamoxifen*: hemolytic-uric syndrome (soon and quickly after the administration the number of blood plates decreases, the red blood cells and the bunch function are deteriorated).