The anti-proliferative effect of turmeric and rooibos extracts on human Hodgkin lymphoma cells

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The Anti-proliferative Effects of Turmeric and Rooibos Extracts on Human Hodgkin Lymphoma Cells

Emily Kogel

March 29, 2016
Hodgkin Lymphoma

- Immune system support
- Lymph
  - Lymphocytes + other white blood cells
  - Antibodies
  - Nutrients
Apoptosis
Cancer Treatment: Options

- Surgery

Hodgkin Lymphoma:
- Radiation therapy
- Chemotherapy

http://www.joshgitalis.com/pre-and-post-surgery-nutrition/
http://www.dailymail.co.uk/health/article-1177248/New-scans-reveal-chemotherapy-working.html
Turmeric & Rooibos

• Natural health products - new cancer treatment options?

Turmeric
• Strong anti-inflammatory agent
• Supports immune system
• Activates intrinsic apoptosis

Rooibos
• Inhibitor of x-ray-induced cell transformation
• Anti-oxidant activity
• Free-radical scavengers, like flavonoids, induce apoptosis in cancerous cells

http://www.healingthebody.ca/healing-benefits-of-turmeric/
http://www.medicinehunter.com/sites/default/files/rooibos_main.jpg?1332955889
Methods of Extraction

- 3 solvents
- Whole, complex mixture
Do the **anti-proliferative properties** of turmeric and **rooibos extracts** in cancer cells translate to **Hodgkin lymphoma cells**, and if they do, is the mechanism of action **selective**?

- Assess toxicity and efficacy *in vivo*
- Discover the mode of action
- Determine selective cytotoxicity
Do the anti-proliferative properties of turmeric and rooibos extracts in cancer cells translate to Hodgkin lymphoma cells, and if they do, is the mechanism of action selective?

**OBJECTIVES**

1. Determine if turmeric and rooibos extracts, both individually and in combination, exhibit cytotoxic effects against Hodgkin Lymphoma cells (*WST-1 assay*)

2. Quantify cytotoxicity (*Trypan blue exclusion assay*)

3. Determine permanency of the drugs’ effects (*Revival assay*)
1. Determining cytotoxicity

- WST-1 Assay

Lower absorbance = Lower viability

https://i.ytimg.com/vi/7u7cMZyKG9E/hqdefault.jpg
http://www.clontech.com/IE/Support/Applications/Cell_Biology/ibcGetAttachment.jsp?cItemId=54709
Cytotoxicity of Turmeric Extracts (KM-H2)

Cold Water

Ethanol (Unfiltered)

Hot Water

Ethanol (Filtered)

Absorbance at 450 nm

% of Control

Absorbance at 450 nm

% of Control

Concentration (mg/ml)

Concentration (mg/ml)
Cytotoxicity of Turmeric Extracts (KM-H2) - ROUND 2

**Cold Water**

<table>
<thead>
<tr>
<th>Concentration (mg/mL)</th>
<th>48 Hours</th>
<th>96 Hours</th>
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**Ethanol (Unfiltered)**

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0.1
Cytotoxicity of Rooibos Extracts (KM-H2)

Absorbance at 450 nm

% of Control

Cold Water

Hot Water

Ethanol (Unfiltered)

Ethanol (Filtered)

Concentration (mg/mL)

Absorbance at 450 nm

% of Control

48 Hours

96 Hours

3/31/2016
Cytotoxicity of Rooibos Extracts (KM-H2) – ROUND 2

Cold Water

Hot Water

Ethanol (Unfiltered)
2. Quantify cytotoxicity

- Trypan blue exclusion assay
2. Quantify cytotoxicity:

Effect of ethanolic turmeric extract on the viability of KM-H2 cells
2. Quantify cytotoxicity:

Effect of rooibos extracts on the viability of KM-H2 cells

Cold water rooibos

![Graph showing viability of KM-H2 cells over time with cold water rooibos extracts at different concentrations.]

Hot water rooibos

![Graph showing viability of KM-H2 cells over time with hot water rooibos extracts at different concentrations.]

Legend:
- **Rooibos 0.1**
- **Rooibos 0.2**
- **Rooibos 0.3**
- **VP16 (0.05)**
- **Control**
2. Quantify cytotoxicity:

Effect of extracts in combination on the viability of KM-H2 cells

Cold water combination

Hot water combination

Number of cells/mL

Time (hours)

Turmeric EtOH 0.05
Rooibos 0.1
Turmeric EtOH 0.05 + Rooibos 0.1
VP16 (0.05)
Control
3. Determine permanency of the drugs’ effects

- Revival assay

<table>
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<th>24h</th>
<th>48h</th>
<th>72h</th>
<th>96h</th>
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Growth of KM-H2 cells in various treatment groups:

PRELIMINARY RESULTS

48 hours (with drug)

24-72 hours (without drug)
Conclusions

- Turmeric: ethanolic extracts exhibit anti-proliferative effects on Hodgkin lymphoma cells that are likely long-lasting

- Rooibos: hot water extracts show cytotoxicity against Hodgkin lymphoma cells

- A combinatorial effect exists when low doses of ethanolic turmeric and cold/hot water rooibos extracts are used together

These natural health products show potential to be studied further for the development of more effective chemotherapeutic agents.
Future Work

• Assess selectivity of extracts by studying cytotoxicity in normal lymphocytes

• Evaluate the mode of cell death induction

• Assess toxicity and efficacy alone and in combination in animal models
Dedication

- Dedicated to the memory of Kevin Couvillon, who lost his battle against Leukaemia in 2010.
Acknowledgments

- Dr. Pandey
- Members of Dr. Pandey’s Lab
- Funding
Thank you!