Pilot Studies of Cherry Juice Concentrate for Gout Flare Prophylaxis

Naomi Schlesinger1*, Ruth Rabinowitz2 and Michael Schlesinger3

1Professor of Medicine, Chief, Division of Rheumatology, Department of Medicine, UMDNJ-Robert Wood Johnson Medical School, New Brunswick, NJ, USA
2Lecturer, Hubert H. Humphrey Center for Experimental Medicine and Cancer Research, The Hebrew University-Hadassah Medical School, Jerusalem, Israel
3Professor of Experimental Medicine and Cancer Research, Hubert H. Humphrey Center for Experimental Medicine and Cancer Research, The Hebrew University Hadassah Medical School, Jerusalem, Israel

Abstract

The management of gout involves treating pain and inflammation associated with acute flares and lowering the uric acid pool. A challenge associated with the successful management of gout is an increased risk of acute flares after initiation of urate-lowering therapy (ULT). Prophylactic anti-inflammatory therapy is recommended to prevent flares and foster compliance with urate lowering therapy.

The aim of our studies was to assess whether use of cherry juice concentrate is useful for gout flare prophylaxis.

We report the results of three studies using cherry juice concentrate for gout prophylaxis. The first is a randomized controlled study comparing the use of cherry juice concentrate versus pomegranate juice concentrate for flare prophylaxis. The second is a retrospective study evaluating flare prophylaxis when cherry juice concentrate was taken over a 4 month period or longer. Lastly, a third study evaluating the effect of cherry juice concentrate compared with pomegranate juice concentrate on secretion of interleukins by human monocytes exposed to monosodium urate (MSU) crystals in vitro.

Ingesting cherry juice concentrate reduced the incidence of flares in gout patients regardless of whether or not they were treated with ULT. The number of flares was further reduced by cherry juice ingestion in patients receiving ULT than in patients not on ULT. We did not find a significant change in serum urate levels from baseline following intake of the cherry juice concentrate for 4 months or longer. Thus, cherry juice concentrate was most likely contributing to a reduction in flares via anti-inflammatory actions. We found cherry juice concentrate to inhibit in vitro secretion of IL-1β by up to 60%.

In conclusion, our studies suggest that, consumption of cherry juice concentrate for a period of 4 months or longer, reduces acute gout flares, via anti-inflammatory actions such as inhibition of IL-1β secretion. Large long-term randomized controlled trials are needed to further evaluate the usefulness of cherries and cherry juice concentrate for gout flare prophylaxis.

Keywords: Acute gout; Cherries; Serum urate

Introduction

Gout is the most common inflammatory arthritis in men and older women. The management of gout involves treating pain and inflammation associated with acute flares and lowering the uric acid pool. A challenge associated with the successful management of gout is an increased risk of acute flares after initiation of urate-lowering therapy (ULT). Prophylactic anti-inflammatory therapy is recommended to prevent flares and foster compliance with urate lowering therapy [1]. This can compromise patient adherence to ULT and hence adversely affect the outcome of treatment [2]. Prophylactic low-dose anti-inflammatory therapy is recommended to prevent flares and foster compliance with ULT.

Prophylactic administration of low doses of colchicine and/or nonsteroidal anti-inflammatory drugs (NSAIDs) is recommended when initiating ULT in order to decrease the risk of gout flares [3]. Although, use of low doses of colchicine has been the standard of care for gout prophylaxis, there is limited data regarding the efficacy of colchicine given chronically for gout prophylaxis [4]. The recommendations for colchicine prophylaxis are based on results from placebo-controlled studies that show patients receiving colchicine prophylaxis to experience fewer flares compared with patients receiving placebo [5-6]. NSAIDs, too, are used for gout prophylaxis [7] although they have not been widely studied in clinical trials. However, safety concerns, contraindications, intolerance or lack of efficacy, limit regular use of NSAIDs or colchicine as prophylaxis in many patients with gout.

The connection of gout and hyperuricemia with gluttony, alcohol and obesity dates from ancient times [8]. Decreasing consumption of meat, seafood and alcoholic beverages as well as increasing consumption of dairy products, and tofu may be helpful in reducing gout [8]. It has been suggested in the popular press and to a lesser extent in the scientific literature that consumption of cherries alleviates arthritic pain and gout. In a 1950 paper stating that "observations made by responsible physicians suggest" that in 12 patients with gout, eating half a pound of fresh cherries or canned cherries decreased serum urate and prevented flares of gout [9]. Only three case histories are described in the paper. Not all three ate cherries persistently. In addition, there have been several anecdotal reports of the use of cherries as an effective treatment of gout; however, these reports have not been confirmed in controlled nutritional studies. The mechanism of action of cherries...
is unclear. Anthocyanins extracted from cherries have shown anti-inflammatory properties and inhibited cyclooxygenase activities [10,11] and modulation of tumor necrosis factor (TNF)-secretion.

The aim of our studies was to assess whether use of cherry juice concentrate is useful for gout flare prophylaxis and whether it does so via a change in serum urate (SU) or via anti-inflammatory effects.

Herein, we report the results of three studies using cherry juice concentrate for gout flare prophylaxis. The first is a randomized controlled (RCT) pilot study comparing the use of cherry juice concentrate versus pomegranate juice concentrate for gout flare prophylaxis as reflected by the number of gout flares, percent of patients with gout flares, SU level and serum creatinine when taken over a 4 month period. The second is a retrospective study evaluating flare prophylaxis when cherry juice concentrate was taken over a 4 month period or longer. Lastly, a third study evaluating the anti-inflammatory effect of cherry juice concentrate compared with pomegranate juice concentrate on the secretion of interleukins by human monocytes exposed to monosodium urate (MSU) crystals in vitro.

Clinical Studies

Materials and methods (studies 1 and 2)

The juice concentrates used in all the studies reported here were: cherry juice concentrate prepared from fruit harvested in northern California, and pomegranate juice concentrate, from fruit harvested in Michigan. Cherries as well as the pomegranate juice concentrates contain no added sugar or sweeteners, no preservatives and no fillers. They are 100% natural juices. Each tablespoon of cherry juice concentrate is the equivalent of 45-60 cherries and each tablespoon of pomegranate concentrate is equivalent to the juice from one pomegranate. The juice concentrates were kept refrigerated until they were consumed.

Study 1: Randomized controlled study of cherry juice concentrate versus pomegranate juice concentrate for gout flare prophylaxis:

Eighteen patients with MSU crystal- proven gout were entered into this study. Four patients dropped out of the study; 2 from the cherry group (n=1 death prior to starting the study; n=1 non compliant), and 2 from the pomegranate group (n=1 severe heartburn, n=1 non compliant). Fourteen patients with crystal- proven gout completed an institutional review board (IRB) approved protocol. They were randomized into two groups: group A (n=9) received a tablespoon of cherry juice concentrate twice daily and group B, the control group, received a tablespoon of pomegranate juice concentrate twice daily. Patients continued use of allopurinol, prophylactic colchicine or NSAIDs if they have been taking these drugs prior to initiating the study. The parameters recorded at baseline and at 120 days were the number of gout flares, percent of patients with gout flares, medications, SU level and serum creatinine.

Study 2: Retrospective study evaluating flare prophylaxis when cherry juice concentrate was taken over a 4 month period or longer:

A retrospective chart review was performed of patients seen in our clinic between 7/1/04 and 5/1/09 with a primary or secondary diagnosis of either gouty arthropathy or Gout NOS, diagnosis codes 274.0 or 274.9. The parameters recorded at baseline and after at least 4 months were the number of gout flares, percent of patients with and without gout flares, medications, SU level and serum creatinine concentration.

Statistical analysis

The Student's t-test was used to compare the different measured parameters. The significance of changes within each group was analyzed by Student's paired t-test. All P values were 2-tailed and values of less than 0.05 indicate statistical significance.

Results

Study 1: Randomized controlled trial (RCT) of Cherry juice concentrate versus pomegranate juice concentrate for gout prophylaxis:

Eighteen patients with MSU crystal- proven gout were entered into this study. Four patients dropped out of the study; 2 from the cherry group (n=1 death prior to starting the study; n=1 non compliant), and 2 from the pomegranate group (n=1 severe heartburn, n=1 non compliant). Fourteen patients with crystal- proven gout completed an institutional review board (IRB) approved protocol. They were randomized into two groups: group A (n=9) received a tablespoon of cherry juice concentrate twice daily and the control group, group B (n=5), received a tablespoon of pomegranate juice concentrate twice daily. (Table 1)

Patient demographics included: age: range: 28-75; (mean ± SE= 56.43 ± 4.10); disease duration: range: 3-41; (mean ± SE= 14.43 ± 3.04); Body mass index (BMI): range: 24.4-34.4; mean ± SE= 30.02 ± 0.84;

<table>
<thead>
<tr>
<th>Patient number</th>
<th>No. of flares before study as reported by patient</th>
<th>No. of flares documented during study</th>
<th>ULT/ Colchicine</th>
<th>NSAIDs</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group A (Cherry)</td>
<td>3</td>
<td>1 per month 0 per 4 months</td>
<td>None</td>
<td>Stopped Celebrex</td>
</tr>
<tr>
<td></td>
<td>4</td>
<td>3 per month 0 per 4 months</td>
<td>None</td>
<td>Stopped Celebrex</td>
</tr>
<tr>
<td></td>
<td>5</td>
<td>3 per month 2 per month</td>
<td>Allopurinol 500mg /d, Colchicine 0.6 mg /d</td>
<td>No change</td>
</tr>
<tr>
<td></td>
<td>7</td>
<td>1 per month 1 per 2 months</td>
<td>None</td>
<td>Stopped Celebrex</td>
</tr>
<tr>
<td></td>
<td>8</td>
<td>1-2 per month 3 per 4 months</td>
<td>None</td>
<td>Stopped indomethacin</td>
</tr>
<tr>
<td></td>
<td>10</td>
<td>1-2 per month 0 per 4 months</td>
<td>Allopurinol 100mg /d</td>
<td>No change</td>
</tr>
<tr>
<td></td>
<td>11</td>
<td>1 per year 0 per 4 months</td>
<td>None</td>
<td>No change</td>
</tr>
<tr>
<td></td>
<td>14</td>
<td>0 per 4 years 0 per 4 months</td>
<td>Allopurinol 300mg /d</td>
<td>No change</td>
</tr>
<tr>
<td></td>
<td>16</td>
<td>2 per year 1 per 4 months</td>
<td>None</td>
<td>Stopped indomethacin</td>
</tr>
<tr>
<td>Group B (Pomegranate)</td>
<td>1</td>
<td>4 per month 3 per month</td>
<td>None</td>
<td>No change</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>1 per month 1 per 4 months</td>
<td>Allopurinol 500mg /d, Colchicine 0.6 mg /d</td>
<td>No change</td>
</tr>
<tr>
<td></td>
<td>6</td>
<td>1-3 per year 1 per 4 months</td>
<td>Allopurinol 500mg /d</td>
<td>No change</td>
</tr>
<tr>
<td></td>
<td>9</td>
<td>2 per year 0 per 4 months</td>
<td>Colchicine 0.6 mg /d</td>
<td>No change</td>
</tr>
<tr>
<td></td>
<td>13</td>
<td>1 per month 1 per month</td>
<td>None</td>
<td>No change</td>
</tr>
</tbody>
</table>

1: This patient stopped taking allopurinol.

Table 1: (Study 1) Consumption of cherry juice decreases the number of gout flares and use of NSAIDs.

Table 2: (Study 2) Consumption of cherry juice decreases the number of gout flares.

<table>
<thead>
<tr>
<th>Patient Group</th>
<th>Number of patients</th>
<th>Flare-free after 4 months of cherry juice</th>
<th>Number of flares /year prior to cherry juice</th>
<th>Number of flares /year after cherry juice</th>
<th>Significance of effect of cherry juice</th>
</tr>
</thead>
<tbody>
<tr>
<td>Not on ULT</td>
<td>11</td>
<td>4/11 (36%)</td>
<td>6.82 ± 2.10</td>
<td>2.64 ± 1.11</td>
<td>P=0.0899</td>
</tr>
<tr>
<td>On ULT</td>
<td>13</td>
<td>8/13 (62%)</td>
<td>6.89 ± 1.83</td>
<td>1.39 ± 0.60</td>
<td>P=0.0086</td>
</tr>
<tr>
<td>Total</td>
<td>24</td>
<td>12/24 (50%)</td>
<td>6.85 ± 1.34</td>
<td>2.00 ± 0.60</td>
<td>P=0.0001</td>
</tr>
</tbody>
</table>

Twelve (50%) of the 24 patients consuming cherry juice concentrate chronically were flare-free at 4-6 months. Four of eleven (36%) patients not taking ULT were flare-free at 4-6 months; while 8/13 (62%) of patients taking ULT were flare-free at 4-6 months. However the difference in the proportion of flare-free individuals between patients taking ULT and those not taking ULT was not statistically significant. The average SU among all patients who were flare-free was 7.8 mg/dl.

Laboratory Study

Study 3: The effect of cherry juice concentrate on the secretion of interleukins by human monocytes exposed to monosodium urate crystals in vitro

The aim of this study was to determine whether exposure of human monocytes in vitro to cherry juice concentrate affects their capacity to secrete interleukin when exposed to MSU crystals.

Materials and methods (study 3)

Before assessing the capacity of juice concentrates to inhibit the secretion of interleukins by human monocytes we determined the cytotoxic effect of the juice concentrates on the cells employing the MTT assay. In this test, the staining of the cells by 3-(4, 5-Dimethylthiazol-2-yl)-2, 5-diphenyltetrazolium bromide was determined using a microplate reader. Only dilution of juice concentrates that did not exhibit cytotoxic activity on monocyctic cells were tested for their capacity to inhibit the secretion of interleukins.

The cells tested in vitro were either monocytes derived from the buffy coat of volunteer blood donors obtained from the blood bank or THP-1 cells of a monocyte-like cell line. The THP-1 cell line is a monocyte-like cell line derived from a patient with acute monocytic leukemia. Cherry juice concentrate and pomegranate juice concentrate manufactured by Brownwood Acres were tested for their capacity to inhibit interleukin secretion. MSU monohydrate crystals used for monocyte stimulation were prepared according to the method described by Schiltz et al. [12].

THP-1 cells (1X10⁶ cells suspended in 3 ml culture medium) or adherent monocytes from buffy coat were introduced into 35 mm Petri culture dishes. After 2 hours of incubation at 37o C in a 5% CO₂ humidified atmosphere the supernatant fluid was removed and 1.5 ml MSU crystal suspension containing 100 to 250 µg/ ml was added. Either culture medium or various concentrations of cherry or pomegranate juice were added to the Petri dishes. The cells in the culture dishes were exposed for 24 hours simultaneously to MSU crystals and to dilutions of juice concentrates at 37o C in a 5% CO₂ humidified atmosphere. The supernatant fluid of the cultures was collected and the in vitro effect of juice concentrates on the secretion of cytokines by monocytes exposed to MSU crystals was determined. The level of both IL-1β, TNF-α in the supernatant fluids was assessed using Enzyme-linked immunosorbent assays (ELISA).

The Immunoassays were carried out with Human IL-1β/IL-1F2
and Human TNF-α/TNFSF1A Quantikine kits obtained from R&D Systems, Abington OX143NB, UK. In short, 50 μL assay diluent was added to each well, followed by 200 μL standard, control or cell culture supernatant samples to each well. After 2 hours of incubation at room temperature the supernatant fluid in each well was aspirated, followed by 4 washes. 200 μL of anti-cytokine conjugate was added to each well, incubated for 1 hour at room temperature. Thereafter the wells were washed 4 times, and 200 μL substrate solution was added to each well incubated for 20 minutes, and 50 μL stop solution was added to each well. The optical density of each well was determined using a microplate reader set to 450 nm.

Results

As assessed by the MTT method, pomegranate juice concentrates were more toxic than cherry juice concentrates for human monocytes. The exposure in vitro to MSU crystals elicited secretion of IL-1β and TNF-α by either peripheral human blood monocytes or by cells of the THP-1 monocyte-like line. At a concentration that had no cytotoxic effect on monocytes (dilutions of 1:1600 or higher) cherry juice concentrate inhibited the secretion of IL-1β by up to 60 % (Table 3a). Non-toxic concentrations of pomegranate juice had either a weak inhibitory effect or a stimulatory effect on IL-1β secretion. Exposure of human monocytes to cherry juice concentrate elicited an inhibitory effect on TNF-α secretion, while pomegranate juice had a relatively weak or no effect on TNF-α secretion (Table 3b).

Discussion

Gout flare prophylaxis is an integral part of chronic gout treatment. Safety concerns and contraindications limit chronic NSAIDs or colchicine use as prophylaxis in many patients with gout. Consumption of cherries and cherry products has been reported to be health promoting, particularly in alleviating arthritic pain and gout [9]. We have found cherry juice concentrate to be well tolerated and efficacious for gout flare prophylaxis.

In our prospective pilot RCT, gout patients consuming cherry juice concentrate had a significant decrease in the number of gout flares within 4 months of initiating ingestion of the cherry juice concentrate, an effect not seen in the control group, treated with pomegranate juice. Fifty five % of patients ingesting cherry juice concentrate were flare-free and stopped their NSAIDs intake. In our retrospective study of patients consuming cherry juice concentrate daily for 4 months; a 50% or greater reduction in gout flares was seen in approximately half of the patients (similar percentage to that seen in our RCT). Importantly, 36% of patients not taking ULT were flare-free at 4-6 months of ingesting cherry juice despite an average SU of 8.7 mg/dl. Thus, although SU level in patients not taking ULT remained higher than 6.8mg/dL, the point of saturation of uric acid, cherry juice consumption significantly reduced their gout flares. Thus, ingesting cherry juice concentrate reduced the incidence of flares in gout patients regardless of whether or not they were treated with ULT.

The intake of cherries by healthy individuals was reported to provoke a significant decrease in SU levels over 5 h post dose, whereas other fruits such as strawberries, grapes and kiwifruit, produced no change [13]. Neither, in our pilot RCT nor in our retrospective study did we find a significant change in SU levels from baseline following cherry juice concentrate consumption for 4 months or longer. It is therefore not a change in SU levels that contributed to the reduced gout flares in patients receiving cherry juice concentrate. However, gout patients receiving ULT were much more responsive to ingestion of cherry juice than those not receiving ULT. In our retrospective study, 36 % of the gout patients who were not on ULT versus 62% who were on ULT were flare-free at 4-6 months. Thus, the number of flares was further reduced by cherry juice ingestion in patients receiving ULT than in patients not on ULT. These results indicate that, cherry juice concentrate was most likely contributed to a reduction in flares via its anti-inflammatory actions.

Both sweet and tart cherries are rich in antioxidants, including anthocyanins (responsible for red skin and flesh color), catechins, chlorogenic acid, flavon glycosides and melatonin. Anthocyanins extracted from cherries have shown anti-inflammatory properties, via inhibition of cyclooxygenase (COX) activities [10,11]. Cherries contain natural COX 1 and COX 2 inhibitors. The COX inhibitory activities of anthocyanins from cherries were comparable to those of ibuprofen and naproxen at 10 ΦM concentrations. In addition, anthocyanins extracted from cherries have shown anti-inflammatory properties, via scavenging of the reactive nitric oxide (NO) radical [14]. Anthocyanins 1 and 2 are present in both cherries and raspberries. Anthocyanins and other phenolics also inhibit NO production in activated macrophages.

Table 3a: (Study 3) The capacity of cherry juice concentrate to inhibit the secretion of IL-1β by human monocytes exposed in vitro to MSU crystals.

<table>
<thead>
<tr>
<th></th>
<th>Control, without juice</th>
<th>Cherry juice 1:1600</th>
<th>Cherry juice 1:4000</th>
<th>Pomegranate juice 1:1600</th>
<th>Pomegranate juice 1:4000</th>
</tr>
</thead>
<tbody>
<tr>
<td>IL-1β (concentration (pg/ml))</td>
<td>83.356</td>
<td>30.696</td>
<td>30.205</td>
<td>157.768</td>
<td>229.073</td>
</tr>
<tr>
<td>Inhibition of IL-1β secretion</td>
<td>63.3 %</td>
<td>63.8 %</td>
<td>-89.3 %</td>
<td>-174.8 %</td>
<td></td>
</tr>
</tbody>
</table>

Table 3b: (Study 3) The capacity of cherry juice concentrate to inhibit the secretion of TNF-α by human monocytes exposed in vitro to 250 μg / ml MSU crystals.

<table>
<thead>
<tr>
<th></th>
<th>Control, without juice</th>
<th>Cherry juice 1:4000</th>
<th>Cherry juice 1:8000</th>
<th>Pomegranate juice 1:8000</th>
</tr>
</thead>
<tbody>
<tr>
<td>TNF-α (concentration (pg/ml))</td>
<td>52.673</td>
<td>28.579</td>
<td>33.521</td>
<td>42.170</td>
</tr>
<tr>
<td>Inhibition of TNF-α secretion</td>
<td>45.74 %</td>
<td>36.36%</td>
<td>19.94%</td>
<td></td>
</tr>
</tbody>
</table>
[14] and modulate tumor necrosis factor (TNF-α) secretion [15]. Various flavons and flavonols were found to either inhibit or induce TNF-α production [15].

It has recently become apparent that gouty inflammation is primarily interleukin 1β (IL-1β) driven [16]. MSU crystals stimulate IL-1 release by monocytes and synovial mononuclear cells [17] as well as the cryopyrin (NLRP3) inflammasome an intracellular, multiprotein complex. Cryopyrin regulates activation of the protease caspase-1 which in turn controls the activation of IL-1β [16]. The release of IL1β [16,17] promotes neutrophil influx into the joint and joint inflammation ensues. The central role of IL-1 in the pathogenesis of gout was further demonstrated by studies of IL-1 inhibitors, showing reduction in the risk of gout flares [18].

As reported herein, we found cherry juice concentrate to inhibit the in vitro secretion of IL-1β by up to 60%. In contrast, non-toxic concentrations of pomegranate juice either had a weak inhibitory effect, or a slight stimulatory effect on IL-1β secretion. Exposure of human monocytes to cherry juice concentrate elicited a weak inhibitory effect on TNF-α secretion, while pomegranate juice had either a weak inhibitory effect or no effect.

There are a number of limitations to our studies. First, in our pilot RCT cherry juice was only given for 16 weeks. A study of a more prolonged period of cherry juice prophylaxis is needed. Secondly, our clinical studies were small, but the combination of two clinical studies reported herein, make for a stronger support of the use of cherry juice concentrate for prophylaxis flare gout.

In conclusion, our studies suggest that, consumption of cherry juice concentrate for a period of 4 months or longer, reduces acute gout flares, via anti-inflammatory actions such as inhibition of IL-1β secretion. Large long-term randomized controlled trials are needed to further evaluate the usefulness of cherries and cherry juice concentrate for gout flare prophylaxis.

Acknowledgment

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References