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Quantitative Assessment of Citric Acid in Lemon Juice, Lime Juice, and Commercially-Available Fruit Juice Products

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Abstract

Background and Purpose—Knowledge of the citric acid content of beverages may be useful in nutrition therapy for calcium urolithiasis, especially among patients with hypocitraturia. Citrate is a naturally-occurring inhibitor of urinary crystallization; achieving therapeutic urinary citrate concentration is one clinical target in the medical management of calcium urolithiasis. When provided as fluids, beverages containing citric acid add to the total volume of urine, reducing its saturation of calcium and other crystals, and may enhance urinary citrate excretion. Information on the citric acid content of fruit juices and commercially-available formulations is not widely known. We evaluated the citric acid concentration of various fruit juices.

Materials and Methods—The citric acid content of 21 commercially-available juices and juice concentrates and the juice of three types of fruits was analyzed using ion chromatography.

Results—Lemon juice and lime juice are rich sources of citric acid, containing 1.44 and 1.38 g/oz, respectively. Lemon and lime juice concentrates contain 1.10 and 1.06 g/oz, respectively. The citric acid content of commercially available lemonade and other juice products varies widely, ranging from 0.03 to 0.22 g/oz.

Conclusions—Lemon and lime juice, both from the fresh fruit and from juice concentrates, provide more citric acid per liter than ready-to-consume grapefruit juice, ready-to-consume orange juice, and orange juice squeezed from the fruit. Ready-to-consume lemonade formulations and those requiring mixing with water contain ≤6 times the citric acid, on an ounce-for-ounce basis, of lemon and lime juice.

INTRODUCTION

Citric acid (2-hydroxy-1,2,3-propanetricar-boxylic acid) is a weak tricarboxylic acid that is naturally concentrated in citrus fruits. At physiologic blood pH, and to a lesser extent in urine, it exists mainly as the trivalent anion. Citric acid is frequently used as a food additive to provide acidity and sour taste to foods and beverages. Citrate salts of various metals are used to deliver minerals in biologically-available forms; examples include dietary supplements and medications. Among fruits, citric acid is most concentrated in lemons and limes,[1] comprising

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as much as 8% of the dry fruit weight. A major source of citric acid *in vivo* results from endogenous metabolism in the mitochondria via the production of ATP in the citric acid cycle. Gastrointestinal absorption of citric acid from dietary sources has been associated with a modest increase in urinary citrate excretion.[2–4]

Urinary citrate is a potent, naturally-occurring inhibitor of urinary crystallization. Citrate is freely filtered in the proximal tubule of the kidney. Approximately 10% to 35% of urinary citrate is excreted; the remainder is absorbed in various ways, depending on urine pH and other intra-renal factors. Citrate is the most abundant organic ion found in urine. Hypocitraturia, defined as <320 mg (1.67 mmol) urinary citrate/day,[5] is a major risk factor for calcium urolithiasis. The activity of citrate is thought to be related to its concentration in urine, where it exhibits a dual action, opposing crystal formation by both thermodynamic and kinetic mechanisms. Citrate retards stone formation by inhibiting the calcium oxalate nucleation process and the growth of both calcium oxalate and calcium phosphate stones, largely by its ability to bind with urinary calcium and reduce the free calcium concentration, thereby reducing the supersaturation of urine. Citrate binds to the calcium oxalate crystal surface, inhibiting crystal growth and aggregation.[6] There is also evidence that citrate blocks the adhesion of calcium oxalate monohydrate crystals to renal epithelial cells.[7] Medical interventions to increase urinary citrate are a primary focus in the medical management of urolithiasis.[8]

The amount of diet-derived citrate that may escape *in vivo* conversion to bicarbonate is reportedly minor.[9] Nonetheless, a prior study reported increased urinary citrate after 1 week on 4 ounces lemon juice per day, diluted in 2 L water, in stone formers with hypocitraturia.[2] Two retrospective studies showed an effect in calcium stone formers of lemon juice and/or lemonade consumption on urinary citrate,[3,4] but a recent clinical trial showed no influence of lemonade on urinary citrate.[10]

The U.S. Department of Agriculture Nutrient Database does not provide information on the citric acid content of foods. While the citric acid content of lemon juice has been reported,[2] no published information exists about the citric acid content of commercially-available beverages in the U.S. Knowledge of the citric acid content of fruit juices and commercially-available lemonade and limeade products may be clinically applicable to patients requiring enhancement or maintenance of therapeutic urinary citrate concentrations. Several methods have been used to measure citrate content of foods, including polarographic, enzymatic, and ion-exclusion chromatography.[11–13]

These techniques are subject to interference, may be relatively insensitive, and can be laborious. In the past decade, ion chromatography using suppressed conductivity has become the method of choice for measuring organic acids and other anions in a variety of matrices.[14] We have used this technique to measure citric acid in different fruit juices.

MATERIALS AND METHODS

Materials

All products were obtained from local supermarkets in Madison, Wisconsin and Winston-Salem, North Carolina, and included fresh fruits, ready-to-consume fruit juices, lemon and lime juice concentrates, and crystallized lemonade formulations. The products purchased are listed in Table 1.

Sample preparation

Samples of the ready-to-consume beverages and juice concentrates were taken directly from their packages. Crystallized lemonade formulations were mixed with water according to package directions. From the fresh fruits, juice was extracted manually. Samples were diluted 1/5000 in water for analysis.

Equipment

Ion chromatography analyses were performed on a system consisting of a Dionex ED50 conductivity detector, a Dionex AS11-HC 2 × 250-mm ion exchange column with a guard column at a controlled temperature of 30°C, and a Dionex ASRS-ULTRA 2-mm suppressor (Dionex Corporation, Sunnyvale, CA).

Chromatographic conditions

A gradient of 20 to 35 mM sodium hydroxide over 10 minutes was used; citrate eluted at 7.6 minutes. Peak areas were related to those of a standard curve for quantifying citric acid concentration.

Statistical methods

We compared the citric acid concentrations of groups of juices and juice products with an analysis of variance (ANOVA). Pair-wise comparisons were made using Fisher's protected least significant difference tests. P-values <0.05 were considered as significant. All analyses were performed using SAS statistical software (SAS Institute Inc., Cary, NC).

RESULTS

The citric acid content of various fruit juices and fruit beverages are listed in Table 1. Comparisons of the citric acid content of the juices and beverages, by group, are shown in Figure 1. The juice of lemons and limes squeezed from the fruits contained the most citric acid (48 and 46 g/L, respectively). There was no difference between the juice of lemons and limes for citric acid content ($P = 0.35$). Lemon and lime juice from concentrate were similar for citric acid content (34–39 g/L). Grapefruit juice and orange juice from ready-to-consume, 100% juice formulations contained 25 and 17 g/L, respectively. There was no difference between the regular orange juice formulation and its "light" counterpart. Orange juice squeezed directly from oranges had a lower citric acid content than ready-to-consume orange juice ($P = 0.003$). Of the commercially-available lemonade and limeade formulations, which are generally formulated to contain 15% real juice or less, those that are ready-to-consume contained more citric acid than the powdered mixes that were prepared by mixing with water according to package instructions ($P = 0.03$). There was no difference between the light and sugar-free ready-to-consume lemonades and their regular counterparts ($P = 0.76$) and no difference between the light and sugar-free lemonades and the crystallized, powdered lemonade mixes ($P = 0.21$).

DISCUSSION

The medical management of calcium urolithiasis depends on manipulating the balance of crystal promoters versus inhibitors. Nutrition therapy that is targeted to an individual patient's risk factors is a basic strategy for kidney stone prevention and an appropriate adjunct to pharmacologic therapy. A cornerstone of prevention is achieving appropriate urine dilution by enhancing urine volume. This reduces the supersaturation of urine, a necessary first step in crystal formation.

A second goal in preventing calcium stone formation is to enhance the concentration of crystal inhibitors. Of these, citrate is the most clinically significant, as it may be manipulated by either diet or pharmacologic therapy or a combination thereof. While 320 mg (1.67 mmol) urinary citrate in a 24-hour urine collection is considered the cutoff for the definition of hypocitraturia,

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[5] some clinicians target a 24-hour urinary citrate concentration of ≥600 mg (3.12 mmol), which is closer to the urinary citrate excretion of healthy, non–stone forming individuals.[15]

Hypocitraturia, if severe and/or persistent, usually requires pharmacologic therapy in the form of potassium citrate, which enhances urine pH and also citrate excretion. The identification and promotion of consumption of fluids that add to the crystal inhibitory potential of urine is appealing, not only to promote fluid intake but to enhance urinary citrate excretion. Citric acid is a naturally-occurring organic acid present in multiple fruits and their juices. Data on the citric acid content of fresh fruit juices and commercially-available fruit juice beverages may therefore prove useful in constructing nutrition therapy regimens for calcium stone formers.

Lemon and lime juice, both from the fresh fruit and from juice concentrates, provide more citric acid per liter than ready-to-consume grapefruit juice, ready-to-consume orange juice, and orange juice squeezed from the fruit. These data concur with those previously reported.[2] As lemon and lime juice contain 38 and 35 mg potassium/oz, respectively, about the same as grapefruit juice and about 60% that of orange juice (potassium content obtained from the U.S. Department of Agriculture, Agricultural Research Service, USDA National Nutrient Database for Standard Reference, Release 19, Nutrient Data Laboratory Home Page: http://www.ars.usda.gov/main/site_main.htm?modecode=12354500, accessed 07/02/2007), ingestion of lemon or lime juice on a daily basis could provide dietary alkali that would decrease renal tubular reabsorption of citrate, resulting in enhanced urinary citrate excretion. The distribution of lemon or lime juice in ample water or other fluid, consumed throughout the day, would also add to the volume of fluids ingested, resulting in enhanced urine output[4] and reduced urine supersaturation.

Further research should determine the bioavailability of dietary citric acid from various sources and characterize the response to dietary citric acid in kidney stone formers who are hypocitraturic, as well as those who are normocitraturic. The impact of diet-derived citrate on urinary concentrations among calcium stone formers consuming different diets (e.g., high fruit/vegetable intake *v* low fruit/vegetable intake; high meat intake *v* low meat intake) should be assessed, as dietary patterns are known to influence urinary citrate concentrations.[9]

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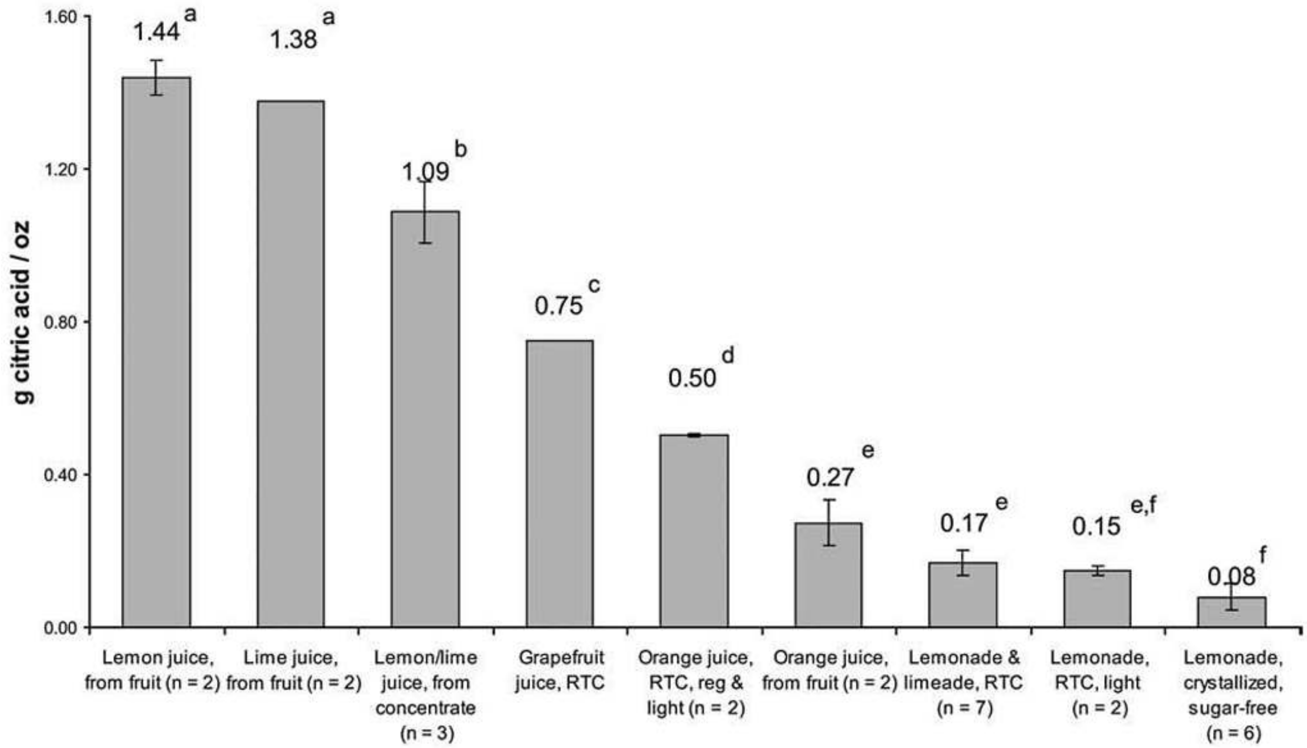

FIG. 1.
Comparison of citric acid concentrations (g/oz.) of juices and juice products by group. Bars, where shown, represent SD for each group. Values above each bar are the mean citric acid content (g/oz.) per group. Groups with same letters are similar ($p > 0.03$). RTC, ready to consume; reg, regular.

Table 1

Citric Acid Content, in Descending Order, of Various Fruit Juices and Commercially-Available Juice Formulations (Grams per Liter)

			Total citric acid	
Product	**Type of product**	***n***	**Mean**	**SD**
Lemon juice	fresh, from fruit	2	48.0	3.82
Lime juice	fresh, from fruit	2	45.8	6.86
Lemon juice, Concord Foods	juice concentrate	1	39.2	
Lime juice, ReaLime 100%	juice concentrate	1	35.4	
Lemon juice, ReaLemon 100%	juice concentrate	1	34.1	
Grapefruit juice, Florida's Ruby Red	ready-to-consume	1	25.0	
Orange juice, Tropicana Pure Premium	ready-to-consume	1	16.9	
Orange juice, Tropicana Light 'n Healthy	ready-to-consume	1	16.7	
Orange juice	fresh, from fruit	2	9.10	1.98
Limeade/limonada, Minute Maid	ready-to-consume	1	7.30	
Lemonade, Newman's Own	ready-to-consume	1	6.70	
Lemonade, Florida's Natural	ready-to-consume	1	6.20	
Lemonade, Minute Maid Light	ready-to-consume	1	5.20	
Raspberry lemonade, Minute Maid	ready-to-consume	1	5.00	
Lemonade, Tropicana	ready-to-consume	4	4.83	0.61
Pink lemonade, Minute Maid	ready-to-consume	1	4.80	
Lemonade, Tropicana Sugar-Free	ready-to-consume	3	4.60	0.44
Lemonade, Minute Maid	ready-to-consume	1	4.40	
Lemonade mix, Crystal Light	drink mix	2	4.20	0.71
Pink lemonade mix, Crystal Light	drink mix	2	3.40	
Raspberry lemonade mix, Crystal Light	drink mix	1	3.10	
Lemonade mix, Kool-Aid Sugarfree	drink mix	1	2.10	
Lemonade mix, Country Time	drink mix	1	1.60	
Crystallized lemon, True Lemon	dry mix	1	0.92	