Journal of Food and Drug Analysis

Volume 8 | Issue 3 Article 2

2000

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Journal of Food and Drug Analysis, Vol. 8, No. 3, 2000, Pages 180-186 藥物食品分析 第八卷 第三期

A Gas Chromatographic Method for Determination of Nicotinamide, Paraben Esters and Caffeine in Commercial Health Drinks, Tonic Drinks and Cold Formulas

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(Received: December 19, 1999; Accepted: March 9, 2000)

ABSTRACT

A simple and rapid method was developed to simultaneously determine the nicotinamide, paraben as well as caffeine by using megapore immediate-polar column (CP-Sil 24CB, 30 m × 0.53 mm, 1.5 μm) with direct injection gas chromatography. Direct quantitative analysis of nicotinamide, paraben esters and caffeine in health drinks, tonic drinks, and cold formulas was carried out without any sample pretreatment procedure. The water soluble compound 1,9-nonanediol was used as an internal standard. The detection limit for nicotinamide, paraben esters and caffeine was 2 μg/mL. Recovery studies were performed using 1 mL of vitamin B drink, tonic drink and cold solution, each spiked with nicotinamide, paraben esters and caffeine at 100.0 μg, respectively. The recoveries for nicotinamide were 96, 102 and 105%, respectively, and the coefficient of variation was less than 7.2%. The recoveries for butylparaben were 105, 95, and 107%, respectively, and the coefficient of variation was less than 8.3%. The recoveries for caffeine were 101, 95 and 104%, respectively, and the coefficient of variation was less than 7.5%. Fifty-two commercial health drinks, tonic drinks and cold solutions were analyzed by the current method. The nicotinamide content was found as: 20.3-241.7, 0-210.7 and 0-246.8 μg/mL, respectively; the isobutylparaben content was found as: 0-54.1, 0-42.9 and 0-134.1 mg/mL; the butylparaben content was found as: 0-106.8, 0-120.1 and 0-184.6 μg/mL; and the caffeine content was found as: 0-433.2, 0-80.7 and 472.3-1742.9 μg/mL. These results indicated that 6 out of 18 commercial health drinks, 2 out of 12 commercial tonic drinks, and 2 out of 22 commercial cold formulas exceeded the Taiwan nicotinamide recommended daily nutrient allowances (RDNA, 14.4 μg) level. Furthermore, 25 out of 52 samples labeled "preservative-free" were detected to be preservative-containing products. The total preservative contents in some test samples exceeded the regulation levels of 100 μg/mL; and 27 out of 30 samples were detected to be caffeine-containing products. These results were inconsistent with the CNS (Chinese National Standard) rules.

Key words: health drink, tonic drink, cold solution, nicotinamide, paraben, caffeine, gas chromatography, quantitative analyses

INTRODUCTION

Tonic drinks are classified under the regulation of non-prescription drugs in Taiwan. By the definition of the Department of Health, a tonic drink contains one adult dose of oral suspension filled in a glass bottle or ampoule, and which the patient can take as intake one dose in one bottle. The regulations are as follows: (1) one unit of the filling volume is limited to lower than 100 mL; (2) the commercial name of the product should be XX tonic drink; (3) the ingredients of this drink should be confined to nutritive and healthy components; (4) the products with the functions of hormones (including steroids), GI-medications, antiscolics, anti-vertigo agents, central nervous system-stimulants, antipyretics, analgesics, antitussives, and anti-expectorants are not allowed to be registered using the name "tonic drink"; and (5) caffeine and its derivatives are not allowed to be added to these products. If the ingredients are not classified in the medicine category or the dosage of the ingredients are within the regulation of food additives, the products are classified as soft drinks (in the food category) and can be retailed in regular stores[(1)].

Vitamin B, amino acids, and hepatic function enhancers are the major components in tonic drinks, which are said to be able to relieve fatigue, promote alertness, restore energy, and protect the liver[(1)]. Owing to the increase in popularity of health drinks and the diversity of tonic drinks, the borderline between both drinks has become ambiguous and difficult to define. There may be a potential health hazard to consumers as a consequence of misuse of these two products.

An excessive or inappropriate use of additives (including preservatives, nutrient supplements, antioxidants, and sweeteners) in foods can damage the health of consumer[(2)]. Therefore, the types of additives and contents used in each food item are always the most important parts for investigation. For instance, nicotinamide, a nutritive supplement, is widely added into commercially available tonic and vitamin B drinks[(2)]. However, an overdose of nicotinic acid and nicotinamide can result in hyperemia symptoms, such as flushed or itchy skin, headaches, peptic ulcers, or even liver damage[(3)]. Nicotinic acid and nicotinamide were thus banned

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in meat products by the Japanese government in 1982. As consumption of tonic and vitamin B drinks (both contain nicotinamide) has recently become increasing popular in Taiwan[1], the development of a precise and accurate analytical method for free nicotinic acid and nicotinamide is necessary. Caffeine, a widespread alkaloid in plant, is a stimulant to the central nervous system and is commonly used as an ingredient in soft drinks as well as in medicine. Lately, numerous reports have indicated that intake of caffeine in an overdose is associated with many clinic diseases such as coronary heart disease, myocardial infarction, cancers (urinary tract, kidney, and pancreas), anxiety, and fibrocystic breast disease[5,6]. In addition, food preservatives are added to tonic and health drinks to inhibit outgrowth of microorganisms. The most commonly used food preservatives in these drinks are paraben esters.

Advertisements for tonic and health drinks always proclaim their ability to relieve fatigue, restore energy, and promote alertness[1]. Caffeine is a component to stimulate alertness in regular soft drinks. Based on the regulation of Department of Health, the concentration of caffeine in a certain soft drink can not exceed 200 ppm except tea, coffee, and cocoa; the label of "containing caffeine" is required if the drink is not caffeine-free. Furthermore, caffeine can not be use as an ingredient in tonic drinks. Because there is no labeling of the presence of caffeine on the commercial tonic and health drinks in Taiwan, we were interested in examining the contents of caffeine in these products. Also, the contents of nicotinamide and preservatives in these products were investigated.

The current analytical methods for nicotinamide[7-13], preservatives[14-20], and caffeine[21-26] include: colorimetric, thin-layer chromatography (TLC), high performance liquid chromatography (HPLC), and gas chromatography (GC). Of the above methods, most reagents used in the colorimetric method possess high toxicity, instability, and low accuracy in analysis. The TLC method always results in low accuracy in analysis and quantitation. The complexity of purification steps of the HPLC method may reduce the precision of analysis. The traditional GC method uses a large amount of organic solvents to extract the target compounds from a sample for analysis and produces a large amount of solvent waste. In addition, derivertization and other purification steps are tedious and may reduce the precision of quantitation.

The GC method is one of the most important modern analytical techniques because of its advantages of high resolution and sensitivity. Owing to years of research using GC, we found that insertion of a ball of glass wool into a liner of the GC injector or application of a guard column (ca. 1-2 cm) could effectively prevent the nonvolatile compounds from getting into the analytical column and, thus, moderate the interference from the contaminants. Also, the tailing effect of each peak could be reduced. We found that the commercially available megapore capillary GC column was superior in water resistance. Even though the samples were in aqueous solution, direct injection of the samples into GC with this column resulted in a comparable repeatability of resolution and retention time to a new column. It was not necessary to frequently clean the retained salts and other impurities in the liner. Under normal circumstances, the liner could be used to analyze more than 100 samples before cleaning. The cleaning procedure is quite simple: The liner should be removed from GC and soaked in hydrochloric acid solution for 10 minutes. The old glass wool is removed from the liner and then washed in water. A ball of new glass wool is inserted into the dried liner, and then placed into the injector for use.

Due to the above advantages, we chose to analyze aqueous samples by direct injection into GC without further treatments, but only spiking an internal standard into the samples before analysis. The purpose of this study was to establish a simple, fast, and accurate method through the use of an adequate GC column and suitable analytical conditions to quantify nicotinamide, paraben esters, and caffeine in tonic drinks, cold formulas and health drinks.

MATERIALS AND METHODS

I. *Materials*

A total of 52 commercial samples, including 18 vitamin B- and amino acid-health drinks (HD), 12 hepatic function protecting-, alcohol detoxifying-, and fatigue reliving-tonic drinks (TD), and 22 cold formulas (CF), were collected from supermarkets and drug stores in the Tainan and Pingtung areas of Taiwan. Nicotinamide, 1,5-pentandiol, 2-methyl-n-pentanoic acid, 2-methyl-1,5-pentanediol, 1,6-hexanediol, 1,4-dihydroxybenzene, paraben eaters, including ethyl paraben, (Et-P), isopropyl paraben, (IPr-P), propyl paraben, (Pr-P), isobutyl paraben, (IBu-P), butyl paraben (Bu-P), caffeine, and 1,9-nonanediol standards (purity>98%) were obtained from TCI Co. (Tokyo, Japan).

II. *Methods*

(I) *Preparation of Standard Compounds and Internal Standard Solutions*

An equal amount (0.2 g) of nicotinamide, Et-P, IPr-P, Pr-P, IBu-P, Bu-P, caffeine, and 1,9-nonanediol (internal standard, IS) was placed individually in a volumetric flask (100 mL) and dissolved in methanol. The volume was then brought to 100 mL with methanol.

(II) *Determination of Relative Response Factors (RRF) of Nicotinamide, Paraben Esters, and Caffeine to 1,9-Nonanediol*

An equal concentration (0.2%, g/mL) of nicotinamide, paraben eaters (Et-P, IPr-P, Pr-P, IBu-P, and Bu-P), or caffeine and IS (0.2%, g/mL) in methanol were mixed together in different ratios (nicotinamide, paraben eaters, or caffeine/IS = 2/1, 1/1, and 1/2). RRF was calculated as the following formula:

$$RRF = (A_S/W_S)/(A_{IS}/W_{IS}) \quad (1)$$

RRF: relative response factors of nicotinamide, paraben eaters, or caffeine to IS.

A_S : GC peak areas of nicotinamide, paraben eaters, or caffeine.

A_{IS} : GC peak area of IS.

W_S : weight (mg) of nicotinamide, paraben eaters, or caffeine.

W_{IS} : weight (mg) of IS.

(III) *Quantitation of Nicotinamide, Paraben Esters, and Caffeine*

One mL of each health drink, tonic drink, and cold formula was placed separately in a 7-mL sample vial, in which 50 μL of 0.2% (g/mL) IS were added (IS: 100 μg). The sample was properly mixed and 0.1 μL was injected into GC for analysis. The amount of nicotinamide, paraben esters, or caffeine in each sample was calculated as the following formula:

$$\text{Nicotinamide, paraben esters, or caffeine } (\mu g/\ mL) = (A_S/A_{IS})(W_{IS}/RRF)/V \quad (2)$$

V: volume (mL) of each sample.

(IV) *Determination of Detection Limits*

Standard solutions (2.0 mg/mL) of nicotinamide, paraben esters, and caffeine were diluted separately with water to concentrations of 40, 20, 10, 5, 2, and 1 μg/mL. One mL of each mixed separately with 50 μL of 0.2% (g/mL) IS. Each concentration of standard solutions was injected into GC in triplicate to determine the detection limit of each standard compound.

V. *Fortification Recovery Test*

Addition of 50 μL of 0.2% (g/mL) standard solutions of nicotinamide, paraben esters, and caffeine were added separately to 1 mL of vitamin B drink, tonic drink, and cold formula (in a 7-mL vial). The blank sample was prepared without addition of the above standard solution. The mixtures were spiked with 50 μL of 0.2% (g/mL) internal standard solution, after mixing, 0.1 μL of each mixture was injected into GC for analysis. The recovery of each standard solution was measured in triplicate.

VI. *GC Conditions*

A GC (GL Science 390B, Tokyo, Japan) equipped with a flame ionization detector (FID) with H_2 flow rate at 30 mL/min and air flow rate at 300 mL/min for this study. The flow rate of carrier gas (N_2) was set at 5 mL/min. The temperatures of injection port and detector were set at 280˚C and 300˚C, respectively. The oven temperature was programmed to initiate at 100˚C and hold for 2 min. The temperature was raised to 180˚C at a rate of 8˚C/min, increased to 250˚C at a rate of 10˚C/min, and finally increased to 290˚C at a rate of 40˚C and hold for 3 min. The injection volume was 0.1 μL with direct injection mode.

RESULTS AND DISCUSSION

I. *Study of GC Conditions*

Without a preparation procedure, the commercial health drink, amino acid drink, and vitamin B drink samples spiked with an internal standard were separately injected into GC for analysis. The objective of this study was to establish a precise, accurate, and fast method to analyze nicotinamide, paraben eaters, and caffeine at the same time with the assistance of a suitable GC column and appropriate analytical conditions.

Wang and Lee[26] quantified caffeine in tea, coffee, and Coca-Cola by the direct injection method with a non-polar DB-1 column (5 m × 0.53 mm, 1.0 μm). However, this DB-1 column was not able to quantify nicotinamide, paraben esters, and caffeine in a same GC run. Therefore, in order to separate nicotinamide, paraben esters, and caffeine, a 30 m column was necessary to increase the resolution. Our current study used a more stable and durable mid-polar CP-Sil 24 CB column (30 m x 0.53 mm, 1.5 μm) to analyze the above compounds. Without a preparation procedure, the standard compounds and the samples of health drink, tonic drink, and cold formula were analyzed by direct injection and with the GC conditions as described in Materials and Methods. The retention times of nicotinamide, Et-P, IPr-P, Pr-P, IBu-P, Bu-P, and caffeine were 12.31, 13.16, 13.34, 14.39, 14.92, 15.58, and 18.32 min. The GC chromatograms for standard compounds, a tonic drink, and a cold formula are shown in Figures 1 and 2. Each analysis took only 20 min.

Selection of the internal standard was conducted by adding a small amount of water soluble standards (1,5-pentanediol, 2-methyl-n-pentanoic acid, 2-methyl-1,5-pentanediol, 1,6-hexandiol, 1,4-dihydroxybenzne, and 1,9-nonanediol) into health drinks, tonic drinks, and cold formulas, respectively, and analyzed under the above GC conditions. The retention times of these six internal standards were 6.36, 6.07, 6.40, 6.71, 9.71, and 11.02 min, respectively. The 1,9-nonanediol peak did not overlap with other ingredient peaks of each drink, thus, 1,9-nonanediol was chosen as the internal standard for this study.

II. *Determination of Relative Response Factor (RRF) of Nicotinamide, Paraben Esters, and Caffeine to 1,9-Nonanediol*

Since 1,9-nonanediol was selected as the internal standard, determination of RRF of nicotinamide, paraben esters, or caffeine to 1,9-nonanediol was necessary to precisely quantify nicotinamide, paraben esters, and caffeine in samples. With RRF, the content of nicotinamide, paraben esters,

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and caffeine in different samples could be calculated using formula (1). The standard curves were obtained by plotting the peak area ratios (Y-axis) of nicotinamide, paraben esters, and caffeine to the internal standard versus the concentration ratios (X-axis) of nicotinamide, paraben esters, and caffeine to the internal standard. The R^2 of each standard curve was more than 0.99. The RRF of nicotinamide, Et-P, IPr-P, Pr-P, IBu-P, Bu-P, and caffeine to 1,9-nonanediol were 0.62, 0.64, 0.69, 0.70, 0.61, 0.60, and 0.37, respectively. The structural differences between 1,9-nonanediol and the analyzed aromatic compounds might result in the wide-ranging RRF (0.37-0.70). However, to accurately quantify these compounds were achievable with the assist of correction curves.

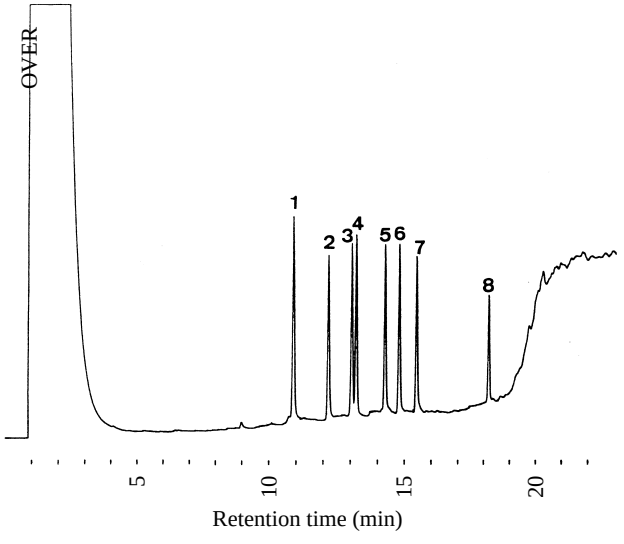

Figure 1. Gas chromatograms of nicotinamide, ethyl-, isopropyl-, propyl-, isobutyl-, butyl-paraben and caffeine authentic standard by direct injection.
GC method. Peaks : 1 = 1,9-nonanediol (IS), 2 = nicotinamide, 3 = ethyl paraben, 4 = isopropyl paraben, 5 = propyl paraben, 6 = isobutyl paraben, 7 = butyl paraben, 8 = caffeine.

III. *The Detection Limits of Nicotinamide, Paraben Esters, and Caffeine on GC-FID*

The standard solutions of nicotinamide, paraben esters, and caffeine were diluted to a series of concentrations and were directly injected into the GC for analysis. The FID detector was set at range: 1 and attenuation: 2. The detection limits of nicotinamide, paraben esters, and caffeine were determined under the GC condition described above, and were approximately 2 μg/mL for all compounds.

IV. *Fortification Recovery Test*

The recoveries of nicotinamide, paraben esters, and caffeine in a vitamin B drink (HD-1), tonic drink (TD-1), and cold formula (CF-1) are shown in Table 1. The results showed that addition of 211.0 and 105.0 μg of nicotinamide to 1 mL of a vitamin B drink, tonic drink, and cold formula produced recoveries of 96, 102, and 105%, respectively, with coefficient of variations below 7.2%; addition of 103.5 μg of paraben esters to 1 mL of a vitamin B drink, tonic drink, and cold formula generated recoveries of 105, 95, and 107%, with coefficient of variations below 8.3%; addition of 100.5 μg caffeine to 1 mL of the same drinks and cold formula, resulted in recoveries of 101, 95, and 104, respectively, with coefficient of variations below 7.5%. As shown in the above results, the direct injection method is a simple, quick, and accurate method.

For analysis of nicotinamide, comparison of the direct injection method[30] developed by our laboratory and the AOAC method (colorimetric method)[9] showed no significant differences. The direct injection GC method was not only reliable, but also simpler and faster as compared to the AOAC method. Cyanogen bromide, a reagent used in the colorimetric method, is highly toxic and under strict regulation in Taiwan. Purchasing this chemical is very difficult since it is banned from importation into Taiwan. In addition,

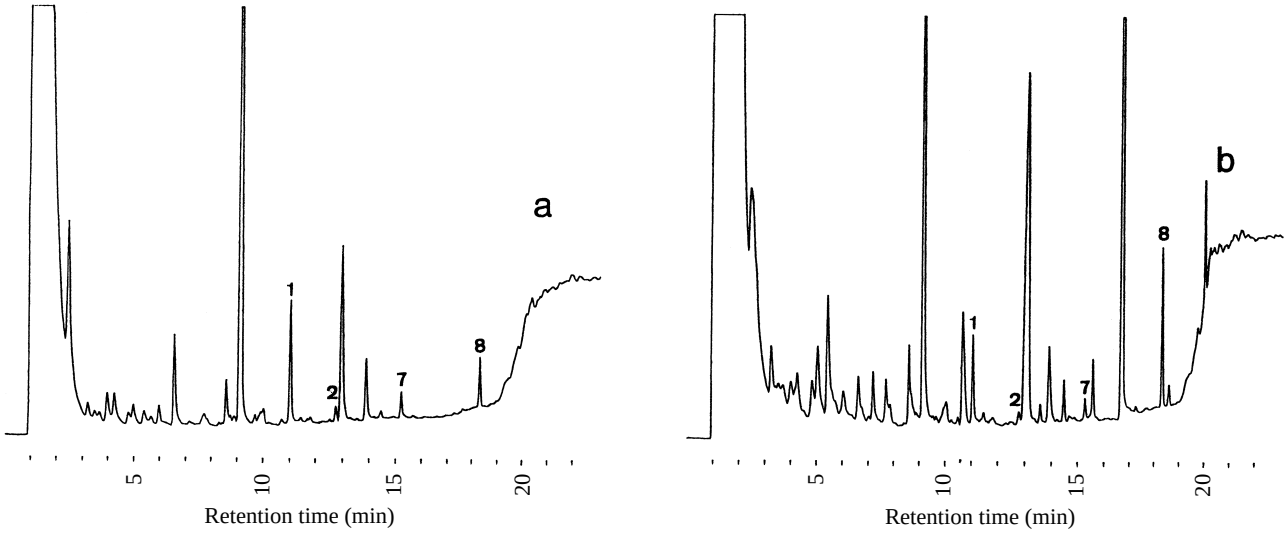

Figure 2. Gas chromatograms of commercial (a) tonic drink and (b) cold formula by direct injection GC method. Peaks : 1 = 1,9-nonanediol (IS), 2 = nicotinamide, 7 = butyl paraben, 8 = caffeine.

Table 1. Recoveries of the nicotinamide, butylparaben and caffeine in spiked commercial vitamin drinks, tonic drinks and cold formulas by the direct injection gas chromatographic method

Sample	Compound spiked	Blank[a] (μg)(A)	Amount Added (μg) (B)	Amount found [b] (μg) (C)	Recovery (%)[c]	CV (%)[d]
HD-1	Nicotinamide	120.7	105.0	217.3	96.3	5.2
TD-1		82.4	105.0	191.2	102.0	6.8
CF-1		12.4	105.0	123.1	104.8	7.2
HD-1	Butyl paraben	22.1	103.5	131.4	104.6	2.7
TD-1		0.0	103.5	98.5	95.2	5.4
CF-1		0.0	103.5	111.2	107.4	8.3
HD-1	Caffeine	298.5	100.5	403.5	101.1	3.9
TD-1		72.4	100.5	163.7	94.7	5.7
CF-1		681.2	100.5	811.8	103.9	7.5

[a] Nicotinamide, butyl paraben and caffeine in 1 mL vitamine drink, tonic drink and cold formula.
[b] Average of triplicate analyses. [c] Recovery (%) = (C − A)/ B × 100%. [d] Coefficient of variation (cv %).

Table 2. Nicotinamide, paraben and caffeine content of various commercial health and tonic drinks

Sample[a]	Nicotinamide	Ibu-P	Bu-P	Caffeine
		μg/mL[b]		
HD1	123.4	ND	21.4	300.2
HD2	241.7	ND	ND	433.2
HD3	72.4	54.1	ND	218.1
HD4	56.3	ND	ND	50.6
HD5	105.8	ND	ND	138.4
HD6	87.4	23.7	ND	176.7
HD7	64.6	ND	30.7	95.2
HD8	103.7	ND	ND	89.4
HD9	49.2	ND	7.7	101.9
HD10	22.1	ND	21.3	34.7
HD11	56.8	ND	42.1	129.5
HD12	21.3	ND	106.8	ND
HD13	18.2	ND	54.5	53.5
HD14	20.3	ND	ND	188.7
HD15	ND	ND	ND	64.6
HD16	11.8	40.5	ND	ND
HD17	10.3	32.7	ND	24.6
HD18	14.7	42.9	36.8	16.8
TD1	81.3	ND	ND	71.3
TD2	102.7	ND	ND	77.5
TD3	34.6	ND	120.1	ND
TD4	137.4	ND	25.9	59.4
TD5	201.9	ND	78.8	58.3
TD6	trace[c]	ND	ND	64.8
TD7	31.5	ND	ND	30.4
TD8	Trace[c]	ND	43.6	51.9
TD9	80.4	ND	ND	80.7
TD10	210.7	ND	ND	21.4
TD11	34.8	ND	35.7	26.4
TD12	52.9	ND	23.9	67.1

[a] HD = health drink, TD = tonic drink.
[b] Average of duplicate analyses. ND = not detected. [c] trace < 2 μg/mL.

Table 3. Nicotinamide, paraben and caffeine content of various commercial cold formulas

Sample[a]	Nicotinamide	IBu-P	Bu-P	Caffeine
		μg/mL[b]		
CF–1	11.1	ND	ND	674.3
CF–2	ND	ND	ND	515.1
CF–3	ND	ND	44.5	942.6
CF–4	15.9	46.3	ND	1078.5
CF–5	5.3	29.1	ND	635.6
CF–6	10.2	ND	ND	528.6
CF–7	10.5	46.8	ND	950.8
CF–8	12.3	ND	ND	1253.3
CF–9	16.6	ND	ND	710.2
CF–10	14.4	37.5	ND	472.3
CF–11	23.9	ND	ND	1630.5
CF–12	242.7	ND	ND	489.4
CF–13	27.5	86.0	ND	1144.6
CF–14	7.3	134.1	ND	1085.9
CF–15	11.7	ND	14.5	1008.2
CF–16	246.8	ND	184.6	1637.1
CF–17	13.9	ND	ND	1742.9
CF–18	11.2	ND	ND	1443.3
CF–19	ND	32.1	ND	1341.4
CF–20	13.6	ND	ND	1458.8
CF–21	16.3	ND	ND	953.3
CF–22	11.2	ND	ND	656.6

[a] CF = cold formula.
[b] Average of duplicate analyses. ND = not detected.

it is hazardous to cope with cyanide bromide, and the waste produced may create a handling problem. Therefore, the direct injection GC method would be a good alternative for nicotinamide analysis.

In analysis by the AOAC method, paraben esters were brought out by steam distillation from a food sample under an acidic condition. The distillate was brought to basic condition and extracted with ether for paraben esters, and the ether extract was then analyzed using a GC. The results obtained from both our direct injection method and the AOAC method were quite agreeable. However, the former method took only 25 min for each analysis and was much easier and faster than the later method, which needed 120 min for each analysis.

Without preparation steps, this direct injection method was capable of quantifying nicotinamide, paraben esters, and caffeine in one analysis, and needed only 20 min for each run. Thus, the procedure of this method was easy and rapid.

V. *Contents of Nicotinamide, Paraben Esters, and Caffeine in Commercial Health Drink, Tonic Drink, and Cold Formula*

All 52 samples, including amino acid drinks, vitamin B drinks, tonic drinks, and cold formulas, were injected separately into a GC to quantify the contents of nicotinamide, paraben esters, and caffeine. As shown in the Tables 1 and 2,

nicotinamide ranged from 20.3-241.7, 0-210.7, and 0-246.8 μg/mL in various health drinks, tonic drinks, and cold formulas, respectively; paraben esters ranged from 0-54.1, 0-42.9, and 0-184.6 μg/ mL; caffeine ranged from 0-433.2, 0-80.7, and 472.3-1742.9 μg/mL, respectively.

For a bottle of health drink (including amino acid drink and vitamin B drink), the volumes were between 160-300 mL, and the contents of nicotinamide were between 6.1-72.5 mg/bottle. The contents in 6 among 18 commercial samples exceeded the Recommended Daily Nutrient Allowances (RDNA) for nicotinamide (14.4 mg)[3]. For various tonic drinks, the volumes ranged from 30-100 mL/bottle, and the amounts of nicotinamide were from 0-21.1 mg/bottle. The quantities in 2 out of 12 samples were higher than RDNA value. For cold formulas (30-60 mL/bottle), the contents of nicotinamide ranged from 0-14.8 mg/bottle, and 2 among 22 samples surpassed RDNA value.

Most commercial health drinks, tonic drinks, and cold formulas contain IBu-P or Bu-P, but a great portion of them are not labeled "contains preservatives". According to the Law for the Control of Food Sanitation, the content of 0.1 g/L of paraben esters is acceptable in non-carbonate drinks. Samples tested in this study violated the regulation, and exceeded the CNS including 1 among 18 health drinks, 1 among 12 tonic drinks, and 2 among 22 cold formulas.

Regarding caffeine content, apart from cold formulas, both health drinks and tonic drinks were not labeled "contains caffeine," but were detected as positive. Based on the CNS, the levels of caffeine in soft drinks should not exceed 200 μg/mL, and all drinks containing caffeine should be labeled except tea, coffee, and cocoa. The CNS for tonic drinks indicates that the addition of caffeine in tonic drinks is not allowed. Most commercial health and tonic drinks do not meet the CNS; among them, caffeine contents in 3 out of 18 health drinks were higher than 200 μg/mL. As shown in Table 3, the caffeine contents in different cold formulas were excessively high; one-half of 22 samples exceeded 1000 μg/mL. Consump-tion of a bottle (60 mL) of cold solution could include an intake of more than 60 mg of caffeine. Therefore, those heavily dependent on cold formulas must be cautious.

ACKNOWLEDGEMENT

We thank Dr. C. Y. Tai for his translation work.

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市售保健飲料、口服液及感冒藥液中菸鹼醯胺、對羥苯甲酸酯類及咖啡因之氣相層析分析

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摘　要

本研究建立了直接注入氣相層析分析市售保健飲料、口服液及感冒藥液等液體樣品中菸鹼醯胺、對羥苯甲酸酯類及咖啡因之快速、簡便之同步測定方法。採用直接注入之方式，以中間極性之CP-Si1 24 CB管柱（30 m × 0.53 mm,1.5 mm）同步分析定量上述液體樣品之菸鹼醯胺、對羥苯甲酸酯類及咖啡因，選擇水溶性之1,9-壬二醇（1,9-nonanedio1）爲內標準，三者之最低檢出濃度約爲2 μg/mL左右。分別添加100 μg之菸鹼醯胺、對羥苯甲酸丁酯及咖啡因於1mL維生素B飲料、口服液及感冒藥液檢體中，直接取樣注入GC分析，其回收率分別爲：菸鹼醯胺，96、102及105%，變異係數在7.2%以下；對羥苯甲酸丁酯，105、95及107%，變異係數在8.3%以下；及咖啡因，101、95及104%，變異係數在7.5%以下。以本方法分析市售不同廠牌之保健飲料、口服液及感冒藥液等液體樣品共52件之菸鹼醯胺、對羥苯甲酸酯類及咖啡因含量。結果顯示，菸鹼醯胺含量分別爲20.3-241.7、0-210.7及0-246.8 mg/mL；對羥苯甲酸異丁酯含量分別爲0-54.1、0-42.9及0-134.1 μg/mL；對羥苯甲酸丁酯含量分別爲0-106.8、0-120.1及0-184.6 μg/mL及咖啡因含量分別爲0-433.2、0-80.7及472.3-1742.9 μg/mL。以上結果顯示，菸鹼醯胺若換算爲每瓶之總含量，則18件保健飲料中有6件，12件口服液中有2件及22件感冒藥液中有2件超過國人每日平均建議攝取量(RNDA=14.4 mg)。在防腐劑含量方面，大部分商品均未標示含“防腐劑”，52件樣品中有25件檢測出含對羥苯甲酸異丁酯或丁酯，其中有4件超過CNS標準（100 μg/mL）；而在咖啡因含量方面，保健飲料及口服液均未標示咖啡因含量，30件樣品中有27件檢測出含咖啡因，顯然不符CNS之規定。

關鍵詞：保健飲料，口服液，感冒藥液，菸鹼醯胺，對羥苯甲酸酯類，咖啡因，氣相層析，定量分析