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Comprehensive detection of 120 additives in food using nontargeted MS data acquisition

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Abstract

The compliance assessment on the labeling of food additives is a hard job, because there are nearly thousand legal food additives can be used in food, and countless illegal additives must also deal with. This study developed a nontargeted data acquisition screening method based on liquid chromatography high resolution mass spectrometry (HRMS) in which a precursor ion and two product ions of each analyte are able to be recorded. The high throughput screening method worked as foodomics that characterized and identified every food components as long as they were ionized in terms of theory. The data acquisition method called data independent acquisition (DIA) was achieved by a full scan form m/z 70–1050, and then followed wide window fragmentations of product ions recording. A full scan and the followed fragmentations generated 21 spectra in 2.6 s contributed about 6 data points for a typical 0.2–0.3 min width peak in HPLC. A detection database list of 120 additives included 79 colorants, 13 sweeteners, 12 preservatives and 7 antioxidants was established. Thirty-three commercial samples including beverages, candies, and sauces were surveyed for testing additives. Sweeteners (rebaudioside A) and flavoring agents (malic acid and fumaric acid) were found the most under declared additives. HPLC column often do not provide adequate retention for highly polar compounds such as organic acids (flavoring agents). In this study they were coeluted, but were able to be separated and determined by HRMS worked as the secondary separation tool. The surveillance results showed there is still room for food manufacturers to improve the connection between their product information and consumers.

Keywords: Additives, DIA, Fast screening, HRMS, Nontargeted

1. Introduction

Food additives are required to be labeled on the package of food. Most direct additives belong to parts of the ingredient labels of food products, and some trace amounts of indirect additives are not required to be labeled. Additives used in food commonly include colorants, sweeteners, preservatives, antioxidants, and flavoring agents. These natural and/or artificial additives need to be regulated and monitored before addition, treating, or processing. Food additives are evaluated by authorities such as European Food Safety Authority’s (EFSA) or Food and Drug Administration (FDA) to be represented as legal additives, while others not listed in food additive status list are recognized as illegal. The regulations of nations applicable to food additives are vary that force exporters to adjust their product compositions to the importers’ markets. Failure to comply may result in adulteration, misbranding, non-compliance and rejection [1]. Therefore, the monitoring and detection of additives in foods has to include both legal and illegal additives depend on their legal states. Besides, some additives such as Sudan dyes, dimethyl yellow and ethoxyquin are illegal or not permitted to use in food for human consumption,
but may be found in contaminated foods [2–4]. It is necessary to develop a simultaneous detection method for broadly screening of additives in foods. However, additives with various functions to foods present differences in their physico-chemical properties, these make the monitoring of a large group of additives difficult in every step for a method development such as sample preparation, separation and detection.

Various analytical methods reported the detections of a single class of additives. Sagdic [5] detected 18 phenolic compounds including antioxidants by high performance liquid chromatography (HPLC) in grape. Two natural antioxidants were monitored in enriched virgin olive oil through capillary electrophoresis method [6]. Seven preservatives were simultaneously determined in cosmetics by dispersive liquid–liquid microextraction coupled with capillary electrophoresis [7]. Multiclass determinations of additives in a single run were developed in recent year. Boyce [8] used micellar electrokinetic chromatography to separate and detect 6 antioxidants and 3 sweeteners in beverages. Ultra performance liquid chromatography (UPLC) coupled with electrospray ionization tandem mass spectrometry (ESI-MS/MS) was applied for rapid determination of 5 preservatives, 6 artificial sweeteners and 9 synthetic dyes in kimchi. The rapid separation was performed in 5.5 min and can be used for rapid quality control [9]. Methods focused on the sample pretreatment and cleanup were applied in accurate determination of preservatives and artificial sweeteners in juice by HPLC to remove matrix interference prior to chromatographic determination [10].

High resolution mass spectrometry (HRMS) such as orbitrap provided full-scan spectra with resolution 70,000 FWHM (full width at half maximum), which gave chance to look into the composition of a sample. The higher resolution made the assigned masses more accurate and background peaks better resolved. Studies had proved HRMS produced similar results in terms of reliability, specificity and sensitivity to the most wildly used low resolution MS/MS [11,12]. A validated approach analyzed 43 additives including sweeteners, flavor agents, antioxidants and preservatives in dairy product by quadrupole orbitrap mass spectrometry. Sample was prepared through quick, easy, cheap, effective, rugged and safe (QuEChERS) method prior to a 15-min fast analysis [13]. Another similar approach described the detection of 69 dyes in wines [14]. The results proved HRMS was a powerful method for screening analytes in foods. Additional capabilities of HRMS beyond the current MS/MS were by confirming the presence of an analyte not included in the target list. The measure of analytes without previous compound-specific tuning and selection enabled retrospective data analysis for a broad list of compounds, so called nontargeted data acquisition [15]. Compared to tandem mass only applicable to around 200 targets [16], nontargeted data acquisition carried out larger compound database, and widely used as metabolic fingerprinting and profiling approaches [17]. Wang [18] generated a non-target data acquisition for target analysis called nDATA workflow for screening of 845 pesticide residues in fruits and vegetables by UHPLC/Q-Orbitrap. The detection was based on the retention time, mass accuracy of a precursor and fragment ions which significantly expanding the number of pesticides currently being screened by traditional MS/MS approaches.

The aim of this study was designed to build a screening method for 120 additives by HRMS. The major areas of routine monitoring of food were pesticide residues, veterinary residues and natural toxins. Methods for these three classes of residues were well developed for detection of multiple residues in a single commodity, and be able to quantity. However, the multi-class analytical method for additives such as dyes, sweeteners, preservatives, antioxidants and flavoring agents was few. A nontargeted method called variable data independent acquisition (vDIA) was developed for comprehensive sample detection of 120 additives in this study.

2. Methods

2.1. Chemicals

Methanol, acetonitrile, ethyl acetate, acetone, fumaric acid and ammonia acetate were purchased from Sigma–Aldrich (St. Louis, MO, US). Reference standards including (name and purity%) allura red AC (98%), alizarin (95%), alizarin green (95%), amaranth (95%), auramine O (95%), azorubine (95%), benzyl violet 4B (95%), brilliant blue FCF (95%), carminic acid (95%), chrysoidine G (95%), citrus Red 2 (95%), crocein orange G (95%), curcumin (95%), diethyl yellow (95%), erythrosine (95%), fast green FCF (95%), indigo carmine (95%), light green SF yellowish (95%), lissamine green B (95%), malachite green (95%), metanil yellow (95%), methyl yellow (95%), α-naphthol orange (95%), naphthol yellow S (95%), new coccin (95%), orange G (80%), orange II (95%), para red (95%), patent blue V (95%), phloxine (95%), ponceau SX (98%), quinoline yellow
WS (95%), rhodamine B (95%), rose Bengal (95%), scarlet GN (95%), solvent green 3 (95%), Sudan black B (95 %), Sudan I (95%), Sudan II (95%), Sudan III (95%), Sudan IV (95%), Sudan orange G (95%), Sudan red 7B (95%), Sudan red G (95%), Sudan red B (95%), sulforhodamine B (95%), sunsent yellow FCF (95%), tartrazine (95%), xylene fast yellow 2G (60%), pigment orange 5 (98%), disperse orange 37 (96%), oil orange SS (98%), red 2G (98%), fumaric acid (99%), succinic acid (99%), L-glutamic acid (99%), tartaric acid (99%), sodium lactate (99%), DL-lactic acid (99%), propyl gallate (98%), nordihydroguaiaretic acid (97%), 4-hexylresorcinol (98%), ethoxyquin (99%), methyl p-hydroxybenzoate (99%), ethyl p-hydroxybenzoate (99%), propyl p-hydroxybenzoate (99%), butyl p-hydroxybenzoate (99%), benzyl 4-hydroxybenzoate (99%), phenyl 4-hydroxybenzoate (98%), n-heptyl 4-hydroxybenzoate (99%), natamycin (99%), sorbic acid (99%), aspartame (98%), neohesperidin dihydrochalcone (98%), neotame (98%), rebaudioside B (95%), sucralose (98%), L-drochalcone (98%), neotame (98%), rebaudioside A (99%), acacia gum (99%), ascorbic acid (99%), ascorbyl palmitate (99%), citric acid (98%), diacetyl tartaric acid ester (99%), dextran (98%), food grade gelatin (98%), maltodextrin (98%), lactose (98%), leucine (98%), lactic acid (99%), maltose (98%), natural vanilla extract (95%), malt extract (98%), sodium alginate (99%), sorbitol (98%), stachyose (98%), starch (98%), sodium caseinate (99%), sucrose (98%), stevioside (100%), sucrose ester (98%), sucrose fatty acid ester (98%), polyglycerol monostearate (98%), sodium tripolyphosphate (99%), xanthan gum (99%), thymol (99%), turmeric (95%), vanillin (99%), xanthan gum (99%), and acesulfame potassium (99%).

2.2. Instrumentation for generating target list of additives

An UHPLC-ESI-Orbitrap MS system consisted of an UltiMate 3,000 pump, a Q Exactive mass spectrometer and a accucore aQ C18 column (2.1 × 150 mm, 2.6 μm particle size, Thermo Fisher Scientific, Rockford, IL, US) were utilized. The column was maintained at 35 °C and the flow rate was set at 0.5 mL/min. A gradient elution containing 0.1% formic acid with 20 mM ammonium acetate (A), and acetonitrile with 0.1% formic acid (B) was applied. The gradient was hold on 1% (B) for first 1 min and then increased from 1 to 99% (B) over the next 8 min. The eluent was retained for the next 7 min, and then the elution was re-equilibrated at 1% (B) for 1 min after each injection. The injection volume was 5 μL. The mass spectrometer was operated at ESI positive (3.5 kV) and negative (2.5 kV) mode. Resolution was set at 70,000 (defined at m/z = 200 and was set at full width at half maximum, FWHM). The precursor list was built by direct infusion mass spectrometry of each individual additive standard at concentration of 1 ppm.

2.3. Evaluation of limits of detection (LODs) by data independent acquisition (DIA)

Candy and carbonated sparkling water were used as blank sample for the evaluation of LODs for 79 colorants. Pork jerky and carbonated sparkling water were used for 13 sweeteners. Twelve preservatives were tested in pork jerky. Seven antioxidants and 9 flavoring agents were evaluated in carbonated sparkling water. Carbonated sparkling water was ultrasonicated for 15 min, and 10 g was transferred into a 100-mL volumetric flask. Additive standard stock solutions were spiked separately at the concentration 0.01, 0.02, 0.05, 0.1, 0.2, 0.5, 1, 5 and 10 mg/kg, and made up to volume with 50% methanol solution (v/v). Solutions were filtered with membrane filters prior to analysis. Candy and pork jerky were ground separately and 10 g each was transferred into a 100-mL volumetric flask. Additive standard stock solutions were then spiked into sample for 30 min, and made up to volume with 50% methanol solution. The volumetric flask was ultrasonicated for 15 min, and then centrifuged at 5,000×g. The filtrate was passed through a polyvinylidene fluoride (PVDF) membrane filter as sample solution.
Mass spectrometer worked in DIA mode and the parameters were as following. A full scan form m/z 70–1,050 was conducted, and then precursors in wide windows were selected to obtain 21 MS/MS spectra. Isolation window 27 amu was applied for m/z 100 to 500 in every 25 amu increment (e.g. 100, 125, 150, 175, 200, 225, 250, 275, 300 amu). Isolation window 104 amu was utilized for m/z 500 to 1,000 in every 100 amu increment. The ions in every collection were then sent to high energy dissociation (HCD) cell for fragmentation, and the followed C-trap prior to Orbitrap analysis. The cycle time for a full scan and 21 fragments spectra was about 2.6 s. The samples were required to run HPLC HRMS/MS twice, one for positive ESI and another for negative ESI due to the long cycle time of data acquisition.

2.4. Surveillance

Candies, beverages and soy sauces in total 33 samples were purchase from local market. Samples were separated into liquid and solid samples, and followed the same procedures described in sample preparation for LODs evaluation. The detection and data processing of non-target data acquisition for target analysis were performed using TraceFinder 3.3 software (Thermo Fisher Scientific). In a positive sample, the precursor ion of an analyte was extracted form full scan spectra, and the product ions were extracted form fragment spectra associated to the precursor segment.

3. Results and discussion

The precursor ion and two product ions along with retention time of each analyte were shown in Table 1. The ionization of general chemicals were either [M+H]+ or [M−H]−. However, for colorants, some were exceptional. [M + H−Na]+, [M−Na]−, [M+H2−Na]+, [M+H3−Na2]+, [M+H3−Na]+, [M−Cl]+ and [M + H−Ca]2+ might happen depended on the chemicals and the pH of sample solution and eluent. The target list was generated in parallel reaction monitoring (PRM) mode of mass spectrometry. A full scan at scan range m/z 70–1,050 was first conducted, and then the scan data was compared with the established precursor list. If there were a match, the mass spectrometer would isolate a target precursor ion (window was 4 amu), then fragmented the target precursor ion in the collision cell (30 eV and 60 eV stored in C-trap), and then sent to Orbitrap for detection. The resulting product ions were obtained as MS/MS spectrum, and all spectra of standard compounds were listed in the supplemental file (https://www.jfda-online.com/cgi/editor.cgi?article=3366&view=additional_files&context=journal). The dead time, no interaction between the sample and the column, was estimated to be 0.7 min [(0.105 cm)2 × π × 15 cm × 70%/0.5 cm2 per min, 70% was an estimate of interstitial void space in porous silica particles]. Most additives obtained good retentions, except flavoring agents such as L-cysteine, fumaric acid, L-glutamic acid, tartaric acid, sodium lactate, DL-malic acid, glucono-δ-lactone, which obtained retention time between 0.6 and 0.8 min. These compounds co-eluted with other no retention food matrix. The mass spectrometer worked as the secondary separation tool. The low-resolution mass spectrometer (LRMS) basically separate two ions differing by one mass unit along the whole range scanned. The high-resolution mass spectrometer (HRMS), such as Orbitrap, measures ion oscillation frequencies and transfer into mass-to-charge ratio (m/z) with resolution 70,000 FWHM. The instrument provides accurate mass with detecting error below 5 ppm which enable the secondary separation of analytes in complex co-elute food matrix. Fig. 1b showed a chromatogram (total ion chromatogram, TIC) of a soy sauce (sample 32) at RT between 0 and 2.5 min. There was a big and broad peak indicated no/poor retention and co-elute in the first 2 min. The ESI negative spectrum of RT = 0.7 was showed in Fig. 1a that dressed there were a few compounds co-elute around the column dead time. These compounds were high polar chemicals performed no retention on a typical C18 column. However, lactate (Fig. 1c) and glutamic acid (Fig. 1d) were able to be separated from the co-elute matrix by selecting theoretical accurate mass of 89.0244 and 146.0459, respectively. HRMS was powerful and enable the secondary separation of analytes from very complex co-elute without interference. In Fig. 1a, obtained accurate mass of lactate m/z 89.02403 and glutamic acid m/z 146.04531 showed differences of −0.15 and −0.04 ppm, respectively, to their assigned value in Table 1. The advantage of HRMS enabled the detection and separation of analytes even in dead time range of chromatographic separation.

3.1. DIA for precursor selection and product ions detection

DIA required no selection of a special precursor in a narrow range of typically 1 amu (conventional MRM), instead a wide window such as 27 or 104 amu in this study was selected. Various wide windows were selected to cover all potential analytes in the range between m/z 100 to m/z 1,000, and the followed fragmentation generated product ions for
| Compound | Elemental composition | Ionization mode | Precursor Ion (theoretical) | Delta mass | Product ions | Retention time | LODs (ppm) | Candy | Beverage |
|----------|----------------------|-----------------|-----------------------------|------------|--------------|----------------|------------|-------|----------|
| 1 Tartrazine C_{16}H_{9}N_{4}Na_{3}O_{9}S_{2} | [M+2H−3Na]− | 466.9973 | 2.7 | 197.9872 | 3.02 | 0.5 | 0.5 |
| 2 Amaranth C_{26}H_{11}N_{2}O_{10}S_{3} | [M+2H−2Na]− | 536.9738 | 2.4 | 316.9696 | 3.17 | 0.5 | 0.5 |
| 3 Indigo carmine C_{16}H_{8}N_{2}O_{2}S_{2} | [M+H−2Na]2− | 209.9867 | 6.7 | 79.9559 | 3.38 | 5 | 5 |
| 4 Cochineal red A, Ponceau 4R C_{20}H_{11}N_{2}O_{10}S_{3} | [M+2H−3Na]2− | 267.9833 | 3.2 | 145.0280 | 3.48 | 5 | 5 |
| 5 Sunset yellow C_{16}H_{10}N_{2}O_{2}S_{2} | [M+H−2Na]− | 407.0013 | 1.4 | 205.9917 | 3.68 | 0.5 | 0.5 |
| 6 Naphthol yellow S C_{16}H_{12}N_{2}O_{2}S | [M+H−2Na]− | 312.9772 | 1.3 | 327.0445 | 3.68 | 0.5 | 0.5 |
| 7 Orange G C_{16}H_{10}N_{2}O_{5}S_{2} | [M+H−2Na]− | 407.0013 | 1.7 | 301.9560 | 3.74 | 0.1 | 0.1 |
| 8 Allura red AC C_{18}H_{14}N_{2}O_{8}S_{2} | [M+H−2Na]− | 451.0275 | 1.6 | 205.9917 | 3.95 | 0.5 | 0.5 |
| 9 Carminic Acid C_{22}H_{22}O_{3} | [M−H]− | 491.0831 | 1.2 | 357.0610 | 4.06 | 0.5 | 0.5 |
| 10 Xylene fast yellow 2G C_{16}H_{10}Cl_{3}N_{2}O_{2}S_{2} | [M+H−2Na]2− | 251.9689 | 1.7 | 170.9996 | 4.1 | 5 | 5 |
| 11 Scarlet GN C_{18}H_{14}N_{2}O_{2}S_{2} | [M+H−2Na]+ | 437.0472 | 2.3 | 210.0454 | 4.23 | 0.1 | 0.1 |
| 12 Azorubine C_{20}H_{4}N_{2}O_{2}S_{2} | [M+H−2Na]− | 457.0170 | 3.9 | 233.0203 | 4.41 | 0.5 | 0.5 |
| 13 Ponceau SX C_{18}H_{14}N_{2}O_{2}S_{2} | [M+H−2Na]− | 435.0326 | 2.8 | 211.0151 | 4.46 | 0.1 | 0.1 |
| 14 Light green SF C_{20}H_{3}N_{2}O_{2}S_{2} | [M+H−2Na]− | 747.1510 | 1.1 | 683.1891 | 4.49 | 0.5 | 0.5 |
| 15 G3 (Fast green FCF) C_{27}H_{3}N_{2}O_{10}S_{3} | [M+H−2Na]− | 763.1459 | 1.4 | 170.0043 | 4.53 | 0.5 | 0.5 |
| 16 Lissamine green B C_{27}H_{3}N_{2}O_{2}S_{2} | [M+H−Na] | 352.0285 | 2.7 | 272.0717 | 5.21 | 0.05 | 0.05 |
| 17 Brilliant blue FCF C_{37}H_{4}N_{2}O_{4}S_{2} | [M+2H−2Na]− | 747.1510 | 2.6 | 288.0666 | 4.77 | 0.1 | 0.1 |
| 18 α-Naphthol orange C_{16}H_{11}N_{2}O_{2}S | [M−Na]− | 327.0445 | 2.4 | 221.0151 | 4.46 | 0.1 | 0.1 |
| 19 Quinoline yellow S C_{18}H_{13}NaNO_{2}S | [M−3H−Na]− | 352.0285 | 2.7 | 272.0717 | 5.21 | 0.05 | 0.05 |
| 20 Sulforhodamine B C_{26}H_{4}N_{2}O_{2}S | [M+H]+ | 559.1567 | 2.5 | 515.0941 | 5.26 | 0.1 | 0.1 |
| 21 Crocein orange G C_{16}H_{11}N_{2}O_{4}S | [M−Na]+ | 327.0445 | 2.6 | 206.9995 | 5.26 | 0.1 | 0.1 |
| 22 Orange II (Acid orange 7) C_{16}H_{11}N_{2}SO_{4} | [M−Na]+ | 327.0445 | 1.5 | 170.0043 | 5.26 | 0.1 | 0.1 |
| 23 Patent blue V C_{27}H_{31}N_{2}O_{8}S_{2} | [M+2H−Na]+ | 561.1724 | 2.8 | 479.1999 | 5.28 | 0.5 | 0.5 |
| 24 Alizarin C_{14}H_{4}O_{4} | [M−H]− | 229.0350 | 3.8 | 211.0151 | 5.42 | 0.5 | 0.5 |
| 25 Alizarin green (Patent green) C_{37}H_{14}ClN_{2}Na_{2}O_{5}S_{2} | [M+2H−Na]+ | 703.1698 | 2.5 | 517.1342 | 5.48 | 0.5 | 0.5 |
| 26 Chrysoidine G C_{13}H_{12}N_{4}·HCl | [M−Cl]+ | 213.1135 | 4.2 | 121.0634 | 5.49 | 0.1 | 0.1 |
| 27 Curcumin C_{25}H_{20}O_{8} | [M−H]− | 367.1187 | 6.7 | 94.0525 | 5.8 | 5 | 5 |
| 28 Benzyl violet 4B C_{30}H_{40}N_{2}O_{5}S_{2} | [M−Na]− | 710.2364 | 2.7 | 630.2810 | 5.8 | 0.1 | 0.1 |
| 29 Auramine O C_{17}H_{2}N_{3} | [M+H]+ | 268.1808 | 1.2 | 147.0916 | 5.99 | 0.1 | 0.1 |
| 30 R7 (Erythrosine) C_{20}H_{24}N_{4}O_{5} | [M+3H−2Na]+ | 836.6623 | 3.1 | 582.8521 | 6.4 | 0.5 | 0.5 |

(continued on next page)
| Compound               | Elemental composition         | Ionization mode | Precursor Ion (theoretical) | Delta mass | Product ions | Retention time | LODs (ppm) | Candy Beverage | Beverage |
|-----------------------|------------------------------|-----------------|-----------------------------|------------|--------------|----------------|-----------|---------------|-----------|
| 31 Sudan orange G     | C_{12}H_{10}N_{2}O_{2}       | [M+H]^+         | 215.0815                    | 2.5        | 93.0573      | 7.09            | 0.1      | 0.1           |           |
| 32 Malachite green    | C_{23}H_{32}ClN_{2}          | [M–Cl]^+        | 329.2012                    | 4.9        | 313.1699     | 7.52            | 0.5      | 0.5           |           |
| 33 Rhodamine B        | C_{28}H_{31}ClN_{2}O_{3}     | [M–Cl]^+        | 443.2329                    | 1.4        | 355.1077     | 7.58            | 0.05     | 0.05          |           |
| 34 Phloxine           | C_{30}H_{34}BrClN_{2}O_{3}   | [M–Na]^+        | 784.5406                    | 4.4        | 658.6242     | 7.72            | 5        | 5             |           |
| 35 Rose bengal        | C_{29}H_{34}ClN_{2}O_{3}     | [M–Na]^+        | 970.4919                    | 2.9        | 762.6936     | 7.75            | 5        | 5             |           |
| 36 Para red           | C_{16}H_{11}N_{3}O_{3}       | [M+H]^+         | 294.0873                    | 1.4        | 277.0843     | 8.33            | 0.5      | 0.5           |           |
| 37 Methyl yellow      | C_{14}H_{13}N_{3}            | [M+H]^+         | 226.1339                    | 2.7        | 208.1120     | 8.88            | 0.1      | --            |           |
| 38 Sudan red G        | C_{17}H_{14}N_{2}O_{2}       | [M+H]^+         | 279.1128                    | 4.0        | 261.0843     | 8.88            | 0.1      | 0.1           |           |
| 39 Citrus Red 2       | C_{18}H_{16}N_{2}O_{3}       | [M+H]^+         | 309.1234                    | 2.6        | 291.0992     | 8.94            | 0.5      | 0.5           |           |
| 40 Sudan I            | C_{16}H_{12}N_{2}O_{3}       | [M+H]^+         | 249.1022                    | 1.2        | 231.0784     | 8.94            | 0.5      | 0.5           |           |
| 41 Dimethyl yellow    | C_{18}H_{19}N_{3}            | [M+H]^+         | 254.1652                    | 2.7        | 236.1425     | 9.05            | 0.1      | 0.1           |           |
| 42 Sudan II           | C_{18}H_{18}N_{2}O_{3}       | [M+H]^+         | 277.1335                    | 3.2        | 259.1092     | 9.95            | 5        | 5             |           |
| 43 Sudan black B      | C_{20}H_{24}N_{6}            | [M+H]^+         | 457.2135                    | 4.5        | 439.1892     | 10.28           | 5        | 5             |           |
| 44 Sudan III          | C_{22}H_{16}N_{4}O_{2}       | [M+H]^+         | 353.1397                    | 2.9        | 335.1158     | 10.69           | 0.5      | 0.5           |           |
| 45 Solvent green 3    | C_{28}H_{22}N_{2}O_{2}       | [M+H]^+         | 419.1754                    | 2.3        | 391.1511     | 10.79           | 0.5      | 0.5           |           |
| 46 Sudan red 7B       | C_{24}H_{21}N_{5}            | [M+H]^+         | 380.1870                    | 2.7        | 352.1631     | 10.95           | 5        | 5             |           |
| 47 Sudan IV           | C_{24}H_{20}N_{4}O_{3}       | [M+H]^+         | 381.1710                    | 3.8        | 363.1468     | 11.15           | 5        | 5             |           |
| 48 Metanil yellow     | C_{18}H_{14}N_{2}O_{2}S_{3}  | [M–Na]^+        | 352.0761                    | 0.9        | 334.0521     | 11.46           | 0.5      | 0.5           |           |
| 49 Eosin Y            | C_{20}H_{18}BrN_{2}O_{3}     | [M+H–2Na]^+     | 646.6992                    | 0.6        | 628.6754     | 11.21           | 10       | 10            |           |
| 50 Fluorescein        | C_{20}H_{12}O_{5}            | [M–H]^+         | 331.0612                    | 0.9        | 313.0372     | 11.66           | 0.2      | 0.2           |           |
| 51 Brilliant yellow   | C_{24}H_{18}N_{3}O_{2}S_{2}  | [M+H–2Na]^−     | 579.065                     | 0.4        | 561.0411     | 11.98           | 10       | 10            |           |
| 52 Disperse orange 3  | C_{12}H_{10}N_{4}O_{2}       | [M–H]^−         | 241.0731                    | 1.5        | 223.0491     | 12.22           | 0.5      | 0.5           |           |
| 53 Gentian violet     | C_{25}H_{30}N_{3}Cl           | [M–Cl]^+        | 372.2434                    | 1.1        | 354.2199     | 7.89            | 0.1      | 0.1           |           |
| 54 Basic fuchsin      | C_{20}H_{20}ClN_{3}          | [M–Cl]^+        | 302.1652                    | 1.0        | 284.1418     | 8.05            | 0.1      | 0.1           |           |
| 55 Astrazon orange G  | C_{22}H_{21}ClN_{2}          | [M–Cl]^+        | 315.1856                    | 1.6        | 307.1617     | 8.11            | 0.1      | 0.1           |           |
| 56 Astrazon orange R  | C_{28}H_{27}ClN_{2}          | [M–Cl]^+        | 391.2169                    | 1.9        | 373.1929     | 8.53            | 0.1      | 0.1           |           |
| 57 Basic violet 2     | C_{22}H_{24}N_{2}O_{2}Cl      | [M–Cl]^+        | 330.1965                    | 1.8        | 312.1726     | 8.93            | 0.1      | 0.1           |           |
| 58 2-Methoxy-4-nitroaniline (Fast red B Base) | C_{24}H_{33}N_{3}O_{2} | [M+H]^+ | 169.0608                     | 1.2        | 151.0374     | 5.26            | 0.2      | 0.2           |           |
| 59 Alizarin yellow GG | C_{13}H_{18}N_{2}O_{2}Na_{2} | [M–Na]^−        | 286.0469                    | 0.9        | 268.0225     | 6.23            | 0.1      | 0.1           |           |
| 60 Fast yellow AB     | C_{12}H_{11}N_{3}O_{5}S_{2}  | [M–H]^−         | 356.0016                    | 0.6        | 338.0776     | 6.73            | 10       | 10            |           |

(continued on next page)
| Compound                        | Elemental composition | Ionization mode | Precursor Ion (theoretical) | Delta mass | Product ions | Retention time | LODs (ppm) | Candy Beverage | Beverage |
|--------------------------------|-----------------------|-----------------|----------------------------|------------|--------------|----------------|------------|----------------|----------|
| 61 Disperse yellow 3           | C₁₅H₁₅N₃O₂            | [M−H]⁻           | 268.1092                   | 0.7        | 134.0611     | 7.31           | 0.1        | 0.1            |          |
| 62 o-Aminoazotoluene           | C₁₄H₁₅N₃              | [M+H]⁺           | 226.1339                   | 1.5        | 121.076      | 7.74           | 0.1        | 0.1            |          |
| 63 Pigment orange 5            | C₁₆H₁₅N₃O₅            | [M−H]⁻           | 337.0578                   | 1.2        | 125.0118     | 8.11           | 0.5        | 0.5            |          |
| 64 1-(Methylamino)-anthraquinone | C₁₅H₁₁N₂O₂        | [M+H]⁺           | 238.0863                   | 0.6        | 167.0098     | 7.8            | 0.2        | 0.5            |          |
| 65 4-aminoazobenzene           | C₁₂H₁₁N₃              | [M+H]⁺           | 198.1026                   | 0.3        | 165.0699     | 6.9            | 0.1        | 0.1            |          |
| 66 Lithol rubine BK            | C₁₈H₁₂CaN₂O₆S        | [M+H]eCa         | 385.05                     | 0.8        | 87.0401      | 5.55           | 10         | 10             |          |
| 67 Sudan red B                 | C₄₂H₃₀N₄O              | [M+H]⁺           | 381.171                    | 1.7        | 224.1196     | 11.29          | 1          | 1              |          |
| 68 Solvent yellow 21           | C₃₄H₂₀C₄N₄O₆          | [M−H]⁻           | 692.123                    | 1.3        | 475.0616     | 6.92           | 1          | 1              |          |
| 69 Solvent orange 62           | C₳₂H₂₀C₄N₄O₆          | [M−H]⁻           | 726.1033                   | 1.5        | 447.0146     | 7.96           | 1          | 1              |          |
| 70 Solvent red 8               | C₳₂H₂₀C₄N₄O₆          | [M−H]⁻           | 729.1268                   | 2.2        | 417.9691     | 7.84           | 1          | 1              |          |
| 71 Solvent yellow 16           | C₁₆H₁₄N₄O             | [M−H]⁻           | 277.1095                   | 1.1        | 117.0458     | 8.58           | 1          | 1              |          |
| 72 Fast brown RR               | C₁₆H₁₄N₄              | [M+H]⁺           | 263.1291                   | 0.7        | 143.0730     | 7.22           | 0.02       | 0.02          |          |
| 73 Oil orange SS               | C₁₂H₁₄N₄O             | [M+H]⁺           | 263.1179                   | 0.4        | 246.1152     | 9.3            | 0.02       | 0.02          |          |
| 74 Red 2G (Azophloxine)        | C₁₆H₁₃N₃Na₂O₈S₂       | [M+H−2Na]⁻       | 464.0228                   | 1.1        | 358.9775     | 4.06           | 2          | 2              |          |
| 75 Cu(II) Chlorin e4           | C₃₃H₃₂CuN₃Na₂O₄       | [M+H−2Na]⁻       | 612.1803                   | 2.3        | 481.1459     | 9.19           | 10         | 20             |          |
| 76 Cu(II) Chlorin e6           | C₃₄H₃₁CuN₃Na₃O₆       | [M+2H−2Na]⁻      | 656.1702                   | 3.1        | 553.1670     | 10.39          | 10         | 20             |          |
| 77 4-amino carminic acid       | C₂₂H₁₅NO₁₂            | [M−H]⁻           | 490.0991                   | 3.7        | 507.1615     | 4.12           | 0.5        | 0.5            |          |
| 78 Disperse orange 37          | C₁₇H₁₅Cl₂N₃O₂         | [M+H]⁺           | 392.0676                   | 4.3        | 326.0670     | 8.2            | 0.1        | 0.1            |          |
| 79 Guinea green B              | C₃₇H₃₅N₂Na₂O₇S₂       | [M−Na]⁻          | 667.1942                   | 4.5        | 165.0784     | 165.0784       | 0.1        | 0.1            |          |
| 80 Saccharin                   | C₂₁H₂₄NO₃S            | [M−H]⁻           | 181.9917                   | 4.2        | 105.9598     | 2.83           | 0.02       | 0.02          |          |
| 81 Cyclamate                   | C₄H₃₄NO₂S            | [M−H]⁻           | 178.0543                   | 4.1        | 79.9568      | 3.02           | 0.1        | 0.1            |          |
| 82 Aspartame                   | C₁₄H₁₈N₄O₅            | [M+H]⁺           | 295.1288                   | 2.3        | 120.0813     | 4.03           | 0.1        | 0.1            |          |
| 83 Stevioside                  | C₃₈H₆₀O₁₈             | [M−H]⁻           | 803.3706                   | 1.6        | 641.3231     | 5.58           | 0.1        | 0.1            |          |
| 84 Acesulfame potassium        | C₂₁H₂₆NO₂S            | [M−H]⁻           | 161.9866                   | 5.2        | 317.2116     | 1.36           | 0.02       | 0.02          |          |
| 85 Dulcin                      | C₂₁H₂₄N₂O₂            | [M+H]⁺           | 181.0971                   | 4.3        | 108.0449     | 4.5            | 0.02       | 0.02          |          |
| 86 Nefosphenid dihydrochalcone | C₂₈H₃₈O₁₅             | [M+H]⁺           | 613.2127                   | 0.9        | 137.0602     | 4.89           | 1          | 1              |          |
| 87 Glycyrrhizin                | C₁₂H₁₆O₁₆             | [M−H]⁻           | 821.3959                   | 0.3        | 259.181      | 5.52           | 0.1        | 0.1            |          |
| 88 Neotame                     | C₂₀H₃₀N₂O₅            | [M+H]⁺           | 379.2227                   | 2.9        | 351.0563     | 5.62           | 0.1        | 0.1            |          |
| 89 Allitame                    | C₁₄H₁₄N₂O₄S          | [M+H]⁺           | 332.1638                   | 3.3        | 179.0708     | 5.52           | 0.1        | 0.1            |          |
| 90 Rebaudioside A              | C₁₄H₇O₂₃              | [M−H]⁻           | 965.4235                   | 2.1        | 803.3755     | 5.13           | 10         | 10             |          |

(continued on next page)
| Compound | Elemental composition | Ionization mode | Precursor Ion (theoretical) | Delta mass* | Product ions | Retention time | LODs (ppm) | Candy Beverage | Pork jerky |
|----------|------------------------|----------------|-----------------------------|-------------|--------------|----------------|-------------|----------------|------------|
| 91       | C_{38}H_{60}O_{18}     | [M−H]^−        | 803.3707                   | 0.7         | 641.3237     | 5.55           | 10          | 10             |            |
| 92       | C_{12}H_{19}Cl_{3}O_{8} | [M+NH_{4}]^+   | 414.0484                   | 1.4         | 198.9290     | 3.88           | 2           | 1              |            |
| 93       | C_{10}H_{12}O_{5}      | [M−H]^−        | 211.0612                   | 3.3         | 216.0629     | 4.95           | 0.1         |                |            |
| 94       | C_{10}H_{14}O_{2}      | [M−H]^−        | 195.0662                   | 3.1         | 95.0128      | 5.43           | 0.1         |                |            |
| 95       | C_{18}H_{22}O_{4}      | [M−H]^−        | 301.1445                   | 0.7         | 122.0362     | 6.29           | 0.5         |                |            |
| 96       | C_{12}H_{19}Cl_{3}O_{8} | [M+NH_{4}]^+   | 414.0484                   | 1.4         | 198.9290     | 3.88           | 2           | 1              |            |
| 97       | C_{10}H_{14}O_{3}      | [M−H]^−        | 235.1703                   | 2.1         | 217.1587     | 7.26           | 10          |                |            |
| 98       | C_{14}H_{19}NO         | [M+H]^+        | 218.1539                   | 0.5         | 174.0913     | 7.41           | 0.2         |                |            |
| 99       | C_{12}H_{19}NO_{2}S-HCl | [M−Cl]^+      | 122.0270                   | 0.4         | 76.0215      | 0.61           | 10          |                |            |
| 100      | C_{4}H_{6}O_{4}        | [M−H]^−        | 115.0037                   | 0.0         | 71.0133      | 0.73           | 10          |                |            |
| 101      | C_{4}H_{6}O_{4}        | [M−H]^−        | 117.0193                   | 0.0         | 73.0290      | 1.08           | 10          |                |            |
| 102      | C_{4}H_{6}O_{4}        | [M−H]^−        | 146.0459                   | 4.0         | 128.0348     | 0.68           | 1           |                |            |
| 103      | C_{4}H_{6}O_{4}        | [M−H]^−        | 149.0092                   | 4.0         | 102.0555     | 0.68           | 10          |                |            |
| 104      | C_{4}H_{6}O_{4}        | [M−H]^−        | 89.0244                    | 4.2         | 59.0133      | 0.79           | 0.5         |                |            |
| 105      | C_{4}H_{6}O_{4}        | [M−H]^−        | 133.0142                   | 3.0         | 71.0133      | 0.72           | 0.5         |                |            |
| 106      | C_{4}H_{6}O_{4}        | [M+H]^+        | 195.0877                   | 2.2         | 107.0497     | 3.93           | 0.1         |                |            |
| 107      | C_{4}H_{6}O_{4}        | [M−H]^−        | 173.0932                   | 4.0         | 155.0821     | 0.90           | 10          |                |            |
| 108      | C_{4}H_{6}O_{4}        | [M+H]^+        | 195.0511                   | 2.6         | 129.0188     | 0.68           | 10          |                |            |

* Difference between the measured mass/charge (m/z) of procurer and the exact m/z of that ion. Expressed as δ ppm.

** Compound is not soluble in water.
the mixed precursors in every wide window. Theoretically, DIA was able to record all chemicals with the required information of precursors and product ions in a sample as long as they were able to be ionized by ESI. The identification of analytes was performed according to SANTA/11813/2017 guidelines [19]. Mass accuracy less than 5 ppm was applied for 2 fragments, and less than 10 ppm was for a precursor. Matched retention time was limited in 0.2 min. Signal was required to have S/N ratio larger than 3 or at least 5 subsequent scans presented (in some case S/N ratio can not be calculated due to S/N ratio = infinity). A total ion chromatogram of sample 22 as an example was shown in Fig. 2a, and the presence of allura red AC was shown in Fig. 2b which data was processed by selecting a product ion \( m/z \) 205.9917 from a group of precursors in the range of \( m/z \) 449–476 (accurate mass of allura red AC was 451.0275). The scan spectrum of RT = 4.05 (from full scan chromatogram, Fig. 2a) was showed in Fig. 2c. There were various compounds co-elute and each one can be a precursor. In DIA, isolation window 27 amu was applied for \( m/z \) 100 to 500 in every 25 amu increment. Therefore, the precursor ion of allura red AC was selected along with other ions presented in the range of \( m/z \) 449 to 476 (Fig. 2d). Multiple precursors were fragmented together, and the mixed product ions were shown in Fig. 2e. Two characteristic ions 79.95695 and 205.99075 indicated the positive identification of allura red AC.

3.2. LODs for the screening method

Colorants were tested in candy and carbonated sparkling water for LODs evaluation. The obtained LODs ranged between 0.02 and 20 part per million (ppm), and mostly 0.5 ppm in Table 1. Antioxidants and flavoring agents observed LODs at 1–10 ppm range. Preservatives were tested in pork jerky.
matrix and obtained LODs between 0.1 and 1 ppm. In the comparison to other multi-residue studies for additives by targeted acquisition method [9], this nontargeted data acquisition provided 10 to 100-fold higher LODs. Conventional targeted method optimized ionization and fragmentation parameters for each analyte such as cone voltage for precursors and collision energy for product ions. However, non-targeted method was not able to optimize MS parameter for “non target”, instead a basic or typical MS parameter was applied for a broad compound screening or called high throughput screening.

Colorants such as rhodamine B observed similar LOD to a targeted MRM method. Cu(II) Chlorin e4 and Cu(II) Chlorin e6 observed LODs as 20 ppm in this study, but a targeted MRM method reported 20 ppb as the LODs and 50 ppb as the LOQs [20]. Therefore, nontargeted method utilizing DIA is able to screen a broad list of analyte, and also suitable for food additive screening in which the detection level is around ppm level. However, for trace level analysis such as pesticide or veterinary drug residues, DIA screening may not applicable in such low detection level (10–100 ppb), and traditional optimized targeted MRM method with sample preparation for limited analytes is still suggested.

3.3. Surveillance results of samples form commercial market

Thirty three foods were tested utilizing the DIA method. The detected additives and labeled
| No. | Product (form)          | Labeled additives                                                                                       | Detected additives                                                                 |
|-----|-------------------------|--------------------------------------------------------------------------------------------------------|---------------------------------------------------------------------------------------|
| 1   | Sparkling water (lemon) | Citric acid, Flavoring (Gum arabic, Sucrose acetoisobutyrate, Medium-chain triglyceride), Sodium citrate, Vitamin C, Cartithamine, Vitamin B6 | Rebaudioside A, DL-Malic acid, Fumaric acid, Glucono-O-Lactone, Sodium Lactate         |
| 2   | Orange soda             | Citric acid, Gum arabic, Flavoring, Glycerol, Vitamin C, Vitamin E, Stevia, Medium-chain triglyceride, Glycerol ester of wood rosin, Carotene, sodium carbonate | Rebaudioside A, DL-Malic acid, Fumaric acid, Glucono-O-Lactone, L-Glutamic acid, Sodium Lactate, Tartaric acid |
| 3   | Sparkling water (grape) | Flavorsing, Citric acid, Vitamin C, Tartaric acid, Sodium citrate, Allura red AC, Brilliant blue FCF | Tartaric acid, Glucono-O-Lactone, Sodium Lactate, DL-Malic acid, Fumaric acid, Succinic acid, Allura red AC, Brilliant blue FCF |
| 4   | Apple soda              | Flavoring, DL-Malic acid, Caramel colors, Stevia                                                                 | DL-Malic acid, Fumaric acid, Glucono-O-Lactone, L-Glutamic acid, Sodium Lactate, Tartaric acid |
| 5   | Berry vinegar drink     | Citric acid, flavoring, Sodium erythorbate, Sodium metaphosphate, Sodium polyphosphate, Anthocyanin, Sodium pyrophoaphate | Rebaudioside A, DL-Malic acid, Fumaric acid, Glucono-O-Lactone, Sodium Lactate, Succinic acid |
| 6   | Apple soda              | Apple flavoring, Caramel colors, Citric acid, Natural apple flavoring                                                                 | DL-Malic acid, Fumaric acid, Glucono-O-Lactone, Sodium Lactate, Succinic acid |
| 7   | Apple vinegar drink     | Citric acid, flavoring, Anthocyanin, Sodium erythorbate, Sodium citrate, Sodium metaphosphate, Sodium polyphosphate, Sodium pyrophoaphate | Aspartame, Acesulfame potassium, Fumaric acid, Sodium Lactate |
| 8   | Soft drink              | Citric acid, Sodium citrate, Vitamin C, Flavoring, Acesulfame potassium, Aspartame, Beta-carotene         | Aspartame, Acesulfame potassium, Fumaric acid, Sodium Lactate |
| 9   | Sparkling grape drink   | Grape flavoring, Citric acid, Grape skin pigment                                                                 | DL-Malic acid, Fumaric acid, Glucono-O-Lactone, L-Glutamic acid, Sodium Lactate, Tartaric acid, Succinic acid |
| 10  | Zero soda               | Flavouring, Citric acid, Erythritol, Vitamin C, DL-Malic acid, Monosodium L-aspartate, Sucralose, Acesulfame potassium, Vitamin B1, Vitamin B2, Vitamin B6 | Aspartame, Acesulfame potassium, Sucralose |
| 11  | Mints (grape)           | Sorbitol, Aspartame, Acesulfame potassium, Malic acid, Grape flavoring, Magnesium stearate, Silicon dioxide, Erythrosine, Brilliant blue | Tartaric acid, DL-Malic acid, Sodium Lactate, Aspartame, Acesulfame potassium, Erythrosine, Brilliant blue FCF |
| 12  | Mints (soda)            | Sorbitol, Aspartame, Acesulfame potassium, Malic acid, Soda flavoring, Magnesium stearate, Silicon dioxide, Brilliant blue | DL-Malic acid, Fumaric acid, Sodium Lactate, L-Glutamic acid, Aspartame, Acesulfame potassium, Brilliant blue FCF |
| 13  | Mints (peach)           | Sorbitol, Aspartame, Acesulfame potassium, Peach flavoring, Malic acid, Magnesium stearate, Silicon dioxide, Erythrosine | DL-Malic acid, Sodium Lactate, Succinic acid, Aspartame, Acesulfame potassium, Erythrosine |
| 14  | Mints (peach, no sugar) | Sorbitol, Acesulfame potassium, Sucralose, Magnesium stearate, Flavoring, DL-Malic acid, Citric acid, Allura red AC | DL-Malic acid, Acesulfame potassium, Sucralose, Allura red AC |
| 15  | Mints                   | Sorbitol, Acesulfame potassium, Sucralose, Magnesium stearate, Flavoring, Lactic acid, Calcium lactate, Indigo carmine | Sodium Lactate, Acesulfame potassium, Sucralose, Indigo carmine |
| 16  | Mints                   | Sorbitol, Acesulfame potassium, Sucralose, Flavoring, Magnesium stearate, Brilliant blue FCF             | Aspartame, Acesulfame potassium, Sucralose, Brilliant blue FCF |

(continued on next page)
Table 2. (continued)

| No. | Product               | Product form | Labeled additives                                                                                           | Detected additives                                                                 |
|-----|----------------------|--------------|------------------------------------------------------------------------------------------------------------|-----------------------------------------------------------------------------------|
| 17  | Mints (lemon)        | Thailand     | Isomalt, Citric acid, Lactic acid, Flavoring, Sucralose, Tartrazine, Brilliant blue FCF                   | Sodium Lactate, Sucralose, Allura red AC, Tartrazine, Brilliant blue FCF           |
| 18  | Mints (berries)      | Thailand     | Isomalt, Flavoring, Citric acid, DL-Malic acid, Sucralose, Aceulfame potassium, Allura red AC, Tartrazine | DL-Malic acid, Sucralose, Acesulfame potassium, Allura red AC                       |
| 19  | Mints (honey)        | Thailand     | Isomalt, Citric acid, Flavoring, Sucralose, Tartrazine                                                   | Sucralose, Tartrazine                                                             |
| 20  | Mints (honey and lemon) | India       | Isomalt, Aspartame, Aceulfame potassium, Flavoring, Citric acid, Betacarotene                             | Aspartame, Acesulfame potassium                                                   |
| 21  | Mints                | India        | Isomalt, Aspartame, Aceulfame potassium, Flavoring, Curcumin, Brilliant blue FCF                          | Aspartame, Acesulfame potassium, Curcumin, Brilliant blue FCF                      |
| 22  | Mints (fruits)       | Australia    | DL-Malic acid, Starch acetate, Flavoring, Sodium citrate, Xanthan gum, Titanium dioxide, Starch sodium succinate, Palm wax, Sunset yellow FCF, Allura red AC, Brilliant blue FCF | DL-Malic acid, Fumaric acid, Tartrazine, Sunset yellow FCF, Allura red AC, Brilliant blue FCF |
| 23  | Lollipop (apple)     | Vietnam      | Lactic acid, DL-Malic acid, Citric acid, Flavoring, Curcumin                                            | Sodium Lactate, DL-Malic acid, Curcumin                                           |
| 24  | Mints (strawberry)   | Vietnam      | Lactic acid, DL-Malic acid, Citric acid, Flavoring, Betarubin                                           | Sodium Lactate, DL-Malic acid                                                     |
| 25  | Chewing gum (mint)   | Taiwan       | Sorbitol, Maltitol, D-Xyitol, D-Mannitol, Aspartame, Acesulfame potassium, Sucralose, Glycerol, Flavoring, Sodium carboxymethyl cellulose, Palm wax, Sodium alginate, BHT, Brilliant blue FCF, Fumaric acid, Succinic Acid | Aspartame, Acesulfame potassium, Sucralose, Brilliant blue FCF                      |
| 26  | Grape C              | Taiwan       | Grape powder (oxidized starch, citric acid, flavoring, grape skin pigment), Vitamin C, Hydropropyl methylcellulose, Citric acid, Cellulose, Grape flavoring, Magnesium stearate, Ferric pyrophosphate, Folic acid, Vitamin B12 (sodium citrate, citric acid, vitamin B12) | Tartaric acid, L-Glutamic acid, Sodium Lactate, DL-Malic acid                       |
| 27  | Mints                | Australia    | Starch acetate, Flavoring, Xanthan gum, Palm way, Starch sodium succinate, Titanium dioxide, Brilliant blue FCF | Fumaric acid, Succinic Acid                                                       |
| 28  | Mints                | Taiwan       | Sorbitol, Maltitol, Maltitol syrup, D-Mannitol, Aspartame, Acesulfame potassium, Flavoring, Sodium carboxymethyl cellulose, Palm wax, Sodium alginate, BHT, Brilliant blue FCF, Fumaric acid, Succinic Acid | Aspartame, Acesulfame potassium, Brilliant blue FCF                                 |
| 29  | Soy sauce            | Taiwan       | Glycyrrhizin                                                                                               | Tartaric Acid, Glucono-Δ-Lactone, DL-Malic Acid, Fumaric Acid, L-Glutamic Acid, Sodium Lactate, Succinic Acid, Glycyrrhizin acid |
| 30  | Soy sauce            | Taiwan       | Acetylated distarch adipate, Corn syrup, Glycin, Sodium succinate                                         | L-Glutamic Acid, Fumaric Acid, DL-Malic Acid, Sodium Lactate, Succinic Acid        |
| 31  | Soy sauce            | Taiwan       | Malitol syrup                                                                                              | L-Glutamic Acid, Fumaric Acid, DL-Malic Acid, Sodium Lactate, Succinic Acid        |
| 32  | Soy sauce            | Taiwan       | None                                                                                                       | Glucono-Δ-Lactone, L-Glutamic Acid, Sodium Lactate, DL-Malic Acid, Succinic Acid |
| 33  | Soft dring           | Taiwan       | Flavoring, Citric acid, Vitamin C, DL-Malic acid, L-Aspartic acid, Vitamin B2, Vitamin B6, Vitamin B1      | Tartaric acid, Glucono-Δ-Lactone, DL-Malic Acid, Fumaric Acid, L-Glutamic Acid, Sodium Lactate, Succinic Acid, Glycyrrhizin acid |
additives were listed in Table 2. Sample 1, 6 were sparkling water and soda found under declared rebaudioside A as sweetener, which might come from the flavoring agent in their ingredients. For other drinks and candies, fumaric acid, glucono-δ-lacton, sodium lactate, DL-malic acid, L-glutamic acid, tartaric acid and succinic acid were the most often flavorings under declared, which might come from natural extracts, flavoring agent mixture or artificial additives in their ingredients. Some colorants such as tartrazine (in sample 18, red candy in appearance) and Brilliant blue FCF (in sample 27, white mints in appearance) were labeled in product package, but not detected. Artificial sweeteners such as acesulfame potassium, sucralose and aspartame which belong to regular sweetener testing list were all clear labeled in the package and detected. This surveillance results showed that all tested samples were all complied with regulation, but there is still room for manufacture to improve clearly declaration of product information to consumer.

4. Conclusions

This study established a nontargeted DIA analytical method and a detection database list of 120 food additives for rapid screening of food products. The database included 79 colorants, 13 sweeteners, 12 preservatives and 7 antioxidants, enclosed the chromatographic retention time, accurate mass of molecular ion, accurate mass of two product ions, LODs in matrix (candy, beverage or jerky). HRMS enabled secondary separation. Some high polar flavoring agent, such as lactate, L-theanine and glutamic acid were able to be separated and determined by HRMS in coelutant. However, due to the normal sample preparation and the neglect of matrix effects, DIA offered higher LODs for most compounds compared to traditional targeted MRM method. Mass spectrometer worked in full scan and wide window fragmentations that recorded all components in theoretical enabled retrospective analysis for unknowns and increasable target lists. A survey of commercial products of beverages, candies and sauces showed this method can efficiently screen food for illegal additives or false labeling. The results revealed sweeteners and flavoring agents were the most under declared additives. For example, no-calorie sweetener rebaudioside A, about 150–400 times sweeter than sugar, was found in some soda. The surveillance results showed the current labeling is good, but still needed for improvement. The detection database will continually be expand to increase the detection range of food additives for this rapid screening method.

Conflict of interest

There is no potential conflict of interest to declare.

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