Preliminary Metabolomic Analysis of Goat Milk from Different Breeds Using Mass Spectrometry

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Abstract— Metabolomics facilitates the identification of numerous metabolites in a sample with mass less than 1 kD. The purpose of this study was to determine the effect of feed on the metabolites in raw goat milk samples obtained from two breeds namely Saanen and Shami that were collected from a farm in Selangor and Negeri Sembilan, Malaysia, respectively. The types of feed given to the goats were recorded. The fat in the samples were removed via centrifugation before analysis using liquid chromatography quadrupole time of flight mass spectrometry (LC-QToF-MS). From the chromatograms obtained the presence of veterinary drug residue, di- and tri-peptides, short and long-chain fatty acids, some components from plants, and insecticide residue were detected using HMP, KEGG, LMP, and METLIN databases identification. It is expected that through metabolomics study it may be possible to formulate the feed and determine the breed that give good quality milk.

Keywords— Metabolomics, goat milk, LC-QToF-MS, databases.

I. INTRODUCTION

In the last 20 years scientists begin to realise that a simultaneous compound or metabolite identification method would assist in confirming health and physiological status of an organism. Metabolites represent a diverse group of low-molecular-weight structures including lipids, amino acids, peptides, nucleic acids, organic acids, vitamins, thiols and carbohydrates [1]. Metabolomics aims to detect, identify, and quantify a total population of low molecular weight compounds to gain functional information in a biological system [2].

Milk is a complex biological fluid secreted for neonate nourishment and development. Milk contains water, lipids, carbohydrates, proteins, vitamins, minerals, and smaller metabolites. Changes in chemical composition of milk affect nutritional, safety, and technological properties of the milk [3]. In addition to the normal nutritional composition, goat milk contains other metabolites that have functional properties and may be unique to type of feed and breed [4], [5]. Sundekilde [6] suggested that metabolomics approach can be used to establish biomarkers in bovine milk as a diagnostic tool for determining quality and technological properties. Metabolites as biomarkers for specific biochemical pathways will indicate different physiological status of the organism [2].

In addition, coupling chromatography to mass spectrometry (MS) offers an excellent solution to complex mixture analyses and has been extensively used in metabolomics. Chromatographic separation of metabolites prior to MS analyses has several advantages: i) reduces matrix effects and ionization suppression, ii) separates isomers, iii) provides additional and orthogonal data (i.e. retention time/factor/index) valuable for metabolite annotation, and iv) allows for more accurate quantification of individual metabolites [7].

Nevertheless it has now become relatively routine to comprehensively compare the levels of thousands of metabolite peaks in one sample group to another in an untargeted manner. This approach, called untargeted metabolomics, has the potential to implicate unexpected pathways with a unique phenotype or disease process [8].

Reaves [9] coined out that one of the uses of metabolomics data is to pick out a few interesting compounds for further study. This was the further emphasized by [10], whereby he classified a high quality milk as having low somatic cell count (SCC), low standard plate count (SPC), no human pathogen, and no antibiotic residues. Hence, this preliminary effort aims at profiling the metabolites of goat milk in selected farms in some parts of Peninsular Malaysia.
A. Milk collection and sample preparation

The method of [11] was followed with modification. Goat milk was collected from a farm in Negeri Sembilan and Selangor and the freshly collected milk samples were immediately cooled in ice and appropriate care was observed to prevent any possible cross-contamination. The individually milked samples from different goats (n = 4) were pooled, collected in sterile bottles and transported in an icebox at temperature to the Faculty of Science and Technology, Universiti Sains Islam Malaysia (USIM), then immediately frozen at -20°C until further analysis. Aseptic techniques were applied, wherein all the equipment were pre-sterilized prior to analysis.

Goat milk was thawed and centrifuged at 3,000 x g for 10 min using centrifuge (Novil) at 4°C to remove high molecular weight compounds. The supernatant were then collected and analyses were done on the same day.

B. Instrumentation

Untargeted metabolomics profile of three goat milk samples (1 = Saanen species, 2 = Shami species) were carried out using LC-QToF-MS (LCMS iFunnel Q-TOF) at Agro Biotechnology Institute (ABI), MOSTI, MARDI, Serdang. Specifications include column type: RRHD Zorbax Eclipse Plus C18; 1.8µ x 2.1mm x 100mm with temperature control on icebox at temperature to the Faculty of Science and Technology, Universiti Sains Islam Malaysia (USIM), then immediately frozen at -20°C until further analysis. Aseptic techniques were applied, wherein all the equipment were pre-sterilized prior to analysis.

An Agilent G6550A Accurate Mass QTOF was used to obtain the MS data. The mobile phases used were highly purified water + 0.1% formic acid (A) and acetonitrile+0.1% formic acid (B). The flow rate was set up at 0.6 mL/min with 30 min total run time. The prepared samples were placed into the LCMS autosampler. The injection volume of sample was 10 µL. The samples were run at gradient 5% B (0-0.5 min); 30% B (0.5-13 min); and 95% B (13–22.0 min). Analysis was performed in positive ion mode with the following settings:-capillary voltage: 3500 V; nozzle voltage: 100 V; fragmentor voltage: 175 V; nebulizer pressure (N): 35 psi; drying gas: 5L/min at 350°C, and sheath gas: 11 L/min at 350°C. The mass range was at 100–1700 m/z.

C. Database Annotation

For compound identification, several databases were referred to including KEGG, HMDB, LMP, and METLIN that were all from Mass Hunter Qualitative analysis software, Agilent Technologies.

III. RESULTS AND DISCUSSIONS

Metabolite levels, although present at low concentrations, can provide information of biochemical status in response to environments or genetic manipulation. These components can have a profound impact on the development and maintenance of metabolic, immunological and physiological processes [2], [12].

Figures 1 and 2 show the extracted compound chromatogram (ECC) of goat milk from both Saanen and Shami breeds at the end of the analysis period (30 min), respectively. The ECC profiles of goat milk were distinct with dissimilar chromatogram patterns suggestive of different breeds. Reference [2] specified that metabolomic analysis could detect changes in the metabolites as a result of genetic influence or feed given [13], [14].

The distinctive feed given to both breeds was barli sprout to Saanen, and indigofera spp. and palm leaves to Shami breeds. The other feed given were quite similar at any other goat farms namely napier leaves and brans.

Table I listed out the known compounds detected, from the smallest to the highest mass present in milk from Saanen (only). However, the overall mass spectrometry analysis identified compounds with masses ranging from 0.099 kD to 19.6 kD and 0.098 kD to 18.8 kD for Saanen (22117 compounds) and Shami (20679 compounds) species, respectively. The ranges above than 1 kD indicate the poor sample preparation for metabolomic analysis. The metabolites were grouped into several categories namely plant secondary metabolites, drugs, naturally occurring metabolites, environmental contaminants, microbial metabolism in diverse environments including microbial secondary metabolites, and ‘others’ for easy classification purposes (Table II). Some microbial metabolites were observed due to both long cold storage and the milking management practices of the milk samples obtained as supported by [15] and [16], correspondingly. Other compounds detected were drug metabolites such as pinacidil, spiramycin 3, deschlorobenzoyl indomethacin, methotrimereprazone sulfoxide and environmental contaminants (trichlorfon). Trichlorfon is an organophosphate which has adverse effects on health [17], [18].

The list of 20 highest concentrations of known compounds detected present in milk from Saanen and Shami are shown in Table II. It was observed that although the milk samples were from different breeds, some similarities existed. Such compounds include asp-phe-arg, maltose, pantothenic acid, 3-butyrl propionic acid, GPEtn(18:1(9Z)/0:0), and, neu5Ac alpha2-6Galbeta1-4Glcbeta-Sp., all of which were categorized as naturally occurring metabolites.

In addition, [19] indicated that valine and glycine were specific to goat milk, talose and malic acid to cow milk, and hydroxylglutaric acid to pasteurized samples via a gas chromatography-mass spectrometry (GCMS). It was noted that LCMS detected amino acids in the form of di- and tripeptides compared to GCMS that can identify individual amino acids [19] envisaging the beneficial effects towards health. For lipids, by-products of metabolism of polyunsaturated fatty acids (PUFA) generated hydroxy fatty acids, o xo fatty acids, conjugated fatty acids, and partially saturated trans-fatty acids as intermediates [20].

IV. CONCLUSIONS

Preliminary study of metabolite profiling of milk by mass spectrometry is able to generate the high quality data needed for further qualitative analysis. Goat milk has compounds related to health benefits in the form of conjugates.

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APPENDIX

Fig. 1: Extracted Compound Chromatogram (ECC) of Goat Milk from Saanen Species.

Fig. 2: Extracted Compound Chromatogram (ECC) of Goat Milk from Shami Species.

TABLE I
PARTIAL SUMMARY (QUALITATIVE) OF THE IDENTIFIED COMPOUNDS USING LC-QTOF-MS DETECTED IN MILK FROM SAANEN THE REFERRED DATABASE.

| Molecular Mass (kD) | Retention Time (Min) | Putative Identification            | Empirical Formula | METLIN Database |
|---------------------|----------------------|------------------------------------|------------------|-----------------|
| 100.0891            | 26.314               | 4-Methylpentanal                   | C6 H12 O         | 6155            |
| 105.0793            | 14.138               | 3-Methylamino-1,2-propandiol       | C4 H11 N O2      | 44718           |
| 112.0278            | 0.914                | Uracil                             | C4 H4 N2 O2      | 258             |
| 118.0635            | 1.427                | α-hydroxyisovalerate               | C5 H10 O3        | 3751            |
| 120.0575            | 2.621                | 4-Hydroxystyrene                   | C8 H8 O          | 7012            |
| 122.0373            | 4.627                | Benzoic acid                       | C7 H6 O2         | 1297            |
| 133.0739            | 16.218               | L-O-Methylthreonine                | C5 H11 N O3      | 44769           |
| % | M.W. | Formula |
|---|------|---------|
| 135.0685 | 3.488 | N-Acetylarylamine | C8 H9 N O | 6107 |
| 136.0391 | 1.094 | Hypoxanthine | C5 H4 N4 O | 83 |
| 136.0528 | 1.32 | Phenylacetic acid | C8 H8 O2 | 129 |
| 139.0269 | 1.602 | 4-Nitrophenol | C6 H5 N O3 | 4100 |
| 140.0473 | 1.32 | 2-METHOXYRESORCINOL | C7 H8 O3 | 44540 |
| 141.0431 | 1.032 | 2-Aminomuconate 6-semialdehyde | C6 H7 N O3 | 3248 |
| 144.0786 | 1.272 | 3-butyl propionic acid | C7 H12 O3 | 35708 |
| 144.0791 | 1.042 | Hydroxycyclohexane-carboxylic Acid | C7 H12 O3 | 44761 |
| 144.0793 | 0.861 | 3-butyl propionic acid | C7 H12 O3 | 35708 |
| 145.1107 | 1.144 | N-METHYLISOLEUCINE | C7 H15 N O2 | 44542 |
| 145.1108 | 0.899 | 2R-aminohexanoic acid | C7 H15 N O2 | 35932 |
| 148.0523 | 7.59 | p-HYDROXYCINNAMALDEHYDE | C9 H8 O2 | 44643 |
| 149.0482 | 8.168 | N-Acetyl-p-henoquinonimine | C8 H7 N O2 | 578 |
| 149.1056 | 1.38 | Trolamine | C6 H15 N O3 | 43365 |
| 151.0494 | 1.364 | 8-Hydroxyadenine | C5 H5 N5 O | 5527 |
| 152.0333 | 1.181 | Xanthine | C5 H4 N4 O2 | 82 |
| 158.0948 | 0.98 | 3-Oxovalproic acid | C8 H14 O3 | 2990 |
| 161.0843 | 3.488 | Indole-3-ethanol | C10 H11 N O | 6932 |
| 163.0633 | 5.061 | 3-Methyldioxyindole | C9 H9 N O2 | 7024 |
| 163.0634 | 3.487 | 4-(3-Pyridyl)-3-butenic acid | C9 H9 N O2 | 6236 |
| 164.0475 | 1.321 | o-Coumaric acid | C9 H8 O3 | 306 |
| 164.1202 | 13.97 | cis-Jasmone | C11 H16 O | 36077 |
| 165.0651 | 1.075 | 1-Methylguanine | C6 H7 N5 O | 3778 |
| 166.0496 | 2.095 | 3-Methylxanthine | C6 H6 N4 O2 | 2820 |
| 166.0636 | 2.307 | Atrolactic acid | C9 H10 O3 | 5462 |
| 170.1307 | 14.491 | 2-decyllic acid | C10 H18 O2 | 34719 |
| 178.0628 | 20.694 | 8S-hydroxy-2-Decene-4,6-dienoic acid | C10 H10 O3 | 35309 |
| 184.0888 | 5.906 | Benzhydrol | C13 H12 O | 1761 |
| 197.0693 | 4.626 | L-DOPA | C9 H11 N O4 | 42 |
| 200.1414 | 17.7 | 2R-hydroxy-10-undecenoic acid | C11 H20 O3 | 35681 |
| 203.1172 | 1.041 | Isopentenyladenine | C10 H13 N5 | 6612 |
| 205.0747 | 3.487 | deschlorobenzoyl Indomethacin | C11 H11 N O3 | 829 |
| 210.0378 | 1.159 | D-Saccharic acid | C6 H10 O8 | 3343 |
| 211.0848 | 5.366 | 3-O-Methyl-L-DOPA | C10 H13 N O4 | 967 |
| 213.1004 | 1.354 | N-(3-oxo-hexanoyl)-homoserine lactone | C10 H15 N O4 | 36733 |
| 214.1572 | 8.963 | 3R-hydroxy-5Z-dodecenoic acid | C12 H22 O3 | 35609 |
| 214.1572 | 22.39 | 12-hydroxy-10-dodecenoic acid | C12 H22 O3 | 35534 |
| 216.1366 | 14.489 | Undecanedioic acid | C11 H20 O4 | 5846 |
| 219.1107 | 1.186 | Pantothenic Acid | C9 H17 N O5 | 241 |
| 220.0713 | 6.737 | Trifluoromethylphenylpropanediol | C10 H11 F3 O2 | 1958 |
| 228.1476 | 2.824 | Ile Pro | C11 H20 N2 O3 | 23858 |
| 228.148 | 3.125 | 1-L-Leucyl-L-Proline | C11 H20 N2 O3 | 4134 |
| 228.1728 | 17.844 | 10-keto tridecenoic acid | C13 H24 O3 | 35736 |
| 230.1634 | 3.419 | Val Ile | C11 H22 N2 O3 | 23923 |
| 230.2251 | 12.033 | 2,2,9,9-tetramethyl-undecan-1,10-diol | C14 H30 O2 | 36474 |
| X      | Y  | Name                                                                 |
|-------|----|----------------------------------------------------------------------|
| 232.1056 | 2.011 | N2-Succinyl-L-ornithine                                               |
| 232.1423 | 1.294 | Thr Ile                                                              |
| 232.1425 | 1.833 | Ethylenediamine-N,N'-di-a-butyr acid                                  |
| 239.066  | 5.793 | Acyclovir (8-hydroxy-9-(2-hydroxythoxymethyl)guanine                 |
| 240.2093 | 26.223 | 6-pentadecenoic acid                                                |
| 241.1316 | 1.525 | 1,2-Propanediol, 3-[(2-(2-methoxyphenoxy)ethyl]aminoo               |
| 242.1883 | 24.746 | 14-hydroxy-12-tetradecenoic acid                                     |
| 244.1676 | 6.427 | 2-methyl-dodecanedioic acid                                          |
| 244.1788 | 3.881 | Leu Leu                                                              |
| 244.2044 | 14.138 | 2-Hydroxymyristic acid                                               |
| 245.1639 | 4.263 | Pinacidil                                                            |
| 246.104 | 2.556 | Met Pro                                                              |
| 246.1473 | 4.082 | 3-Hydroxydodecanedioic acid                                          |
| 248.1265 | 2.926 | 8-CYCLOPENTYLTHEOPHYLLINE                                            |
| 249.0169 | 1.248 | Alendronate                                                          |
| 250.0524 | 1.207 | Silver sulfadiazine                                                  |
| 252.1148 | 2.04  | Cimetidine                                                           |
| 253.0817 | 5.815 | Monapterin                                                           |
| 253.1339 | 1.146 | 4-(2-hydroxy-3-isopropyl-aminopropyl)benzoic acid                   |
| 254.1267 | 2.621 | MIDODRINE                                                           |
| 255.922  | 0.719 | Trichlorfon                                                         |
| 256.1043 | 12.207 | Dinorpromazine                                                      |
| 258.0885 | 1.429 | 4'-Hydroxyfenoprofen                                                 |
| 260.0504 | 13.971 | Tiaprofenic acid                                                    |
| 260.1376 | 1.198 | Glu Leu                                                             |
| 260.1735 | 1.265 | carisoprodol                                                        |
| 261.1581 | 1.555 | Pinacidil-N-Oxide                                                   |
| 262.0583 | 0.806 | 7-Carboxynalidixic acid                                             |
| 262.0802 | 1.216 | Asp Glu                                                             |
| 264.1475 | 3.759 | Phe Val                                                             |
| 264.1477 | 4.338 | 2H-Indol-2-one, 1,3-dihydro-4-(2-hydroxy-3-((1-methylethyl)aminopropoxy)- |
| 266.1267 | 2.074 | Phe Thr                                                             |
| 266.2249 | 14.488 | 8-heptadecynoic acid                                               |
| 267.0973 | 8.217  | Vidarabine                                                          |
| 267.0975 | 7.206 | Zidovudine                                                          |
| 268.0848 | 14.047 | 2,4-imidazolidinedinedione, 5-(7-oxabicyclo[4.1.0]hepta-2,4-dien-3-yl)-5-phenyl- |
| 268.1536 | 1.177 | Leu His                                                             |
| 268.2404 | 17.706 | 9-heptadecynoic acid                                               |
| 270.085  | 1.604 | Chloramphenicol alcohol                                             |
| 270.2196 | 23.371 | 3-keto palmitic acid                                               |
| 271.1645 | 1.514 | Pro Arg                                                             |
| 272.2355 | 16.203 | 4-hydroxy palmitic acid                                             |
| 273.2671 | 20.115 | C16 Sphinganine                                                     |
| 276.1213 | 7.254  | 4'-Hydroxyxymethoprim                                              |
| 276.1717 | 14.044 | cyclandelate                                                       |
| Mass      | Charge | Molecular Formula                        | MW   | Reference |
|-----------|--------|-----------------------------------------|------|-----------|
| 278.1268  | 2.765  | Tyr Pro                                 | C14 H18 N2 O4 | 23901    |
| 278.1636  | 5.664  | Phe Leu                                 | C15 H22 N2 O3 | 23831    |
| 278.2251  | 13.086 | 9,12,14-octadecatrienoic acid           | C18 H30 O2 | 34823    |
| 280.1421  | 4.027  | Tyr Val                                 | C14 H20 N2 O4 | 23784    |
| 280.2407  | 16.786 | 13E,17-octadecadienoic acid             | C18 H32 O2 | 34967    |
| 282.0186  | 0.727  | 2-Naphthaleneacetic acid, 6-(sulfooxy)- | C12 H10 O6 S | 1381    |
| 282.2561  | 21.022 | 5-octadecenylic acid                    | C18 H34 O2 | 34752    |
| 284.1624  | 4.884  | Dihydroartemisinin                      | C15 H24 O5 | 1119     |
| 284.2357  | 17.894 | 2-oxo-heptadecanoic acid                | C17 H32 O3 | 35798    |
| 286.2151  | 15.503 | Hexadecanedioic acid                    | C16 H30 O4 | 5642     |
| 288.1475  | 11.491 | N-(3-Indolylacetyl)-L-isoleucine         | C16 H20 N2 O3 | 34533    |
| 288.2301  | 22.298 | 10,16-dihydroxy-palmitic acid           | C16 H32 O4 | 35689    |
| 289.1533  | 6.609  | 4-Amino-6,7-dimethoxy-2-(1-piperazinyl)quinazoline | C14 H19 N5 O2 | 2460 |
| 290.1885  | 14.519 | 8-hydroxy-17-octadecene-10,12-dinoic acid | C18 H26 O3 | 35647    |
| 294.1217  | 0.248  | Phe Glu                                 | C14 H18 N2 O5 | 23833    |
| 294.1583  | 3.548  | Ile Tyr                                 | C15 H22 N2 O4 | 23691    |
| 294.1584  | 4.218  | Tyr Ile                                 | C15 H22 N2 O4 | 23905    |
| 294.2196  | 15.517 | 12-oxo-9-octadecynoic acid              | C18 H30 O3 | 35849    |
| 296.1008  | 2.021  | Asp Tyr                                 | C13 H16 N2 O6 | 23882    |
| 296.1191  | 4.577  | Met Phe                                 | C14 H20 N2 O3 S | 23682 |
| 296.1263  | 13.402 | Vomitoxin                               | C15 H20 O6 | 41207    |
| 296.2352  | 20.887 | 13S-hydroxy-9E,11Z-octadecadienoic acid | C18 H32 O3 | 35636    |
| 297.1078  | 2.094  | 1-Methylguanosine                       | C11 H15 N5 O5 | 6324    |
| 297.2667  | 17.366 | (Z)-N-(2-hydroxyethyl)hexadec-7-enamide | C18 H35 N O2 | 3715 |
| 298.1418  | 7.242  | Idebenone Metabolite (Benzenehexanoic acid, 2,5-dihydroxy-3,4-dimethoxy-6-methyl-) | C15 H23 O6 | 772     |
| 298.2146  | 14.805 | 8E-Heptadecenedioic acid                | C17 H30 O4 | 35999    |
| 298.2491  | 18.572 | 8R-hydroxy-9Z-octadecenoic acid         | C18 H34 O3 | 35650    |
| 300.2669  | 15.1   | DL-11-hydroxy stearic acid              | C18 H36 O3 | 35441    |
| 301.0618  | 7.859  | 3-Hydroxy-7-aminoclonazepam             | C15 H12 Cl N3 O2 | 1886 |
| 302.2091  | 14.448 | 10-hydroxy-hexadecan-1,16-dioic acid    | C16 H10 O5 | 36003    |
| 303.1795  | 2.121  | Thr Ile Ala                             | C13 H25 N3 O5 | 17758 |
| 307.0996  | 1.398  | Cys Trp                                | C14 H17 N3 O3 S | 23806 |
| 309.1321  | 4.189  | Ser Phe Gly                             | C14 H19 N3 O5 | 16806    |
| 309.1688  | 1.417  | Tyr Lys                                | C15 H23 N3 O4 | 23796    |
| 310.2138  | 14.082 | 16-hydroperoxy-9Z,12,14E-octadecatrienoic acid | C18 H30 O4 | 35359    |
| 311.1844  | 1.852  | Pro Val Pro                             | C15 H25 N3 O4 | 21539    |
| 311.2826  | 26.275 | (±)-3-Azaprostanolic acid               | C19 H37 N O2 | 43397    |
| 312.2301  | 16.302 | 8,13-dihydroxy-9,11-octadecadienoic acid | C18 H32 O4 | 35506    |
| 312.2302  | 15.328 | 9-hydroxy-10-oxo-12-octadecenoic acid   | C18 H32 O4 | 35510    |
| 314.2459  | 15.131 | Octadecanedioic acid                   | C18 H34 O4 | 5748     |
| 316.2618  | 12.121 | 7,8-dihydroxy stearic acid             | C18 H36 O4 | 35465    |
| 317.1947  | 2.348  | Val Leu Ser                            | C14 H27 N3 O5 | 17706 |
| 317.2932  | 12.175 | Phytosphingosine                       | C18 H39 N O3 | 7066     |
| 320.1339  | 1.14   | Asn Ser Thr                            | C11 H20 N4 O7 | 17468   |
| 321.1792  | 7.922  | Phe Arg                                | C15 H23 N5 O3 | 23741   |
| M/z   | Ret Time | Peak Area | Name                                                                 | Formula | MW  | Conc  |
|-------|----------|-----------|----------------------------------------------------------------------|---------|------|-------|
| 325.2259 | 14.216   | 10-nitro,9Z,12Z-octadecadienoic acid | C18 H31 N O4 | 35951 |
| 326.1339 | 6.214    | His Gly Asn | C12 H18 N6 O5 | 18496 |
| 326.2082 | 13.596   | 2,3-dinor-11β-PGF2α | C18 H30 O5 | 36093 |
| 326.2093 | 13.563   | 2,3-Dinor-8-iso-PGF2α | C18 H30 O5 | 36202 |
| 326.2102 | 14.52    | 2R-hydroperoxy-9Z,12Z,15Z-octadecatrienoic acid | C18 H30 O5 | 36034 |
| 327.1431 | 1.19     | Pro Pro Asp | C14 H21 N3 O6 | 19777 |
| 328.1419 | 5.026    | Phe Tyr | C18 H20 N2 O4 | 23928 |
| 328.2251 | 12.944   | 9S,11R,15S-trihydroxy-2,3-dinor-13E-prostaenoic acid-cyclo[8S,12R] | C18 H32 O5 | 36203 |
| 330.1675 | 5.651    | Iridodial glucoside | C16 H26 O7 | 41180 |
| 330.2405 | 12.662   | 9,10,13-trihydroxy-11-octadecenoic acid | C18 H34 O5 | 35508 |
| 330.2424 | 14.489   | 9,12,13-trihydroxy-10-octadecenoic acid | C18 H34 O5 | 35509 |
| 332.2563 | 15.316   | 18-hydroxy-9S,10R-dihydroxy-steaeric acid | C18 H36 O5 | 36012 |
| 335.1842 | 6.201    | Leu Phe Gly | C17 H25 N5 O4 | 17301 |
| 339.1546 | 12.164   | Ser His Pro | C14 H21 N5 O5 | 20096 |
| 339.1907 | 2.089    | Ala Ile His | C15 H25 N5 O4 | 15676 |
| 341.2933 | 13.028   | N-Acetylsphingosine | C20 H39 N O3 | 411 |
| 342.1116 | 2.977    | Maltose | C12 H22 O11 | 413 |
| 342.2043 | 9.125    | 2,3-dinor, 6-keto-PGF1α | C18 H30 O6 | 36156 |
| 342.2044 | 14.412   | 2,3-Dinor-TXB2 | C18 H30 O6 | 3829 |
| 342.2773 | 20.27    | Eicosanedioic acid | C20 H38 O4 | 35987 |
| 343.2479 | 5.551    | Val Ile Leu | C17 H33 N3 O4 | 15845 |
| 343.3085 | 20.211   | Dihydroceramide C2 | C20 H41 N O3 | 43402 |
| 344.1548 | 0.879    | Methotrexate sulfoxide | C19 H24 N2 O2 S | 1192 |
| 344.2046 | 4.17     | Asn Val Ile | C15 H28 N4 O5 | 19356 |
| 344.2172 | 13.087   | Ile Gly Arg | C14 H28 N6 O4 | 19906 |
| 344.2563 | 19.902   | sn-1,2-Dioctanoylglycerol | C19 H36 O5 | 414 |
| 345.1551 | 2.601    | Thr Pro Glu | C14 H23 N3 O7 | 18357 |
| 345.19  | 1.425    | Glu Val Val | C15 H27 N3 O6 | 19647 |
| 345.2261 | 5.845    | Leu Leu Thr | C16 H31 N3 O5 | 15718 |
| 346.2352 | 19.928   | 9,10-dihydroxy-Octadecanedioic acid | C18 H34 O6 | 35985 |
| 346.2353 | 13.971   | 9-hydroperoxy-12,13-dihydroxy-10-octadecenoic acid | C18 H34 O6 | 35355 |
| 347.0632 | 1.072    | Zidovudine monophosphate | C10 H14 N5 O7 P | 3068 |
| 347.1689 | 2.903    | Thr Asp Ile | C14 H25 N3 O7 | 20490 |
| 348.1649 | 6.213    | Asp Lys Ser | C13 H24 N4 O7 | 22423 |
| 348.2008 | 2.455    | Lys Thr Thr | C14 H28 N4 O6 | 18228 |
| 348.2513 | 12.23    | Satisvic acid | C18 H36 O6 | 35482 |
| 348.2517 | 12.969   | Dodecyl glucoside | C18 H36 O6 | 34493 |
| 349.1749 | 13.967   | His Pro Pro | C16 H23 N5 O4 | 16950 |
| 349.2001 | 6.502    | Ile Ala Phe | C18 H27 N3 O4 | 15688 |
| 351.1893 | 6.761    | His Val Pro | C16 H25 N5 O4 | 20213 |
| 352.1884 | 13.99    | Idebenone Metabolite (QS-10) | C19 H28 O6 | 758 |
| 353.1218 | 2.081    | Asp Tyr Gly | C15 H19 N3 O7 | 23386 |
| 354.2395 | 14.161   | 13,14-dihydro-15-keto-PGF2α | C20 H34 O5 | 36105 |
| 354.2774 | 22.734   | 1-(9Z,12Z-octadecadienoyl)-rac-glycerol | C21 H38 O4 | 4252 |
| Mass   | Charge | Compound                                                                 | Formula     | PDB Code | Chain | Codons  | Score |
|--------|--------|--------------------------------------------------------------------------|-------------|----------|--------|---------|-------|
| 355.3452 | 26.149 | N-(2-hydroxyethyl)icosanamide                                            | C22 H45 N O2 | 3723     |        |         |       |
| 356.2562 | 22.568 | 13,14-dihydro-PGF2alpha                                                  | C20 H36 O5  | 36150    |        |         |       |
| 356.2927 | 15.823 | 1-(11E-octadecenoyl)-rac-glycerol                                        | C21 H40 O4  | 4250     |        |         |       |
| 356.3278 | 16.236 | 13-hydroxy-docosanoic acid                                               | C22 H44 O3  | 35576    |        |         |       |
| 358.2717 | 14.946 | 9,13-dihydroxy-12-ethoxy-10-octadecenoic acid                           | C20 H38 O5  | 35499    |        |         |       |
| 359.2053 | 2.223  | Glu Leu Val                                                              | C16 H29 N3 O6 | 19672   |        |         |       |
| 360.1321 | 8.169  | NITRENDIPINE                                                             | C18 H20 N2 O6 | 44354   |        |         |       |
| 360.178  | 3.585  | Trp Val Gly                                                               | C18 H24 N4 O4 | 18336   |        |         |       |
| 361.1302 | 1.215  | Asp Met Pro                                                               | C14 H23 N3 O6 S | 21062  |        |         |       |
| 365.1945 | 6.199  | Ile Ala Tyr                                                               | C18 H27 N3 O5 | 19800   |        |         |       |
| 366.19   | 2.485  | Lys Tyr Gly                                                               | C17 H26 N4 O5 | 22856   |        |         |       |
| 367.1377 | 2.248  | Gly Tyr Glu                                                               | C16 H21 N3 O7 | 19325   |        |         |       |
| 367.3086 | 14.078 | PGE2alpha dimethyl amine                                                 | C22 H41 N O3 | 36173   |        |         |       |
| 368.1642 | 13.973 | 3-beta-hydroxyandrost-5-en-17-one sulfate                                  | C19 H28 O5 S | 4095    |        |         |       |
| 368.1643 | 13.171 | Testosterone sulfate                                                     | C19 H28 O5 S | 3558    |        |         |       |
| 368.181  | 8.478  | Asn Val His                                                               | C15 H24 N6 O5 | 18456   |        |         |       |
| 368.2171 | 12.158 | Pro Pro Arg                                                               | C16 H28 N6 O4 | 21182   |        |         |       |
| 370.1524 | 6.176  | N-[(diphenylmethoxy)acetyl]-Glutamine                                     | C20 H22 N2 O5 | 2322    |        |         |       |
| 370.1968 | 12.887 | Lys Ser His                                                              | C15 H26 N6 O5 | 20296   |        |         |       |
| 370.2327 | 13.437 | Arg Val Pro                                                               | C16 H30 N6 O4 | 18734   |        |         |       |
| 370.3083 | 19.143 | Docosanedioic acid                                                       | C22 H42 O4  | 35989   |        |         |       |
| 371.14   | 22.565 | Levobunolol sulfate                                                      | C17 H25 N O6 S | 935    |        |         |       |
| 372.2522 | 12.633 | 8,8a-Deoxyoleandolide                                                   | C20 H36 O6  | 40994   |        |         |       |
| 372.3249 | 16.464 | 13,14-dihydroxy-docosanoic acid                                          | C22 H44 O4  | 35577   |        |         |       |
| 373.1071 | 4.611  | Cys Asp His                                                              | C13 H19 N5 O6 S | 19299  |        |         |       |
| 373.1746 | 2.429  | His Ala Phe                                                              | C18 H23 N5 O4 | 15997   |        |         |       |
| 374.2824 | 14.865 | 3beta-Hydroxycholesterol-4-en-24-oic Acid                                | C24 H38 O3  | 42785   |        |         |       |
| 379.1044 | 0.802  | Asp Met Asp                                                              | C13 H21 N3 O8 S | 18369  |        |         |       |
| 382.1969 | 12.606 | Asn His Leu                                                              | C16 H26 N6 O5 | 20737  |        |         |       |
| 382.343  | 12.87  | 24-hydroxy-10Z-tetracosenoic acid                                        | C24 H46 O3  | 35581   |        |         |       |
| 383.1415 | 7.857  | N-acetylglactosamine                                                     | C14 H25 N O11 | 4231   |        |         |       |
| 384.2122 | 14.312 | Thr His Lys                                                               | C16 H28 N6 O5 | 19678   |        |         |       |
| 384.231  | 14.136 | 9a-Fluoroalloetetrahydrocortisol                                         | C21 H33 F O5 | 2717    |        |         |       |
| 384.359  | 18.438 | 24-hydroxy-tetracosanoic acid                                            | C24 H48 O3  | 35579   |        |         |       |
| 388.1632 | 10.844 | Nisoldipine                                                              | C20 H24 N2 O6 | 1551    |        |         |       |
| 388.2309 | 4.621  | Glu Ile Lys                                                               | C17 H32 N4 O6 | 16755   |        |         |       |
| 388.2438 | 11.38  | Arg Leu Thr                                                              | C16 H32 N6 O5 | 22752   |        |         |       |
| 390.1511 | 6.209  | Loganin                                                                  | C17 H26 O10 | 41146   |        |         |       |
| 390.1763 | 1.343  | Asp Lys Glu                                                               | C15 H26 N4 O8 | 21250   |        |         |       |
| 390.2776 | 12.586 | 12beta-Hydroxy-3-oxo-5beta-cholan-24-oic Acid                            | C24 H38 O4  | 42745   |        |         |       |
| 391.2465 | 8.053  | Leu Ile Phe                                                              | C21 H33 N3 O4 | 16864   |        |         |       |
| 393.1898 | 5.086  | Phe Ile Asp                                                               | C19 H27 N3 O6 | 21641   |        |         |       |
| 394.1497 | 4.97   | Phe Asn Asp                                                              | C17 H22 N4 O7 | 23445   |        |         |       |
| 398.1916 | 13.066 | Asp His Lys                                                              | C16 H26 N6 O6 | 22066   |        |         |       |
| 400.0169 | 1.173  | Octulose-1,8-bisphosphate                                                | C8 H18 O14 P2 | 157     |        |         |       |
| M.W.   | Retention Time | Name                                                                 | Molecular Formula | Mass  | Log P |
|-------|---------------|----------------------------------------------------------------------|-------------------|-------|-------|
| 400.2072 | 12.832        | Arg Pro Glu                                                           | C16 H28 N6 O6     | 18852 |       |
| 402.0963 | 6.212         | 4-O-Demethyl-13-dihydroadriamycinone                                   | C20 H18 O9        | 664   |       |
| 402.261  | 8.381         | Lys Gin Lys                                                           | C17 H34 N6 O5     | 22767 |       |
| 402.2959 | 22.093        | Lys Lys                                                               | C18 H38 N6 O4     | 18451 |       |
| 404.0019 | 1.637         | Uridine diphosphate (UDP)                                              | C9 H14 N2 O12 P2  | 97    |       |
| 404.2058 | 1.364         | Flunanizine                                                           | C26 H26 F2 N2     | 2731  |       |
| 405.2268 | 14.042        | Lisinopril                                                            | C21 H31 N3 O5     | 1009  |       |
| 407.1978 | 4.582         | Asn Gln Phe                                                           | C18 H25 N5 O6     | 20970 |       |
| 407.2047 | 2.035         | Leu Glu Phe                                                           | C20 H29 N3 O6     | 15774 |       |
| 408.2857 | 14.462        | 3beta,6alpha,7beta-Trihydroxy-5beta-cholan-24-oic Acid               | C24 H40 O5        | 42672 |       |
| 408.2874 | 16.041        | 3beta,6beta,7alpha-Trihydroxy-5beta-cholan-24-oic Acid               | C24 H40 O5        | 42673 |       |
| 410.1431 | 21.11         | 2-hydroxy-hexacosanoic acid                                           | C26 H52 O3        | 35584 |       |
| 410.1741 | 14.876        | ROTENONIC ACID, METHYL ETHER                                          | C24 H26 O6        | 44195 |       |
| 412.1918 | 18.877        | 17alpha,20beta-Hydroxyprogesterone sulfate                            | C21 H32 O6 S      | 3562  |       |
| 412.3903 | 21.11         | 2-hydroxy-hexacosanoic acid                                           | C26 H52 O3        | 35584 |       |
| 414.2272 | 10.031        | Pro Trp Leu                                                           | C22 H30 N4 O4     | 16187 |       |
| 416.1585 | 8.421         | Dehydronomodiopine                                                    | C21 H24 N2 O7     | 1543  |       |
| 417.1642 | 2.148         | Phe His Asp                                                           | C19 H23 N5 O6     | 20877 |       |
| 417.235  | 1.9           | Arg Lys Asp                                                           | C16 H31 N7 O6     | 17233 |       |
| 418.1845 | 5.696         | Val Asp Trp                                                           | C20 H26 N4 O6     | 23096 |       |
| 418.3081 | 25.607        | (24S)-1alpha,24-dihydroxy-22-oxativamin D3 7/(24S)-1alpha,24-dihydroxy-22-oxacholesterol | C26 H42 O4        | 41998 |       |
| 420.2476 | 2.279         | Val Arg Phe                                                           | C20 H32 N6 O4     | 19820 |       |
| 420.3202 | 13.378        | (5Z)-4,4-difluorovitamin D3 7/(5Z)-4,4-difluorocholecalciferol         | C27 H42 F2 O      | 42074 |       |
| 422.0821 | 0.881         | alpha,alpha'-Trehalose 6-phosphate                                   | C12 H23 O14 P     | 3529  |       |
| 423.2   | 4.065         | Tyr Glu Ile                                                           | C20 H29 N3 O7     | 15868 |       |
| 424.2543 | 4.669         | His Arg Ile                                                           | C18 H32 N8 O4     | 7641  |       |
| 427.029  | 1.231         | Zidovudine diphasate                                                  | C10 H15 N5 O10 P2 | 3069  |       |
| 432.1527 | 6.536         | BMPN-benzoic acid glucuronide                                         | C21 H24 N2 O8     | 682   |       |
| 436.2055 | 26.78         | Asp Phe Arg                                                           | C19 H28 N6 O6     | 17786 |       |
| 437.17  | 14.377        | 7-Hydroxydooxazosin                                                   | C22 H23 N5 O5     | 2456  |       |
| Breeds                        | Shami                              | Saanen                            |
|------------------------------|------------------------------------|------------------------------------|
| Drugs                        | Pinacidil                          | Pinacidil                          |
|                              | Spiramycin 3                       | Spiramycin 3                       |
|                              | Deschlorobenzoyl Indomethacin      | Deschlorobenzoyl Indomethacin      |
|                              | Methotrimeprazine sulfoxide        |                                    |
| Microbial metabolism in      | 3-Methylxanthine                   | 3-Methylxanthine                   |
| diverse environments         | Isopentenyladenine                 | Isopentenyladenine                 |
| including microbial          | -                                  | Atrolactic acid                    |
| secondary metabolites        | -                                  | Isopentenyladenine                 |
|                              | -                                  | Benzoic acid                       |
|                              | -                                  | D-Saccharic acid                   |
| Environmental contaminants   | Trichlorfon                        | Trichlorfon                        |
| Naturally occurring           | Asp Phe Arg                        | Asp Phe Arg                        |
| metabolites                  | Selenocysteine                     | -                                  |
|                              | Maltose                            | Pantothenic acid                   |
|                              | 3-butyrl propionic acid            | 3-butyrl propionic acid            |
|                              | GPEtn(18:1(9Z)/0:0)                | GPEtn(18:1(9Z)/0:0)                |
|                              | GPGroP(16:0/18:1(9Z))              | -                                  |
|                              | Neu5Ac alpha2-6Galbeta1-4Glcbeta-Sp| Neu5Ac alpha2-6Galbeta1-4Glcbeta-Sp|
|                              | 1,4-Methylimidazoleacetic acid     |                                    |
| Plant secondary metabolites  | 4-Hydroxystyrene                   | -                                  |
| Others                       | 3-Hydroxydodecanedioic acid (Disorder)| - | 13S-hydroxy-9E,11Z-octadecadienoic acid (Human skin tanning) |
|                              |                                   | 13,14-dihydro-15-keto-PGF2 alpha (hormone) |
|                              |                                   | 9,12,13-trihydroxy-10-octadecenoic acid (Anti-stress substance/antifungal) |
|                              | Laminaribiose (plant saccharide)  | -                                  |
|                              | 5′ guanylate diphosphate (Purine metabolism in microbes) | - |