Supplementary Table 1.

Table… The predicted metabolites of compounds found in BM using BioTarnsformer 3.0

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Bacopasaporin C Metabolites** | | | | |
| Predicted Result (structure and name) | Reaction Type | Reaction information | MWt | Biotransformation Reaction |
| 5-[3,4-dihydroxy-5-(hydroxymethyl)oxolan-2-yl]oxy-6-[[16-hydroxy-2,6,6,10,16-pentamethyl-17-(2-methylprop-1-enyl)-19,21-dioxahexacyclo[18.2.1.01,14.02,11.05,10.015,20]tricosan-7-yl]oxy]oxane-3,4-diol | Glycoside hydrolysis | Enzyme: Glycosidase  BioSystem: HUMAN | 736.4397 |  |
| 6-(hydroxymethyl)oxane-2,3,4,5-tetrol | Glycoside hydrolysis | Enzyme: Glycosidase  BioSystem: HUMAN | 180.0633 |  |
| 4,5-dihydroxy-6-({4-hydroxy-2-[(2-{[16-hydroxy-2,10,16-trimethyl-17-(2-methylprop-1-en-1-yl)-19,21-dioxahexacyclo[18.2.1.0¹,¹⁴.0²,¹¹.0⁵,¹⁰.0¹⁵,²⁰]tricosan-7-yl]oxy}-4-{[6-(hydroxymethyl)oxan-2-yl]oxy}-5-methyloxan-3-yl)oxy]-5-(hydroxymethyl)oxolan-3-yl}oxy)oxane-2-carboxylic acid | Alkyl-OH-glucuronidation | Enzyme: UDP-glucuronosyltransferase  BioSystem: HUMAN | 1074.524 |  |
| 3,4,5-trihydroxy-6-({5-[(5-hydroxy-2-{[5-hydroxy-5,11,15,15,19-pentamethyl-4-(2-methylprop-1-en-1-yl)-2,22-dioxahexacyclo[19.1.1.0¹,⁶.0⁷,²⁰.0¹⁰,¹⁹.0¹¹,¹⁶]tricosan-14-yl]oxy}-4-[(2,3,4,5-tetrahydroxycyclohexyl)oxy]oxan-3-yl)oxy]-2-(hydroxymethyl)-4-methyloxolan-3-yl}oxy)oxane-2-carboxylic acid | Alkyl-OH-glucuronidation | Enzyme: UDP-glucuronosyltransferase  BioSystem: HUMAN | 1074.524 |  |
| 3-({3,4-dihydroxy-5-[(5-hydroxy-2-{[16-hydroxy-2,6,6,10,16-pentamethyl-17-(2-methylprop-1-en-1-yl)-19,21-dioxahexacyclo[18.2.1.0¹,¹⁴.0²,¹¹.0⁵,¹⁰.0¹⁵,²⁰]tricosan-7-yl]oxy}-4-{[3,4,5-trihydroxy-6-(hydroxymethyl)oxan-2-yl]oxy}oxan-3-yl)oxy]oxolan-2-yl}methoxy)-4,5,6-trihydroxycyclohex-1-ene-1-carboxylic acid | Alkyl-OH-glucuronidation | Enzyme: UDP-glucuronosyltransferase  BioSystem: HUMAN | 1074.524 |  |
| ({3,4-dihydroxy-5-[(5-hydroxy-2-{[16-hydroxy-6,6,16-trimethyl-17-(2-methylprop-1-en-1-yl)-19,21-dioxahexacyclo[18.2.1.0¹,¹⁴.0²,¹¹.0⁵,¹⁰.0¹⁵,²⁰]tricosan-7-yl]oxy}-4-{[3,4,5-trihydroxy-6-(hydroxymethyl)oxan-2-yl]oxy}oxan-3-yl)oxy]oxolan-2-yl}methoxy)sulfonic acid | Sulfation of primary alcohol | Enzyme: Alcohol sulfotransferase  BioSystem: HUMAN | 978.4494 |  |
| **Bacopaside II Metabolites** | | | | |
|  |  |  |  |  |
| **Bacoside A Metabolites** | | | | |
| 7-hydroxy-1-(2-hydroxy-6-methylhept-5-en-2-yl)-9a-(hydroxymethyl)-6,6-dimethyl-hexadecahydro-1H-cyclopenta[a]phenanthren-2-one | Glycoside hydrolysis | Enzyme: Glycosidase BioSystem: HUMAN | 474.3709 |  |
| 6-ethyl-5-[(3,4,5-trihydroxyoxan-2-yl)oxy]oxane-2,3,4-triol | Glycoside hydrolysis | Enzyme: Glycosidase BioSystem: HUMAN | 312.1056 |  |
| **Bacopaside X Metabolites** | | | | |
| 5-{[3,4-dihydroxy-5-(hydroxymethyl)oxolan-2-yl]oxy}-6-{[16-hydroxy-2,6,6,10,16-pentamethyl-18-(2-methylprop-1-en-1-yl)-21-oxahexacyclo[18.2.1.0¹,¹⁴.0²,¹¹.0⁵,¹⁰.0¹⁵,²⁰]tricosan-7-yl]oxy}oxane-3,4-diol | Glycoside hydrolysis | Enzyme: Glycosidase  BioSystem: HUMAN | 736.4397 |  |
| 6-(hydroxymethyl)oxane-2,3,4,5-tetrol | Glycoside hydrolysis | Enzyme: Glycosidase  BioSystem: HUMAN | 180.0633 |  |
| 6-({7-[(3-{[3,4-dihydroxy-5-(hydroxymethyl)oxolan-2-yl]oxy}-5-hydroxy-4-{[3,4,5-trihydroxy-6-(hydroxymethyl)oxan-2-yl]oxy}oxan-2-yl)oxy]-2,6,6,10,16-pentamethyl-18-(2-methylprop-1-en-1-yl)-19,21-dioxahexacyclo[18.2.1.0¹,¹⁴.0²,¹¹.0⁵,¹⁰.0¹⁵,²⁰]tricosan-16-yl}oxy)-3,4,5-trihydroxyoxane-2-carboxylic acid | Alkyl-OH-glucuronidation | Enzyme: UDP-glucuronosyltransferase BioSystem: HUMAN | 1074.52 |  |
| 3,4,5-trihydroxy-6-({4-hydroxy-2-[(5-hydroxy-2-{[16-hydroxy-2,6,6,10,16-pentamethyl-18-(2-methylprop-1-en-1-yl)-19,21-dioxahexacyclo[18.2.1.0¹,¹⁴.0²,¹¹.0⁵,¹⁰.0¹⁵,²⁰]tricosan-7-yl]oxy}-4-{[3,4,5-trihydroxy-6-(hydroxymethyl)oxan-2-yl]oxy}oxan-3-yl)methyl]-5-(hydroxymethyl)oxolan-3-yl}oxy)oxane-2-carboxylic acid | Alkyl-OH-glucuronidation | Enzyme: UDP-glucuronosyltransferase  BioSystem: HUMAN | 1074.524 |  |
| 3,4,5-trihydroxy-6-({4-[(5-hydroxy-2-{[5-hydroxy-5-methyl-3-(2-methylprop-1-en-1-yl)-2,22-dioxahexacyclo[19.1.1.0¹,⁶.0⁷,²⁰.0¹⁰,¹⁹.0¹¹,¹⁶]tricosan-14-yl]oxy}-4-{[2,3,4-trihydroxy-5-(hydroxymethyl)cyclohexyl]methyl}oxan-3-yl)oxy]-2-(hydroxymethyl)cyclopentyl}oxy)oxane-2-carboxylic acid | Alkyl-OH-glucuronidation | Enzyme: UDP-glucuronosyltransferase  BioSystem: HUMAN | 1074.524 |  |

Supplementary Figure 1.



Supplaemenatry figure 1. HPLC confirmation of BM. **A** HPLC analysis of standards Bacoside A3, Bacopaside II, Bacopaside X and Bacopasaponin C, With peaks for each between 20 and 25 minutes **B** HPLC analysis of BM showing matching peaks between 20 to 25 minutes retention time.