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LC-ESI-MS/GC-MS Based Metabolite Profiling of Chlorophytum comosum (Thunb.) Jaques and evaluation of its antioxidant and antiproliferative effects on lung and breast cancer cell lines

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2	(Thunb.) Jaques and evaluation of its antioxidant and antiproliferative effects on
3	lung and breast cancer cell lines
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26 LC-ESI-MS/GC-MS Based Metabolite Profiling of *Chlorophytum comosum*27 (Thunb.) Jaques and evaluation of its antioxidant and antiproliferative effects on
28 lung and breast cancer cell lines

29 Abstract

Background *Chlorophytum comosum* popularly known as Spider Ivy is an important medicinal plant in traditional Chinese medicine utilized in the treatment of many ailments, however its detailed chemical composition and biological activity is not much explored. The present study aims to identify different chemical constituents present in roots and leaves of *Chlorophytum comosum* and investigates its antioxidant, antiproliferative and haemolytic effects on breast (MCF-7) and lung cancer cell lines (A549, H1299) as compared to normal lung (L-132) cell lines.

37 Methods Chemical constituents from aqueous roots and leaves extracts were identified 38 using LC-ESI-MS/GC-MS. The identified compounds were annotated based on match of 39 mass spectral database with the literature using NIST 14 and METLIN databases. 40 Antioxidant activity was checked using DPPH, FRAP and TPC assays. The 41 antiproliferative effects of ethanolic roots and leaf extracts of Chlorophytum comosum 42 were measured by MTT assay on breast (MCF-7), lung cancer (A549 & H1299) and 43 normal lung (L-132) cell lines. The toxicity studies of the extracts were carried out using 44 Haemolytic assay.

45 **Results** GC-MS analysis identified 34 new metabolites in roots and 17 from leaves, while 46 as 17 compounds from roots and 7 from leaves were detected by LC-ESI-MS. Significant 47 antiproliferative effects were observed on MCF-7 & A549 cell lines with IC₅₀ values 48 ranging from $31.94 \mu g/ml$ to $77.84 \mu g/ml$ while no marked response was observed against 49 normal cell line. Haemolysis studies revealed no significant toxicity of the extracts 50 towards the biological system.

51 Conclusion Our study is the first preliminary report on the detailed chemical composition 52 and antiproliferative potential of *Chlorophytum comosum*, indicating significant specific 53 antiproliferative activities against lung (A549) and breast (MCF-7) cancer cell lines. 54 However, further studies are required to understand the mechanism involved in cytotoxic 55 properties of *Chlorophytum comosum*.

56 Keywords: GC-MS, LC-ESI-MS, *Chlorophytum comosum*, A549, MCF-7,
57 Phytochemicals, Cytotoxicity.

58 Background

59 Chlorophytum comosum (Thunb.) Jacques commonly known as Spider Ivy belongs to 60 family Lilliaceae and genus Chlorophytum. It is a popular ornamental plant and is 61 considered to be an excellent air purifier and air detoxifier [1-2]. In traditional Chinese 62 medicine, it is used for the treatment of respiratory ailments such as Bronchitis, Asthma 63 [3-4], Fractures and Burns [3]. Saponins isolated from roots of Chlorophytum comosum 64 have been found to exhibit cytotoxic and antitumour promoter activity in selected cancer 65 cell lines [5-6]. However the detailed information on plant phyto-composition is still 66 lacking. Despite having the ethnobotanical background validated by previous findings, 67 not many efforts have been taken to explore this plant for its therapeutic active 68 constituents. In this study we reported for the first time the comparative and 69 comprehensive chemical profiling of *Chlorophytum comosum* through GC/MS, LC-ESI-70 MS method and evaluated their antioxidant and antiproliferative potential using in vitro 71 assays. The newer compounds identified were matched using mass spectral database 72 search by NIST and METLIN [7] .The antiproliferative activity of ethanolic roots 73 (CCRE) and leaf (CCLE) extracts were checked by MTT assay on breast (MCF-7) and 74 lung (A549 & H1299) and normal lung cell line (L-132).

76 Methods

77 Plant Material

Plant material was obtained from the herbal garden of Jamia Hamdard. The harvested roots and leaves were fully mature, healthy and free from disease. The plant samples were identified by the botanist Dr. Sunita Garg (NISCAIR, New Delhi) and the voucher specimens bearing no. NISCAIR/RHMD/CONSULT/ 2016/2975-02 were deposited in the herbarium.

83 Chemicals and Reagents

84 All the solvents used for the study i.e. Methanol, n-Butanol, Petroleum Ether, Water, 85 Formic Acid, Acetonitrile were of HPLC/LC-MS grade and were purchased from Merck 86 (Darmstadt, Germany). Ethanol and chemicals (DPPH, TPTZ, L-Ascorbic acid, FeCl₃, 87 and Na₂CO₃) were of the highest purity and were purchased from the local commercial 88 supplies. Cell culture media DMEM with phenol red (#1932403), Fetal Bovine Serum (# 89 10438034), Trypsin –EDTA solution with phenol red (#1897336) and antibiotic solution 90 PenStrep (#192493) were purchased from Gibco USA. MTT reagent (#MICB8173V), 91 Standard Vinblastine, Folin Ciocalteau reagent were obtained from Sigma Aldrich USA. Hydrogen peroxide, Gallic acid and DMSO were purchased from Thomas Baker India. All 92 93 the cell lines used in the study were procured from ATCC (USA).

94 **Preparation of Plant Extracts**

95 *Chlorophytum comosum* leaves aqueous (CCLA), ethanolic (CCLE) and *Chlorophytum* 96 *comosum* roots aqueous (CCRA), ethanolic fractions (CCRE) were prepared using soxhlet 97 extraction method. Briefly, the plant materials were washed to remove debris, air-dried and 98 grinded. The grinded plant material (10gm) was defatted using petroleum ether (1:15 w/v), 99 suspended in water/ethanol (150mL) and extracted for 24 hrs at 50^oC. The extracts were 100 then collected, filtered, and processed to become colourless using n-butanol, concentrated 101 on a rotary evaporator at 55°C and 55mbar pressure until the semi-dried substance was
102 obtained. The semi-dried substance was further lyophilized to remove the traces of solvent.

103 The samples were then stored in airtight vials at 4^{0} C till further use.

104 Gas Chromatography/Mass Spectrometry (GC-MS) Analysis

105 To conduct the GC-MS analysis, aqueous leaf and root extracts (5 mg each) were dissolved 106 in 1 mL of methanol and filtered using 0.22-micron filter to utilize in further analytical 107 procedures. GC-MS analysis was carried out on Shimadzu GCMS QP-2010 plus system 108 using chromatographic separation column (Rtx-5 Sil MS column; 30 m×0.32 mm id 109 x0.25µm film thickness). The operating conditions of the column included oven 110 temperature program from 80°C to 210°C at 4°C/min withhold time of 2 min and from 111 210°C to 300°C at 15°C/min withhold time of 5 min, and the final temperature was kept 112 for 20 min. The injector temperature was maintained at 270°C, the volume of injected 113 sample was 0.3 µL, pressure 85.4kPa, total flow 76.8 mL/min, column flow 1.21 mL/min, 114 linear velocity 40.5 cm/sec, purge flow 3.0 mL/min, split ratio: 60.0, ion source 115 temperature 230°C, scan mass range of m/z 40-600 and interface line temperature 280°C. 116 The peak area based on retention time was expressed as percentage composition of the 117 crude extract. The identification and characterization of compounds were performed by 118 comparing their mass spectra with data from NIST 14 (National Institute of Standards and 119 Technology, US) and WILEY 8 library.

Liquid Chromatography Mass Spectrometry (LC-ESI-MS) analysis in positive ion mode

122 LC-MS is the high-end technology which is increasingly employed in plant analytical 123 research [7]. LC-MS enables the separation, identification and characterization of 124 phytoconstituents present in complex crude plant extracts by giving high chemical 125 specificity and sensitivity. Qualitative or quantitative estimation of known/unknown

126 compounds present in the crude mixture can be deciphered using LC-MS [7]. Briefly 127 aqueous extracts CCLA and CCRA were prepared in LCMS grade methanol and analysed on QSM-LC-MS electrospray ionization (ESI) single quadrupole mass spectrometer 128 (Waters, Milford-MA) coupled with Synapt G2 using BEH C18, 1.7µm - 2.1 x 50 mm 129 column on positive ion mode (ES+) with following mobile phases: (A) 0.1% formic acid 130 131 (B) Acetonitrile (C) Methanol (sample input and acquisition; 5µL /min flow rate and 250 132 µL injection volume). The parameters for analysis were as follows: source temperature 1000°C, cone voltage 40 eV, capillary voltage 3 kV, desolvation temperature 3500°C, cone 133 gas flow 50 l/h, and desolvation gas flow 900 l/h. Mass spectra were detected in the ESI 134 135 positive ion mode between m/z 100-1000 with a scan time of 0.40/sec for 20 minutes. 136 Leucine Enkephalin was used as a reference material for mass spectrometer tuning and calibration. The data generated by LC-ESI-MS was analysed using an open source platform 137 138 XCMS software combined with METLIN [8].

139 DPPH Free Radical Scavenging Activity

140 The free radical scavenging activity of *Chlorophytum comosum* aqueous leaf (CCLA) and 141 roots extract (CCRA) were measured following methodology given by [9] with slight 142 modifications. Briefly, 200 mmol L⁻¹ of DPPH solution was prepared by dissolving 1.57g 143 of DPPH in 20 mL of methanol and was stored in dark. Different concentrations (1000 144 µg/mL-62.5 µg/mL) were prepared separately on a 96 wells microplate by serial dilution 145 (Stock conc. 100mg/mL, working conc. 1mg/mL). Blank was prepared using methanol. 146 Control was prepared by adding 200 µL of DPPH in 50 µL of water. The reaction was 147 initiated by adding 200 µL of DPPH in 50 µL of the extract of varied concentrations followed 148 by an incubation period of 60 minutes. The absorbance was recorded on ELISA microplate 149 reader at 515nm. Ascorbic acid was taken as standard and tested with the same 150 concentrations as extracts. The experiments were repeated 3 times and % DPPH free radical 151 scavenging activity of the extracts was measured using the following formula and values 152 were expressed as mean \pm SD.

153 % DPPH scavenging = $[(C - S) \times 100] / C.... (Eq.-1)$

154 C= Absorbance of control;

155 S= Absorbance of samples.

156

157 Ferric Reducing Antioxidant Power Assay (FRAP)

158 FRAP is an electron transfer based assay which monitors the reaction of Fe²⁺ with TPTZ to 159 form a violet-blue color from colorless oxidized Fe^{3+.} The reducing ability of the extracts 160 were measured by the method given by [10] with some modifications. Briefly different 161 concentrations of the extracts were prepared by serial dilution (1000 μ g/mL to 62.5 μ g/mL) 162 on 96 wells microplate (Stock conc. 100mg/mL, working conc. 1mg/mL). The standard 163 curve was prepared using Ascorbic acid. FRAP working solution was prepared by adding 164 10 volume of acetate buffer (300 mM, pH- 3.6, 20mL) with 1 volume of TPTZ (40mM 165 prepared in dil. HCL, 2mL) and 1 volume of FeCl₃ (20mM, 2mL) solution. The reaction 166 mixture were mixed well and stored at 37^oC. Deionized milli q water served as blank while 167 control contained FRAP solution alone. 50 μ L of the extract were mixed with 240 μ L of 168 FRAP solution on 96 well microplate and incubated in dark for 30 minutes. The absorbance 169 was measured at 593nm. FRAP values were obtained by comparing the absorption change 170 in test mixture (triplicate values) with the standard.

171 Determination of Total Phenolic Contents (TPCs)

Total phenolic contents of the aqueous extracts were measured as described by [9] with slight modifications. Folin Ciocalteau (FC) reagent was diluted in 1:1 ratio (v/v) in double distilled water (DDW). 25 μ L of each sample (stock conc. 100mg/mL) was loaded on to the 96 well plate and diluted 4 times by adding 75 μ L of DDW. To the diluted samples, 176 25 μ L of the FC reagent was added and incubated for 6 min, then 100 μ L of 75 g L⁻¹ 177 Na₂CO₃ was added. The solution was placed in the dark for 30 min at room temperature 178 for color development. After incubation the absorbance were taken at 765 nm. Acetone 179 served as blank. Gallic acid was used as a standard (1000 μ g/mL-62.5 μ g/mL) to produce 180 a calibration curve (average R² = 0.9990). Each sample was run in duplicate. Total 181 phenolic concentration was expressed as mg gallic acid equivalent (GAE) g⁻¹ dried 182 sample.

183 MTT Assay

184 The antiproliferative effects of ethanolic roots (CCRE) and leaf extracts (CCRE) were 185 measured using MTT assay. The cytotoxic effect of CCLE and CCRE on MCF-7(Breast) 186 and A549, H1299 (Lung) cancer cell lines were investigated and compared against normal 187 human epithelial cell line (L-132) for the selective response. Cell lines were procured from 188 ATCC, USA and were maintained at 37°C with 5% CO₂ in an incubator using DMEM 189 complete medium with 10% FBS and 1% antibiotic solution. Briefly, cells were seeded with 190 a cell density of 40,000 cells per well into 96 well culture plate. Cells were treated with 191 varying extract concentrations (10-320 µg/mL) for 24hrs. Vinblastine was used as positive 192 control in the study and was exposed to cells at varying concentrations from 3.12-100 µM. 193 After 24 hrs of the treatment, cells were treated with 10 µL MTT solution (0.5mg/mL) and 194 further incubated for 4 hrs, the medium was then discarded and the formazan crystals were 195 dissolved using 100 µl of DMSO solution. The absorbance was measured at 570nm using 196 Spectra max spectrophotometer. The % cell inhibition were measured by using the following 197 formula (Eq.-2). The concentration at which the test drug inhibited cell growth by 50% i.e. 198 inhibitory concentration (IC_{50}), is generated from the dose-response curves using Graph Pad 199 Prism software (8.1). The final concentration of DMSO was maintained at 0.1% in all the 200 test preparations. 1 % DMSO containing cells served as vehicle control.

201

% Cell Inhibition by CCLE /CCRE extracts = $[(C - S) \times 100] / C \dots (Eq.-2)$

202

C= Absorbance of control; S= Absorbance of samples.

203 Haemolysis Assay

204 The haemolytic activity of the extracts was measured by colorimetric assay as described 205 previously by [11] at 594 nm. Briefly, 5mL of fresh human blood was collected in K₃ EDTA 206 vacutainer tubes and was centrifuged at 1000 rpm for 10 min at 4^oC. The supernatant 207 containing plasma was carefully removed and the white buffy layer was aspirated with 208 precautions and discarded into 3% hydrogen peroxide solution. The erythrocytes were then 209 washed thrice with 1X PBS, pH 7.4. Washed erythrocytes were diluted twice (1:2) with 1X 210 PBS. 50 µL of the diluted erythrocytes suspension were taken into 2 mL Eppendorf tube and 211 100 µL of ethanolic root and leaves extract of different concentrations ranging from 10 212 µg/mL-320 µg/mL were added. 100 µL of 1X PBS was used as negative control while 100 213 µL of 1% SDS served as positive control The reaction mixture was incubated at 37° C for 214 60 min. After incubation the volume was made upto 1 mL by adding 850µL of 1X PBS. 215 Finally centrifuged at 300 rpm for 3 min and the resulting haemoglobin in supernatant was 216 measured at 540 nm by spectrophotometer to determine the concentration of haemoglobin. 217 The percent haemolytic activity of the extracts were expressed using (Eq.-3).

- 218 % Haemolytic activity of extract = $[(C S) \times 100] / C.....$ (Eq.-3).
- 219 C= Absorbance of control;
- 220 S= Absorbance of samples.

221 Statistical Analysis

222 IC₅₀ values for cytotoxicity tests were derived from nonlinear regression analysis (curve 223 fit) based on sigmoid dose-response curve (variable) and computed using Graph Pad Prism 224 8.1 (San Diego, CA, USA). The data were expressed as mean \pm SD of three independent 225 experiments. Differences were considered statistically significant at p < 0.05.

226 **Results**

227 Metabolite profiling of leaf and roots of Chlorophytum comosum by GC-MS

228 The qualitative and quantitative analysis of the active constituents present in aqueous roots 229 and leaf extracts of Chlorophytum comosum were performed using GC/MS. Thirty four 230 new compounds were identified in leaf extracts (CCLA) while 17 metabolites were 231 observed in root extracts (CCRA). The identified phytoconstituents belonged to different 232 classes of phenols, triterpenes, sterols, alcohols, aldehydes, fatty acids, saponins, organic 233 acids and hydrocarbons. Significant differences in chemical composition between roots 234 and leaf extracts were observed. Phenolic compounds such as Guaiacol (1.38%), 4vinylguaiacol (1.28%), Syringol (1.08%), and 3-hydroybenzyl alcohol (7.59%) were 235 236 observed in the leaf extract (Table 1). On the contrary the saponins of various class such 237 as Tigogenin tosylate (4.43%), Neotigogenin (16.25%), Yuccagenone (14.05%) were 238 found as the major portion of the root part (Table 2). However, saponins like 239 Sarsasapogenin (7.54%) and Diosgenin (1.27%) were detected in leaf as well. An 240 important plant sterol β - Sitosterol was found to be present in higher amounts in roots 241 (7.79%) as compared to leaf (1.24%). Similarly, the fatty acids were also present in higher 242 amounts in roots as compared to leaves. Polysaccharides were also identified in leaf but 243 found to be absent in roots. The % content of metabolites was expressed in terms of peak 244 area depending upon retention time. Identification of the components was achieved based 245 on the retention indices match and fragmentation pattern with the database available in 246 NIST14 library.

247 Metabolite profiling of leaf and roots of *Chlorophytum comosum* by LC-ESI-MS in 248 positive ion mode

LC-ESI-MS analysis of CCLA and CCRA extracts led to the tentative identification ofcompounds detected in positive ion mode using METLIN database batch search with an

251 accuracy of 10ppm. The identified compounds were annotated based on match of mass 252 spectral database with the literature and were selected from the database based on 253 possibility of availability and solubility in crude mixture. 17 metabolites were detected in 254 roots among which Purpurogallin [12], Ginsenoyne B [13], Sagecoumarin [14] Gnidicin 255 [15] possess important pharmacological functions. In leaf 7 metabolites annotated, were 256 fatty acids like Oleyl Anilide a known ACAT inhibitor [16], 6-Bromo-5E,9Z,13Z-257 docosatrienoic acid, 3-(2-Heptenyloxy)-2-hydroxypropyl undecanoate, 5,6-Dichloro-1,3-258 cyclohexadiene, alkaloid Serratanidine, Salicylic acid beta-D-glucoside and Methyl (9Z)-259 6'-oxo-6,5'-diapo-6-carotenoate. The peak pattern of metabolites separated in positive ion 260 mode [M+H] in roots and leaves are depicted in Table (3 & 4). LC-ESI-MS total 261 chromatogram is presented in (Figure 1).

262 Antioxidant activity by Chlorophytum comosum

263 Antioxidants have been found to be a stabilizing factor in cancer prevention [17]. In the 264 present study the antioxidant potential of Chlorophytum comosum aqueous extracts 265 (CCLA & CCRA) were determined by DPPH and FRAP assay. The scavenging effects 266 were checked on 5 different concentrations (62.5-1000 µg/ml). Leaf extract exhibited 267 maximum response in both DPPH assay (IC₅₀ 3.08 μ g/ml) and FRAP (311.2 μ g/ml) assay 268 as compared to root extracts IC₅₀ 5.8 µg/ml in DPPH & 548.3 µg/ml in FRAP). % 269 scavenging of DPPH followed a dose dependent increase with slighter variation at 125 270 µg/ml, similar response were observed in FRAP assay where the absorbance indicating 271 the reducing ability of the extracts increased dose dependently (Figure 2a and 2b).

272 Quantification of total phenolic contents (TPCs)

In agreement with our GC-MS finding the total phenolic content in the leaf extract werefound to be higher than the root extract The total phenolic content of roots and leaf extract

275 was found to be 2.0.2 and 3.38 mg/g respectively of the gallic acid equivalent (GAE) g^{-1}

dried sample (Figure 2c) Each sample was run in duplicates. The standard curve was calibrated using 5 different concentrations (62.5-1000 μ g/ml) and the spectra was recorded at 795nm.

279 Anti-proliferative activity on breast cancer cell lines (MCF-7)

280 The anti-proliferative potential of ethanolic leaf (CCLE) and roots (CCRE) extracts on 281 MCF-7 cancer cells versus L-132 normal cells were determined by MTT assay. Cells 282 were treated with the extracts (CCLE, CCRE) in dose dependent manner (10µg/ml-283 320µg/ml) for 24 hours. The % cell inhibition of MCF-7 cells was observed from 8.62 % 284 to 91.2 % in roots (CCRE) while in leaves (CCLE) it was observed from 3.45 % to 88.45 285 %. The cytotoxic effect of the CCRE was higher as compared to the CCLE extract with 286 an IC₅₀ value of 31.94 µg/mL, while as for the CCLE it was 57.8 µg/mL (Figure 3). Most 287 importantly at the similar doses, these extracts did not show significant reduction on the 288 viability of L-132 cells thereby suggesting that their cytotoxicity is specific towards 289 cancer cells. The IC₅₀ value ranges were in accordance with NCI guidelines where the 290 suitability of a drug candidate with less than 100 μ g/ml is taken into consideration. 291 Standard vinblastine exhibited the similar level of inhibitory effects with IC₅₀ value of 6.9 292 µM in MCF-7 and 80.1 µM in L-132 cell lines as shown in (Figure 3).

293 Anti-proliferative activity on lung cancer cell lines (A549 & H1299)

In response to 24 hrs dose dependent treatment of ethanolic leaf and root extracts on lung cancer cell lines, significant cytotoxicity with varying response on each cell line was observed. In A549 cell line, dose dependent % cell inhibition was observed from 9.32 % to 92.7 % (CCRE) while as, in CCLE it dropped from 8.32% to 91.7 % with an IC₅₀ values of 62.25 μ g/mL and 77.84 μ g/mL respectively. In case of H1299 no significant anti-proliferative effects of roots extract was seen as the maximum % cell inhibition achieved was from 4.32 % to 58.14 % while on the other hand % cell inhibitory trend of

301 9.17 % to 90.68% was observed in leaves extract. Among both the cell lines A549 cell 302 line showed maximum response with an IC₅₀ value of 62.25 μ g/mL (CCRE) and 77.84 303 μ g/mL(CCLE) as compared to H1299 where the calculated IC₅₀ values were 173.7 μ g/mL 304 (CCRE) and 82.63 μ g/mL (CCLE). No inhibitory response was seen on L-132.Selective 305 response of the extracts on different cancer cell lines based on IC₅₀ values are depicted in 306 (Figure 3).

307 In vitro haemolytic effects of Chlorophytum comosum on human erythrocytes

308 The haemolysis of red blood cells (RBCs) is a major concern for the clinical development 309 of therapeutic agents [18]. Therefore to determine the safety profile, the haemolytic 310 activities of the CCRE and CCLE were also evaluated. The haemolytic activity of the 311 extracts were found to be insignificant and did not raise beyond 30% at the selected 312 concentrations however the % haemolytic activity was observed in dose dependent 313 manner. This indicates that even at maximum inhibitory concentration, the samples 314 CCLE and CCRE did not show any significant haemolytic activity on RBC's thus CCLE 315 and CCRE can be considered safe for pharmaceutical studies. Percent (%) haemolytic 316 active at different concentrations of CCRE and CCLE is depicted in (Table 5).

317 **Discussion**

318 Chlorphytum comosum (Thunb). Jacques (Spider Ivy) is a well-known plant for its 319 ornamental value worldwide however based on ethanopharmacological evidence it has 320 documented to be used in traditional Chinese medicine preparations for treatment of 321 respiratory ailments, fracture and burns [3-4] In India roots part of Chlorophytum 322 comosum is available under the common name of 'Safed Musli' and is believed to be 323 utilized as a substitute/ adulterant in preparation of important class of drug 'Rasayna' in 324 Ayurveda [19]. Even though earlier studies have isolated few steroidal saponins from root 325 extracts of Chlorophytum comosum that have found to possess anti-tumor cytotoxic

326 properties [5-6], not much attempts were made further to investigate this plant for its 327 pharmaceutical potential. With reference to the previous studies and based on 328 ethanomedical uses specifically with respect to its role in treatment of respiratory ailments 329 we decided to perform the comprehensive chemical profiling of various parts of 330 Chlorophytum comosum and revealed the significant antiproliferative activity of 331 ethanolic roots and leaves extracts of *Chlorophytum comosum* against the cancer cells 332 lines of two different origin i.e. Lung (A549, H1299) and Breast (MCF-7). Our study also 333 noted that the selective and specific response of the crude extracts were towards cancer 334 cell lines as no cell inhibitory effects were seen on normal Lung (L-132) cell line when 335 tested comparatively. The maximum antiproliferative effects with high selectivity were 336 seen on MCF-7 and A549 cell lines with IC₅₀ values $< 100 \mu$ g/ml (Figure 3). In case of 337 H1299 cell line no significant antiproliferation was recorded in roots extracts (IC₅₀ value> 338 100µg/ml) however leaves extract showed moderate activity towards H1299 cell line 339 $(IC_{50} \text{ value} = 82.63 \,\mu\text{g/ml})$. These results are in accordance of the previous studies where 340 Butanolic root fractions of Chlorophytum comosum yielded significant anti-tumor 341 promoter activity against tumor promoter induced phospholipids metabolism of HeLa cell 342 lines[5] In another study conducted by Matushita et al., apoptosis induction by Butanolic 343 extracts in four human cancer cell lines (HeLa, CCRF-HSB-2,HL-60 and U937) were 344 seen [4]. Chinese herbs are the excellent source of bioactive natural products that have 345 been found to inhibit proliferation, induce apoptosis, suppress angiogenesis, retard 346 metastasis and enhance chemotherapy, exhibiting anti-cancer potential both in vitro and 347 in vivo [20]. To discover new potential sources of anticancer plants, initial in vitro 348 cytotoxic screening and chemical profiling of the plant is an important step and provides 349 basic evidence of availability of active metabolites which needs to be further validated by 350 isolation and characterization studies followed by in vivo screening. Our study provides

the initial data for the selection of this plant for its consideration towards the isolation ofnovel anticancer compounds.

Saponins are the known cytotoxic agents and possess strong haemolytic activity [21].GC-353 354 MS findings from our studies revealed saponins as major constituents in roots portion 355 while phenols were mainly found to be present in leaves extracts. To rule out the 356 possibility of toxic effects of the extracts, we measured the anti-haemolytic activity of the 357 extracts using haemolysis assay. No significant haemolytic studies were observed in both 358 leaves and roots extract which may suggest the possibility of mechanism of action of 359 these phytoconstituents at the synergistic level. Similarly high phenolic content of leaves 360 extract as indicated by GC-MS and TPC measurement might have been responsible for 361 the better antioxidant effects in leaves extract as compared to root extracts. In a study 362 conducted by Deore et al. methanolic roots extract possessed the strongest antioxidant 363 activity among various species of Chlorophytum [22]. A plethora of evidences supports 364 phenolic components as a strong antioxidant and anticancer agents [23-24]. Compounds 365 like Purpurogallin as recorded in LC-MS data is a suggested anticancer compound which 366 inhibits mitogen-activated protein kinase kinase 1/2(MEK1/2) signaling 367 pathway[25]. Thus, to best of our knowledge our study is the first systematic and 368 comparative profiling of the plant marking the presence of other anticancer active 369 molecules than saponins, however this is the initial screening which needs to be further 370 validated by isolation and characterization studies to find out the lead molecules involved 371 in anticancer therapy.

372 Conclusion

We conclude that *Chlorophytum comosum* possess diversified active constituents of therapeutic potential (chiefly phenols and saponins) that in synergy possess significant antioxidant and antiproliferative potential against the breast (MCF-7) and lung cancer (A-

- 376 549) cell lines. Our study also justifies the traditional use of *Chlorophytum comosum* in
- 377 treatment of lung associated disorders.

378 List of Abbreviations

- 379 CCLA : Chlorophytum comosum leaf aqueous extract
- 380 CCRA : *Chlorophytum comosum* roots aqueous extract
- 381 CCLE : Chlorophytum comosum leaf ethanolic extract
- 382 CCRE : Chlorophytum comosum roots ethanolic extract
- 383 TPCs : Total Phenolic Contents
- 384 DPPH : 2, 2-diphenyl-1-picrylhydrazyl
- 385 FRAP : Ferric Reducing Antioxidant Power
- 386 GC-MS: Gas Chromatography Mass Spectrometry
- 387 LC-ESI-MS: Electrospray Ionization Mass Spectrometry
- 388 Declaration
- 389 Ethics Approval and Consent to Participate
- **390** Not Applicable
- **391 Consent for Publication**
- 392 Not Applicable
- 393 Availability of Data and Materials

394 All data generated or analysed during this study are included in this published article [and

- its supplementary information files].
- **396 Competing Interests**
- 397 "The authors declare that they have no competing interests"
- 398 Author's Contribution

399 The study conceptualization and design was done by Farooqi H. The experiment

400 execution, data collection, data analysis and manuscript drafting was completed by

401 Adhami S, Abdin M.Z. provided the necessary facilities for the study and Malik A.A.
402 provided with the suggestions for improved manuscript.

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Peak No.	Identified Compound Name (IUPAC)	Common Name	Compound Class	CAS No.	Pub Chem CID	Molecular Weight	Molecular Formula	% area	Retention Time
1	1-methylpiperidine	N-Methylpiperidine	t-Amines	626-67-5	12291	99.177	$C_6H_{13}N$	0.42	5.768
2	2-methyl-N-(2- methylbutyl) butan-1- imine	1-Butanamine, 2- methyl-N-(2- methylbutylidene)-	Aliphatic Imines	54518-97-7	521517	155.285	$C_{10}H_{21}N$	0.79	7.840
3	3-methyl-N-(3- methylbutyl)butan-1- imine	Isopentylidene isopentylamine	Amines	35448-31-8	118823	155.285	$C_{10}H_{21}N$	1.67	8.048
4	2-methoxyphenol	Guaiacol	Phenols	90-05-1	460	124.139	$C_7H_8O_2$	1.38	9.021
5	Undecane	Undecane	Alkane Hydrocarbon	1120-21-4	14257	156.313	$C_{11}H_{24}$	2.44	9.219
6	butan-2-yloxy-butyl- dimethylsilane	2butyl(dimethyl)silylo xybutane	Organosilicon	NA	582888	188.386	$C_{10}H_{24}OSi$	2.42	10.743
7	butoxy(trimethyl)silane	1-Butanol, trimethylsilyl ether	Organosilicon	1825-65-6	519537	146.305	C ₇ H ₁₈ OSi	5.81	12.335
8	2-tert-butyl-5- (hydroxymethyl)-5- methyl-1,3-dioxolan-4- one	2-t-Butyl-5- hydroxymethyl-5- methyl-[1,3]dioxolan- 4-one	Oxalidinone	NA	554337	188.223	$C_9H_{16}O_4$	41.05	12.895
9	4-ethenyl-2- methoxyphenol	4-vinylguaiacol	Phenols	7786-61-0	332	150.177	$C_9H_{10}O_2$	1.28	13.310
10	2,6-dimethoxyphenol	Syringol	Phenols	91-10-1	7041	154.165	$C_8H_{10}O_3$	1.08	13.933

Table 1 Identified chemical constituents and their % content from leaves of Chlorophytum comosum by GC/MS

Peak No.	Identified Compound Name (IUPAC)	Common Name	Compound Class	CAS No.	Pub Chem CID	Molecular Weight	Molecular Formula	% area	Retention Time
11	3-(hydroxymethyl)phenol	3-Hydroxybenzyl alcohol	Phenols	620-24-6	102	124.139	$C_7H_8O_2$	7.59	14.582
12	dodecane-1-thiol	1-dodecanethiol	Thiols	112-55-0	8195	202.4	$C_{12}H_{26}S$	0.66	17.032
13	2-tert-butyl-4- methoxyphenol	(1,1Dimethylethyl)-4- methoxyphenol	Aromatic Ether	25013-16-5	8456	180.247	$C_{11}H_{16}O_2$	1.68	17.332
14	2-cyclohepta-2,4,6-trien- 1-yl-1H-pyrrole	2-cyclohepta-2,4,6- trien-1-yl-1H-pyrrole	Pyrrole Alkaloids	NA	593678	157.216	$C_{11}H_{11}N$	0.66	17.646
15	Cyclopenta[C]Pyran-1-ol, 1,4a,5,6,7,7a-Hexahydro- 4,7-Dimethyl acetate, [1S- (1.Alpha.,4a.Alpha.,7.Beta .,7a.alpha.)	(1S)-1 Acetooxymyodesert- 3-ene	Unknown	NA	NA	210	$C_{12}H_{18}O_3$	0.19	19.107
16	tetradecyl prop-2-enoate	2-Propenoic acid,tetradecyl ester	Carboxyester	21643-42-5	88984	268.441	$C_{17}H_{32}O_2$	2.35	19.183
17	(2R,5S,6S)-2,6,10,10- tetramethyl-1- oxaspiro[4.5]decan-6-ol	2,6,10,10- Tetramethyl-1- oxaspiro[4.5]decan-6- ol	Terpenoid	77981-89-6	23624156	212.333	$C_{13}H_{24}O_2$	0.34	20.426
18	2,2-dimethyl-3-(2- methylprop-1- enyl)cyclopropane-1- carboxylic acid	Chrysanthemic acid	Cyclopropane carboxylic acid	10453-89-1	2743	168.236	$C_{10}H_{16}O_2$	0.86	20.468

Peak No.	Identified Compound Name (IUPAC)	Common Name	Compound Class	CAS No.	Pub Chem CID	Molecular Weight	Molecular Formula	% area	Retention Time
10	bis(2-methylpropyl)								
19	benzene-1,2-	Phthalic acid,	phthalate ester	84-69-5	6782	278.348	$C_{16}H_{22}O_4$	0.4	21.447
	dicarboxylate	diisobutyl ester							
20	6-methylhepta-1,6-dien- 2-ylbenzene	(5-Methyl-1- methylene-5- hexenyl)benzene	Unknown	NA	576233	186.298	$C_{14}H_{18}$	1.11	22.982
21	6-methylhepta-1,6-dien- 2-ylbenzene	(5-Methyl-1- methylene-5- hexenyl)benzene	Unknown	NA	576233	186.298	$C_{14}H_{18}$	1.71	23.018
22	Trimethylsilyl hexadecanoate	Palmitic acid, TMS	Fatty acid	55520-89-3	521638	328.612	C ₁₉ H ₄₀ O ₂ Si	0.78	23.695
23	1-Methyl-5- phenylbicyclo[3.2.0]Hepta ne	1-Methyl-5- phenylbicyclo[3.2.0] Heptane	Unknown	NA	576531	186.298	$C_{14}H_{18}$	1.03	23.812
24	3-(2-phenylaziridin-1- yl)propane nitrile	3-(2-Phenyl-1- aziridinyl)propane nitrile	Aziridine	NA	563034	171.231	$C_{11}H_{12}N_2$	0.76	24.115
25	methyl (Z)-octadec-9- enoate	Methyl Oleate	Fatty acid methyl ester	112-62-9	5364509	296.495	$C_{19}H_{36}O_2$	0.73	24.365
26	methyl octadecanoate	Methyl Sterate	Fatty acid methyl ester	112-61-8	8201	298.511	$C_{19}H_{38}O_2$	0.63	24.664
27	ethyl (E)-3-(4- methylphenyl)prop-2- enoate	Ethyl-4-methyl cinnamate	Methyl ester of Cinnamic acid	20511-20-0	641318	190.242	$C_{12}H_{14}O_2$	0.79	26.29

Peak No.	Identified Compound Name (IUPAC)	Common Name	Compound Class	CAS No.	Pub Chem CID	Molecular Weight	Molecular Formula	% area	Retention Time
28	bis(2-ethylhexyl) hexanedioate	DEHA	Diester	103-23-1	7641	370.574	$C_{22}H_{42}O_4$	4.08	27.558
29	5,5bis (heptylsulfanyl)pentane- 1,2,3-triol	D-Ribose, 2-deoxy- bis(thioheptyl)- dithioacetal	Polysaccharides	NA	575898	380.646	$C_{19}H_{40}O_3S_2$	3.24	28.931
30	bis(2-ethylhexyl) benzene-1,2- dicarboxylate	Bis(2- ethylhexyl)phthalate	Ester of phthalic acid	117-81-7	8343	390.564	$C_{24}H_{38}O_4$	1.54	29.51
31	(3S,8S,9S,10R,13R,14S,17 R)-17-[(2R,5R)-5-ethyl-6- methylheptan-2-yl]- 10,13-dimethyl- 2,3,4,7,8,9,11,12,14,15,16 ,17-dodecahydro-1H- cyclopenta[a]phenanthre n-3-ol	β-Sitosterol	Phytosterols	83-46-5	222284	414.718	C ₂₉ H ₅₀ O	1.24	38.051
32	(E)-5-[(1S,2R,4aR,8aR)- 1,2,4a,5-tetramethyl- 2,3,4,7,8,8a- hexahydronaphthalen-1- yl]-3-methylpent-2-en-1- ol	Kolavenol	Diterpenoids	19941-83-4	6442554	290.491	C ₂₀ H ₃₄ O	0.49	39.571

Peak No.	Identified Compound Name (IUPAC)	Common Name	Compound Class	CAS No.	Pub Chem CID	Molecular Weight	Molecular Formula	% area	Retention Time
33	(1R,2S,4S,5'S,6R,7S,8R,9S, 12S,13S,16S,18R)- 5',7,9,13- tetramethylspiro[5- oxapentacyclo[10.8.0.02,9 .04,8.013,18]icosane-6,2'- oxane]-16-ol	Sarsasapogenin	Steroids	126-19-2	92095	416.646	C ₂₇ H ₄₄ O ₃	7.54	42.569
34	(1S,2S,4S,5'R,6R,7S,8R,9S, 12S,13R,16S)-5',7,9,13- tetramethylspiro[5- oxapentacyclo[10.8.0.02,9 .04,8.013,18]icos-18-ene- 6,2'-oxane]-16-ol	Diosgenin	Sapogenins	512-04-9	99474	414.63	C ₂₇ H ₄₂ O ₃	1.27	43.393
NA:	Not Available, Database	- Wiley 8. Lib. / N	IST14.lib						

485 Table 2 Identified chemical constituents and their % content from roots of *Chlorophytum comosum* by GC/MS

Peak No.	Identified Compound Name (IUPAC)	Common Name	Compound Class	CAS No.	PubChem CID	Molecular Weight	Molecular Formula	% Area	Retention Time
1	Dodecane	Dodecane	Alkane Hydrocarbon	112-40-3	8182	170.34	$C_{12}H_{26}$	9.37	9.213
2	4a,8-dimethyl-2-prop-1-en-2-yl- 2,3,4,5,6,7-hexahydro-1H-naphthalene	Naphthalene	Aromatic Hydrocarbon	41071-31-2	605019	204.357	$C_{15}H_{24}$	0.64	14.949
3	Diethyl benzene-1,2-dicarboxylate	Diethyl phthalate	Benzoic acid esters	84-66-2	6781	222.24	$C_{12}H_{14}O_4$	2.29	17.725
4	4-trimethylsilyloxycyclohexan-1-ol	Cis -1,4- cyclohexanediol,o- (trimethylsilyl)	Fatty Alcohols	54725-69-8	554465	188.342	C ₁₉ H ₂₀ O ₂ Si	1.94	18.672
5	(3E,5E)-2,6-dimethylocta-3,5-diene	2,7-dimethylocta- 3,5-diene	Alkenes	28980-73-6	17939649	138.254	$C_{10} H_{18}$	0.91	19.126
6	(6S) 6-methyloctan-1-ol	6- Methyl-1-octanol	Aliphatic Hydroxy Compound	110453-78-6	520908	144.258	$C_9 H_{20}O$	0.77	19.206
7	Trimethylsilyl hexadecanoate	Palmitic acid	Fatty Acids	55520-89-3	521638	328.612	$C_{19}H_{40}O_2Si$	2.98	23.705
8	Methyl 9,10-Dideutero-9-Octadecenoate	Methyl Elaidate	Fatty Acids	1937-62-8	5280590	298.00	$C_{19}H_{34}D_2O_2$	2.95	24.371

9	Methyl 15-methylhexadecanoate	Hexadecanoic acid, 15-methyl-,methyl ester	Fatty Acids	6929-04-0	522345	284.484	$C_{18}H_{36}O_2$	2.95	24.669
10	bis(2-ethylhexyl) hexanedioate	Diethylhexyl adiapate	Esters	103-23-1	7641	370.574	$C_{22} H_{42} O_4$	18.9	27.56
11	N-[4-(2-tert-butyl-5-oxo-1,3-dioxolan-4- yl)butyl]formamide	Formamide	Amide	NA	582477	243.303	$C_{12}H_{21}NO_4$	1.35	28.651
12	bis(2-ethylhexyl) benzene-1,2- dicarboxylate	Bis(2-ethylhexyl) phthalate	Esters	117-81-7	8343	390.564	C ₂₄ H ₃₈ O ₄	7.35	29.514
13	Sarsasapogenin 3-tosylate	Tigogenin tosylate	Steroidal Glycoside	NA	587000	570.829	$C_{34}H_{50}O_5S$	4.43	36.693
14	(3S,8S,9S,10R,13R,14S,17R)-17-[(2R,5R)- 5-ethyl-6-methylheptan-2-yl]-10,13- dimethyl-2,3,4,7,8,9,11,12,14,15,16,17- dodecahydro-1H- cyclopenta[a]phenanthren-3-ol	β-Sitosterol	Phytosterols	83-46-5	222284	414.718	$C_{29}H_{50}O$	7.79	38.062
15	2,2,4a,7a-Tetramethyl-1,2a,3,4,5,6,7,7b- octahydrocyclobuta[e]inden-5-ol	2,2,4a,7a- Tetramethyldecahy dro-1H- cyclobuta[e]inden- 5-ol	Sesquiterpenes	NA	573804	222.372	C ₁₅ H ₂₆ O	4.97	39.568
16	Neotigogenin	25S-Tigogenin	Triterpenoids	470-01-9	12304433	416.646	$C_{27}H_{44}O_3$	16.2	42.566
17	(1R,2S,4S,5'R,6R,7S,8R,9S,12S,13S,18S)- 5',7,9,13-tetramethylspiro[5 oxapentacyclo[10.8.0.02,9.04,8.013,18]ic osane-6,2'-oxane]-16-one	Yuccagenone	Steroidal Glycoside	470-07-5	101692	414.63	C ₂₇ H ₄₂ O ₃	14.0	43.376

486 NA: Not Available, Database - Wiley 8. Lib. / NIST14.1

487 Table 3 Tentatively identified metabolites in roots of *Chlorophytum comosum* by LC-ESI-MS (+ ion mode)

488	Analyte	Tentative allotment of compounds based on	Parent ion	Peak Intensity	Mass Recorded	METLIN ID
489	No.	METLIN	(m/z)		(METLIN)	
490 491	1.	5,6-Dichloro-1,3-cyclohexadiene	148.99	6.263e6	147.98	69011
492	2.	5-Fluoroindole-2-Carboxylic Acid	180.04	4.115e6	179.03	44300
493 494	3.	4,5-Dichloro-3H-1,3-dithiol-2-one	186.88	2.011e6	185.87	89030
495	4.	Purpurogallin	221.04	1.375e6	220.03	43934
496 497	5.	Ginsenoyne B	295.14	8.843e5	294.13	93731
498 499	6.	Salicylic acid beta-D-glucoside	301.09	5.089e6	300.08	95685
500 501	7.	gamma-Glutamyl-beta-(isoxazolin-5-on-2-	302.09	9.296e5	301.09	95006
502 503	8.	11beta-Chloromethylestradiol	321.16	7.740e5	320.15	69957
504	9.	Heneicosanedioic acid	357.30	1.749e6	356.29	35988
505 506	10.	Oleyl Anilide	358.31	4.706e6	357.30	44935
507 508	11.	Hexadecyl Acetyl Glycerol	359.31	1.121e6	358.30	43452
509 510	12.	6-Bromo-5E,9Z,13Z-docosatrienoic acid	413.20	1.289e7	412.19	96831
511 512 513	13.	7,8,3',4'-Tetramethoxy-6 6''-dimethylpyrano [2'' 3'''5 6)flavone	425.15	2.830e6	424.15	49820
514 515	14.	Isosyringinoside	535.20	7.687e5	534.19	95276
	15.	Sagecoumarin	537.09	1.345e6	536.09	89672
516	16.	4-(4-Hydroxyphenyl)-	625.18	1.132e6	624.18	93899
517		2-butanone O- [2-galloyl-6-p-coumaroylglucoside]				
518	17	Gnidicin	629.23	8.064e5	628.23	67423

						520
Analyte	Tentative allotment of	Parent ion (m/z)	Peak Intensity	Mass Recorded	METLIN ID	
No.	compounds based on METLIN	Positive ion mode [M+H]		(METLIN)		521
1.	5,6-Dichloro-1,3- cyclohexadiene	148.9924	7.904e6	147.98	69011	522
2.	Serratanidine	296.1872	2.612e6	295.1784	67995	523
3.	Salicylic acid beta-D-glucoside	301.0908	6.044e6	300.0845	95685	524
4.	3-(2-Heptenyloxy)-2- hydroxypropyl undecanoate	357.2993	2.186e6	356.2927	89668	525 526
5.	Oleyl Anilide	358.3117	6.640e6	357.3032	44935	520
6.	Methyl (9Z)-6'-oxo-6,5'-diapo- 6-carotenoate	393.2399	4.100e6	392.2351	88088	528
7.	6-Bromo-5E,9Z,13Z- docosatrienoic acid	413.2067	4.100e6	412.1977	96831	529
						530

519 Table 4 Tentatively identified metabolites in leaves of *Chlorophytum comosum* by LC-ESI-MS (+ ion mode)

535 Table 5 Dose dependent % haemolytic activity observed in *Chlorophytum comosum* extracts.

Samples	Treatment (μg//ml)	% Haemolysis 537
Control	PBS	0.00 538
Positive control	1% SDS	85.75
	10	3.25 539
	20	5.30 540
CCLE	40	9.11 541
CCLE	80	17.18 542
	160	19.08 5/13
	320	28.11
		544
	10	3.40
	20	7.52 545
CCDE	40	14.01
CCRE	80	19.24
	160	24.94
	320	30.01

1 X PBS buffer served as control. % haemolytic activity were measured against 1% SDS which served as positive control.

- **Figure 1** Total ion chromatogram of *Chlorophytum comosum* aqueous root (a) and leaf extract (b) by LCESI- MS.



550 Selected peaks in roots were identified as (d) Purpurogallin, (f) Salicylic acid beta-D-glucoside, (g) Heneicosanedioic acid, (h) Oleyl Anilide and in

b

- 551 leaf as (a) 5,6-Dichloro-1,3-cyclohexadiene, (h) Oleyl Anilide (i) 6-Bromo-5E,9Z,13Z-docosatrienoic acid when compared with XCMS Metlin
- 552 database in positive ion mode. Note: At some peaks no metabolites were recorded in database.

⁵⁵⁷ Figure 2 Measurement of antioxidant potential of *Chlorophytum comosum* by DPPH, FRAP and TPC assays



mean \pm SD of three independent experiments with a correlation coefficient ($R^2 = 0.9921$) and p value < 0.05. (c) Total phenolic content available

in different parts of *Chlorophytum comosum*. (c-a) standard curve of gallic acid, (c-b) % phenolic content in roots and leaves of *C. comosum* Total
 phenolic content was expressed as mg gallic acid equivalent (GAE) per gram of dried sample.

Figure 3 MTT cell viability assay.



573 Antiproliferative activity of ethanolic roots and leaf extract were evaluated in a dose dependent exposure (10µg/ml-320µg/ml) of test samples for

574 24hrs. Vinblastine was used as control Dose dependent cell growth inhibitory effects of CCLE and CCRE on different cell lines and response of

- 575 standard vinblastine on cancer versus normal cell line L-132 is represented with their respective IC50 values. In L-132 IC50 values were not
- 576 calculated for CCLE and CCRE due to lesser percentage inhibition (<50%). The data is statistically significant with P <0.05.

Figures



b

Figure 1

Total ion chromatogram of Chlorophytum comosum aqueous root (a) and leaf extract (b) by LCESI-MS. Selected peaks in roots were identified as (d) Purpurogallin, (f) Salicylic acid beta-D-glucoside, (g) Heneicosanedioic acid, (h) Oleyl Anilide and in leaf as (a) 5,6-Dichloro-1,3-cyclohexadiene, (h) Oleyl Anilide (i) 6-Bromo-5E,9Z,13Z-docosatrienoic acid when compared with XCMS Metlin database in positive ion mode. Note: At some peaks no metabolites were recorded in database.



Figure 2

Measurement of antioxidant potential of Chlorophytum comosum by DPPH, FRAP and TPC assays (a) Dose dependent % scavenging of DPPH by aqueous leaf (CCLA) and roots (CCRA) extract of Chlorophytum comosum. (b) Total reducing capacity of leaf and roots of Chlorophytum comosum as observed with dose dependent decrease in absorbance by FRAP assay. The values are mean ± SD of three independent experiments with a correlation coefficient (R2= 0.9921) and p value < 0.05. (c) Total phenolic content available in different parts of Chlorophytum comosum. (c-a) standard curve of gallic acid, (c-b) % phenolic content in roots and leaves of C. comosum Total phenolic content was expressed as mg gallic acid equivalent (GAE) per gram of dried sample.



Figure 3

MTT cell viability assay. Antiproliferative activity of ethanolic roots and leaf extract were evaluated in a dose dependent exposure ($10\mu g/ml-320\mu g/ml$) of test samples for 24hrs. Vinblastine was used as control Dose dependent cell growth inhibitory effects of CCLE and CCRE on different cell lines and response of standard vinblastine on cancer versus normal cell line L-132 is represented with their respective IC50 values. In L-132 IC50 values were not 575 calculated for CCLE and CCRE due to lesser percentage inhibition (<50%). The data is statistically significant with P <0.05.

Supplementary Files

This is a list of supplementary files associated with this preprint. Click to download.

- Table1AdhamietalGCMSANALYSISOFLEAVESOFCC.docx
- Table3AdhamietalLCESIMSROOTSCC.docx
- Table2AdhamietalGCMSANALYSISOFROOTSOFCC.docx
- Table4AdhamietalLCESIMSLEAVESCC.docx
- Table5AdhamietalHaemolysisbyCC.docx