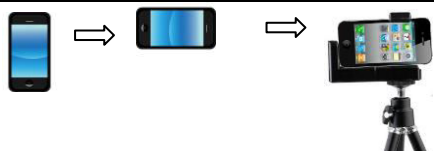


## Script to prepare video clip(s)

Manuscript Title:	Enhanced cytotoxic potential of <i>Orthosiphon stamineus</i> extract in MCF-7 cells through suppression of nucleolin and bcl <sub>2</sub>		
ID: 32337	LC-MS/MS		
Material:	Time:	Date:	Location: Your Lab

### Position of your smart phone



Take the video using your smart phone in horizontal position instead of vertical

Smart phone on the tripod reduces the shaking of picture

### Set-up of LC-MS/MS

Materials and Reagents

LC-MS/MS, acetonitril, water, acetic acid

Action	Subtitle
<b>1</b> Add HPLC grade acetic acid to two separate bottles containing acetonitrile and water. Final concentration in each bottle will be 1%. Close the cap and mix the solvent properly. Open the cap of each bottle and place them at the top of the UHPLC. One end of the delivery tube with the filter will be placed at the bottom of each bottle	
<b>2</b> Switch on all the buttons of UHPLC one by one	Wide shot
<b>3</b> Clicking the icon of the HyStar 3.2 software in the desktop computer	
<b>4</b> Click the "On" button to start the UHPLC	
<b>5</b> Set the parameters like flow rate, column temperature, ratio of each mobile phase, injection volume, negative mode, MS/MS scan, mass range (m/z 50-1500), collision energy (10 eV), nitrogen gas flow rate (6 L/min), UV detector (260 nm), and others	Close view

### Preparation of the Sample

Materials and Reagents

Fraction 3 of *O. stamineus* extract, Disposable syringe (1 mL), Syringe filter (0.45 µm), Eppendorf tube (1.5 mL), LC-MS amber glass vial with seal (1.5 mL)

Action	Subtitle
<b>1</b> Dissolve the extract using the mobile phase	
<b>2</b> Take 1 mL of the extract into the syringe	
<b>3</b> Place syringe filter at the tip of the syringe	

<b>4</b>	Push the plunger of the syringe to that the extract comes out.
<b>5</b>	Collect the filtered extract in the Eppendorf tube
<b>6</b>	Check that the solution is clear
<b>7</b>	Keep 0.5 mL of the filtered extract into the LC-MS amber glass vial
<b>8</b>	Close the vial top by a screwed cap with central silicone septum
<b>9</b>	Place the vial into the autosampler rack
<b>10</b>	Check the number of the vial position. The same number must be typed in the software, vial section

### Injection of the Sample and Data Collection

Materials  
and  
Reagents

[LC-MS/MS, printer](#)

Action	Subtitle
<b>1</b>	Set the run time using the computer
<b>2</b>	Click the RUN button
<b>3</b>	Data collection can be stopped at any time by either clicking on the Stop button
<b>4</b>	When data collection is complete, either through a manual stop run or when the preset run time is complete, a report will be generated.
<b>5</b>	Show the peak
<b>6</b>	Show the mass of that peak
<b>7</b>	Printer the data in a A4 size paper