Response to Reviewer 2 Comments

The presented article deals with interesting and important issues related to the acquisition of valuable bioactive compounds from natural sources. Nevertheless, the article requires some important amendments.

The main focus is on carelessness in the editing of the text, the necessary is deletion of unnecessary spaces, Latin genre names and prefixes like t- should be written in italic and there is no lower index for numbers in some formulas of chemical compounds. Moreover, in several places (lines 52 and 354) there is no reference to the list of quoted literature. The size, description and position of all graphs should be unified.

Additionally, several parts of the text require the Authors' comment:

**Point 1:** line 65: Has the TPP method been already used for polysaccharides?

**Response 1:** Yes. As we have discussed in conclusions section. In order to understand it cleanly, we inserted “Currently, TPP is also applied to extract and purify tapioca starch and tapioca starch derivatives [16], chitosan from shrimp shell [17], alginates from *Dunaliella salina* [18], levan and hydrolyzed levan from several levansucrase microorganisms, among them *Zymomonas mobilis*, a mobile Gram-negative bacterium [19], aloe polysaccharides [20] and *Corbicula fluminea* polysaccharides [21]. More recently, Wang *et al.* reported that TPP is utilized to separate and purify polysaccharide–protein complexes (PSP) from *C. fluminea* [22]. The highest extraction yield of PSP was 9.0% under the following optimal conditions: 20% (w/v) ammonium sulfate concentration, 1.5: 1.0 (v/v) t-butanol to crude extract ratio, 30 min, and 35 °C. The purified PSP also exhibited strong radical scavenging capacities and antioxidant activities *in vitro*. TPP have been applied in the fields of plant, animal and microorganism. However, its use for separation polysaccharides from edible and medicinal fungi has not yet been reported.” in line 65.

**Point 2:** line 72: How can you explain the efficiency of over 100% (for tomato) for the example cited?
Response 2: The author of the example cited explains, “The unusual enzyme recovery of 183% needs explanation. It has been often observed (Dennison & Lovrein 1997) that three-phase partitioning leads to simultaneous activation of the enzyme which (if the enzyme recovery is high) results to such an apparently observed value of 183% yield. Recently, we have found that enzyme activation frequently observed during three-phase partitioning may be the result of increased flexibility in the enzyme molecule. X-Ray diffraction studies show three-phase partitioning treated proteinase K has an unusually high B-factor (Singh et al. 2001).”

Point 3: line 235: Will ethanol not also extract lipids? Would not it be better to use a different solvent, less polar at an earlier stage to remove lipids?

Response 3: Yes, ethanol can extract and remove most of the lipids and micro molecular compounds, including colour ingredients, free amino acids, free sugars, phenols, and so on. Some organic solvents can be used to extract lipids such as petroleum ether, ethyl acetate, acetone and chloroform. For example, Wang et al. report that C. fluminea was treated thrice with refluxing petroleum ether for 6 h each treatment to remove lipids and pigments. [Wang YY, Qiu WY, Wang ZB, et al. Extraction and characterization of anti-oxidative polysaccharide–protein complexes from Corbicula fluminea through three-phase partitioning. RSC Adv., 2017, 7:11067-11075]. Shao et al. reported that 100 g of plant material from Tripterygium wilfordii was extracted with 200 mL ethyl acetate in a Soxhlet extractor for 12 h to remove lipids. [Shao D, Dunlop WD, Lui EMK, et al. Immunostimulatory and anti-inflammatory polysaccharides from Tripterygium wilfordii: comparison with organic extracts. Pharmaceutical Biology, 2008, 46(1-2):8-15]. Zhang et al. reported 100 g powder of sclerotia and 20 g powder of mycelia were defatted with diethyl ether and acetone for 4 h. [Zhang M, Zhang L, Cheung PCK, et al. Molecular weight and anti-tumor activity of the water-soluble polysaccharides isolated by hot water and ultrasonic treatment from the sclerotia and mycelia of Pleurotus tuber-regium. Carbohydrate Polymers, 2004, 56(2):123-128.].

Many documents use ethanol to remove lipids because ethanol is efficient, safe, non-toxic and cheaper. For example, Zhang et al. reported the fruiting bodies of H. erinaceus were first exhaustively extracted with ethanol under reflux for 12 h to remove lipids. [Zhang AQ, Zhang JS, Tang QJ, et al. Structural elucidation of a novel fucogalactan that contains 3-O-methyl rhamnose isolated from the fruiting bodies of the fungus, Hericium erinaceus.
Fan et al. reported Dried *C. comatus* mycelium (325.15 g) was extracted 3× with 95% of ethanol EtOH for 1 h under reflux to remove lipid. [Fan JM, Zhang JS, Tang QJ, et al. Structural elucidation of a neutral fucogalactan from the mycelium of *Coprinus comatus*. *Carbohydrate Research*, 2006, 341(9):1130-1134.]

**Point 4:** line 265: The content of the chapter indicates rather the analysis of the composition and not the properties of the polysaccharides, so the title of the chapter is inadequate to the content.

**Response 4:** We changed the title of this chapter to “Preliminary study on the polysaccharide composition of IOPS”.