Lithiation of white button (*Agaricus bisporus*) mushrooms using compost fortified with common lithium salts – accumulation, processing effects and *in vitro* release of lithium

Summary of the doctoral dissertation

The role of the metallic element Lithium (Li) in human nutrition has not yet been identified and it is currently classed as non-essential. However, some of its salts have been used for some decades as therapeutic drugs in the prevention of bipolar disorder, treatment of the active phase of affective disorders and the enhancement of antidepressant and anti-psychosomatic treatment. Nevertheless, there is no deep understanding of the mechanism of this therapeutic effect. Lithium is slightly more toxic than other alkali metals, although small amounts of lithium are not considered harmful - unless the subject is on a sodium-restricted diet. Lithium carbonate (tablets) is the treatment that is most often administered orally to patients to prevent or reduce manic depression, at a daily dose in the range of 600 to 1800 mg. Some patients have been known to suffer a range of side effects to this treatment and additionally, the vast majority (over 95%) of the administered dose is excreted through the urinary system. Therefore safer and more effective ways of administering Li are being sought, which would provide an alternative to the use of pure salts of this element, for example, by lithiation of commonly consumed foods such as mushrooms where the element can be incorporated during cultivation.

The aim of this course of this doctoral project was to: (i) investigate the possibility of lithiation of the white button mushroom (*Agaricus bisporus*) (J.E. Lange) Imbach cultivated in a commercial substrate fortified with different Li salts (Li₂CO₃, LiNO₃, LiOH and Li₂O); (ii) examine the influence of selected culinary processing methods (blanching, pickling, maceration, freezing and blanching, freezing and pickling) on the Li content in preserves made from such enriched mushrooms; (iii) investigate the rate of release (accessibility) of Li from mushrooms in the human gastrointestinal tract *in vitro*; (iv) investigate whether the lithiation process influenced the accumulation of other naturally occurring commercial substrate elements such as Ag, Al, As, Ba, Cd, Co, Cr, Cu, Hg, Mg, Mn, Ni, Pb, Rb, Sb, Se, Sr, Tl, U, V

and Zn in fruiting bodies; (vi) understand the similarities and differences in the composition (Li and other elements) of white bottom mushrooms (*Agaricus bisporus*) processed under industrial conditions in Poland.

The available scientific literature does not provide information on the effect of culinary processing on the Li content of mushrooms. Dried and then macerated and blanched mushrooms lost on average 83% by wet weight (ww) of accumulated Li. Maceration of dehydrated mushrooms resulted in a massive loss of the constitutional dry matter into the water phase, i.e., at around 90%, and possibly also minerals. Thus, maceration transfers most of the organic matter and associated material into the macerate, which after condensation or dehydration, could result in a product that is rich in Li, but also other minerals and organic compounds that were present in the dried mushrooms (but have not been studied here). When blanched, the measured loss of Li from fresh mushrooms - lithiated or non-lithiated - was 33 and 41%, respectively, in terms of dry matter (41 and 50% ww), and when blanching of frozen mushrooms was 55% and 63%. The results obtained suggest that the caps appear to be more prone to loss of Li than the stems. The greater loss of Li from the caps compared to the stalks due to blanching can be explained by the difference in texture (firmness and hardness) between these morphological parts of the fruiting body.

The effect of blanching followed by pickling (marinating) on lithiated fresh mushrooms was the loss of Li by 77% dry weight (dw) and 87% ww, and in the case of non-lithiated, respectively 47 and 72%. The greater loss of Li from mushrooms through blanching and pickling than just blanching appears to be due to acetic acid's chelating and acidifying effects. The acidification to promote an increased Li leaching capacity from the blanched product. For non-lithiated mushrooms, which were deep-frozen initially and then blanched and marinated, the loss was 57% dw and 51% ww.

Knowing the degree of release of an element from food in the human gastrointestinal tract may affect the practice of assessing the benefit or toxicological risk of various elements ingested with food, including those contained in edible mushrooms. The available scientific literature does not provide information on the degree of Li release from the button mushroom and its preserves under the influence of digestive juices or the absorption of the element from the intestinal lumen into the blood. The scale of Li release from lithiated caps as a result of simulated gastrointestinal digestion was from 16 ± 1 to $20\pm2\%$, in the case of non-lithiated caps from 4.4 ± 1.7 to $33\pm2\%$, and in the case of stipes from 17 ± 1 to $21\pm2\%$ and from 5.6 ± 0.8 to $23\pm9\%$. In the case of caps and stipes of non-lithiated mushrooms - fresh after blanching and blanched and then pickled - the concentration of released Li in the digestive juices was below

the limit of quantification of the method (indicating low release); therefore, the release rate was not determined. The obtained results suggest that Li seems to be released to a greater extent from dried mushrooms than those subjected to other household processing, although the quantity of material tested (number of samples of control and lithiated mushrooms - different levels of fortification - for each process) was too small to be confirmed through statistical analysis.

Mushrooms grown on both substrates fortified with Li salts and unfortified substrates accumulated certain amounts of bio-essential elements (Cu, Zn, Se, Mn, Co), toxic elements (Ag, As, Ba, Cd, Hg, Pb, Tl, U), and others (Al, Cr, Cs, Ni, Rb, Sr, V). Nevertheless, the measured contents of these elements in the fortified mushrooms did not exceed the amounts usually observed in cultivated mushrooms as reported from various regions of the world. Preserved mushrooms available on the domestic market were found to be poorer in basic bio-elements but also poorer in toxic elements than fresh (raw) mushrooms. Due to the low concentration values of toxic elements, it can be assessed that preserved mushrooms (marinated or in brine) available on the domestic market are a negligible source of these contaminants to the daily ration of food.