SUMMARY

(1) In the present investigation, the physiological and biochemical aspects of Agaricus trisulphuratus, Rhodocybe subgliva and Agrocybe praecox have been studied making use of fermentation technology for the production of mycelial biomass. These test-fungi are some of the common edible species in West Bengal which are consumed by the local people during rainy season and whose cultivation methods are not yet available.

(2) Pileus tissue cultures from the basidiocarps of A. trisulphuratus, R. subgliva and A. praecox collected from different areas in West Bengal were prepared aseptically and maintained on 3% malt extract agar medium. To study the physiological and biochemical aspects of the test-fungi, a simple synthetic liquid medium namely glucose-asparagine basal medium (Lilly and Barnett, 1951) was selected.

(3) Screening tests for toxins, specially the most deadly cyclopeptides, were performed on the test-fungi and the negative results confirm that the mushrooms chosen for the study are safe for human consumption and would present no toxic hazard.

(4) The optimum incubation period for the submerged growth of mycelium and production of protein by
A. trisulphuratus and A. praecox was found to be 16 days while it was 20 days in case of R. subgliva. Regarding temperature, 25°C was the optimum for growth and protein production by the mycelium of A. trisulphuratus, whereas it was 35°C in case of both R. subgliva and A. praecox.

(5) From the experimental results it was found that the optimum pH for both mycelial growth and yields of protein by A. trisulphuratus, R. subgliva and A. praecox were 5.5, 5.0 and 5.0 respectively.

(6) Starch, glucose and maltose were found to be the best carbon sources both for growth and protein production by A. trisulphuratus, R. subgliva and A. praecox respectively. The three test-fungi failed to grow in presence of sodium citrate when used as a sole source of carbon.

(7) Among the inorganic, organic and complex nitrogen sources used, the complex type was observed to have more stimulatory effect on the submerged growth and protein production by the test-fungi, followed by asparagine, an organic nitrogen source. For A. trisulphuratus, L-asparagine was found to be the best source of nitrogen while it was yeast-extract for R. subgliva and A. praecox.

(8) Carbon/Nitrogen ratio in the medium varied depending upon the strain used for investigation. The optimum C:N ratios
for net mycelial yield and protein content of *A. trisulphuratus*, *R. subaliva* and *A. praecox* were 40:2 of starch; asparagine, 40 : 3 of glucose : yeast extract and 40 : 1 of maltose : yeast extract respectively.

(9) Combinations of trace elements always gave better growth yields in comparison to those of individuals. Furthermore, it was noted that no one element alone sufficed but simultaneous effect of the individual elements was required for the maximum growth and yield of protein.

(10) Findings on vitamin nutrition of the test-fungi revealed that medium containing all the vitamins under study excepting pyridoxine favours the growth of *A. trisulphuratus* whereas *R. subaliva* and *A. praecox* gave maximum yields in media deficient in thiamine and PABA respectively indicating the test-fungi to be auxoautotrophic for the respective vitamins.

(11) At a dilution grade of 25 ppm, all the four hormones used had been found to have an inhibitory effect on both growth and protein content of the test-fungi. Maximum yields of the three test-organisms were obtained in the medium without hormones which serves as control.

(12) From the analytical experiments, it might be concluded that total sugar content as well as nitrogen concentration of the
media decreased gradually during the fermentation period of the test-fungi. Furthermore, the pattern of fluctuation of $\text{pH}$ and titrable acidic contents of the three media analyzed correspond directly with the utilization of the different components of the respective media.

(13) The ash content ($\text{g}/100 \text{ g dry wt.}$) of the mycelia of $A.\text{trisulphuratus}$, $R.\text{subgliva}$ and $A.\text{praecox}$ were estimated to be 7.92%, 3.61% and 5.81% respectively under optimal conditions while it is 4.60%, 1.98% and 3.20% respectively under basal conditions. The test-organisms contained significant quantities of magnesium, a lesser amount of calcium and a very low amount of iron in their ash samples.

(14) Relatively high crude fibre contents of 10.4%, 11.8% and 8.5% (on a dry wt. basis) had been obtained on analysis of the mycelia of $A.\text{trisulphuratus}$, $R.\text{subgliva}$ and $A.\text{praecox}$ grown in optimal synthetic media. The data obtained for enzyme digestibility of dried mycelia showed that the amount of indigestible residue left after pancreatin digestion was less than that found after pepsin digestion in the test-fungi.

(15) The submerged-culture mycelia of $A.\text{trisulphuratus}$, $R.\text{subgliva}$ and $A.\text{praecox}$ had been observed to be fairly good sources of folic acid, niacin and vitamin C. But none of the test-organisms assayed contained vitamin B12 (Cyanocobalamin).
The amount of crude protein in the mycelium varied with the culture and the medium used for growth. The average crude protein content of the dry mycelia of *A. trisulphuratus*, *R. subgliva* and *A. praecox* was found to be 25.07%, 28.11%, 25.76% respectively under basal conditions while it was 31.88%, 35.51%, 33.60% in the same order under optimal conditions. The mycelial protein of the test-organisms was found to be a good source of essential amino acids, although the values for sulphur containing amino acids were comparatively much lower.

In vitro evaluation of the nutritive value of the test-fungi indicated that proteins of *A. trisulphuratus*, *R. subgliva* and *A. praecox* had high biological value in comparison to other microbial proteins. The predicted Biological Value was determined for *A. trisulphuratus*, *R. subgliva*, *A. praecox* as 46.34, 57.36, 41.27 and 70.25, 84.18, 58.30 based on the whole egg and FAO reference proteins respectively. Considering S-amino acids (methionine and cystine) as limiting, the Amino Acid Score had been estimated for the test-fungi as 31.85, 27.30 and 36.40 respectively based on FAO reference protein.

From the experimental data, it was observed that fungal mycelial biomass of the test-fungi produced on a large scale through fermentation technology, could be used as an additional source of food. No toxic effects were found in the mycelia of these test-fungi. This might partly neutralize the acute protein malnutrition problem of the majority of the Indian population.