CHAPTER – V

CHAPTER V

MINERAL ELEMENTS REQUIREMENT OF *Saccharomyces cerevisiae*A100 FOR BIOSORPTION OF Hg^{++}

Living cells require the nutrition for their growth, multiplication and biological activity. The organisms differ in their nutritional requirement. The mineral elements play significant role in the living cell. Deficiency of the essential minerals result in malfunction of the various biological activities. Like other organisms *Saccharomyces cerevisiae*A100 require the supplementation of several minerals like phosphate, sodium, potassium, magnesium, calcium, zinc, manganese, iron etc. in the synthetic medium for its optimum growth and activity.

The role of minerals in the nutrition of fungi started receiving attention with the classical researches of Raulin in 1869(235). Like other organisms yeast exhibit two types of mineral requirements (236-246)

a. Those, which are required in comparatively larger amount. These are called **macro elements** e.g., Phosphorus (P), Potassium (K), Sulphur (S), Magnesium (Mg) and Calcium (Ca).
b. Those, which are required in trace amount. These are called **trace elements** or **micro-nutrients** e.g., Iron (Fe), Zinc (Zn), Copper (Cu), Manganese (Mn), Molybdenum (Mo), Cobalt (Co), Vanadium (V) etc.

The specific term of macro and micro nutrient only define the amount of the element require for the growth and activity of the organism but not the essentiality of their requirement by the organism. Deficiency of some trace element may even inhibit the growth completely. Pirt expressed the relationship between the growth yield and growth rate(222). Minerals form an integral part of many enzyme systems or a constituent of certain pigments. Fungi have generally large requirements for phosphorus, potassium, sulphur, chloride and magnesium but smaller requirements for at least 5 micro-elements viz. zinc, copper, iron, manganese and molybdenum(247).

Trace element requirement is generally low and it ranges from 10^{-9} mol L^{-1} to 10^{-6} mol L^{-1}. The concentrations higher than optimum range show an extreme toxicity over growth of microorganism(248,249).

The essentiality of phosphorus in the fungal nutrition was recognized as early as the nineteenth century (235). According to Cockefair sugars could not be oxidized and nitrates could not be reduced to amino acids without an adequate supply of phosphorus (250). It was reported that phosphorus content was higher in younger mycelium as compared to lower content in older fungal mycelium (251). The report also suggested that phosphorous content in the mycelium could be varied by changing its concentration in culture medium. Spores contain higher amounts of phosphorous than the mycelium. However their inorganic as well as organic phosphate tend to leach out easily, as was reported by Bajaj et. al.(252).

Fungi, generally utilize phosphorous in the form of phosphate. Both inorganic as well as organic phosphates may be utilized by fungi for meeting their phosphorous requirements. Generally inorganic phosphates are incorporated into the culture medium in the form of potassium salt, and most of the potassium phosphates like potassium orthophosphate (K_3PO_4), potassium metaphosphate (KPO_3), potassium pyrophosphate ($K_4P_2O_7$), potassium mono-hydrogen phosphate (K_2HPO_4) and potassium dihydrogen phosphate (KH_2PO_4) have been found to be generally utilized by fungi. Potassium monohydrogen and dihydrogen phosphates are more frequently incorporated in culture media, because besides furnishing utilizable phosphate and potassium ions, there salts also act as useful buffers and exert a controlling influence over the pH changes in the medium caused by fungal growth and metabolism (253). Fungi utilize the organic and inorganic phosphate sources by hydrolysis process with the help of enzyme phosphatases.

According to Engl and Kunz Phosphate source in the growth medium is responsible for presence of P-ligands on the cell surface of the organism that plays an important role in metal biosorption. Cd^{++} biosorption is significantly reduced in phosphate limited medium (254). Ag^{++} biosorption by *Saccharomyces cerevisiae* is also found to be effected in absence of phosphate source(255). Besides the growth, in case of fungi phosphorous also affects some important physiological process such as; nitrogen assimilation, vitamin synthesis, oxygen consumption, participation in most of the

steps of EM pathway either in the form of substrate or enzyme or coenzyme. Some metabolic processes such as rate of glucose utilization were found to be dependent on phosphorous concentration of fungal cell (236).

Potassium is present in large amount in both mycelium and spores, and concentrations as low as 0.001 – 0.004 M of this metal is adequate for most fungi (256-258). Usually potassium is incorporated in the culture media in the form of phosphate and / or nitrate. Rennerfelt reported that sub optimum level of this metal interferes with sugar utilization (251). Complete absence of potassium in culture media causes increased accumulation of oxalic acid (259,260). Muntz reported that potassium and ammonium ions exert stimulatory effect upon glucose fermentative activity of yeast enzymatic extracts (261). So, considering the importance of potassium in fungal metabolism, it has to be added in the nutritional media in a adequate amount.

Magnesium was an essential metal for fungi and it could not be replaced by any other metal (262,263). Generally magnesium is provided in the medium as sulphate at about 0.001 M concentration and it was found that the growth of many fungal species were proportional to the magnesium content of the medium(256,264,265). Nicholas and Fielding observed that *A. niger* attained best mycelial growth at 20 mg/L of magnesium(266). Generally, the *Aspergillus* species show a higher utilization of magnesium in surface culture than in shake culture (267,268). Magnesium plays an active role in fungal metabolism by stimulating various enzyme systems (269,270). It has been reported that magnesium has some activating effect over oxidative metabolism of carbohydrate (271,272). Magnesium also known to play its role in ion antagonism against various toxic metals like boron, aluminium and copper (267,273,274).

Young and Bennett were first reported that calcium is essential for fungal nutrition (275). They observed that absence of calcium in culture medium affects the growth of fungi. Calcium is also playing a non nutritional role in fungi by altering its ion antagonist effect against certain toxic monovalent cations like hydrogen, sodium, potassium etc.

Thus, considering the importance of macro-elements viz., phosphorous, sodium, potassium, magnesium and calcium for physiological reason such as growth and

metabolism of *Saccharomyces cerevisiae,* these elements are to be supplemented in adequate concentrations in the biosorption medium.

Requirements of microelements in fungi include iron (Fe^{2+}), zinc (Zn^{2+}), manganese (Mn^{2+}), molybdenum (Mo^{6+}), cobalt (Co^{2+}), copper (Cu^{2+}) and vanadium (V^{5+}). The functions of each vary from serving in coenzyme functions to catalyze many reactions, vitamin synthesis, and cell wall transport. The requirements are generally in very low quantity.

It was established by many workers that iron is an essential trace metal for fungi (276-278). Iron forms an integral part of the fungal protoplasm. It is associated with various enzyme system like cytochromes, cytochrome oxidases, catalase etc. Most of the fungi can detect only 0.1 – 0.3 ppm of iron and some of the could respond to the presence of as little as 0.1 μg iron in 60 ml of the medium (279). In various studies it was found that sub-optimal concentration of iron and absence of it in media cause poor growth and sporulation of fungus (280-282). These effects are thought to be due to impaired synthesis of iron containing enzymes under these conditions (283). Iron also plays an effective role as an ion antagonist against some toxic ions like zinc and copper (284).

It is now almost established that all fungi require zinc as a micronutrient. Essentiality of zinc for fungal growth was first recognized by Raulin (235) and since then its necessity in fugal nutrition has been confirmed by various investigations (285-288). It is incorporated in the fermentation medium usually as $ZnSO_4.7H_2O$. Zinc concentration of the medium influences the quantity of many primary and secondary metabolites. This metal acts as toxic in higher concentration and behave as an activator even at sub-optimal concentration. Role of zinc was found to be through activating various enzyme systems (289). Deficiency of this metal influence various metabolic processes like carbohydrate metabolism, organic acid fermentation (288,290,291), amino acid metabolism, nucleic acid metabolism (292-294). Concentration of zinc ion in the fermentation medium affects the amount of citric acid produced by *Aspergillus niger* (295,296).

Fungi needed manganese as minor element in their nutrition. Generally this metal is required in minute amount (0.005 – 0.01 ppm). Some investigators recorded a little

change in mycelial dry weight of *Saccharomyces cerevisiae* when manganese was completely omitted from the culture media (266,289). However other workers observed a 50% reduction in growth of *A. niger* (297,298), 50 – 75% decrease in the mycelial yields of *A. niger* in Mn – deficient medium (299). According to Tandon and Chandra manganese deficiency symptoms become more and more pronounced as the inoculum quantity is reduced(300-304). Thus it have been concluded that fungi are able to detect and utilize even very minute amount of manganese, which is carried along with the inoculum. Role of manganese has been noted in the biosynthesis of some primary and secondary metabolites. Manganese at 0.2×10^{-5} M concentration inhibits citric acid synthesis by *Aspergillus niger* (296), but manganese ion was required upto 0.1×10^{-5} M concentration and proved inhibitory only at 1.0×10^{-5} M concentration (305).

It was recognized by some researchers that micro amounts of copper was required by fungi for growth (237,306-310). Steinberg reported that sporulation of fungi is affected by concentration of copper, however different fungi seem to differ in their response (311,312). The most important aspect of its toxic or stimulating influence depends on its concentration. In contrast to its nutritive role in minute quantities, its fungicidal effect on the majority of fungi may be cited. So, it is a component of various copper fungicides, at higher concentrations (313). The toxic effect of copper is almost counteracted by Fe^{2+} and Mn^{2+} ions but zinc ion enhances the toxicity of copper (284). A number of enzymes have been shown to require copper as their co-factor, e.g. tyrosinase and laccase. According to Yoshimuria catalase production by *Aspergillus* was dependent upon the presence of various trace metals including copper(314). Copper is also known to exert a striking influence on pigmentation of coloured spores. Javillies and Mulder observed that the fungal spore colour changes from yellow to brown and ultimately to black with increase in copper ion concentration(315,316).

Molybdenum was recognized as essential trace metal for nitrogen metabolism for fungi, bacteria and higher plants including those able to fix atmospheric nitrogen(317,318). Molybdenum requirement of fungus was first recognized by Steinberg who found that its requirement was prominent when grown on nitrogen containing medium than when ammonium nitrogen source was used(249,319). This

metal is needed for fungus in extremely minute amount ranging from 0.1 ppb to 10.0 ppb(279,320). Microbes can detect infinitesimal quantity of molybdenum. Nicholas and Fielding observed an increase in mycelial growth in *A. niger* even at very negligible concentration of molybdenum (266). It is now well established that molybdenum acts as a functional constituent of nitrate reductase system (321,322).

Fungal requirement of cobalt was reported by Marson and Ballantine (323,324). In bacteria, actinomycetes and animals, cobalt is essential for synthesis of vitamin B_{12} group compounds (cobalamines) in which cobalt forms a constituent part of tetrapyrole ring of these vitamins. However, vitamin B_{12} synthesis in fungi require more investigations to confirm its essentiality. Some reports suggested that cobalt is not essential for some fungal species (325).

Essentiality of vanadium in fungal physiology is yet to be confirmed. There were few reports regarding the requirement of vanadium and its possible role in nutrition and cell growth which stated that very little amount may have effective contribution(281,326).

Thus, it was considered necessary to study the effect of both macro and microelements on the nutrition and growth of *Saccharomyces cerevisiae*A100 and subsequently on the biosorption of Hg^{++} from the growth medium.

Material & Methods:

Surface culture biosorption was carried out for the determination of optimum concentrations of macro elements viz., suitable phosphate source, sodium, potassium, calcium and magnesium. For this purpose 48 hours old culture of *Saccharomyces cerevisiae*A100 with 2 ml volume having spore density of 1.7×10^6 per ml used as an inoculum for 50 ml biosorption medium. Hg^{++} ion concentration of the medium was adjusted to 30ppm and biosorption of Hg^{++} was carried out at pH 5.0 for 48hours at 30^0C ($\pm$ 0.5^0C) in B.O.D. incubator. The composition of the biosorption medium was : glucose (5 %), urea (0.15%), $MgSO_4$,7 H_2O (0.05%), KH_2PO_4 (0.1%), KCl (0.05%). For the objectives mentioned above, all the factors were kept constant except the one, which was to be optimised.

For determination of effects of trace elements, the biosorption medium should be free from those trace metals. For this purpose chemicals were made free from metal

impurities by chloroform extraction procedure (328). According to this method the components of the medium were dissolved in triple distilled water separately. They were then transferred to a separating funnel and 0.1 gm of 8 – hydroxy quinolone (8 – HQ) was added. 5 ml of chloroform was then added and the pH adjusted to 7.2 with appropriate acid or alkali. The mixture was vigorously shaken by hand for 4 minutes, 3 minutes, 2 minutes and 1 minute, each time adding 5 ml of chloroform subsequently and separating the chloroform layer from the mixture after each time interval. 0.1 gm of 8 – HQ and 5 ml of chloroform was again added and the pH adjusted to 5.2. Similar shaking was done as before and finally the mixture was heated for a considerable period of time in a water bath to remove all trace of chloroform. After purification, all solutions were sterilized in autoclave for 15 minutes at 121°C temperature.

The Chemicals used for biosorption medium were of analytical grade, which were further purified from trace elements. Mineral requirements of *Saccharomyces cerevisiae*A100 was determined by adding the inorganic salt of the metal in graded amounts to obtain the optimum concentration. Macro element requirement of the fungus was assessed by studying the role of potassium (supplied as KCl), phosphorous (added as KH_2PO_4 or K_2HPO_4), calcium (added as $CaCl_2,H_2O$), sodium (added as NaCl), magnesium (added as $MgSO_4,7\ H_2O$),boron (as H_3BO_3), EDTA in different concentration.

Trace element requirement of the fungus was assessed by studying the role of iron (added as $FeSO_4,7H_2O$), copper (added as $CuSO_4,5H_2O$), zinc (added as $ZnSO_4,7H_2O$), manganese (added as $MnSO_4,H_2O$), nickel (added as $NiSO_4,7H_2O$), cobalt (supplied as $CoCl_2,6H_2O$), molybdenum (added as $Na_2MoO_4,2H_2O$), vanadium (added as NH_4VO_3). All trace elements were added at a concentration of 1 – 20 μg/ml. other cultural conditions remained the same as before (Chapter 4, of the Thesis).

Initially, the basal medium did not contain the trace elements to be tested. The mineral salt, under investigation, was addded in different amounts to the above biosorption medium to obtain the optimum biosorption of Hg^{++} from the surrounding medium. The results obtained after successive supplementation of macro and micro elements into the biosorption medium are discussed below.

Result & Discussion:

A. Effect of Macro Element On Biosorption of Hg^{++}

i) Effect of Different Concentration of phosphate source on biosorption of Hg^{++} by *Saccharomyces cerevisiae*A100

TABLE 3: OPTIMIZATION OF SUITABLE PO_4 SOURCE FOR MAXIMUM CELL GROWTH AND BIOSORPTION OF Hg^{++}

Phosphate Source	Concentration (%)	Cell Growth (mg/L)	Biosorption (%)
K_2HPO_4	0.05	21.94	70.4 ± 1.16
	0.075	22.00	72.6 ± 1.17
	0.10	22.90	74.0 ± 1.11
	0.15	23.86	75.2 ± .70
	0.20	22.78	70.6 ± 1.30
	0.25	21.06	67.2 ± 1.22
	0.30	20.60	64.2 ± 1.26
KH_2PO_4	0.05	21.67	62.8 ± .97
	0.075	22.41	68.8 ± .58
	0.10	22.59	71.6 ± .53
	0.15	22.26	70.6 ± .68
	0.20	20.78	65.2 ± .87
	0.25	19.45	58.1 ± .71
	0.30	17.04	55.4 ± .59

Biosorption Values are expressed as mean ± Standard Deviation

All values of cell growth and biosorption are biologically significant ($p < 0.001$).

Phosphate plays essential role for the growth of yeast, and among other functions this element plays important role in carbohydrate metabolism (328). Between KH_2PO_4 and K_2HPO_4, K_2HPO_4 exhibit remarkable positive effect on cell growth of the organism and biosorption of Hg^{++} as well. At lower concentration differences between two salts are not prominent, but as the concentration increases K_2HPO_4 is proved to be more suitable PO_4^{3-} source for the organism. K_2HPO_4 helps to maintain the pH of the biosorption medium at a more suitable range for biosorption of Hg^{++}. The acidic nature of KH_2PO_4 drops the pH down resulting declination in biosorption rate . The optimum concentration of K_2HPO_4 for biosorption is found to be 0.15% (Table.3.).

ii) Optimization of $MgSO_4,7H_2O$ concentration for Hg^{++} biosorption:

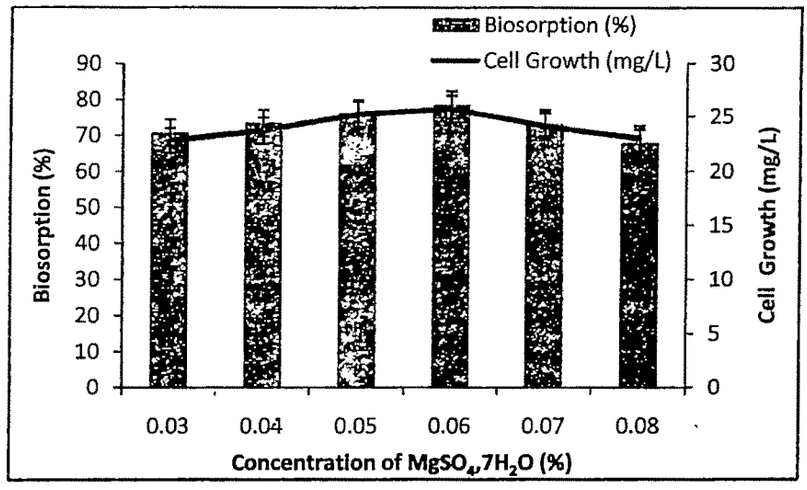

FIGURE 22: EFFECT OF DIFFERENT $MgSO_4$, $7H_2O$ CONCENTRATION ON BIOSORPTION OF Hg^{++}AND CELL GROWTH

Role of magnesium in yeast metabolism is chiefly through its activation influence over various enzyme system, including those of biosorption. Magnesium ion is a very useful co-factor in kinase reaction(156) . The optimum concentration of $MgSO_4$, $7H_2O$ for Hg^{++} biosorption is found to be 0.06% (Fig.22).

iii) Effects of Varying Concentration of NaCl, KCl and $CaCl_2,H_2O$ on biosorption of Hg^{++} by *Saccharomyces cerevisiae*A100

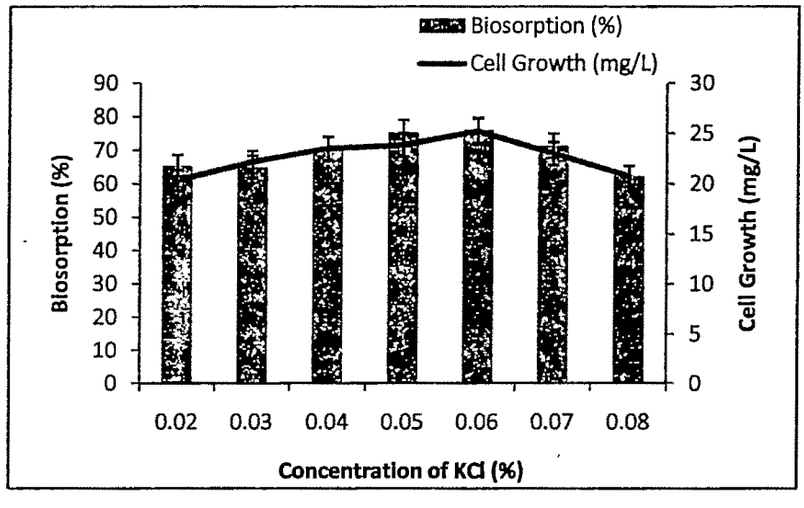

FIGURE 23: EFFECT OF DIFFERENT KCl CONCENTRATION ON BIOSORPTION OF Hg^{++} AND CELL GROWTH

Different concentration of KCl were tested as chloride source for *Saccharomyces cerevisiae*A100.From Fig.23. it is found that 0.06% KCl is optimum for maximum cell growth and biosorption of Hg^{++}.

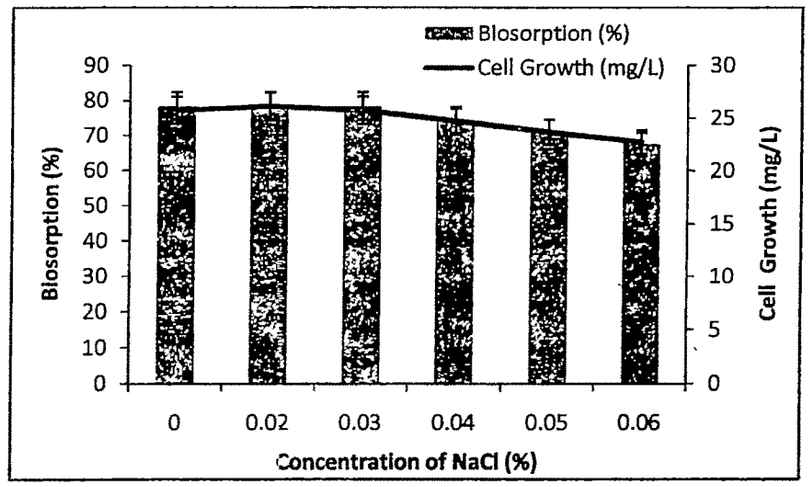

FIGURE 24: EFFECT OF DIFFERENT NaCl CONCENTRATION ON BIOSORPTION OF Hg^{++} AND CELL GROWTH

Fig.24.shows that at lower concentration NaCl does not have any significant effect on Hg^{++} biosorption. When NaCl concentration is increased a little slow down at biosorption is noticed.

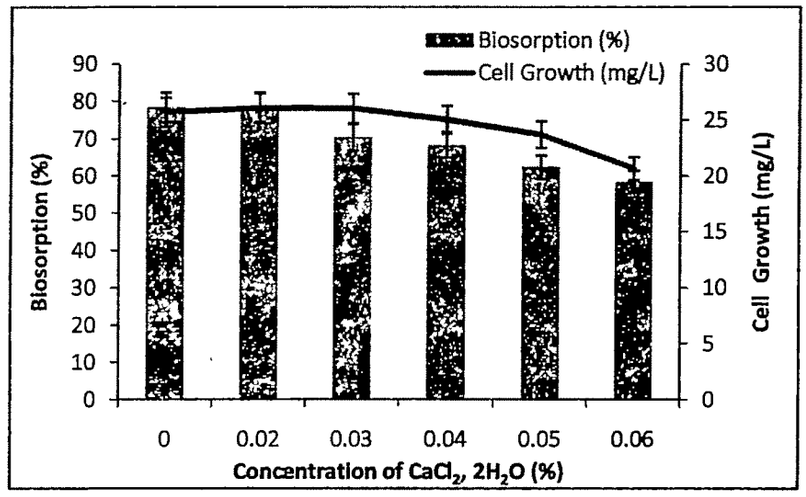

FIGURE 25: EFFECT OF DIFFERENT $CaCl_2$, $2H_2O$ CONCENTRATION ON BIOSORPTION OF Hg^{++} AND CELL GROWTH

Fig.25 indicates that though $CaCl_2$, $2H_2O$ does not show any significant effect on cell growth of *S.cerevisiae*A100 (a very little effect at higher concentration), it significantly declines the biosorption of Hg^{++} at higher concentration.

Chloride ion is essential for fluid and electrolyte balance in the cell. NaCl and $CaCl_2$, $2H_2O$ were also tested as an associate chloride source along with the optimized concentration of KCl. Neither NaCl nor $CaCl_2$, $2H_2O$ shows any promising result on biosorption experiments.The possible reason for such results is potassium seems to be essential for yeast growth while Na and Ca do not appear to be very important in this purpose (328).

Chlorine is considered to be a powerful oxidizing agent that causes harm to microbial cell. Supply of excess chloride salt (KCl & NaCl) hence decreases the cell growth as well as biosorption (226).

iv) Effect of H_3BO_3 on biosorption of Hg^{++}:

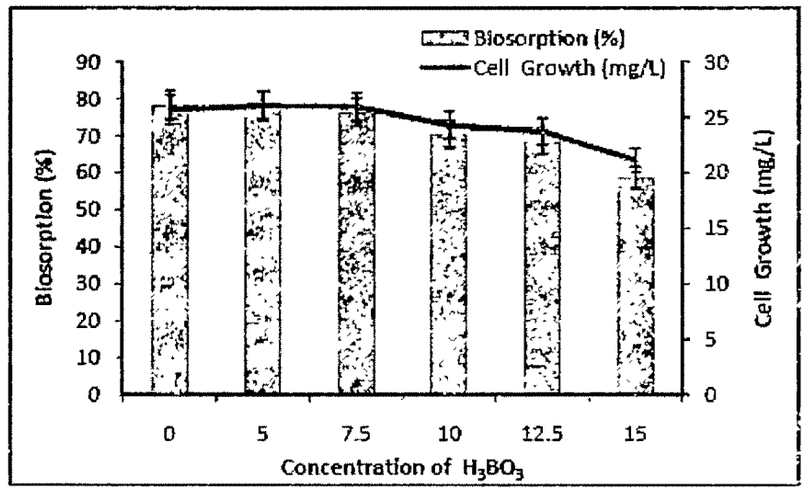

FIGURE 26: EFFECT OF DIFFERENT H_3BO_3 CONCENTRATION ON BIOSORPTION OF Hg^{++} AND CELL GROWTH

From Fig.26 it is concluded that H_3BO_3 has very little effect on cell growth of *S.cerevisiae*A100 at higher concentration and it significantly slows down the biosorption rate higher concentration.

v) Effect of EDTA on biosorption of Hg^{++}:

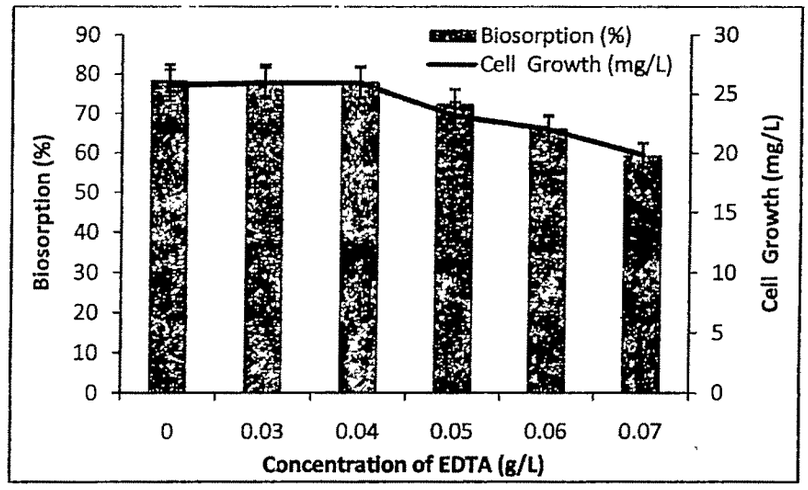

FIGURE 27: EFFECT OF DIFFERENT EDTA CONCENTRATION ON BIOSORPTION OF Hg^{++} AND CELL GROWTH

Fig.27 depicted that at higher concentration EDTA shows negative effect on cell growth of *S.cerevisiae*A100 and biosorption of Hg^{++}.

B. Effect of Trace Element On Biosorption of Hg^{++}

Effects of several trace elements on cell growth of *Saccharomyces cerevisiae*A100 as well as biosorption of Hg^{++} were studied by adding the trace metals in biosorption medium on zero day at concentration varying from 1 to 20 µg/ml .

i) Effect of Different Concentration of Ferrous Ion (Fe^{2+}) :

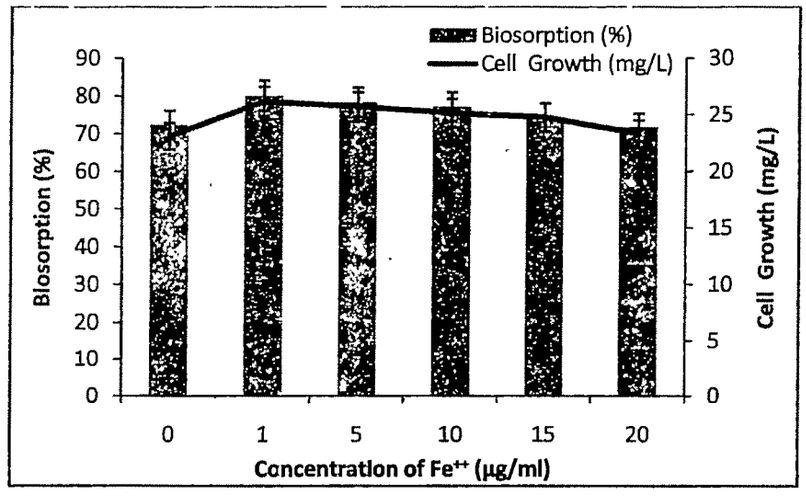

FIGURE 28: EFFECT OF DIFFERENT Fe^{++} ION CONCENTRATION ON BIOSORPTION OF Hg^{++} AND CELL GROWTH

Iron is essential constituent of yeast protoplasm. Association of iron with various enzymes including cytochrome oxidase, catalase and others have lent much support to the possible role of iron(328). In our experiment Fe^{++} ion had a positive effect on biosorption of Hg^{++} and cell growth of yeast at concentration of 1µg/ml (Fig. 28).

ii) Effect of different concentration of Zn^{++} ion as $ZnSO_4,7H_2O$:

TABLE 4: EFFECT OF DIFFERENT Zn^{++} ION CONCENTRATION ON BIOSORPTION OF Hg^{++} AND CELL GROWTH

Concentration of Zn^{++} ion (µg/ml)	Cell Growth (mg/L)	Biosorption (%)
Control	26.14	80.00 ± 0.37
1	23.32	72.60 ± 0.62
5	22.44	67.10 ± 0.23
10	20.05	65.80 ± 0.34
15	19.20	63.20 ± 0.39
20	18.68	60.22 ± 0.44

Biosorption Values are expressed as mean ± Standard Deviation

All values of cell growth and biosorption are biologically significant ($p < 0.001$).

From Table.4 it is observed that Zn^{++} ion has only a little effect on cell growth of *S.cerevisiae*A100 and biosorption of Hg^{++} at higher concentration.

iii) Effect of different concentration of Mn^{++} ion as $MnSO_4,H_2O$:

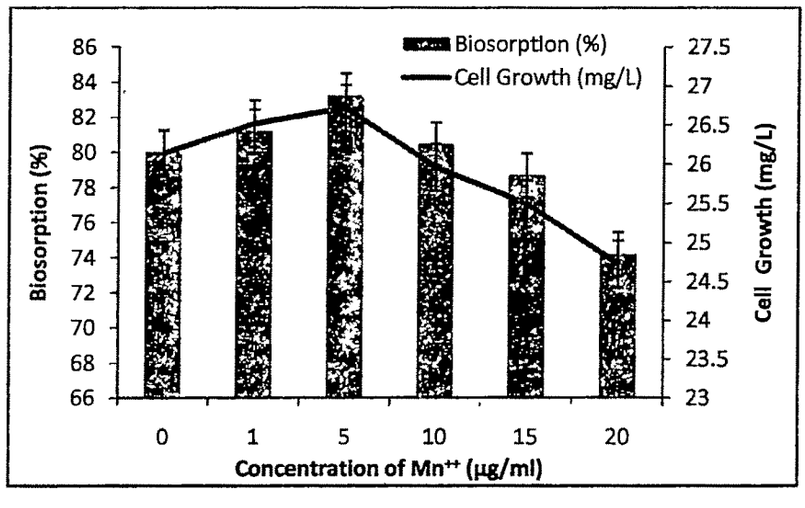

FIGURE 29: EFFECT OF DIFFERENT Mn^{++} ION CONCENTRATION ON BIOSORPTION OF Hg^{++} AND CELL GROWTH

Manganese is known to effect the cellular concentration as well as activity of various enzymes. It is essential for various reductase enzymes as a co-factor(328). Mn^{++} ion had a positive effect upto a concentration of 5µg/ml on Hg^{++} biosorption and cell growth (Fig.29).

iv) Effect of different concentration of Cu^{++} ion as $CuSO_4,5H_2O$:

TABLE 5: EFFECT OF DIFFERENT Cu^{++} ION CONCENTRATION ON BIOSORPTION OF Hg^{++} AND CELL GROWTH

Concentration of Cu^{++} ion (µg/ml)	Cell Growth (mg/L)	Biosorption (%)
Control	26.71	83.26 ± 0.52
1	22.52	70.86 ± 0.64
5	20.76	66.10 ± 0.33
10	17.66	51.20 ± 0.47
15	16.45	46.30 ± 0.62
20	14.05	40.72 ± 0.36

Biosorption Values are expressed as mean ± Standard Deviation

All values of cell growth and biosorption are biologically significant ($p< 0.001$).

Table.5 indicates that Cu^{++} ion has a very adverse effect on growth of *S.cerevisiae*A100 and biosorption of Hg^{++}.

v) Effect of different concentration of Ni^{++} ion as $NiSO_4,7H_2O$:

TABLE 6: EFFECT OF DIFFERENT Ni^{++} ION CONCENTRATION ON BIOSORPTION OF Hg^{++} AND CELL GROWTH

Concentration of Cu^{++} ion (µg/ml)	Cell Growth (mg/L)	Biosorption (%)
Control	26.71	83.26 ± 0.52
1	24.00	78.26 ± 0.32
5	22.20	69.00 ± 0.50
10	19.60	56.27 ± 0.64
15	16.74	50.00 ± 0.35
20	12.60	42.20 ± 0.44

Biosorption Values are expressed as mean ± Standard Deviation

All values of cell growth and biosorption are biologically significant ($p< 0.001$).

From Table.6 it is observed that Ni^{++} ion exhibits a toxic effect to the cell growth of *S.cerevisiae*A100 and as a result biosorption of Hg^{++} declines significantly.

vi) Effect of different concentration of Co^{++} ion as $CoCl_2,6H_2O$:

TABLE 7: EFFECT OF DIFFERENT Co^{++} ION CONCENTRATION ON BIOSORPTION OF Hg^{++} AND CELL GROWTH

Concentration of Cu^{++} ion (µg/ml)	Cell Growth (mg/L)	Biosorption (%)
Control	26.71	83.26 ± 0.52
1	24.80	77.50 ± 0.33
5	24.00	73.30 ± 0.65
10	22.02	66.60 ± 0.59
15	20.44	60.30 ± 0.46
20	17.01	54.70 ± 0.60

Biosorption Values are expressed as mean ± Standard Deviation

All values of cell growth and biosorption are biologically significant ($p< 0.001$).

Table.7 shows that at higher concentration Co^{++} ion has negative influence on cell growth of *S.cerevisiae*A100 and biosorption of Hg^{++}.

vii) Effect of different concentration of Mo^{6+} ion as $Na_2MoO_4,2H_2O$:

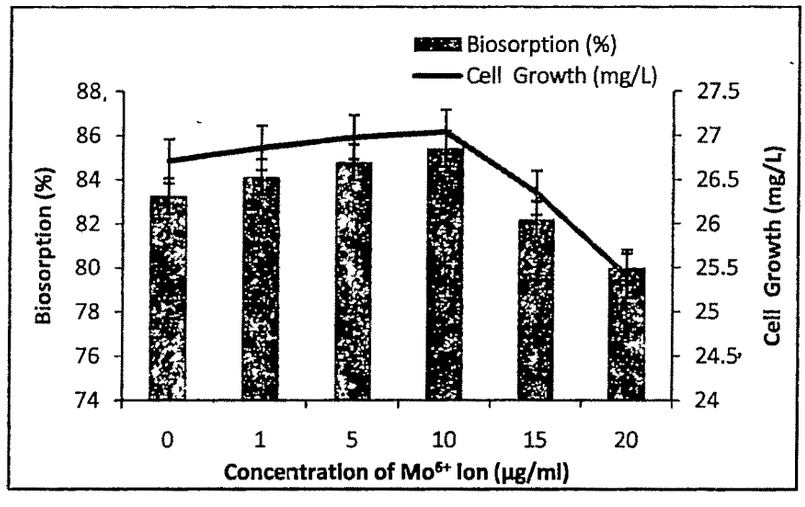

FIGURE 30: EFFECT OF DIFFERENT Mo^{6+} ION CONCENTRATION ON BIOSORPTION OF Hg^{++} AND CELL GROWTH

Molybdenum acts as co-factor in some oxido-reductase enzymes and takes part in electron transfer (156). The positive effect of Mo^{6+} upto a concentration of 10µg/ml on Hg^{++} biosorption and cell growth is revealed in our experiments (Fig.30).

viii) Effect of different concentration of V^{5+} ion as NH_4VO_3:

TABLE 8: EFFECT OF DIFFERENT V^{5+} ION CONCENTRATION ON BIOSORPTION OF Hg^{++} AND CELL GROWTH

Concentration of V^{3+} ion (µg/ml)	Cell Growth (mg/L)	Biosorption (%)
Control	27.04	85.40 ± 0.25
1	26.62	81.21 ± 0.41
5	25.11	79.25 ± 0.30
10	24.32	75.60 ± 0.42
15	22.20	68.60 ± 0.53
20	21.80	63.30 ± 0.40

Biosorption Values are expressed as mean ± Standard Deviation

All values of cell growth and biosorption are biologically significant ($p < 0.001$).

It is observed from Table.8 that V^{5+} ion is toxic to the cell and has negative influence on biosorption of Hg^{++} at higher concentration.

Thus, after optimizing all those factors, the resulting synthetic biosorption medium consisted of glucose (5%), urea (0.15%), K_2HPO_4 (0.15%), $MgSO_4,7H_2O$ (0.06%), KCl (0.06%), Fe^{++} (1µg/ml), Mn^{++} (5µg/ml), Mo^{6+} (10µg/ml).