2 *Egypt. J. Bot.*, **Vol. 54**, No, **1**. pp. 13-27 (2014)

Optimization of Biosorption Conditions of Hexavalent Chromium by *Aspergillus niger* and *Spicaria silvatica* Mycelial Mats, Using Taguchi Orthogonal Array

[1]N.A. Tharwat, [1]T.M. A. Abdel-Rahman, [2]A.H Mostafa and [2]M.A. Ahmed

[1]Botany department, Faculty of Science, Cairo University and [2]National Center for Research on Housing and Building, Giza, Egypt.

IN ORDER to maximize the biosorption efficiency, eighteen experiments were designed to investigate the potentialities of fungal biomass as a resistant and cost effective Cr^{+6} biosorbent. The influences of individual parameters as well as the interaction between them were studied using Taguchi statistical design. A group of statistical analysis that can define the relationships between the responses of independent variables were carried out. In the current study, it was detected that the biosorbed Cr^{+6} by *A. niger* and *S. silvatica* varied greatly among the 18 media used indicating the importance of optimization experiments in bioprocesses. In case of *A. niger* the ANOVA analysis of variance revealed that temperature, mycelium status, $NaNO_3$ and agitation were the most significant factors influencing the Cr^{+6} biosorption .In *S. silvatica*, temperature, mycelium status, agitation and $MgSO_4$ $7H_2O$ were the highly significant variables influencing Cr^{+6} biosorption .The design detected that the experimental values of biosorbed Cr^{+6} were in reasonable agreement with the predicted values and the model was significant. The optimized predicted medium from the software design for Cr^{+6} biosorption by *A. niger* and *S. silvatica* was (A) dead mycelium (B) sucrose (15 gl^{-1}) (C) $NaNO_3$ (1.5 gl^{-1}) (D) $MgSO_4$ $7H_2O$ (0.1 gl^{-1}) (E)temperature (18 ºC) (F) pH (5) (G) cultivation (static)(H) incubation period (5days).

Keywords: Biosorption, Hexavalent chromium, *Aspergillus niger*, *Spicaria silvatica*, Tannery pollution.

The tanning industry forms the backbone of the Egyptian leather industry. The total numbers of tanneries in Egypt are more than 300, of which more than 85% adopt the chromium tanning process (Ibrahim. and Shalaby, 1991) because of its processing speed, low costs, and light color of leather and greater stability of the resulting leather. Tannery effluent containing chromium is one of the most recognized problems in leather industry. Tanning process using chromium compounds is the most common methods for processing of hides (Sreeram, and Ramasami, 2003).

Biosorption is such a technology for treatment of industrial wastewater by interaction with live or dead biological matter and is the most practical and widely used approach for bioremediation of toxic metals and radio nuclides (Volesky, 1990). Biosorption is also defined as a process that utilizes inexpensive dead biomass to sequester toxic heavy metals and to remove contaminants from industrial effluents (Ting *et al*., 1991 and Singh *et al*. 1998). The use of biological materials for heavy metal removal and recovery technologies had gained important credibility, because of the good performance, low cost of this complex material and could be considered as an eco-friendly complementary device to the existing high cost technologies. The natural affinity of biological compounds for metallic elements could contribute to economically purifying heavily metal-loaded wastewater (Ferraz and Teixera, 1999). Many microorganisms had been examined for their biosorptive properties. Fungi, Yeasts, bacteria, and algae had been identified as superior candidates for bioremediation owing to their ability to sequester cationic and anionic metallic species from their aqueous environment (Luef *et al*. 1991; Mclean *et al*. 1994; Tobin *et al.* 1994; Volesky and Holan, 1995 and Volesky and May-Philips, 1995 and Anaemene, 2012). Most studies of biosorption for metal removal had involved the use of either laboratory-grown microorganism or waste biomass generated by the pharmacology and food processing industries or wastewater treatment units (Rao *et al*. 1993). Both living and dead biomass could be used to remove metals, but maintaining a viable biomass during metal adsorption was difficult, because it required a continuous supply of nutrients and avoidance of metal toxicity to the microorganisms (Spinti *et al*. 1995). Use of dead biomass could avoid these problems.

The bioadsorptive capacity of the heat-inactivated cells might be greater, equivalent or less than that of living cell. However, the use of heat inactivated biomass in industrial application might offer some advantages over living cells, such as less sensitive to heavy metal ions concentration and adverse operating conditions (i.e., pH and temperature) (Gadd and White, 1989 and Kapoor *et al*. 1999). Also dead biomass could be regenerated and reused for many cycles. However, the use of dead biomass in powdered form had some problems, such as; difficulty in the separation of biomass after adsorption, mass loss after regeneration, low strength and small particle size, which made it difficult to use in column applications (Kapoor and Viraraghavan, 1998 and Yan and Viraraghavan, 2000). Dead biomass could be immobilized in a granular or polymeric matrix to improve the mechanical strength of the biosorbent. The immobilized biomass was ideal for use in a conventional ion-exchange column or adsorption column (Volesky, 1990).

The traditional “one factor-at-a time” technique used for optimizing a multivariable system is not only time waste, but also require carrying out a number of experiments to determine the optimum levels when the interaction is significant. Statistical optimization design could eliminate these drawbacks by

optimizing all the affecting parameters collectively (Doehlert, 1970). Response service methodologies (RSM) involve four major steps: i) performing the statistically designed experiments, ii) estimating the coefficient in a mathematical model, iii) predicting its response and iv) checking the adequacy of the model (Dasu and Panda, 2000; Reddey *et al*. 2008). Statistically designed experiments minimize the error in determining the effect of parameters in an economical manner (Sharma and Satyanarayan, 2006).

Process optimization plays a vital role in industrial production process in which even small improvement would be decisive for commercial success. In any bioprocess, the small improvement in productivity of any metabolite such as enzymes, hormones, toxins would be achieved through manipulation of the nutritional and physical parameters (Reddy *et al*. 2008).

The objective of this work is to optimize the biosorption conditions of hexavalent chromium by *Aspergillus niger* and Spicaria silvatica mycelial mats, using Taguchi Orthogonal Array (Prasad et al., 2005).

Material and Methods

Test organisms

Two fungal species isolated from Cr^{3+} and Cr^{6+} polluted soil with tannery water (Tharwat et al., 2013).

Estimation of adsorbed and absorbed Cr^{+6} by fungal species

Definite weight of fresh mycelia were washed 3 times with 1.0 M HCL and the washes were pooled and used for determination of the adsorbed chromium on the surface of fungal mycelia . The absorbed chromium inside mycelia was determined in dry biomass after grounding with mortar and pestle. 0.1 g of crushed mycelia was digested in 1 ml of concentrated nitric acid and heated in boiling water bath for 1 hr. After cooling the digest was diluted with an appropriate volume of ddH_2O. Chromium content of diluted mycelial extracts and culture filtrates were determined spectrophotometricaly after complication of the metal ions with 1, 5- diphenyle carbazide at 540 nm (Bartlett and James., 1996).

Optimization of the biosorption conditions of hexa valent chromium by Aspergillus niger and Spicaria silvatica mycelial mats

The size of experimentation was represented by symbolic arrays L18 which indicates 18 experimental trails). Seven indicates 18 experimental trails). 5) factors have been assigned with three levels (7^3) except mycelium was assigned with 2 levels (1^2) with a layout of L18 ($1^2 \times 7^3$) (Table 2).

TABLE 1. Selected variables and their assigned levels for optimization process using Taguchi software design expert 8 was used in this optimization system (Prasad *et al.*, 2005).

variables	Level 1	Level 2	Level 3
(A)Mycelium	Dead (250mg %)	Alive (2disc %)	---
(B) Sucrose (g/L)	15	20	30
(C)Sodium nitrate (g/L)	1.5	3	4.5
(D) MgSO4 7H2O (g/L)	0.1	0.5	1
(E) Temperature (ºC)	18	28	38
(F) pH	5	7	9
(G) Agitation(rpm)	Static	120	200
(H) Incubation(day)	5	7	10

TABLE 2. Fractional factorial design of L-18 (1^2X7^3) orthogonal array used for optimization process.

Experiments	Designed Experiments							
No.	1	2	3	4	5	6	7	8
1	1	1	1	1	1	1	1	1
2	1	1	2	2	2	2	2	2
3	1	1	3	3	3	3	3	3
4	1	2	1	1	2	2	3	3
5	1	2	2	2	3	3	1	1
6	1	2	3	3	1	1	2	2
7	1	3	1	2	1	3	2	2
8	1	3	2	3	2	1	3	1
9	1	3	3	1	3	2	1	2
10	2	1	3	3	3	2	2	1
11	2	1	1	1	1	3	3	2
12	2	1	2	2	2	1	1	3
13	2	2	1	2	3	1	3	2
14	2	2	2	3	1	2	1	3
15	2	2	3	1	2	3	2	1
16	2	3	1	3	2	3	1	2
17	2	3	2	1	3	1	2	3
18	2	3	3	2	1	2	3	1

Eighteen media were designed. The composition of them was recorded in Table 3.

TABLE 3. Composition of The 18 designed media from the interactions of 3 levels of the eight factors.

Media	Mycelium (A) mg%	Sucrose (B) g/L	$NaNO_3$ (C) g/L	MgSO 4 7H2O (D) g/L	Temp (E) °C	pH (F)	Agitation (G) Rpm	Incubation (H) Days
1	Dead	15	1.5	0.1	18	5	Static	5
2	Dead	15	3	0.5	28	7	120	7
3	Dead	15	4.5	1	38	9	200	10
4	Dead	20	1.5	0.1	28	7	200	10
5	Dead	20	3	0.5	38	9	Static	5
6	Dead	20	4.5	1	18	5	120	7
7	Dead	30	1.5	0.5	18	9	120	7
8	Dead	30	3	1	28	5	200	5
9	Dead	30	4.5	0.1	38	7	Static	7
10	Alive	15	4.5	1	38	7	120	5
11	Alive	15	1.5	0.1	18	9	200	7
12	Alive	15	3	0.5	28	5	Static	10
13	Alive	20	1.5	0.5	38	5	200	7
14	Alive	20	3	1	18	7	Static	10
15	Alive	20	4.5	0.1	28	9	120	5
16	Alive	30	1.5	1	28	9	Static	7
17	Alive	30	3	0.1	28	5	120	10
18	Alive	30	4.5	0.5	18	7	200	5

Analysis of experimental data and prediction of performance

The obtained experimental results were fitted in Taguchi (Prasad *et al*., 2005) software (Design-Expert 8- or-Qualitek4) to analyze the influences of individual and interactive factors. ANOVA used to determine the optimum culture conditions for biosorption and to detect the contribution of each selected factor for biosorption activity of fungi .The Qualitek-4 software was equipped to use L-4 to L-64 arrays along with selection of 2 to 63 factors with two, three and four levels to each factor. The automatic design option allows qualitek-4 to select the array used and assign factors to the appropriate flasks. Software operation for optimization was performed at (bigger is better) performance characteristics for all the cases.

Results and Discussion

The data in Table 4 indicated that the experimental actual values of the biosorbed Cr^{+6} by *A. niger* and *S. silvatica* assessed significant variation among the 18 designed media. It varies from 2.80 to 45.20 µg ml^{-1} in *A. niger* and from 2.20 to 40 µg ml^{-1} in *S. silvatica*. This indicated the importance of cultural optimization in metal biosorption by the microorganisms. The optimum medium for *A. niger* was medium (2) containing (dead mycelium, sucrose, 15 g/l,

NaNO3, 3 g/l, Mg SO4 $7H_2O$, 0.5 g/l, temperature 28 ºC, pH 7, agitation speed 120 rpm, incubation period 7 days).

while the optimum medium for Cr6+ biosorption by *S. silvatica* was medium (3) containing **(**dead mycelium, sucrose, 15 g/l, NaNO3, 4.5 g/l, Mg SO4 $7H_2O$, 1.0 g/l, temperature 38 ºC, pH 9, agitation speed 200 rpm, incubation period 10 days).

TABLE 4. Optimization of biosorption conditions of hexavalent chromium (experimental values) by *Aspergillus niger* and *Specaria silvatica* mycelial mats using Taguchi Orthogonal Array (OAs) Design.

Exp. no	*Aspergillus niger*	*Spicaria silvatica*
	Biosorbed Cr^{+6}(µg/ml)	**Biosorbed Cr^{+6}(µg/ml)**
1	5.2	5.6
2	45.2	11.2
3	40	34.2
4	16.8	23
5	30.4	5.8
6	3.8	15
7	13.2	8.2
8	23.4	40
9	6.2	20
10	14	8.8
11	2.8	2.2
12	14.6	17.6
13	13.4	3.2
14	5.2	4.2
15	3.2	6.8
16	8.8	9.4
17	40	16.2
18	5.4	7

The diagnostic case statistics of Cr^{+6} biosorption by *A. niger* and *S. silvatica* detected that the actual experimental values were in reasonable agreements with the predicted values in 11 of 18 media .The residual values, however, exceeded limits (4 $\mu g ml^{-1}$) in 7 of 18 media which were of numbers 1,3,7,10,13,15 and 16 in case of *A. niger* and 5,7,8,9,12,14&15 in case of *S. silvatica* (Table 5).

From ANOVA analysis of variance (Table 6) the F-value of the source model is equal 15.24 & 15.75 for *A. niger* and *S. silvatica*, respectively. This implies that the source models are significant. Furthermore, the P- value of less than 0.05 indicates that the model variables are significant. In case of *A. niger* $NaNO_3$ and temperature are the most significant variables affecting biosorption of Cr^{+6}. Mycelium status, sucrose and agitation came next in significance. In case of *S. silvatica*, mycelium status, $NaNO_3$, $MgSO_4$ $7H_2O$, temperature and agitation are highly significant variables in Cr^{+6} biosorption where the P values were less than 0.05.

TABLE 5. Diagnostic case statistics of chromium biosorption by *A. niger* and *S. silvatica*.

	Actual value (µg ml^{-1})		Predicted value (µg ml^{-1})		Residual value (µg ml^{-1})		Student· s (t-test)	
	A. niger	*S. silvatica*	*A. niger*	*S. silvatica*	*A. niger*	*S. silvatica*	*A. niger*	*S. silvatica*
1	5. 20	5.6	12.08	4.10	-6.88*	1.50	-2.86	0.28
2	45.20	11.20	42.91	12.20	3.29	-1.00	1.210	-0.459
3	40.00	34.20	31.91	33.32	8.09*	0.88	2.14	0.193
4	16.80	23.00	20.26	25.37	-3.46	-2.37	-0.662	-0.400
5	30.40	5.80	27.30	10.77	3.10	-4.97*	0.423	-0.877
6	3.80	15.00	1.59	13.47	2.21	1.53	6.302	0.255
7	13.20	6.20	9.35	1.24	5.85*	6.96*	1.524	1.242
8	23.40	40.00	26.81	34.15	-3.41	8.85*	-1.496	1.184
9	6.20	20.00	6.64	14.24	-0.44	5.76*	-0.154	1.037
10	14.00	8.80	20.49	8.78	-6.49*	0.22	-2.160	0.005
11	2.80	2.20	1.24	-0.43	1.56	2.63	0.262	0.824
12	14.60	17.60	17.35	11.60	-2.75	6.00*	-0.300	1.034
13	13.40	3.20	19.39	6.24	-5.99*	-3.04	-1.066	-0.739
14	5.20	40.20	9.16	12.87	-3.96	-8.67*	-0.602	-1.771
15	3.20	6.80	9.76	15.67	-6.59*	-8.87*	-1.294	-1.732
16	8.80	8.80	-3.13	8.78	10.93*	0.022	4.11	0.005
17	40.00	16.20	38.27	14.84	1.73	1.36	0.364	0.227
18	5.40	7.00	2. 18	10.94	3.22	-3.94	0.537	-0.679

(*) Exceed limits.

TABLE 6. ANOVA analysis of variance for selected factorial model.

Variables	Sum of squares (R^2)		Degree of freedom (df)		Mean square		F value		P value	
	A. niger	*S. silvatica*	*A. niger*	*S. silvatica*	*A. niger*	*S. silvatica*	*A. niger*	*S. silvatica*	*A. niger*	*S. silvatica*
(A) mycelium status	213.39	611.41	1	1	213.39	611.41	13.60	16.25	0.0942	0.004
(B) sucrose	424.54	656	2	-	212.27	328	3.59	8.25	0.0772	0.061
(C) sodium nitrate	835.73	682	2	-	417.87	341	7.06	4.62	0.0171	0.003
(D) Mg SO4	-	460.58	-	2	-	230.29	-	6.12	-	0.0184
(E)Temperature	745.21	444.18	2	2	372.60	222.09	6.26	5.90	0.0228	0.0203
(F) Agitation	459.30	386.16	2	10	229.65	37.62	3.88	3.22	0.0664	0.0192
Sourcmodel (Significant model)	2794.6	1513.55	9	7	310.5	216.22	15.24	15.75	0.0145	0.0070

The regression analysis revealed that the observed model errors are very low in both fungal species which indicated the accuracy of experimentation (Table 7) .The standard deviation values were 3.69 and 3.13 for *A. niger* & *S.silvatica* with values of R-square 0.8551 and 0.8009, respectively. Furthermore the predicted R-squares of 0.6646 & 0.6493 were in reasonable agreement with adjusted R-squares 0.6920 and 0.6615 in *A. niger* and *S. silvatica*, respectively. The adequate precision of 7.330 and 8.455 indicate an adequate signal for Cr^{+6} biosorption by *A. niger* and *S. silvatica*, respectively, as a ratio greater than four is desirable in Taguchi system (Prasad *et al.*, 2005).

TABLE 7. The regression analysis values.

Fungal species	Std.dev.	Mean	Coefficient variance	Precission	R-square	Adj R-square	Pred.R-square	Adeq precision
A.niger	3.69	16.20	47.50	3056.68	0.8551	0.6920	0.6646	7.330
S.silvatica	3.13	13.24	46.31	1097.48	0.8009	0.6615	0.6493	8.455

TABLE 8. The coefficient estimate with standard error.

Variables	Coefficient estimate		Standard error		Coefficient Indix (95%)	
	A. niger	***S. silvatica***	***A. niger***	***S. silvatica***	***A. niger***	***S. silvatica***
Intercept	14.13	9.21	2.84	1.77	7.58	5.18
(A)	-5.41	-7.42	2.84	1.84	-11.98	-11.52
B(1)	8.43	-	1.09	-	15.77	-
B(2)	-3.16	-	-9.27	-	2.95	-
C(1)	0.41	-	-7.41	-	8.23	-
C(2)	9.07	-	1.84	-	16.30	-
D(1)	-	-1.77	-	-6.35	-	2.81
D(2)	-	-5.24	-	-9.81	-	-0.66
E(1)	-8.78	-7.04	-15.35	-11.61	-2.21	-2.46
E(1)	-0.58	3.93	-6.81	-0.65	5.65	8.51
G(1)	-7.51	-3.64	-13.75	-8.21	-1.28	0.94
G(2)	-0.46	-3.04	-2.85	-7.61	9.76	1.54

The estimate model coefficient and their associated statistics were reported in (Table 8).

Final equation in terms of coded factors:

Biosorbed Cr^{+6} by (*A. niger*) =+14.13-5.41+8.43-3.16 +0.41+9.07-8.78-0.58 -7.51-0.46

Biosorbed Cr^{+6} by (*S. silvatica*) = +9.21-7.42-1.77-5.24 -7.04+3.93-3.64-3.04

The optimized predicted medium from the Taguchi designed for Cr^{+6}biosorption by *A. niger* & *S. silvatica* was composed of: (A)Mycelium status (dead), (B) Sucrose (15 gL^{-1}), (C) $NaNO_3$(1.5 gL^{-1}), (D) $MgSO_4$ $7H_2O$ (0.1 gL^{-1}), (E)Temperature (18ºC), (F) pH (5), (G)Agitation (static), (H) Incubation period (5 days).

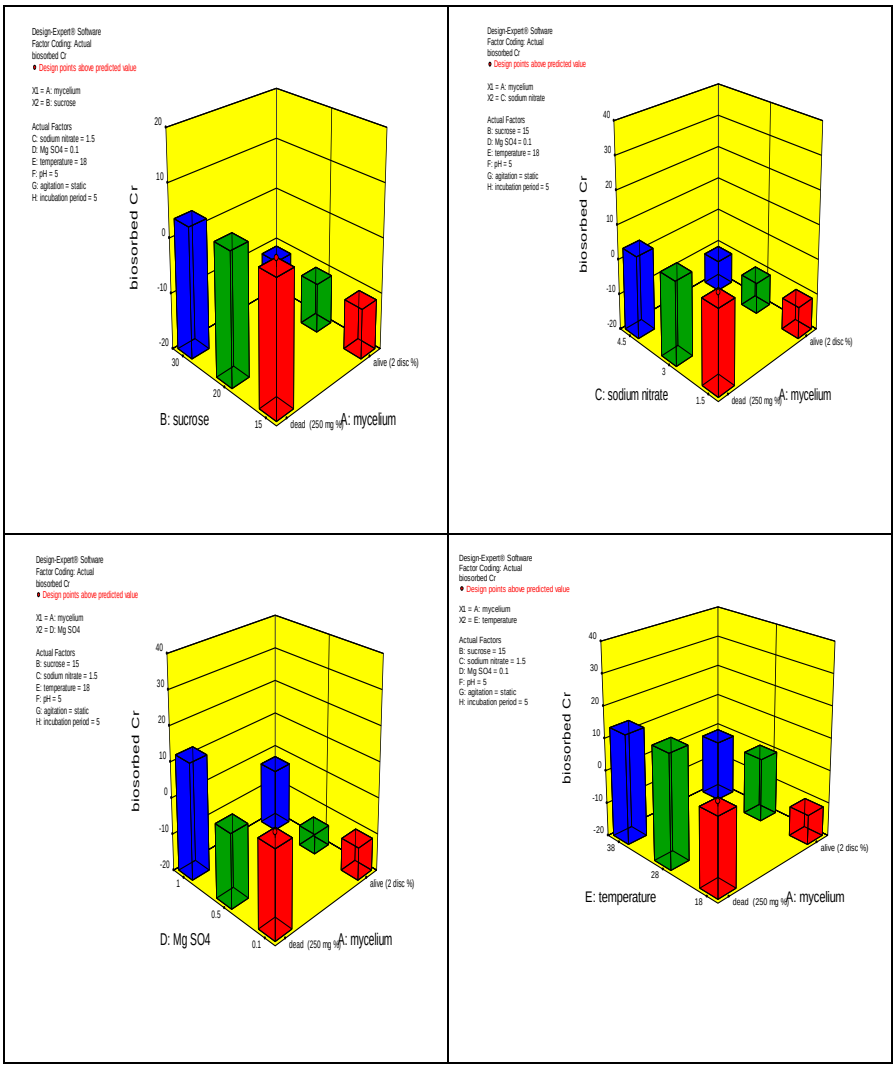

Fig. 1a. Variable interactions with the mycelium (dead & alive) of *S.silvatica* in Cr^{+6} biosorption.

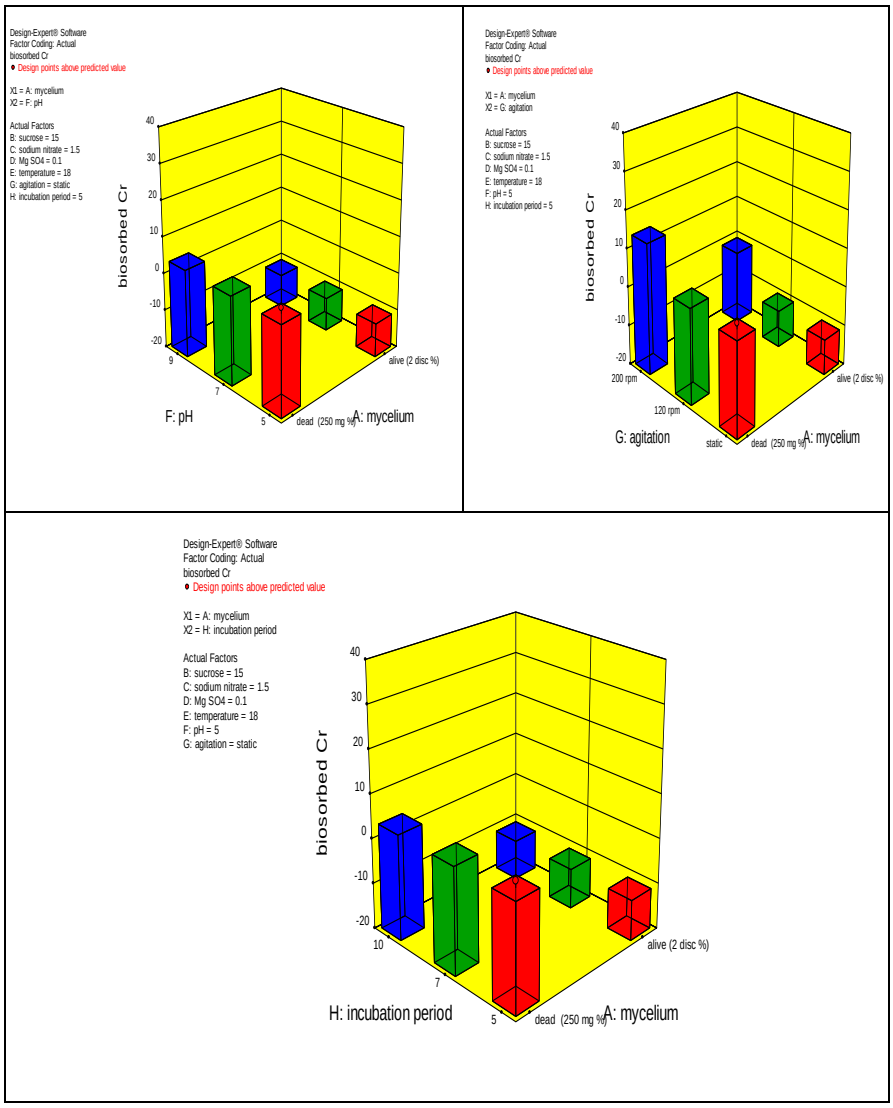

Fig. 1 b. Variable interactions with the mycelium (dead & alive) of *S.silvatica* in Cr^{+6} biosorption.

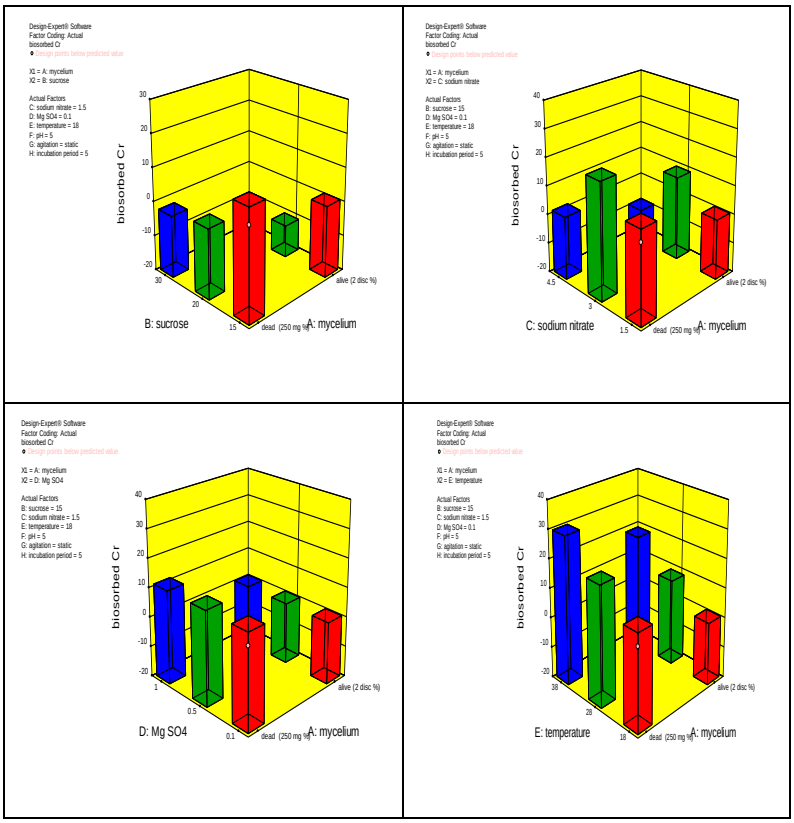

Fig. 2 a. Variable interactions with the mycelium (dead & alive) of *A.niger* in Cr^{+6} biosorption.

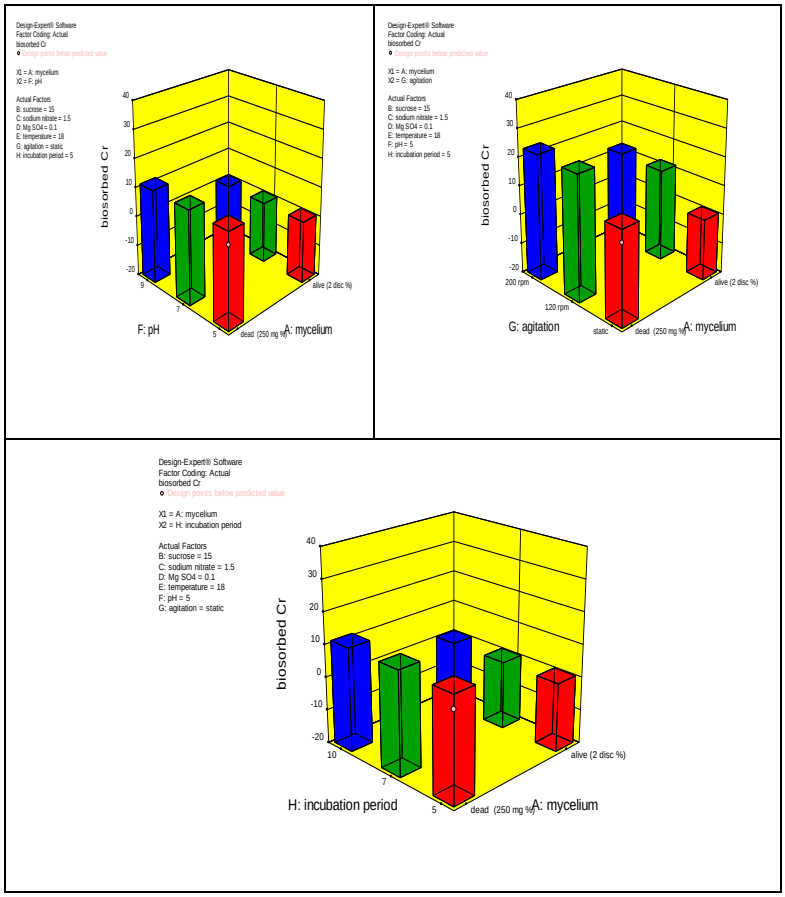

Fig. 2 b. Variable interactions with the mycelium (dead & alive) of *A.niger* in Cr^{+6} biosorption

References

1. **Anaemene, I. A. (2012)** The use of *Candida* sp. In the biosorption of heavy metals from industrial effluent. *European Journal of Experimental Biology*, **3**, 484-488.

2. **Bartlett, R.J. and James, B. R. (1996)** Chromium. In *"Methods of Soil Analysis"*. D. L. Sparks (Ed.), Part3.SSSA, Madison , p. 683-701.

3. **Dasu, V. V. and Panda, T. (2000) Optimization** of microbiological parameters for enhanced griseofulvin production using response surface methodology. *Bioprocess Engineering*, 22, 45–49.

4. **Doehlert, D. H. (1970)** Uniform shell designs. *Applied Statistics*, **19**, 231-239.

5. **Ferraz, A. I. and Teixera, J. A. (1999)** The use of flocculating brewer's yeast for Cr(III) and Pb(II) removal from residual wastewater. *Bioprocess and Biosystems Engineering*, **21,** 431 – 437.

6. **Gadd, G. M. and White, C. (1989)** Removal of thorium from simulated acid proc streams by fungal biomass. *Journal of Chemical Technology & Biotechnology*, **33,** 592–7.

7. **Ibrahim, M. and Shalaby, Z. (1991)** Productivity Improvement for the Egyptian NationalEconomy, with emphasis on the Industrial Sector: Measurements and Assessment of Productivity in the Textiles, Fabrics and Leather Industries. National Research Institute, Egypt.

8. **Kapoor, A. and Viraraghavan, T. (1998)** Biosorption of heavy metals on *Aspergillus niger*: Effect of pretreatment. *Bioresource Technology*, **63,**109-113.

9. **Kapoor, A., Viraraghavan, T. and Cullimore, D. R. (1999)** Removal of heavy metals using the fungus *Aspergillus niger*. *Bioresource Technology*, **70,** 95-104.

10. **Luef, E., Prey, T. and Kubicek, C. P. (1991)** Biosorption of zinc by fungal mycelial wastes. *Applied Microbiology Biotechnology*, **34,** 688-692.

11. **Mclean, R. J. C., Campbell, A. M., Khu, P. T., Persaud, A. T., Bickerton, L.E. and Beau C. D. (1994)** Repeated use of *Bacillus subtilis* cell walls for copper binding. *World Journal of Microbiology and Biotechnology*, **10,** 472-474.

12. **Prasad, K. K., Mohan, S. V., Rao, R. S., Pati, B. R. and Sarma, P. N. (2005)** Laccase production by *Pleurotus ostreatus* 1804 Optimization of submerged culture conditions by Taguchi DOE methodology. *Journal of Biochemical Engineering*, **24**,17-26.

13. **Rao, C. R. N.; Iyengar, L. and Venkobachar, C. (1993)** Sorption of copper (II) from aqueous phase by waste biomass. *Journal of Environmental Engineering*, **119,** 369 – 377.

14. **Reddy, L. V. A., Wee, Y., Yun, J. and Ryu, H. (2008)** Optimization of alkaline protease production by batch culture Bacillus sp. RKY3 through Plackett–Burman and response surface methodological approaches. *Bioresource Technology*, **99**(7), 2242–2249.

15. **Sharma, D. C. and Satyanarayana, T. (2006)** A marked enhancement in the production of a highly alkaline and thermostable pectinase by Bacillus pumilus dcsr1 in submerged fermentation by using statistical methods. *Bioresource Technology*, **97**, 727–733.

16. **Singh, S.; Pradhan, S. and Rai, L. C. (1998)** Comparative assessment of Fe^{3+} and Cu^{2+} biosorption by field and laboratory grown *Microcystis*. *Process Biochemistry*, **33,** 495–504.

17. **Spinti, M.; Zhuang, H. and Trujillo, E. M. (1995)** Evaluation of immobilized biomass beads for removing of heavy metals from wastewaters. *Water Environment Research*, **67,** 943 – 954.

18. **Sreeram, K. J. and Ramasami, T. (2003)** Sustaining tanning process through conservation, recovery and better utilization of chromium. Resources, *Conservation and Recycling*, **81**, 185-212.

19. **Tharwat, N. A., Mostafa, A. H., Abdel-Rahman, T. M. A. and Ahmed, M. A. (2013)** Biosorption of hexavalent chromium tannery water using some fungi isolated from tannery effluent. *Egyptian J. Botany*, (Under publication).

20. **Ting, Y. P., Lawson, F. and Prince, I. G. (1991)** Uptake of cadmium and zinc by the alga *Chlorella vulgaris*: II. Multi-ion situation. *Biotechnology and Bioengineering*, **37,** 445–455.

21. **Tobin, J. M. (2001)** Fungal metal biosorption, In: "*Fungi in Bioremediation*", Gadd, pp. 424-444 G.M, Cambridge University Press, Cambridge, New York.

22. **Volesky, B. (1990)** Biosorption by fungal biomass .In: *Biosorption of Heavy Metals* Volesky, B. (Ed.),pp.140-171.CRC press,Florida.

23. **Volesky, B. and Holan, Z. R. (1995)** *Biosorption of Heavy Metals – Review*. *Biotechnology* Progress, **11,** 235-250.

24. **Volesky, B. and May-Philips, H. A. (1995)** Biosorption of heavy metals by *Saccharomyces cerevisiae*. *Applied Microbiology Biotechnology*, **42,** 797-806.

25. **Yan, G. and Viraraghavan, T. (2000)** Effect of pretreatment on the bioadsorption of heavy metals on *Mucor rouxii*. *Water SA*, **26,** 119-123.

(*Received* 18/6/2013;
accepted 26/8/2013)

التحكم فى الامتزاز الحيوى للكروم السداسى بواسطة الاغزال الفطرية لفطرى Aspergill usnigh spicania, silvatca بإستخدام طريقة تاجوشى

نجوى ثروت[1] ، تهانى عبدالرحمن[1] ، أمير مصطفى[2] و مروة أحمد[2]
[1]قسم النبات – كلية العلوم – جامعة القاهرة و [2]المركز القومى لبحوث البناء والإسكان – جيزة – مصر.

لتحقيق أقصى قدر من الكفاءة فى الامتصاص الحيوى تم تصميم ثمانية عشر تجربة للتحقق من قدرة الكتلة الحيوية الفطرية كمادة فعالة وغير مكلفة للامتصاص الحيوى لعنصر الكروم السداسى التكافؤ .تمت دراسة تأثيربعض العوامل فضلا عن دراسة التفاعل بينها باستخدام التصميم الإحصائي تاجوشى .وتم تنفيذ مجموعة من الأساليب الإحصائية التي يمكن أن تحدد العلاقات بين استجابات المتغيرات المستقلة. وفي الدراسة تم الكشف عن أن الامتصاص الحيوى للكروم السداسى التكافؤ بواسطة فطرة اسبرجلس نيجر و سبيكاريا سلفاتيكا قد اختلف كثيرا فى الاوساط الغذائية المستخدمة مما يدل على أهمية التجارب الأمثل في العمليات الحيوية. بالنسبة لفطرة اسبرجلس نيجرفقد بين تحليل التباين (الانوفا) أن درجة الحرارة ، وحالة الغزل الفطرى و نترات الصوديوم والتقليب كانت أهم العوامل التي اثرت على الامتصاص الحيوى للكروم السداسى التكافؤ. اما فى حالة فطرة سبيكاريا سلفاتيكا فقد كانت درجة الحرارة وحالةالغزل الفطرى و التقليب و كبريتات الماغنسيوم من اهم العوامل التى اثرت على الامتصاص الحيوى للكروم السداسى التكافؤ. و فى تصميم الكشف كانت القيم التجريبية للامتصاص الحيوى للكروم السداسى التكافؤمتوافقة بقدر معقول مع القيم المتوقعة. و كان التركيب المثالى للوسط الغذائى الذى يحقق اعلى امتصاص حيوى للكروم السداسى التكافؤ بواسطة الفطرتين المختبرتين هو : الغزل الفطرى الجاف و السكروز(15 جرام فى اللتر) و نترات الصوديوم و كبريتات الماغنسيوم و درجة حرارة 18 درجة مئوية و درجة حموضة 5 و الزراعة الثابتة لمدة 5 ايام.