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Higher production of lipase enzyme from different microorganisms grown in local natural culture media

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Abstract

Microbial lipase (triacyl glycerol acyl hydrolase) is a very important enzyme in many industrial applications. The production of enzyme from different microorganisms was semi-quantatively screened by producing clear zone around microbial colonies on agar plates which were compatible with the results of quantitative screening for specific activity of lipase produced by most strains. The significant and higher productive lipase strains were only 3 from 42 strains of *Lactobacillus* spp. (*L. plantarum*, *L. acidophilus* and *L. casei*), 1 from 2 strains of *Bacillus subtilis*, 1 from 4 strains of *B. cereus*, 1 from 5 strains of *Pseudomonas aeruginosa*, 1 from 3 strains of *Serratia marcescens*, 1 from 2 strains of *Aeromonas* spp. 1 from 5 strains of *Aspergillus niger*, 1 from *Candida albicans* and 1 from 2 strains of *Saccharomyces cerreviseae*, all these strains have been selected for further experiments. In order to improve the production of this enzyme, four natural local media were used. When the specific activity of lipase produced by selected strains grown in these media compared to optimized synthetic medium tributyrin broth with same environmental conditions for all of them, the highest enzyme activity from different microorganisms were achieved in *Cucurbita pepo* seed medium (Leqa 1) more than synthetic media and other three natural seed media (*Cucumis melo*, sun flower and *Citrullus lanatus*). Maximum specific activity of this enzyme isolated from *L. acidophilus* grown in *Cucurbita pepo* seed medium was reached to 189.84 U/mg. Our results suggest a novel strategy for maximum production of lipase enzyme from different microorganisms by using low cost *Cucurbita pepo* natural seed medium for all previous species or other three natural media for some highly productive species instead of high cost synthetic medium.

Keywords: Lipase, Microorganisms, Natural media, Higher production.

Introduction

Microbial lipases (triacylglycerol acylhydrolases, EC 3.1.1.3) catalyze the hydrolysis and synthesis of esters formed from glycerol and long- chain fatty acids, It is one of the most important enzymes used in biotechnological processes including food, pharmaceutical industries, leather, detergents, cosmetic and industrial wastes management (Sharma et al., 2001). In order to provide a good environment for isolation of lipase producing microorganisms the oily environment may be used. Lipases have emerged as key enzymes in recent times, in growing biotechnology(Gupta et al., 2004). Many microorganisms such as bacteria, yeast and fungi are known to secret lipases, they have been found in diverse habitats such as factories, vegetable oil processing dairies, industrial wastes and soil contaminated with oil (Sztajer et al., 1998). Lipases occur widely in nature, but only microbial lipases are commercially significant (Saxena et al., 1999). The farthest powerful strategy in a biological process is

trying to investigate for the best medium conditions in order to improve and increase the efficiency of the process without increasing the cost (Qamsari et al., 2011)

Some plant seed oils could be use as a rich substrate for lipase enzyme. *Cucurbita pepo* seed oil is a rich source of many fatty acids (47.03%) is comprised mainly of (linoleic, oleic, palmitoleicic stearic, myristic, palmitic caffeic and two caffeic derivatives sinapic, phenolic and vanillic acids), vitamin E including α, β, γ, and δ) and contain protein (35.95%), carbohydrate (6.37%) in addition to some minerals like, Ca, Mg, Mn, Fe, Cu, Zn, Cu and P (Stevenson et al., 2007; Kostalova et al., 2009; Kreft et al., 2009 and Fürnkranz et al., 2012). *Cucumis melo* seeds crude oil contain 37.8%, crude protein 28.58, while the moisture, fiber and total sugar contents were 4.2%, 24.7% and 6.9%, respectively. The main fatty acids were linoleic, oleic, palmitic, and stearic acids (61.10%, 18.75%, 10.37% and 9.18%, respectively, and have essential minerals (K, Ca, Mg, Fe, Cu, Zn, and P. Seed oil also contained a good level of tocopherols, β-sitosterol was found in oil of total sterols (Azhari et al., 2014).

Watermelon seeds contain 50% oil, total unsaturation contents of the oil was 81.6%, with linoleic acid which being the dominant fatty acid (68.3%), the protein content of the oil-free residue was 47% (Moaddabdoost and Safi Kurdi _2010). Sun flower seeds contained three types of commonly used sunflower seeds: linoleic, high oleic and has its own unique levels of monounsaturated, saturated, and polyunsaturated fats. Sunflower oil (linoleic oil) is high in polyunsaturated fatty acids (68%) and low in saturated fats, such as palmitic and stearic acid. dried whole protein (42%), all B and E vitamins, high levels of the dietary minerals: manganese, magnesium, phosphorus, iron and zinc *(Zabaniotou, A.* and *Kantarelis. 2008)*. This study was designed to use different plant seeds as a natural local media instead of synthetic media conditions to improve and maximize the production of lipase from different microorganisms and choose a higher productive strains in order to use them in many important industrial applications in the future.

Materials and Methods

Isolation and identification of microorganisms.

Bacterial species

1- In order to isolate *Lactobacillus* spp. local food samples(cheese, milk, meat and vegetables) were used. Isolated strains of this genus were identified to their species by using Api-50 Bio- Merieux.
2- Soil samples were used to isolate *Burkholdria* and *Serratia* spp. Then identified by VITEK system.
3- Food samples were used to isolate *Bacillus* spp. and identified to their species by morphological and biochemical tests using Bergeys manual of determinative bacteriology
4- Clinical samples to isolate *Pseudomonas aeruginosa* strains which identified by VITEK system.
5- Fresh fish samples to isolate *Aeromonas* spp. then identified by VITEK system.

Fungus and yeast species.

1- Five strains of *Aspergillus niger* and one strain of *Aspergillus oryzae* were taken from graduate students laboratories.
2- One strain of *Candida albicans* was taken from graduate students laboratories.
3- One strain of *Candida cylindraceae* was taken from lab. of science and technology ministry.
4- Two strains of *Saccharomyces cerevisiae* were taken from graduate students laboratories.

Preparation of natural culture media.

A hundred grams of each (sun flower, *Cucurbita pepo*, *Cucumis melo* and *Citrullus lanatus*) seeds were crushed in 1000ml distilled water to make individually slurries, after boiling for 10 minutes the slurries were filtered through muslin cloth, then four natural culture media were prepared after adding 1% sodium nitrate and 0.5% NaCl for each of them, the pH adjusted to 6 for production of lipase from fungi and yeast strains and 7 for bacterial strains, these cultures were distributed in to 500 ml flasks(100 ml for each of them) then autoclaved at 121˚ C for 10(min). Culture media conditions were same for all, these media were named:
Leqa1: *Cucurbita pepo* seeds medium
Leqa2: *Cucumis melo* seeds medium
Leqa3: *Citrullus lanatus* seeds medium
Leqa4: Sun flower seeds medium

Preparation of bacteria and yeast inoculums for lipase production.

The cell concentration was determined microscopically using 1ml of cell suspension after making the serial dilution 1% vol/vol. of cell suspension containing 1×10^6 cell/ml was used as the inoculums in 100ml of the prepared medium Leqa1, Leqa2, Leqa3 and Leqa4 in Erlenmeyer flask, culture media were shacked for 48h at 200 rpm at 30 Cº,

after that they were centrifuged at 12000 rpm for 5(min), the supernatant was used as a source of extracellular enzyme for bacteria but for yeast strains culture media were shacked at 220 rpm for72h at 28C°.

Preparation of fungi inoculums for lipase production.

The sterilized media were inoculated with spores 10^7/ml for a seven days old culture and the flasks containing 100ml of media were incubated at 30 C° in a rotary shaker at 100 rpm for 8 days. The mycelium was harvested by filtration under vacuum and later centrifuged at 12000 rpm for 5(min), the clarified supernatant was used as a source of extracellular enzyme.

Semi-Quantitative (preliminary) screening assay for lipase production by microorganisms.

For preliminary screening of lipase production from microorganisms tributyrin agar was used, all isolated strains were inoculated in to tributyrin agar plates containing 0.5% (w/v) peptone, 0.3% (w/v) yeast extract, 1% (v/v) Tributyrin and 2% agar, pH 7.0 (Sarada S. B. et al., 1998) and incubated at 37C° for 24h for bacterial strains, at 28C° for 48h for yeast strains and at 30C°for 72h for fungi strains. A clear zone around the colonies indicates the production of lipase.

Quantitative screening(lipase assay) for lipase production by microorganisms.

Lipase activity was measured spectrophotometrically (410 nm, pH 7) using p-nitro-phenyl palmitate (PNPP) as a substrate of lipase enzyme as described by Wrinkler and Stukman (1979). One unit of lipase activity was defined as the amount of enzyme that lebarated 1µ mol of p-nitro-phenol(PNPP) per min under the assay condition. Winkler, U. K. and Stukman, M. (1979). Specific activity was expressed in terms of units per milligram of protein concentration by using Lowry method (Lowry *et al*.,1951).

Results

Isolation and identification of microbial strains

Different strains of microorganisms(bacteria, fungi and yeasts) were isolated and identified to their species using an approved scientifically precise methods, for the purpose of investigating the ability of these strains to produce lipase enzyme in order to select the significant and higher productive isolates of them. Table 1 explain the different sample sources and identification procedures used with number of identified strains. The number of *Lactobacillus* (42) strains(which contain six species) were more than others like *Bacillus* (6), *Aspergillus* (6), *Pseudomonas* (5), *Burkholderia* (4), *Aeromonas* (2), *Serratia* (3) *Saccharomyces* (2) and *Candida* (2) spp.

Table 1: Source, number and identified strains of microorganisms

Sample source	No.of isolated strains	Identification procedure used	Identified species and no. of strains
Soil	4	VITEK	*Burkholderia cepacia* . 4
Cheese, dairy, meat and vegetables	42	Api-50 Bio-Merieux.	*Lactobacillus plantarum* (12) *L. acidiphilus* (8). *L. reuteri* (8). *L. buchneri* (5). *L. casei* (5) and *L. delbreukii* (4)
Different foods	6	Morphological and biochemical tests	*Bacillus subtilis*. (2) isolates and *B.cereus*. (4)
Clinical samples	5	VITEK	*Pseudomonas aeruginosa*
Fresh fish	2	VITEK	*Aeromonas* spp.
Soil	3	VITEK	*Serratia marcescens*
Graduate students laboratories	6	-	*Aspergillus niger*. (5) .(1)
Graduate students laboratories	1	-	*Candida albicans*. (1)
Lab. science and technology	1	-	*Candida cylindraceae*. (1)
Graduate students laboratories	2	-	*Saccharomyces cerevisiae*. (2)

Preliminary screening for lipase production from different microorganisms.

The results show in table 2 different diameters of clear zones produced by different microorganisms which increase by increasing lipase production using tributyrin agar plate medium. The strain numbers which produced largest zoon were selected to grow them in natural media in order to improve lipase production from these strains. The largest zone 32 mm was produced by strain no. 5 of *Lactobacillus plantarum,* but *L. reuteri*, *L. buchneri* and *L. delbreukii* strains were couldn't produce clear zones, therefore they neglected. The higher productive strain of each species was selected for further tests

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Table 2: preliminary screening for lipase production from microbial strains on tributyrin agar plate

Identified species	Clear zone diameter mm range	Higher lipase productive strain no.	Identified species	Clear zone diameter mm range	Higher lipase productive strain no.
Burkholderia cepacia	7-22	2	*Pseudomonas aeruginosa*	7-23	5
Lactobacillus plantarum	5-32	5	*Serratia marcescens*	12-27	1
L. acidiphilus	6-26	8	*Aeromonas* spp.	17	2
L. reuteri	-	-	*Aspergillus niger*	7-21	4
L. buchneri	-	-	*Aspergillus oryzae*	16	1
L. casei	17	1	*Candida albicans*	22	1
L. delbreukii	-	-	*Candida cylindraceae*	13	1
Bacillus subtilis	15	2	*Saccharomyces cerevisiae*	10-18	2
B.cereus	19	1			

Quantitative screening for lipase production by selected microorganisms in synthetic tributyrin broth medium.

The results show in table 3 the specific activity of lipase produced by *L. acidiphilus* (151.22 U/mg protein) was more than other species of bacteria fungi and yeast strains. The same pH and temperature for optimized synthetic and natural media were used in this research.

Table 3: Specific activity of lipase produced by selected microorganisms in tributyrin broth medium

Higher productive selected strains	Specific activity(U/mg)	Higher productive selected strains	Specific activity(U/mg)
Burkholderia cepacia	89.67	*Serratia marcescens*	102.95
Lactobacillus plantarum	91.54	*Aeromonas* spp.	95.39
L. acidiphilus	151.22	*Aspergillus niger*	29.87
L. casei	130.37	*Aspergillus oryzae*	84.11
Bacillus subtilis	53.44	*Candida albicans*	132.51
B.cereus	73.69	*Candida cylindraceae*	73.68
Pseudomonas aeruginosa	89.15	*Saccharomyces cerevisiae*	101.91

Growing of selected strains in natural media conditions.

1- *Cucurbita pepo* seeds medium (Leqa 1).

The results in table 4 show significant and higher activity of lipase enzyme produced by the selected strains when they grown in Cucurbita pepo seed medium, that the highest spesific activity reached to189.84 U/mg protein produced by *L. acidiphilus*, then 142.66 U/mg protein produced by *L. casei* and 138.69 U/mg protein produced by *Candida albicans*

Other species produced significant activities for lipase production in this perfect medium which it was better than synthetic medium for production higher amounts of lipase enzyme from different microorganisms.

Table 4: Lipase specific activity produced by selected strains grown in *Cucurbita pepo* and *Cucumis melo* seeds media

Higher productive selective strains	*Cucurbita pepo* seeds medium		*Cucumis melo* seeds medium	
	Enzyme activity (U/ml)	Specific activity (U/mg)	Enzyme activity (U/ml)	Specific activity (U/mg)
Burkholderia cepacia	19.6	97.17	11.42	57.5
Lactobacillus plantarum	35.11	117.46	32.55	113.53
L. acidiphilus	28.8	189.84	21.91	135.08
L. casei	21.6	142.66	20.59	109.4
Bacillus subtilis	16.53	85.51	11.44	61.77
B. cereus	22.98	90.294	20.63	82.65
Pseudomonas aeruginosa	31.16	106.78	29.88	105.35
Serratia marcescens	28.34	117.54	22.1	88.29
Aeromonas spp.	29.11	108.74	25.46	101.92
Aspergillus niger	4.54	40.553	4.12	35.364
Aspergillus oryzae	9.16	87.90	8.88	81.244
Candida albicans	15.77	138.69	15.42	128.39
Candida cylindraceae	31.15	109.87	16.5	83.03
Saccharomyces cerevisiae	18.9	89.40	30.36	111.57

2- *Cucumis melo* seeds medium (Leqa 2).

Lipase producing microorganisms produce significant amounts of enzyme in *Cucumis melo* seeds medium (Leqa 2) as show in table 4, but they were less than that in Leqa1 and synthetic media except activity of lipase produced by *Saccharomyces cerevisiae* which it was more than that in both *Cucurbita pepo* seeds and synthetic media. *Candida albicans* produce good activity in this medium is very suitable for production of lipase from *Candida albicans*

3- *Citrullus lanatus* seeds medium (Leqa3).

The results in table 5 describe enzyme specific activity produced in Leqa 3 medium by different selected strains which were lower than the specific activity of lipase in previous media, but it was good medium for *Aeromonas* spp. and *Saccharomyces cerevisiae*.

4- Sun flower seeds medium (Leqa4).

Results show that sun flower seeds was a good medium in the production of lipase from bacterial strains especially for *Lactobacillus* spp., *Pseudomonas aeruginosa*, *Serratia marcescens*, *Burkholderia cepacia* and for *Saccharomyces cerevisiae* yeast.

Table 5: Lipase specific activity produced by selected strains grown in *Citrullus lanatus* and sun flower seeds media

Higher productive selective strains	*Citrullus lanatus* seeds medium		Sun flower seeds medium	
	Enzyme activity(U/ml)	Specific activity(U/mg)	Enzyme activity(U/ml)	Specific activity(U/mg)
Burkholderia cepacia	1.28	15.75	12.91	93.01
Lactobacillus plantarum	9.45	47.155	29.33	104.71
L. acidiphilus	6.33	55.139	19.33	111.86
L.casei	11.41	79.62	16.52	116.91
Bacillus subtilis	2.51	19.81	13.77	81.52
B.cereus	5.17	26.82	15.81	89.93
Pseudomonas aeruginosa	13.14	65.21	25.64	97.08
Serratia marcescens	10.05	55.74	20.66	95.82
Aeromonas spp.	18.28	95.407	21.49	97.504
Aspergillus niger	1.03	10.218	3.75	32.75
Aspergillus oryzae	1.26	13.93	3.65	33.94
Candida albicans	0.44	3.90	6.31	55.01
Candida cylindraceae	1.93	11.71	5.41	31.41
Saccharomyces cerevisiae	21.17	89.85	24.22	81.87

Discussion

Isolation and identification of microbial strains

Different microorganisms were isolated from different sample sources using selective media for isolation of lipase productive strains and for the purpose of selection higher lipase productive strains of bacteria, fungi and yeasts. Most isolated strains were from bacterial species because they have a vital role in production of lipase enzyme and in commercial ventures.

Preliminary screening for lipase production from different microorganisms.

From the results of preliminary screening we conclude that some strains had a high capacity for lipase production like one strain of each species: *Lactobacillus plantarum, L. acidiphilus, Burkholderia cepacia, Pseudomonas aeruginosa, Serratia marcescens, B. cereus, Aspergillus niger, Candida albicans* and *Saccharomyces cerevisiae*. Other selected species had a good enzyme activity like *L.casei, Aeromonas spp., Bacillus subtilis, Aspergillus oryzae* and *Candida cylindraceae,* thus all these strains were selected for further tests.

Quantitative screening for lipase production by selected strains in synthetic tributyrin broth medium.

After optimization of pH and temperature in this medium, the specific activity of lipase produced by selected strains was measured in tributyrin broth medium, the results were compatible with preliminary screening, that all selected strains were active in producing lipase in this synthetic media except *Aspergillus niger* which had low specific activity (29.87 U/mg protein) when it compared with other species, may be this medium was not suitable for lipase production from this species, but it was suitable for *Aspergillus oryzae* which the specific activity reached to(84.11U/mg protein).

Growing of selected strains in natural media conditions.

1- *Cucurbita pepo* seeds medium (Leqa 1).

This natural medium was perfect for producing the highest lipase activity from different selected species when we compared the specific activity of lipase produced by all strains in five natural and synthetic media.

When we investigate for pumpkin seeds components from published researches, many researchers like (*Stevenson et al., 2007; Kostalova et al., 2009; Kreft et al*., 2009 and *Fürnkranz* et al., 2012) found that it has a promising material for microbial growth. The presence of these components cooperating with each other were able to produce optimum conditions for growing and stimulate all selected microorganisms to produce the enzyme efficiently. Depending on the results obtained, we recommend using this natural and simple preparing medium instead of high-cost other industrial media for robust production of microbial lipase enzyme industrially.

2- *Cucumis melo* seeds medium (Leqa 2).

The results in table 4 show that this natural medium was very suitable for highly production of lipase enzyme from different microbial strains, especially from *Saccharomyces cerevisiae, Lactobacillus* spp., *Candida albicans, Pseudomonas aeruginosa* and *Serratia marcescens* which they efficiently produced lipase enzyme on this medium, because it contains components encourages the production of this enzyme.

Some researchers were analyzed for their physiochemical properties and chemical composition of the oil, Thus depending on our results and previously published researches we concluded, all these contents may produce synergistic effects for growth and induction for highly production of lipase from most selected strains. So we recommend for using this natural chea medium too instead of a high cost synthetic media.

3- 3- *Citrullus lanatus* seeds medium(Leqa3).

This natural medium was good for increasing enzyme activity from *Saccharomyces cerevisiae* and *Aeromonas* spp. that the specific activity of enzyme reached to (89.85 and 95.407 U/mg protein) respectively, but it was not perfect like other three natural media especially for *Candida albicans, Candida cylindraceae, Aspergillus niger, Candida cylindraceae, Burkholderia cepacia* and *Bacillus subtilis* , because lipase activity was lower than that produced in other media as showed in table 5.

Some researchers (Moaddabdoost, Z. and Safi Kurdi, A. A. 2010) stated that watermelon seeds contain 50% oil, total unsaturation contents of the oil was 81.6%, with linoleic acid which being the dominant fatty acid (68.3%), the protein content of the oil-free residue was 47%. Depending on our results and previously published researches we concluded that these components were suitable for highly production of lipase from *Saccharomyces cerevisiae* and *Aeromonas* spp. But not for other selected species.

Thus we recommend using this natural low cost medium just for two previously mentioned species

4- Sun flower seeds medium (Leqa4).

The results of lipase produced by selected species grown in this natural medium described that all bacterial strains and *Saccharomyces cerevisiae* yeast had a high specific activity of lipase produced by them. But this medium was not perfect for *Aspergillus* and *Candida* spp. as showed in table 5.

We investigate sunflower seeds components through published research *(Zabaniotou, A.* and *Kantarelis. 2008)* on this topic which contained three types of commonly used sunflower seeds: linoleic, oleic and has its own unique levels of monounsaturated, saturated, and polyunsaturated fats. dried whole protein (42%), all B vitamins and vitamin E, high levels of the dietary minerals: manganese, magnesium, phosphorus, iron and zinc. The results mean that these components were activated the production of lipase from bacteria and *Saccharomyces cerevisiae* yeast, because of synergistic effect action of these components for enhancing enzyme activity for these strains, thus we recommend using this natural low cost medium for highly production of lipase from these species as an alternative to the industrial media in the future.

Conclusion

From the results of this study, the following conclusions can be made:-

1-Different isolated groups of microorganisms (bacteria, fungi and yeasts) could produce lipase enzyme.
2- Microbial isolates vary in their ability and efficiency in the production of the enzyme.
3- Maximum production of lipase enzyme from different microorganisms can be obtained
using four low cost natural local culture conditions as a novel strategy, instead of high cost synthetic media.
4- Natural *Cucurbita pepo* seeds medium (Leqa 1) was perfect for producing the highest lipase activity from different selected species when we compared the specific activity of lipase produced by all strains in five natural and synthetic media.
5- *Cucumis melo* seeds medium (Leqa 2) and Sun flower seeds medium(Leqa4) were very suitable for highly production of lipase enzyme from most selected strains.
6- *Citrullus lanatus* seeds medium (Leqa3) was good for increasing enzyme activity from *Saccharomyces cerevisiae* and *Aeromonas* spp. but it was not perfect for other species.

References

1. Azhari, S., Xu YS, Jiang QX, Xia WS. 2014. Physicochemical properties and chemical composition of Seinat (*Cucumis melo* var. *tibish*) seed oil and its antioxidant activity. *Grasas Aceites* 65 (1): e008. doi: http://dx.doi.org/10.3989/gya.074913. This is an

2. Fürnkranz, Michael; Lukesch, Birgit; Müller, Henry; Huss, Herbert; Grube, Martin; Berg, and Gabriele. (2012). "Microbial Diversity Inside Pumpkins: Microhabitat-Specific Communities Display a High Antagonistic Potential Against Phytopathogens". *Microbial Ecology*. 63 (2): 418–428.
3. Gupta, R., Gupta, N., Rathi. P., (2004). Bacterial lipases: an overview of production, purification and biochemical properties. *Appl Microbiol Biotechnol*. (2004) 64: 763–781.
4. Košťálová, Zuzana; Hromádková, Zdenka; Ebringerová, Anna (August 2009). "Chemical Evaluation of Seeded Fruit Biomass of Oil Pumpkin (Cucurbita pepo L.var. Styriaca)". *Chemical Papers* (Springer Versita for Institute of Chemistry) 63 (4): 406–413.
5. *Kreft, Samo; Kreft, Marko (2007).* "Physicochemical and Physiological Basis of Dichromatic Colour" *(PDF).* Naturwissenschaften *(Springer Science+Business Media) 94 (11): 935–939*
6. Lowry, O.H.; Rosenbourgh, N.J.; Farr, A-L. and Randall, R.J. (1951). Protein measurement with Folin-Phenol reagent. J. Biol. Chem., 193: 265-275.
7. Moaddabdoost, Z., and Safi Kurdi, A. A.(2010). Characteristics and Composition of Watermelon Seed Oil and Solvent Extraction Parameters Effects. J. American Oil Chemists' Society. 87(6):667-671.
8. Qamsari, E., Mobarak., Kermanshahi, R., Kasra and Negad, Z. Mosavi. (2011). Isolation and identification of a novel, lipase-producing bacterium, *Pseudomnas aeruginosa* KM110. Iran J. Microbiol.; 3(2): 92-98.
9. Sarada Sarkar, B., Sreekanth, Shreya Kant, Rintu Banerjee and B.C. Bhattacharyya, 1998. Production and optimization of microbial lipase. Bioprocess Engineering, 19: 29-32.
10. Saxena, R.K., Ghosh, P.K., Gupta, R., Davinson, W.S., Bradoo, S., Gulati, R. (1999). Potential biocatalysis and future industry. Curr Sci.;77:110–115.
11. Sharma, R., Chisti, Y., Banerjee, U.C. (2001). Production, purification, characterization, and applications of lipases. Biotechnol Adv.;19:627–662.
12. Stevenson, D. G.; Eller, F. J.; Wang, L.; Jane, J.; Wang, T.; Inglett, G. E. (2007). "Oil and Tocopherol Content and Composition of Pumpkin Seed Oil in 12 Cultivars". *Journal of Agricultural and Food Chemistry* 55: 4005–4013.
13. Sztajer, H., Maliszewska, I., Wieczorek, J. (1998). Production of exogenous lipase by bacteria, fungi and actinomycetes. Enzyme Microb. Technol.;10:492–7.
14. Winkler, U. K. and Stukman, M. (1979). Glycogen hyaluronate and some other polysaccharides greatly enhance the formation of exolipase by *Serratia marcescens*. J. Bacteriol.; 139,663-67.
15. Zabaniotou, A. A., Kantarelis. (2008). The odoropoulos "Sunflower shells utilization for energetic purposes in an integrated approach of energy crops: Laboratory study pyrolysis and kinetics". *Bioresource Technology* 99 (8): 3174–3181.

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