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Research Article

Effect of Nickel Sulfate on Yeast Growth and Cell Division

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Abstract

Environmental factors influence carcinogenesis by interfering with a variety of cellular targets. Quantitative evaluation of genetic effects of atmospheric pollutant; ozone, cigarettes smoke and metals, have been performed in yeast system by many workers. In this work, the effect of nickel sulfate on growth and cell division was investigated in yeast culture (*Saccharomyces cerevisiae*). The results revealed that, there are significant changes in the yeast dry weight and cell number in samples treated with different concentrations (0, 4, 10, 20, 100, 200 ppm) for different time (4, 10, 24, 48h). These changes were concentration and time dependent. In addition, there were changes in size and shape of yeast cells and in colony size.

Keywords: Nickel Sulfate; *Saccharomyces cerevisiae*; Dry Weight; Cell Division

Introduction

Nickel is a metal of widespread distribution in the environment. Contact with soluble and insoluble nickel compounds can cause a variety of side effects on human health. Human exposure to Ni may occur through food, water or air [1]. At higher concentrations, nickel ion can be very toxic to both eukaryotic and prokaryotic cells, it potentially inhibits synthesis of macromolecules such as RNA and protein [2,3]. The binding of nickel ions to cellular protein, followed by reaction with endogenous hydrogen peroxide formed by cellular metabolism, could lead to oxygen radical generation and chromosome breakage, which could cause mutation and activation of proto-oncogenes into activated oncogenes [4-7]. Nickel has been classified as a human carcinogen based on epidemiological evidence, which shows high incidence of nasal and lung cancers in refinery workers [8-11]. The results of many in vivo and in vitro researches suggest that nickel and nickel oxide nanoparticles are responsible for lung toxicity, inflammation, oxidative stress and apoptosis [12,13]. Although the toxicity and carcinogenicity of nickel compounds in humans and experimental animals are well demonstrated, the underlying mechanisms of their action remain unclear [14,15]. The mutagenicity and recombinogenicity of environmental pollutant were investigated in yeast system [16-23]. Saccharomyces cerevisiae, commonly known as baker's yeast, has been a preferred organism for genetic research since the mid-twentieth century. The expermintal value of this single-celled eukaryotic species lies in its simple life cycle, alternating haploid and diploid phases, short generation time, and easy-to-identify meiotic products. Recent technological advances enable yeast biologists to use genetic analysis to gain a deep understanding of the organization and regulation of eukaryotic cells [24].

The aim of the present work was to investigate the cytotoxic effects of nickel sulfate on cell division and dry weight using yeast as biological system.

Materials and Methods

Nickel sulfate (from BDH chemicals Ltd. Poole – England.) was chosen for the present investigation. Commercial yeast (*Saccharomyces cerevisiae,* S.I. Lesaffre-France) was grown in a complex medium (Malt Extract Broth, from OXOID Ltd., Basingstoke, Hamp-

shire, England), with pH adjusted to 5.2. The media were enriched with nickel sulfate, in such quantities that initial nickel sulfate concentrations were 4, 10, 20, 100 and 200 ppm) Sterile media (40 ml/treatment) was placed in conical flasks, inculated with 1ml of pure yeast culture and left in a shaking incubator (30ºC, 150 rpm) for defferent time exposure (4, 10, 24, 48 hours). Three replicates of each treatment were taken. Yeast growth was monitored by measuring the yeast cell number per ml using Haemacytometer (Improved Neubauer, R.S. 749. Supplied by Gallenkamp) as in [25]. The dry weight of yeast culture was measured as in Pasternakiewicz (2006). Then the data were statistically tested. The effect of Ni concentration in the medium on yeast growth was estimated using Two-way Analysis of Variance (ANOVA) which was calculated with SPSS computer package.

Results, Discussion and Conclusion

Microorganism show sigmoidal growth due to eventual limitations of space or nutrient accumulation of end product but the over all growth of any organism includes an early exponential phase in optimized normal media. Any changes in rate of growth had to deal with interfering of tested substances with bioactivity of organism or the cell. The rate of growth can be measured by many growth parameters such as volume, biomass (Dry weight), cell number and protein content [22,26-28]. The effect of different concentrations of nickel sulfate and also different time intervals on yeast cell number are represented in figure 1.

The results showed that there are significant differences within treatments ($P < 0.01$). Natural increase of yeast cell number in the control from $1.85x10^7$ at 4h to $8.68x10^7$ at 48h. Treated cells with 4ppm stimulate yeast cell division at 4hr exposure time ($2.046x10^7$). On other hand, there was no significant change observed in cell number in samples treated for 10, 24, 48hr (6.791, 7.177 and 8.780 $x10^7$ respectivily). There were decrease in cell number in samples treated with high concentrations (100, 200ppm) in all exposure times. In meanwhile, 20ppm induce an increase in cell number at all exposure times. 10ppm cause increase in cell number in samples treated for 4hr and 48hr, slight decrease observed in 10, 24hr (7.058, 8.815 $x10^7$ respectivily).

Over all, the effect of nickel sulfate on the cell number was increase by increase time of treatment and depend on nickel sulfate concentration {P(0.015)} comparing to the control. There were highly significant mean differences at (0.05 level) between the effect of high concentrations (100 and 200 ppm) and the other used concentrations ($P > 0.001$). All treatment times had significant effect on the cell number represented by decrease or increase in total cell number, which is an evidence of nickel sulfate genotoxicity [29].

The effects of different concentrations of nickel sulfate and different time intervals on yeast dry weight are represented in figure 2. The results showed that there are significant differences within treatments ($P < 0.01$). Low concentration of nickel sulfate (4, 10, 20 ppm) did not cause significant change in biomass of treated Saccharomyces cerevisiae cells comparing to the control, wherease high concentrations (100, 200 ppm) cause higher significant effect on dry weight of treated cells comparing to the control and low nickel sulfate concentrations.

Figure 1: Number of dividing cells in yeast culture treated with different NiSO4 concentration for different time.

Figure 2: Dry weight of yeast culture treated with different NiSO4 concentration for different time.

In the condition present in this experiment, the cell number stongly decreased with high concentration of nickel sulfate treatment (100 and 200 ppm). The achieved results suggested that the decrease in dry weight may result of inhibition of cell division. The observed data agree with the results of Dovgaliuk and others (2001) who found that nickel sulfate inhibition the root growth and reduce mitotic activity of merestimic cells [30]. Pasternakiewicz (2006) found out that the biomass yield of *Saccharomyces cerevisiae* was lower in medium enriched with cadmium and this action was concentration dependent [22]. Both the specific growth rate and the biomass concentration were more inhibited in the bioaccumulation media containing Ni(II) ions; where the increase of Ni(II) concentration led to a drastic decrease in microbial growth for *Candida utilis* [28]. The decrease in dry weight and cell division may result of the inhibition of protein synthesis [31].

Nickel sulfate caused cell and colony abnormalities represented by changes in their size and shape. The size of normal yeast cell in this experiment under optimized conditions were 8 to 14µ depend on stage of cell division. Change in cell size was observed in culture treated with high concentrations of nickel sulfate to the size ranged between 7 to 10µ, change in the shape of the cells were observed also at the same concentrations (Figure 3). Recultivation of treated cells on solid medium give abnormal size of yeast cells and colonies compare to the control with decrease in the colonies number at the same dilution, which is an evidence of the effect of nickel sulfate on cell division (Figure 4).

Figure 3: Changes in size and shape of cells in yeast cultures treated with high concentrations of nickel sulfate for long time exposure.

Cytoskeleton and cell protein could be a cell target for nickel sulfate [30,32,33]. Change of the protein structure cause a decrease in enzyme activity which lead to inhibition of cell growth or induction of mutation. The nickel binding to protein induced modification in its secondary structure [21] which lead to negative impact in cell growth as result of mutation. Nickel toxicity may lead to damage the membrane integrity result in aberrant cell [34,35].

The experiment showed the negative influence of nickel sulfate on the growth of *Saccharomyces cerevisiae*. Decrease in yeast cell number at high nickel concentrations and long time exposure indicated the inhibition of yeast cell reproduction. Higher nickel concentrations cause drastically decreased dry weight which suggested that nickel in high concentration in the media impaired the synthesis of protein which lead to the decrease of cell number and dry weight.

Figure 4: Abnormal size of yeast cells and colonies after reinoculated of treated cells with different NiSO4 concentrations for different exposure time.

Bibliography

1. Genchi G., *et al*. "Nickel: Human Health and Environmental Toxicology (Revew)". *International Journal of Environmental Research* 17.3 (2020): 679.

2. Guha C and Mookerjee A. "Effect of nickel ion on macromolecular synthesis in *Escherichia coli* K-12". *Nucleus* 22 (1979): 45-47.

3. Ke,Q., *et al*. "Alterations of Histone Modifications and Transegene Silencing by Nickel Chloride". Carcinogenesis 27.7 (2006): 1481- 1488.

4. Landolph JR. "Molecular and cellular mechanisms of transformation of C3H/10T1/2 Cl 8 and diploid human fibroblasts by unique carcinogenic, nonmutagenic metal compounds A review". *Biological Trace Element Research* 21: 65-78.

5. Landolph JR. "Neoplastic transformation of mammalian cells by carcinogenic metal compounds: cellular and molecular mechanisms". In: Foulkes,E.E. (Ed) Biological Effects of Heavy Metals. (1990): 1-18.

6. Landolph JR. "Role of free radicals in metal-induced carcinogenesis". In: Sigel.A. and Sigel,H. (Eds.) Metal Ions in Biological Systems: Interrelations Between Free Radicals and Metal Ions in Life Processes (1999): 485-493.

7. Landolph JR., *et al*. "Molecular biology of deregulated gene expression in transformed C3H/10T1/2 mouse embryo cell lines induced by specific insoluble carcinogenic nickel compounds". *Environmental Health Perspectives* 110.5 (2002): 845-850.

8. Coogan T.P., *et al*. "Toxicity and carcinogenicity of nickel compounds". *Critical Reviews in Toxicology* 19 (1989): 341 -384.

9. Goyer RA. Toxic effects of metals. (1991): 623 -680.

10. Cangual Hakan. "Hypoxia Upregulates The Expression of The NDRG1 Gene Leading to Its Overexpression in Various Human Cancers". BMC GENETICS (2004): 1-5.

11. IARC (The International Agency for Research on Cancer). Nickel and nickel compounds. IARC Monogr. Eval. Carcinog. Risk Hum. 100 (2012): 169-218.

12. Latvala S., *et al*. "Nickel release, ROS generation and toxicity of Ni and NiO micro- and nanoparticles". PLoS One 11 (2016): e0159684.

13. Sutunkova MP., *et al*. "The most important inferences from the Ekaterinburg nanotoxicology team's animal experiments assessing adverse health effects of metallic and metal oxide nanoparticles". *Toxicology Reports* 5 (2018): 363-376.

14. SundermanFWJr., *et al*. "Increased lipid peroxidation in tissue of nickel chloride-treated rats". *Annals of Clinical & Laboratory Science* 15 (1985): 229 -236.

15. Stohs SJ and Bagchi D. "Oxidative mechanisms in the toxicity of metal ions". *Free Radical Biology and Medicine* 18 (1995): 321 -336.

16. Broday., *et al*. "Nickel compounds are novel inhibitors of histone H4 acetylation". *Cancer Research* 60 (2000): 238-241.

17. Dönmez G and Aksu Z. "Bioaccumulation of copper(ii) and nickel(ii) by the non-adapted and adapted growing CANDIDA SP". *Water Research* 35.6 (2001): 1425-1434.

18. Soares EV., *et al*. "Toxic effects caused by heavy metals in the yeast Saccharomyces cerevisiae: a comparative study". *Canadian Journal of Microbiology* 49.5 (2003): 336-343.

19. Lichtenberg-Fraté H., *et al*. "A yeast-based method for the detection of cyto and genotoxicity". *Toxicology in Vitro* 17.5-6 (2003): 709-716.

20. Schmitt M., *et al*. "Phenotypic yeast growth analysis for chronic toxicity testing". *Ecotoxicology and Environmental Safety* 59.2 (2004): 142-150.

21. Romero CS., *et al*. "Structural and functional implications of the hexokinase-nickel interaction". *Journal of Inorganic Biochemistry* 99.12 (2005): 2395-2402.

22. Pasternakiewicz A. "The growth of Saccharomyces cerevisiae yeast in cadmium enriched media". *Acta Scientiarum Polonorum Technologia Alimentaria* 5.2 (2006): 39-46.

23. Chen CC., *et al*. "Genetic analysis of ionizing radiation-induced mutagenesis in Saccharomyces cerevisiae reveals TransLesion Synthesis (TLS) independent of PCNA K164 sumoylation and ubiquitination". *DNA Repair* 5.12 (2006): 1475-1488.

24. Hartwell LH., *et al*. "Genetics: from Genes to Genomes". Mc Graw Hill, Boston(2004) .

25. Colome JS., *et al*. "Laboratory Exercises in Microbiology". West Publishing Company (1986).

26. Leopold AC and Kriedemann PE. "Plant growth and development" (1975).

27. Kaim W and Schwederski B. "Bioinorganic chemistry: Inorganic elements in the chemistry of life". *Wiley Chichester*(1994).

28. Mersin G and Açikel U. "Production of Candida Biomasses for Heavy Metal Removal from Wastewaters". *Trakya University Journal of Natural Sciences* 22.1 (2021): 67-76.

29. Egito LCM., *et al*. "Cytotoxic and genotoxic potential of surface water from the Pitimbu river, northeastern/RN Brazil". *Genetics and Molecular Biology* 30.2 (2007): 435-441.

30. Dovgaliuk AI., *et al*. "Assessment of phyto- and cytotoxic effects of heavy metals and aluminum compounds using onion apical root meristem". *Tsitol Genet* 35.1 (2001): 3-9.

31. Kim JC and Bendixen EL. "Effect of haloxyfop and CGA-82725 on cell cycle and cell division of oat (Avena sativa) root tips". *Weed science* 35 (1987): 769-774.

32. Bemba-Meka PM., *et al*. "Nickel compound-induced DNA single-strand breaks in chromosomal and nuclear chromatin in human blood lymphocytes in vitro: Role of oxidative stress and intracellular calcium". *Mutation Research/Genetic Toxicology and Environmental Mutagenesis* 586.2 (2005): 124-137.

33. Engwa GA., *et al*. "Mechanism and Health Effects of Heavy Metal Toxicity in Humans". Poisoning in Modern World (2019): 1-23.

34. Joho M., *et al*. "Nickel ion resistance mechanisms in yeasts and other fungi". *Indian Journal of Microbiology* 14 (1995): 164-168.

35. Brady D and Duncan JR. "Cation loss during accumulation of heavy metals cations by Saccharomyces cerevisiae". *Biotechnology Letters* 16.5 (1994): 543-548.