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The effect of nanofiber on the biological traits of *Drosophila Melanogaster*

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ABSTRACT

Today nanofiber components (polyacrylonitrile dimethyl formamide etc.) are used in many engineering fields and food industry. Possible effects are need to be identified on the environment and non-target organisms. *Drosophila melanogaster* Meigen (Diptera: Drosophilidae) has been one of the most studied organisms, working as a model in developmental biology and environmental studies. In this study, adults were fed with nanofiber coated diet (PAN-DMF; produced by the electrospinning, range of diamer 260 nm), eggs were obtained from adults, and biological traits were observed until the adult stage. The effect of nanofiber on survivorship and development ratio of *D. melanogaster* were investigated. Nutrient surface and viability were visualized by using SEM. According to the obtained results, PAN-DMF is suited with the nutrient surface compared to controls; and was found to have no adverse effect on the development of insect. Nanofibers can be used in food contact organisms, but more detailed studies would be needed to understand the effects of these components on environment and non target organisms.

Keywords: Nanofiber, *Drosophila melanogaster*, Nutrition, Development

Drosophila Melanogaster'in Biyolojik özelliklerine nanofiberin etkisi

ÖZ

Günümüzde nanofiber komponentler (poliakrilonitril dimetil formamit gibi) bir çok mühendislik alanında ve gıda sektöründe kullanılmaktadır. Çevre ve hedef olmayan organizmalar üzerindeki muhtemel etkilerin belirlenmesi gerekmektedir. *Drosophila melanogaster* Meigen (Diptera: Drosophilidae) gelişim biyolojisi ve çevre çalışmalarında çok çalışılan model bir organizmadır. Bu çalışmada, erginler nanofiber (elektrospinle üretilen 260 nm çap aralığına sahip PAN-DMF) kaplı diyetle beslenerek yetiştirilmiş, ergin evreye kadar biyolojik özellikler gözlenmiştir. Nano fiberin etkisi ile *D. melanogaster*'in yaşama gelişme ve hayatta kalma oranları araştırılmıştır. Besin yüzeyi ve canlılık SEM kullanarak görüntülenmiştir. Elde edilen sonuçlara göre, PAN-DMF kontrolle karşılaştırıldığında besin yüzeyinin uygun olduğu görülmüş; ve böceğin gelişim sürecini kötü etkilemediği bulunmuştur. Besinle ilişkili organizmalarda nanofiberler kullanılabilir, fakat bu bileşenlerin çevre ve hedef olmayan organizmalar üzerindeki etkilerinin anlaşılabilmesi için çok detaylı çalışmalara gerek duyulmaktadır.

Anahtar Kelimeler: Nanofiber, *Drosophila melanogaster*, Beslenme, Gelişim

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1. INTRODUCTION

Nanoparticles (NP) such as proteins present in biological systems are common in nature. These particles are biocompatible materials having surface modifications, but not biodegradable. In recent years, the production of nanomaterials (NM<100 nm) by NP and nanotechnological developments has increased [1]. NPs are classified as metal-based (transition metals and metal oxides), carbon-based (graphene, single- and multi-wall carbon nanotubes and fullerenes), nanocrystals, dendrimers (nano-scale polymers) and cadmium quantum particles [1, 2, 3]. NPs have been produced and used since the 1990s, because of their specific structural properties [4, 5]. Nanoparticles used in medicine and cosmetics products, packaging products (paper, plastic, metal, glass / ceramics), engineering, industrial products and applications (solar batteries, gas sensors, oxygen pumps, polishing, electronics), household appliances, textiles, food products (additive) and biomedical, bioengineering, in vivo applications including tissue repair, magnetic resonance imaging, immunoassay, detoxification of biologic fluids [6, 7, 8].

In use NMs have many advantages but can cause (the lack of standardized risk assessment and toxicological classification) health problems [1, 9, 10, 11]. Due to the widespread use and production of NM, it is very important to evaluate the reliability of these particles before they can be used in model organisms. *Drosophila melanogaster* Meigen is commonly used a model organism in in vivo toxicity studies for understanding vertebrate [10, 12, 13, 14, 15, 16]. In this study have been determined possible effects of nanofibers produced by the electrospinning in *Drosophila* biology and morphology.

2. MATERIALS AND METHODS

D. melanogaster (Wild type) stocks were cultured ($25 \pm 2^\circ\text{C}$, 60-70% relative humidity and 12/12 h L/D photoperiod) with an artificial diet [17, 18]. Nanofiber polyacrylonitrile dimethylformamide (PAN-DMF, diameter 260 nm, 10% range) was produced by electrospinning (26°C , 20 kV, 1-3 ml/h syringe pump, 12 cm collectors distance, Figure 1) [19].

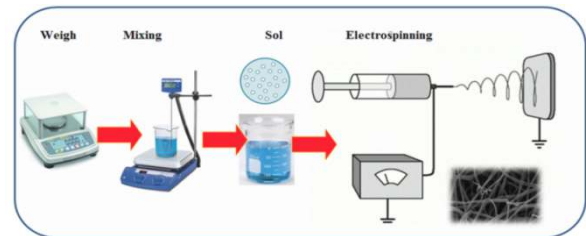


Figure 1. Synthesis Technique of PAN-DMF

The culture diet was coated with PAN-DMF (only the upper surface), and newly emerged adults (6 female: 2 male) were transferred to medium (Figure 2). Eggs were feed with PAN-DMF until adult stage, and biological traits (survivorship, development and sex ratio) of 3rd instar larvae, puparium, female and male were investigated.

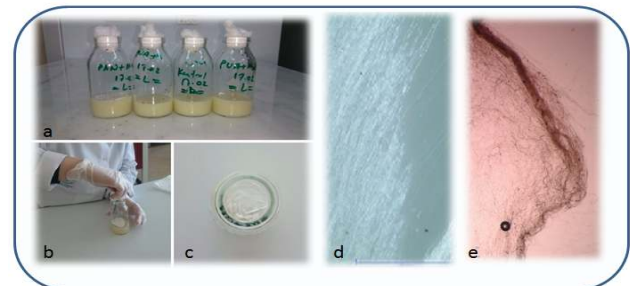


Figure 2. The experimental setup; a. Culture design, b. Food coating, c. The first coating instantly, Microscopic image of Nanofiber d. 1 mm, e. x40.

The experiments were performed four times. Experimental data were expressed as means \pm S.E. The data were subjected to statistical analysis by one-way analysis of variance (ANOVA) was followed by least significant difference (LSD) test to determine significant differences between means. Data on survivorship were compared by chi-squared test [19]. A values of $p < 0.05$ was considered significant (SPSS, 1997).

3. RESULTS AND DISCUSSION

Nutrient surface and viability were visualized by using SEM. According to the obtained results, PAN-DMF is suited with the nutrient surface compared to controls; and was found to have no adverse effect on the development of insect (Figure 3).

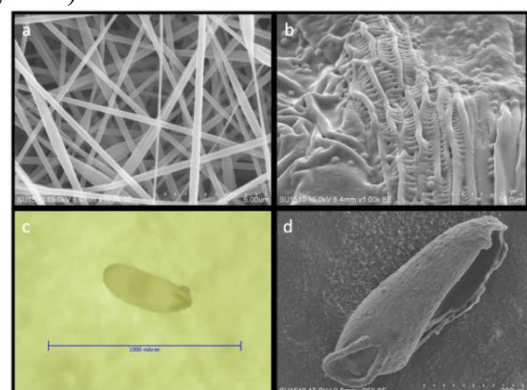


Figure 3. The SEM photos of nutrient surface before (a) and after (b) nutrition experiment with PAN-DMF, c.

Microscopic image of egg (1 mm scale), d. SEM image of the egg (diameter 150 um, height 420 um)

Drosophila is influenced by nutrition [21].

Nutrition has influences on development, fertility, longevity, immune defense in variety of animals [22, 23]. The diets containing the PAN-DMF didn't influence the development time compared to controls. It was found that this application increased the survival rate of the insects by about 14%, and did not changed the ratio of female and male compared to control (Table 1). It is stated that *Drosophila* may be an increase in survival rate and developmental time (CeO₂, CuO) without any interaction with various NPs (Multi-Walled Carbon Nanotubes, silica, p-aminobenzoic acid) [24, 25, 26, 27, 28].

Table 1. Effects of PAN-DMF on survival, development and sex ratio of the *D. melanogaster*

PAN-DMF %	Survival to third instar.	Time to third instar.	Survival to pupal stage.	Time to pupal stage.	Survival to adult stage.	Time to adult stage.	Sex ratio (%)	
	(%) (Mean ± S.E) [§]	(day) (Mean ± S.E) [§]	(%) (Mean ± S.E) [§]	(day) (Mean ± S.E) [§]	(%) (Mean ± S.E) [§]	(day) (Mean ± S.E) [§]	Male (Mean ± S.E) [§]	Female (Mean ± S.E) [§]
0.00 [§]	85.75 ± 2.1a	3.52 ± 0.2a	85.75 ± 3.0a	4.65 ± 0.2a	84.04 ± 1.7a	7.50 ± 0.2a	40.00 ± 1.0a	60.00 ± 1.0a
%10	99.75 ± 0.7b	3.01 ± 0.1a	98.00 ± 0.5b	4.30 ± 0.1a	90.00 ± 1.2b	8.12 ± 0.4a	33.00 ± 1.0a	67.00 ± 1.0a

*Four replicates with 100 larvae per replicate, ^bValues followed by the same letter are not significantly different from each other, P > 0,05 (χ² test, LSD Test). [§]Control.

Gold NP has been reported to cause phenotypic disturbances, reducing life span and sexual productivity in *Drosophila* [29, 30]. In our study, although all stages of development appear to be morphologically similar to controls, males that use of PAN-DMF have been increased size and seen genital wounds (at least 50 individuals, Figure 4). It is expressed in studies where exposure of various NPs may caused developmental retardation (Ag, CdSe, Magnetite), decreased egg production (TiO₂) and survival rate -body pigmentation in *Drosophila* [1]. *Drosophila* protect themselves from harmful effects of environmental stress depending on their detoxification capacities [31]. Also the absence of genital wounds in female may indicate that female subjects are more resistant to environmental stress.

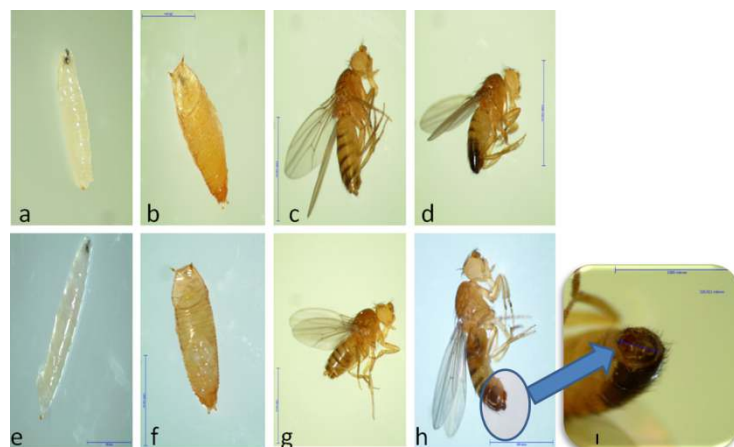


Figure 4. Morphological images of experimental subjects; Control (a) larvae, (b) pupae, (c) female, (d) male, and PAN-DMF (e) larvae, (f) pupae, (g) female, (h) male, (i) genital wound.

4. CONCLUSIONS

Drosophila is a recommended and used model for biological parameters, pigmentation, physiological, metabolic and morphological disorders in the toxicity studies NPs can produce for NM production. The findings of this study showed that nanofiber positively affected on survival and development properties of insect but not morphological. Nanofibers can be used in food contact organisms, but more detailed studies would be needed to understand the effects of these components on environment and non target organisms.

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