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Katyonik Gemini Surfaktant (10-2-10), (12-2-12) ve Ticari Surfaktant CTAB'nın *Allium cepa* L. Üzerindeki Fitotoksik Etkisi

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Özet

Bu çalışmada, alkanedil-a-ω-bis (alkildimetilamonyum) dibromür tipi, m-2-m ile gösterilen farklı alkil gruba sahip (m= 10 ve 12) cationic Gemini surfaktantlar ve ticari katyonik surfaktant hekzadesiltrimetilamonyumun bromürün (CTAB) *Allium cepa* L. üzerinde fitotoksik etkileri incelenmiştir. Dimerik surfaktantlar laboratuarımızda sentezlendi ve saflaştırıldı. *Allium cepa* L. nın kök büyümesi üzerine surfaktantların fitotoksik etkisi laboratuvar şartlarında 7 gün boyunca gözlendi. Çalışmada kullanılan surfaktantlar 0.624, 0.312 and 0.156 g/L olmak üzere üç farklı konsantrasyonda hazırlandı. Fitotoksite neticeleri % 50 lık azaltılan kök büyümesi ile ifade edilen (EC_{50}) etkili konsantrasyona dayanır. Çalışılan üç farklı surfaktant arasında farklı etkiler gözlenmiştir. En büyük inhibasyon etki (10-2-10) Gemini surfaktantda oluşurken en az inhibasyon etki ise (12-2-12) Gemini surfaktantın 0.156 g/L konsantrasyonunda gözlenmiştir. Bütün katyonik surfaktantların artan konsantrasyonu ile inhibitör etkinin arttığı görülmüştür.

Anahtar Kelime: *Allium cepa* L, Katyonik Gemini surfaktant, CTAB**,** Surfaktant etkisi, (10-2-10), (12-2-12**).**

Phytotoxic Effects of Cationic Gemini Surfactants (10-2-10), (12-2-12) and Conventional Surfactant CTAB on *Allium cepa* L.

Abstract

In this study, the phytotoxic effects of cationic Gemini surfactants of the alkanediyl-a- ω -bis (alkyldimethylammonium) dibromide type, with different alkyl groups, referred to as ''m-s-m''(m = 10 and 12) and conventional surfactant hexadecyltrimethylammonium bromide, CTAB) on *Allium cepa* L. were examined. These dimeric surfactants were synthesized and purified in our laboratory. During the 7 days the phytotoxic effects on root growth were observed at the laboratory conditions. Surfactants used in study were prepared at three different surfactant concentrations, 0.624, 0.312 and 0.156 g/L (w/v) concentrations. The phytotoxicity results were based on the effective concentration that reduced root growth by 50 % (EC_{50}). Some differences were observed between the effects of three types of cationic surfactants. While the largest inhibitory effects occur in Gemini (10-2-10), the smallest inhibitory effects occurred in Gemini (12-2-12) at 0.156 g/L concentration. . It was observed that inhibitory effects increased with increasing concentrations of in all cationic surfactants.

Keywords: *Allium cepa* L, Cationic Gemini surfactant, CTAB**,** Effect of surfactant, (10-2-10), (12-2-12**).**

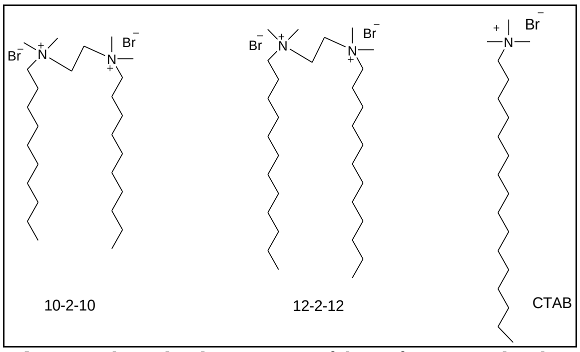
INTRODUCTION

Surfactants are one of the most widely used additives in agriculture. They essentially reduce surface and interfacial tensions by accumulating at the interface immiscible fluids. Surfactants are amphiphilic molecules that contain a non-polar hydrophobic tail and a polar hydrophilic head group. In aqueous solutions, this dual character can lead to self - association or micellization of the surfactant. Surfactants can be classified into ionic (anionic or cationic) and non-ionic depending on hydrophilic head group charge. It is well known that ionic surfactants show higher toxicity value than the non-ionic ones, and those cationic surfactants are more potent then their anionic counterparts [1, 2]. Gemini or dimeric surfactants are novel surfactants that have become of considerable interest in the academic and industrial arena. These surfactants are consist of two amphiphilic moieties connected at the level of the head groups by a spacer group of varying nature widely hydrophobic, hydrophilic, rigid or flexible [3, 4]. Due to their different molecular structure with respect to conventional single tailed surfactants they have superior properties for special purposes [5-9]. Gemini surfactants show very high bacterial activity to depend on the nature and length of spacer and the nature of hydrophobic tail [10-12]. These surfactants are used in skin care formulations, antipollution protocols, analytical separations, nanoscale technology. Furthermore, surfactants are well known to inhibit the growth or to kill bacterial [13-15] and fungal cells [16], or to increase the efficacy of antimicrobial agents [17]. The most commonly studied surfactants have been those containing nonionic surfactants and there are a few studies about the effects of nonionic surfactants on plants. In our previous study, we have reported the phytotoxic effect on root growth for three different octylphenol series non-ionic surfactant, Triton X-114, Triton X-100, Triton X-405 under laboratory conditions using onion (*Allium cepa* L.) [18]. In another study, the effects of polyoxyethylene type nonionic surfactants (POE) on root elongation of *Allium cepa* L. have determined [19]. It was found that the stimulatory effect on root elongation becomes stronger when the number of ethylene oxide group was increased in the POE surfactant. However, there has been not enough research on the practical properties of dimeric ammonium salts for plants. In recent research, it was indicated that the biological effects strongly depend on the structure of surfactants [20]. The aim of this study are to investigate the phytotoxic effects of cationic gemini surfactants with different alkyl groups (10-2-10, 12-2-12) and conventional surfactant hexadecyltrimethylammonium bromide, CTAB on *Allium cepa* L. under laboratory conditions.

MATERIAL and METHOD

Material

The meric cationic surfactant, cetlytrimethyl ammonium bromide (CTAB) has supplied from Merck and used without purification. Dimeric cationic surfactants (or gemini), (10-2-10) and (12-2-12) have been synthesized, purified and characterized in our laboratory [14]. The original materials for the synthesis of the cationic geminis, alkanediyl-α-ω-bis (alkyldimethylammonium) dibromides: 1-dibromodecane, (97 %, Fluka), 1-bromododecane, (95 %, Fluka) and N, N, N', N'-tetramethylethylenediamine (98 %, Fluka) were used without further purification. Surfactant purities were checked by nuclear magnetic resonance (NMR) and surface tension, all with excellent results. 1H NMR spectra were recorded in $CDCl_3$ solution with a Varian Mercury Plus 300 MHz spectrometer. 13C NMR spectra were recorded at 75 MHz. [19]. The characterizations of the surfactants used in this study are given in Table 1 and their molecular structures are shown in Scheme 1.



Scheme 1. The molecular structures of the surfactants used in this study.

Method

In this study, onion (*Allium cepa* L) was selected as a test material because it is a good specimen for study and has many primordial (adventitious) roots that are growing rapidly. *Allium cepa* L. represent the third largest fresh vegetable industry in same places. Healthy and equal-sized bulbs of common onion were selected. Firstly, the outer dry scales of the bulbs were removed and the ring of the root primordial was left intact. Twelve onions composed an experimental set including three of them were control. Secondly, onions were placed individually in 60 mL vessels containing different surfactant solutions. All solutions were prepared with tap water. The temperature of the laboratory was kept at 16 ± 0.5 ºC. At three different surfactant concentrations (0.624, 0.312 and 0.156 g/L), the surfactants solutions were prepared from stock solution (5 g surf./L) for each surfactant. The control groups were treated only with tap water. Experiments were replicated 3 times. Root lengths were measured using a millimeter ruler starting at the onset of incubation, then after 1st, 2nd, 3rd, 4th, 5th, 6th and 7th days. At the end of the 7th day, the total root lengths were measured. Means and standard deviations of the length of onion root tips were determined. The phytotoxicity results were based on the effective concentration that reduced root growth by 50 % (EC_{50}).

RESULTS AND DISCUSSION

The effects of two cationic gemini surfactants, (10-2-10, 12-2-12) and conventional surfactant, CTAB on root elongations of *Allium cepa* L. after 7-days are shown in Figure 1. As can be seen in Fig.1 the phytotoxic effects were observed for three surfactants at all concentrations. Some differences were found between the root elongations of three types of surfactants. Inhibitory effect or toxicity of surfactants occurred in all surfactants. As seen as Fig.2 (a), (b) and (c) inhibitory effects increased with increasing surfactant concentration. There are many studies about inhibitory effects of surfactants. However there isn't any study similar to our study. This is an original research about inhibitory effects of gemini series. In order to evaluate the effect of alkyl chain length variation (m= 10 and 12) on inhibory effect, we kept the spacer chain length (s=2) and head groups the same in two geminis. The m-2-m gemini is a double-headed dicationic surfactant with a short ethylene spacer, which means that its head group has a high charge density. The stability of the (m-2-m) surfactants monolayer increases as the hydrophobic chain length increasing, in direct relation to increasingly stronger hydrophobic interactions. When two gemini surfactants compared, it is seen that (10-2-10) was more toxic than (12-2-10) gemini surfactant. This means that the toxicity of surfactants is depend on the hydrophobic state of the surfactant. Also when gemini (12-2-12) and CTAB compared, gemini surfactant has got lower toxicity than CTAB. Also CTAB has alkyl chain with sixteen carbons, but it was meric surfactant. The highest phytotoxic effects were observed for gemini (10-2-10) at 0.312 and 0.156 g/L surfactant concentration. When we compared the phytotoxic effect of three surfactants doses, the biggest surfactant concentration 0.624 g/L was found to be very toxic.

Table 1. Characterization of the experimental surfactants

Surfactants	Surfactant symbols	Classification	Structural formula
N,N'didecyl-N,N,N',N-tetramethyl N,N-ethanediyl-diammonium dibromide	(10-2-10)	Cationic Gemini	$C_{10}H_{21}N^{+}(CH_3)_2$-$CH_2$-($CH_2$-$OCH_2)_2$-$CH_2$-$^{+}(CH_3)_2C_{10}H_{21}$, 2Br-
N,N'didodecyl-N,N,N',N-tetramethyl N,N-ethanediyl-diammonium dibromide	(12-2-12)	Cationic Gemini	$C_{12}H_{25}N^{+}(CH_3)_2$-$CH_2$-($CH_2$-$OCH_2)_2$-$CH_2$-$^{+}(CH_3)_2C_{12}H_{25}$, 2Br-
Cetyltrimethyl ammonium bromide	CTAB	Conventional Cationic	$C_{16}H_{33}N^{+}(CH_3)_3Br^{-}$

As seen as Fig.1, (EC_{50}) concentration of gemini (12-2-12) was found to be 0.156 g/L dose. (EC_{50}) concentration of gemini (10-2-10) and CTAB wasn't found.

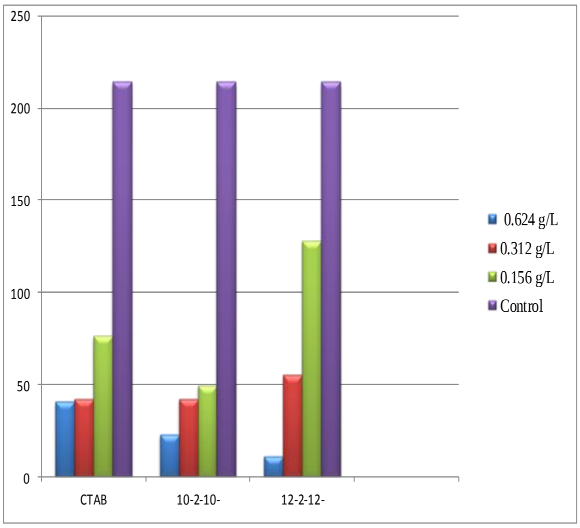
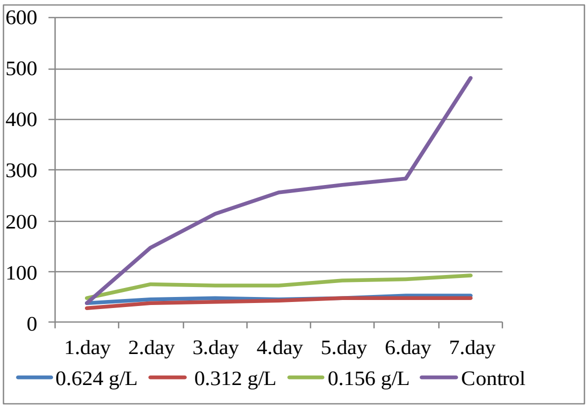
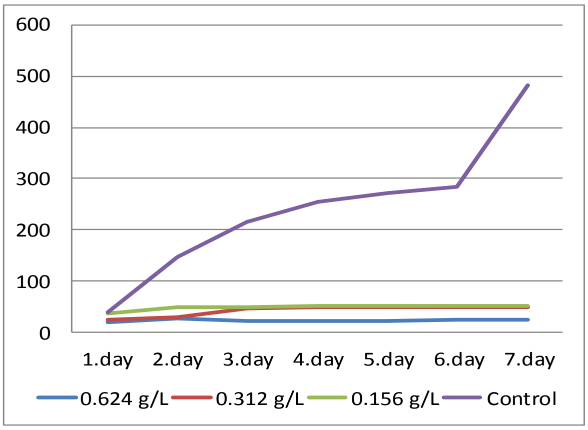


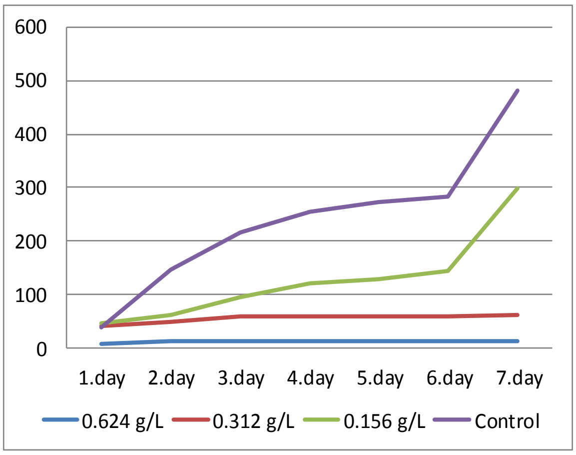
Figure 1. Mean of root elongations effected by three surfactant solutions after 7 days.



(a). CTAB



(b). Gemini (10-2-10)



(c). Gemini 12-2-12

Figure 2. The mean of root elongations affected by three cationic surfactant solutions after 7 days (a). (CTAB), (b). (10-2-10), (c). (12-2-12).

CONCLUSION

In this study, the inhitory effect of three different cationic surfactans was investigated at all studied doses. The lowest phytotoxic effects were observed for gemini (12-2-12) at 0.156 g/L surfactant concentration. At this dose, (EC_{50}) concentration of gemini (12-2-12) was found to be 0.156 g/L. More studies are required to get enough information for detail. Consequently, the effects of surfactants have a marked impact on human health care, bio-technology, environmental protection and agrochemistry. It's important that determine (EC_{50}) concentration, which are close to control values. In addition it must be selected appropriate surfactant, which has minimal toxicity and maximal benefits for each purpose.

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