



INTISARI

Latar Belakang: Biofilm didefinisikan sebagai komunitas bakteri yang dilindungi oleh matriks eksopolisakarida dan melekat pada permukaan biotik atau abiotik. *S. sanguinis* dan *S. mutans* dapat berkoagragasi untuk membentuk biofilm. Nanokitosan yang berasal dari kulit udang galah memiliki sifat khusus sehingga dapat dikembangkan sebagai antibiofilm. Penelitian ini bertujuan untuk mengetahui potensi nanokitosan kulit udang galah dalam menghambat pembentukan dan mendegradasi biofilm *S. sanguinis* dan *S. mutans*.

Metode Penelitian: Penentuan nilai KHM nanokitosan terhadap *S. sanguinis* dan *S. mutans* dengan metode makrodilusi. Uji penghambatan pembentukan biofilm dilakukan dengan menginkubasi *S. sanguinis* dan *S. mutans* dalam media BHI+sukrosa 2% bersamaan dengan nanokitosan konsentrasi 0,5%, 0,25%, dan 0,125%, kontrol positif (khlorheksidin), kontrol negatif (PBS) selama 24 dan 48 jam. Uji degradasi biofilm dilakukan pada kultur biofilm *S. sanguinis* dan *S. mutans* 24 dan 48 jam. Nanokitosan konsentrasi 0,5%, 0,25%, dan 0,125%, kontrol positif (khlorheksidin), kontrol negatif (pbs) ditambahkan dalam kultur biofilm *S. sanguinis* dan *S. mutans* 24 dan 48 jam. Setelah 24 jam, biofilm diwarnai dengan kristal violet 0,1% dan dianalisis dengan *microplate reader* pada $\lambda=540\text{nm}$. Persentase penghambatan pembentukan biofilm dan degradasi biofilm kemudian dihitung. Hasil uji degradasi kemudian diamati menggunakan *Confocal Laser Scanning Microscopy* (CLSM). Data selanjutnya dianalisis statistik dengan *One Way ANOVA* dan *Post Hoc LSD*.

Hasil: Nanokitosan menunjukkan nilai KHM 0,5% terhadap *S. sanguinis* sedangkan 0,25% terhadap *S. mutans*. Uji *One Way ANOVA* menunjukkan terdapat pengaruh nanokitosan dalam menghambat pembentukan biofilm dan mendegradasi biofilm *S. sanguinis* dan *S. mutans* ($p=0,000$). Uji *Post Hoc LSD* menunjukkan nanokitosan 0,5% merupakan konsentrasi yang paling efektif dalam menghambat pembentukan biofilm dan mendegradasi biofilm *S. sanguinis* dan *S. mutans*. Hasil pengamatan CLSM setelah perlakuan nanokitosan 0,5% menunjukkan proporsi kematian sel yang lebih kecil dibandingkan kontrol positif serta tedapat penurunan ketebalan biofilm dibanding kontrol negatif.

Kesimpulan: Nanokitosan dari kulit udang galah dapat menghambat pembentukan biofilm serta dapat mendegradasi sebagian biofilm *S. sanguinis* dan *S. mutans* secara *in vitro*. Nanokitosan 0,5% adalah konsentrasi paling optimal dalam menghambat pembentukan biofilm dan mendegradasi sebagian biofilm *S. sanguinis* dan *S. mutans*.

Kata Kunci: Biofilm, Degradasi, Nanokitosan, Penghambatan, *S.sanguinis*, *S.mutans*



ABSTRACT

Background: Biofilms are defined as bacterial communities confined in self-generated exopolysaccharide matrices and attached to biotic or abiotic surfaces. *S. sanguinis* and *S. mutans* have ability to aggregate to form biofilms. Nanochitosan derived from giant prawn shells has a special characteristic and thus may be developed as antibiofilms. This study aimed to determine the potential of giant prawn shells nanochitosan in inhibiting and degrading *S. sanguinis* and *S. mutans* biofilms.

Research Methods: Minimum inhibitory concentration of nanochitosan against *S. sanguinis* and *S. mutans* was determined using macrodilution method. Biofilm inhibition assay was conducted by incubating *S. sanguinis* and *S. mutans* in BHI supplemented with 2% sucrose together with various concentration of nanochitosan (0.5%, 0.25%, and 0.125%), chlorhexidine (positive control) and PBS (negative control) for 24 and 48 h. Biofilm degradation assay was performed on 24 and 48 hours culture of *S. sanguinis* and *S. mutans*. Various concentration of nanochitosan (0.5%, 0.25%, and 0.125%), chlorhexidine (positive control), pbs (negative control) were added to 24 and 48 hours culture of *S. sanguinis* and *S. mutans* biofilm. After 24 h, the biofilm was then stained using 0.1% crystal violet and measured using microplate reader at $\lambda=540$ nm. The percentage of biofilm formation inhibition as well as percentage of biofilm degradation were then calculated. Biofilm degradation was further observed using Confocal Laser Scanning Microscopy (CLSM). Data were statistically analyzed by One Way ANOVA and Post Hoc LSD.

Results: MIC value of nanochitosan against *S. sanguinis* was 0.5% while MIC value of nanochitosan against *S. mutans* was 0.25%. One Way ANOVA showed significant differences among groups, indicating that nanochitosan affected *S. sanguinis* and *S. mutans* biofilms formation as well as degradation ($p=0.000$). Post Hoc LSD test showed that 0.5% nanochitosan was the most effective concentration in inhibiting and degrading *S. sanguinis* and *S. mutans* biofilms. CLSM observation after 0.5% nanochitosan treatment showed a smaller proportion of cell death compared to positive control and a decrease in biofilm thickness compared to negative control. **Conclusion:** Nanochitosan of giant prawn shells inhibits and partially degrades the formation of *S. sanguinis* and *S. mutans* biofilms in vitro. Nanochitosan concentration of 0.5% is the most optimal concentration in inhibiting and degrading the *S. sanguinis* and *S. mutans* biofilm.

Keyword: Biofilm, Degradation, Inhibition, *S. sanguinis*, *S. mutans*, Nanochitosan