

# Pengaruh Paparan Nitrat Asal Daun Arugula terhadap Pertumbuhan Bakteri *Rothia* spp. pada Biofilm Saliva Perokok Usia Dewasa Muda = Nitrate Originating from Arugula Leaves in Increasing the Number of *Rothia* spp. in the Salivary Biofilm of Young Adult Smokers

Namira Adelia, author

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## Abstrak

Latar belakang: Penelitian terdahulu telah menunjukkan peran penting bakteri *Rothia* spp. dalam keseimbangan mikrobioma oral. Mikrobioma oral yang seimbang berperan terhadap pencegahan berbagai penyakit rongga mulut. Namun, pengaruh pemberian nitrat terhadap pertumbuhan bakteri tersebut masih belum sepenuhnya dipahami, terutama pada kelompok individu yang memiliki kebiasaan merokok. Merokok dapat mengubah komposisi mikrobiota oral dan memengaruhi metabolisme bakteri, sehingga penting untuk menyelidiki bagaimana paparan nitrat dapat memodulasi pertumbuhan bakteri ini dalam konteks tersebut. Dasar dilakukannya penelitian ini adalah untuk mengeksplorasi potensi paparan nitrat yang berasal dari sayur-sayuran dalam meningkatkan jumlah bakteri pereduksi nitrat pada biofilm saliva subjek perokok usia dewasa muda. Tujuan: Melihat potensi nitrat larutan daun arugula dalam meningkatkan pertumbuhan bakteri *Rothia* spp. pada perokok usia dewasa muda. Metode: Sampel saliva hasil pooling dikelompokkan berdasarkan pH supernatant saliva dan kebiasaan merokok. Kemudian, sampel diberikan paparan larutan daun arugula dengan konsentrasi nitrat berbeda (6,25 M dan 3,25 M) dan durasi paparan berbeda (5 dan 9 jam) untuk melihat perubahan terhadap pH supernatant saliva, kadar nitrit saliva dan jumlah bakteri *Rothia* spp. Pengukuran pH supernatant saliva dilakukan menggunakan kertas strip pH. Pengujian kadar nitrit saliva dilakukan dengan uji griess pada sampel yang merepresentasikan kadar nitratnya. Metode Total Plate Count digunakan untuk menghitung jumlah bakteri *Rothia* spp. yang tumbuh pada medium spesifik. Untuk memastikan benar bakteri *Rothia* spp. yang tumbuh, dilakukan pewarnaan gram dan PCR konvensional dengan primer *Rothia*. Hasil: Tidak terdapat perbedaan yang signifikan antara paparan larutan daun arugula terhadap kadar nitrit saliva dan jumlah bakteri *Rothia* spp. Terdapat pola peningkatan kadar nitrit saliva berdasarkan durasi paparan dengan hasil yang berbanding lurus dengan jumlah bakteri *Rothia* spp. Kesimpulan: Hasil penelitian menunjukkan paparan larutan daun arugula mampu mempertahankan dan mengurangi asidifikasi pH supernatant saliva. Secara umum, potensi nitrat asal daun arugula konsentrasi 3,25 M memiliki potensi yang lebih tinggi dibandingkan dengan nitrat asal daun arugula konsentrasi 6,25 M dalam meningkatkan pertumbuhan bakteri *Rothia* spp.

.....Background: Previous research has shown the important role of *Rothia* spp. in maintaining the balance of oral microbiome. A balanced oral microbiome plays a crucial role in preventing various oral diseases. However, the effects of nitrate administration on the growth of these bacteria are not fully understood, especially in individuals who smoke. Smoking can alter the composition of oral microbiota and affect bacterial metabolism, making it important to investigate how nitrate exposure can modulate the growth of these bacteria. The basis of this research is to explore the potential of nitrate exposure derived from

vegetables in increasing the number of nitrate-reducing bacteria in the salivary biofilm of young adult smokers. Objectives: The objective of this study is to investigate the potential of nitrate originating from arugula leaves in increasing the number of *Rothia* spp. bacteria in young adult smokers. Methods: Saliva samples were pooled based on supernatant pH and smoking habit. The samples were then exposed to arugula leaf solution with different nitrate concentration (6,25 M dan 3,25 M) and different exposure durations (5 and 9 hours) to observe changes in supernatant pH, nitrite levels, and the viability of *Rothia* spp. bacteria. Supernatant pH was measured using pH strips. The nitrite levels in saliva were tested using the Griess assay. The total plate count method was used to calculate the viability of *Rothia* spp. bacteria growing on specific media. To confirm that the bacteria growing were indeed *Rothia* spp., gram staining and conventional PCR with *Rothia* primers were performed. Results: There is no significant difference between the exposure of arugula leaf solution regarding the level of nitrite in saliva and the viability of *Rothia* spp. This study shows a pattern of increased nitrite levels in saliva according to the duration of exposure, which are linear with the viability of *Rothia* spp. Conclusion: The results of the study indicate that exposure to arugula leaf solution can maintain and reduce the acidification of supernatant pH. In general, the nitrate potential of arugula leaves at a concentration of 3,25 M has a higher potential compared to the nitrate potential of arugula leaves at a concentration of 6,25 M in enhancing the growth of *Rothia* spp. bacteria.