

Telaah Aktivitas Antiaging secara in vitro pada Ekstrak dan Isolat Teridentifikasi dari *Rubus fraxinifolius* dan *Rubus rosifolius* = Study of In Vitro Anti-aging Activity on Extracts and Identified Isolates from *Rubus fraxinifolius* and *Rubus rosifolius*

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Abstrak

<p>*Rubus fraxinifolius* dan *R. rosifolius* merupakan tanaman *Rubus* yang dapat ditemukan di daerah pegunungan Indonesia. Kedua tanaman memiliki morfologi buah yang mirip yaitu berbentuk berry merah dan edible, serta mengandung senyawa golongan triterpenoid, polifenol dan flavonoid. Beberapa spesies *Rubus* dilaporkan memiliki aktivitas sebagai antiaging yaitu antielastase, antioksidan, dan antitirosinase. Penelitian ini bertujuan untuk menelaah aktivitas antiaging secara in vitro pada ekstrak dan isolat dari tanaman *R. fraxinifolius* dan *R. rosifolius*. Batang, buah, dan daun kedua tanaman diekstraksi menggunakan alat Soxhlet serta dilakukan skrining aktivitas anti elastase dan antioksidan. Selanjutnya terhadap ekstrak terpilih dilakukan pemisahan, fraksinasi dan isolasi senyawa. Isolat yang didapat diidentifikasi menggunakan spektrometri FTIR, ¹H-NMR, ¹³C-NMR, DEPT, HSQC, HMQC, HMBC, dan LC-MS, serta diuji aktivitas antielastase, antitirosinase dan sitotoksitas pada sel fibroblast secara in vitro. Hasil ekstraksi bertingkat menunjukkan bahwa ekstrak metanol daun *R. fraxinifolius* memiliki aktivitas antielastase dan antioksidan tertinggi dengan masing-masing IC₅₀ 57,45 dan 4,33 µg/ml. Terhadap fraksi metanol daun *R. fraxinifolius* (DFM) dilakukan pemisahan menggunakan kromatografi cair vakum dan diperoleh 11 fraksi. Uji antielastase fraksi menunjukkan fraksi paling aktif adalah DFM8. Selanjutnya dilakukan isolasi lebih lanjut terhadap DFM8 dan diperoleh 3 isolat. Hasil elusidasi struktur menunjukkan bahwa ketiga isolat merupakan suatu triterpen pentasiklik tipe ursan. Hasil telaah data pengujian DEPT, HMQC, HSQC, HMBC serta IR dan MS disimpulkan senyawa DFM 8a adalah asam 2,3-glikol, 19 \hat{I} -hidroksi-12-ursen-23,28-dioat ($C_{32}H_{48O_7}$, BM 544); DFM8b asam 2,3-propanandiol, 19 \hat{I} -hidroksi-12-ursen-28-oat ($C_{33}H_{52O_5$, BM 528,38); dan DFM8c asam 2,3-glikol-19 \hat{I} -hidroksi-23,24-nor-12-ursen-28-oat ($C_{30}H_{46O_5$, BM 486,33). Ketiga senyawa hasil isolasi ini merupakan senyawa baru dan belum pernah ditemukan sebelumnya. Uji aktivitas antielastase senyawa DFM8a, DFM8b, dan DFM8c memiliki IC₅₀ berturut-turut adalah 122,199; 98,22; dan 54,33 µg/ml, serta antitirosinase dengan IC₅₀ 207,8; 221,5; dan 335,9 µg/ml. Uji toksisitas menunjukkan bahwa ekstrak DFM, fraksi DFM8, dan isolat DFM8b tidak toksik terhadap sel fibroblas NIH/3T3.</p><hr /><p>*Rubus fraxinifolius* and *R. rosifolius* are *Rubus* genus, which can be found in the mountain of Indonesia. Both plants have similar fruit morphology: red and edible berries and contain triterpenoid, polyphenols, and flavonoids. Some species of *Rubus* are reported to have antiaging activity, antielastase, antioxidant, and antityrosinase. This study aims to examine the in vitro antiaging activity of extracts and isolated compounds from *R. fraxinifolius* and *R. rosifolius*. The stems, fruits, and leaves of both plants were extracted and screened for

antielastase and antioxidant activity. Furthermore, the selected extracts were separated, fractionated, and isolated to yield isolates. The obtained isolates were identified using FTIR spectrometry, ^1H -NMR, ^{13}C -NMR, DEPT, HSQC, HMQC, HMBC, and LC-MS, and also were tested for antielastase, antityrosinase, and cytotoxicity activities in fibroblast cells. The continuous extraction results showed that the methanol extract of *R. fraxinifolius* leaves had the highest antielastase and antioxidant activity with IC₅₀ 57.45 ppm and 4.33 ppm, respectively. The methanol fraction of *R. fraxinifolius* (DFM) leaves were separated using vacuum liquid chromatography and obtained 11 fractions. The antielastase assay of fractions gave the most active fraction was DFM8. Then, further isolation of DFM8 was carried out, and three isolates were obtained. The structural elucidation showed that the three isolates were ursane-type of pentacyclic triterpenes. The results of DEPT, HMQC, HSQC, HMBC, IR and MS spectrometry test concluded that the compound DFM 8a was 2,3-glycol, 19 $\hat{\beta}$ -hydroxy-12-ursen-23,28-dioic acid ($\text{C}_{32}\text{H}_{48}\text{O}_7$, MW 544); DFM8b 2,3-propanandiol, 19 $\hat{\beta}$ -hydroxy-12-ursen-28-oic acid ($\text{C}_{33}\text{H}_{52}\text{O}_5$, MW 528.38); and DFM8c 2,3-glycol-19 $\hat{\beta}$ -hydroxy-23,24-nor-urs-12-en-28-oic acid ($\text{C}_{30}\text{H}_{46}\text{O}_5$, MW 486.33). All isolated compounds are new compounds and have never been found before. The IC₅₀ of antielastase activity of DFM8a, DFM8b, and DFM8c were 122.199; 98.22; and 54.33 ppm, respectively, and the IC₅₀ of antityrosinase activity were 207.8; 221.5; and 335.9 ppm, respectively. Toxicity tests showed that the DFM extract, the DFM8 fraction, and the DFM8b were not toxic to NIH/3T3 fibroblast cells.