

KARAKTERISASI SEL PUNCA SPESIFIK C-KIT DAN SCA-1 HASIL *MAGNETIC ACTIVATED CELL SORTING*: ANALISIS PROFIL MIKRO-RNA DAN EKSPRESI *TRANSCRIPTION FACTORS*

Shoma Adhi Wijaya
(14/372708/PBI/1323)

Abstrak

Cardiac resident stem cell (CRSC) merupakan sel punca yang potensial dalam terapi sel punca jantung. Populasi *cardiac resident stem cell* yang berpotensi digunakan untuk terapi yaitu populasi C-kit dan Sca-1. Populasi tersebut memiliki kemampuan proliferasi yang baik dan dapat berdiferensiasi menjadi jaringan kardiomyosit jantung yang diregulasi oleh *transcription factor* yaitu Nkx2.5, Gata4, Akt2, dan Mef2. Selain itu MikroRNA (miR) berperan dalam diferensiasi dan proliferasi sel. MikroRNA-1 berperan dalam proses diferensiasi dan miR-133a berperan dalam proses proliferasi. *Sorting* sel menggunakan *Magnetic Activated Cell Sorting* (MACS). Tujuan penelitian ini adalah untuk melihat efektivitas metode *sorting* sel, analisis profil miR-1 dan miR-133a, serta ekspresi *transcription factor*. Persentase sel dianalisis dengan menggunakan *flow cytometer*. Konsentrasi RNA diukur dengan *Nano drop spectrophotometer*. Profil mikroRNA dan ekspresi *transcription factor* diukur dengan menggunakan *real-time PCR*. Data yang didapat dianalisis dengan menggunakan *Microsoft Excel*. Hasil menunjukkan persentase sel C-kit yaitu 0% sampai dengan 0,05%. Persentase sel Sca-1 adalah 0,05% sampai dengan 0,25%. Profil miR pada populasi CRSC, C-kit, dan Sca-1 tidak jauh berbeda. Standar deviasi miR sampel nomor 48, 54, dan 64 adalah miR-1 ($n \pm 3,2$; $n \pm 2,6$; $n \pm 2,9$) dan miR-133a ($n \pm 2,3$; $n \pm 3,1$; $n \pm 2,2$). Ekspresi *transcription factor* populasi CRSC, C-kit, dan Sca-1 tidak jauh berbeda. Standar deviasi sampel nomor 48, 54, dan 64 berturut-turut adalah Mef2c ($n \pm 3,1$; $n \pm 1,6$; $n \pm 1,5$), Nkx2.5 ($n \pm 2,7$; $n \pm 2,0$; $n \pm 1,8$), Akt2 ($n \pm 0,7$; $n \pm 1,1$; $n \pm 0,6$), Gata4 ($n \pm 3,1$; $n \pm 1,1$; $n \pm 0,7$). Ekspresi *transcription factor* yang paling tinggi yaitu Akt2 disusul Gata4, Mef2c, dan Nkx2.5. Peningkatan profil miR dan ekspresi *transcription factor* terjadi seiring dengan pertumbuhan sel dari *flask* T75 menuju ke *flask* T150.

Kata kunci: *Cardiac stem cell*, *miR-1*, *miR-133a*, *transcription factor*, *RT-PCR*

MAGNETIC ACTIVATED CELL SORTED C-KIT AND SCA-1 CHARACTERIZATION: MICRO-RNA PROFILE AND TRANSCRIPTION FACTORS EXPRESION ANALYSIS

Shoma Adhi Wijaya
(14/372708/PBI/1323)

Abstract

C-Kit and Sca-1 are two populations that reside in the *cardiac resident stem cell* (CRSC) isolated from cardiac tissue. They are potential to be used as myocardial infarction therapy. Those populations have adequate capabilities to proliferate and differentiate into cardiomyocytes which are regulated by transcription factors such as Nkx2.5, Gata4, Akt2, and Mef2c. Proliferation and differentiation of C-Kit and Sca-1 are also influenced by *microRNA* (miR). miR-1 plays an important role in cell differentiation as well as miR-133a in cell proliferation. Cell sorting was conducted using *Magnetic Activated Cell Sorting* (MACS). This research aims to discover the effectivity of cell sorting method, miR-1 and miR-133a profile analysis, as well as expression of transcription factors. Cell percentage was analyzed using *flow cytometer*. RNA concentration was measured by *Nano Drop Spectrophotometer*. MicroRNA profile and transcription factors expression were analyzed using *real-time* PCR. All data obtained was calculated using *Microsoft Excel*. Results show that percentage of C-Kit population is 0% until 0,05%. Percentage of Sca-1 population is 0,5% until 0,25%. MicroRNA profile of CRSC population, C-kit, and Sca-1 are not significantly different. Standart deviation of sampel number 48, 54, and 64 are miR-1 ($n \pm 3,2$; $n \pm 2,6$; $n \pm 2,9$) and miR-133a ($n \pm 2,3$; $n \pm 3,1$; $n \pm 2,2$). Transcription factors expression of CRSC population, C-kit, and Sca-1 are not significantly different. Standart deviation of sample number 48, 54, and 64 are Mef2c ($n \pm 3,1$; $n \pm 1,6$; $n \pm 1,5$), Nkx2.5 ($n \pm 2,7$; $n \pm 2,0$; $n \pm 1,8$), Akt2 ($n \pm 0,7$; $n \pm 1,1$; $n \pm 0,6$), Gata4 ($n \pm 3,1$; $n \pm 1,1$; $n \pm 0,7$). Akt2 showed the highest expression, followed by Gata4, Mef2c, and Nkx2.5. Escalation of miR profile and transcription factor expression is consistent with the cell growth obtained from flask T75 to flask T150.

Key Word: *Cardiac stem cell, miR-1, miR-133a, transcription factor, RT-PCR*