

INTISARI

Latar Belakang : *Glutathione S-transferase M1 (GSTM1)* adalah salah satu gen penting yang berperan dalam detoksifikasi produk stress oksidatif. Delesi dari *GSTM1* dapat dijumpai bervariasi dalam populasi. Terjadinya delesi pada *GSTM1* dapat menyebabkan hilangnya aktivitas katalitik enzim sehingga dapat memicu penurunan kemampuan sel dalam mendetoksifikasi agen xenobiotik dan genotoksik sehingga terjadi peningkatan stress oksidatif berupa *reactive oxygen species (ROS)*. Makrofag diketahui sebagai komponen sistem imun bawaan yang penting bagi tubuh dan menjalankan berbagai fungsi penting. Dalam menjalankan beragam fungsinya, makrofag perlu melakukan migrasi dan proliferasi. Akumulasi stress oksidatif yang berlebihan diketahui dapat berdampak pada disfungsi metabolik. Namun, adanya efek defisiensi gen detoksifikasi seperti *GSTM1* dengan tingkat kemampuan makrofag dalam bermigrasi dan berproliferasi belum pernah diteliti sebelumnya.

Tujuan : Mengkaji pengaruh *knocked out GSTM1* terhadap migrasi dan proliferasi sel makrofag RAW 264.7 sebagai model sel makrofag *in vitro*.

Metode : *Knocked out* gen *GSTM1* pada sel makrofag RAW 264 sebagai model sel makrofag *in vitro* menggunakan metode CRISPR/Cas9. Sel dikultur dan ditumbuhkan pada medium FBS 10%. Migrasi dan proliferasi sel diamati menggunakan metode *scratch wound healing assay*. Persentase *wound closure* dari sel makrofag RAW 264.7 dengan *knocked out* gen *GSTM1* dan tanpa *knocked out* dibandingkan untuk menilai kemampuan migrasi dan proliferasi sel makrofag RAW 264.7 pada kondisi ada maupun tidak adanya *GSTM1*.

Hasil : Tingkat migrasi dan proliferasi sel makrofag RAW 264.7 dinilai melalui hasil rerata persentase penutupan goresan pada hasil metode *scratch wound healing assay*. Ditemukan rerata persentase penutupan goresan pada sel makrofag RAW 264.7 dengan *knocked out GSTM1* tidak bernilai signifikan secara statistik dibandingkan sel makrofag normal. Akan tetapi, terdapat kecenderungan sel makrofag RAW 264.7 dengan *knocked out* memiliki rerata persentase penutupan goresan yang lebih rendah dibandingkan sel normal.

Kesimpulan : Sel makrofag RAW 264.7 dengan *GSTM1 knocked out* hanya memiliki kecenderungan kemampuan migrasi dan proliferasi menjadi lebih rendah dibandingkan dengan sel makrofag RAW 264.7 normal.

Kata kunci : Migrasi ; proliferasi ; *GSTM1 knocked out* ; makrofag RAW 264.7 ; *scratch wound healing assay*

ABSTRACT

Background: Glutathione S-transferase M1 (GSTM1) is an important gene that plays a role in detoxifying oxidative stress products. Deletion of GSTM1 can be found to vary in populations. Deletion of GSTM1 can cause loss of catalytic enzyme activity, which can trigger a decrease in the cell's ability to detoxify xenobiotic and genotoxic agents, resulting in increased oxidative stress in the form of reactive oxygen species (ROS). Macrophages are known to be an important component of the innate immune system for the body and carry out various important functions. In carrying out various functions, macrophages need to migrate and proliferate. Excessive accumulation of oxidative stress is known to impact metabolic dysfunction. However, the effect of deficiency in detoxification genes such as GSTM1 on the ability of macrophages to migrate and proliferate has never been studied before.

Objective: To examine the effect of GSTM1 knockout on the migration and proliferation of RAW 264.7 macrophage cells as an in vitro macrophage cell model.

Methods: Knocked out the GSTM1 gene in RAW 264 macrophage cells as an in vitro macrophage cell model using the CRISPR/Cas9 method. Next, the cells were cultured and grown in 10% FBS medium. Cell migration and proliferation were observed using the scratch wound healing assay method. The percentage of wound closure of RAW 264.7 macrophage cells with GSTM1 gene knockout and without knockout was compared to assess the migration and proliferation ability of RAW 264.7 macrophage cells in the presence or absence of GSTM1.

Results: The level of migration and proliferation of RAW 264.7 macrophage cells was assessed through the results of the average percentage of scratch closure in the results of the scratch Wound Healing Assay method. It was found that the mean percentage of scratch closure in RAW 264.7 macrophage cells with GSTM1 knock out was not statistically significant compared to normal macrophage cells. However, there was a tendency for RAW 264.7 macrophage cells with knock out to have a lower mean percentage of scratch closure compared to normal cells.

Conclusion: RAW 264.7 macrophage cells with GSTM1 knocked out only tend to have lower migration and proliferation abilities compared to normal RAW 264.7 macrophage cells.

Keywords: Migration; proliferation; GSTM1 knocked out; RAW 264.7 macrophage ; scratch wound healing assay