

Aspek hipoksia pada regenerasi jaringan: Ekor cecak rumah (Hemidactylus platyurus) yang memiliki daya regenerasi tinggi sebagai model = Aspects of hypoxia on tissue regeneration: The tail of common House gecko (Hemidactylus platyurus) which has high Regeneration as a model.

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Abstrak

Latar Belakang: Pada proses regenerasi jaringan terjadi ketidakseimbangan antara kebutuhan oksigen dengan suplai oksigen yang menyebabkan jaringan mengalami hipoksia relatif. Keadaan hipoksia diduga memiliki peran penting dalam proses regenerasi jaringan. Pada penelitian ini, dianalisis ekspresi protein dan gen yang berperan mengatasi keadaan hipoksia (HIF-1 α ; dan HIF-2 α), protein yang berperan dalam suplai oksigen (Cygb), protein yang menstimulasi biogenesis mitokondria (PGC-1 α) serta enzim yang mampu menangkal radikal bebas (MnSOD) pada regenerasi jaringan. Regenerasi jaringan ekor pada cecak rumah (Hemidactylus platyurus) digunakan sebagai model dalam penelitian ini, karena merupakan hewan yang paling dekat secara taksonomi dengan mamalia yang memiliki daya regenerasi tinggi, dibandingkan vertebrata lain dengan kemampuan sama. Diharapkan penelitian ini dapat menjadi model dalam mempelajari proses regenerasi dalam upaya pengembangan terapi penyembuhan luka.

Metode: Penelitian bersifat eksperimental deskriptif menggunakan jaringan 30 ekor cecak hasil regenerasi pada hari ke 1; 3; 5; 8; 10; 13; 17; 21; 25; dan 30 setelah autotomi dengan 3 kali pengulangan pada setiap pengamatan. Cecak diperoleh dari lingkungan laboratorium Zoologi Puslit Biologi LIPI Cibinong. Analisis ekspresi gen dilakukan dengan metoda qRT-PCR; analisis ekspresi protein dinilai dengan metoda Western Blot dan imunohistokimia, serta dilakukan analisis struktur histologi jaringan dengan pewarnaan hematoxilin-eosin. Penelitian dilakukan di Laboratorium Departemen Biokimia & Biologi Molekuler FKUI; laboratorium Histologi FKUI, laboratorium PRVKP FKUI, dan laboratorium Patologi Anatomi FKH IPB. Penelitian dalam kurun waktu tahun 2015-2018.

Hasil penelitian: Grafik pertumbuhan jaringan ekor cecak menghasilkan pola pertumbuhan yang lambat pada 13 hari pertama, pertumbuhan yang sangat cepat hari ke 13 sampai ke 21, dan kembali lambat sampai hari ke 30. Pada awal pertumbuhan, ekspresi HIF-1 α ; dan HIF-2 α ; tinggi menunjukkan jaringan dalam keadaan hipoksia. Tingginya ekspresi Cygb selama proses regenerasi jaringan dari awal hingga akhir pengamatan menunjukkan perannya untuk mengakomodasi oksigen selama proses regenerasi berlangsung. Ekspresi PGC-1 α ; yang tinggi di awal proses dan tetap dipertahankan sampai akhir pengamatan berperan untuk mempertahankan agar energi untuk proses regenerasi dapat terpenuhi melalui biogenesis mitokondria. Tingginya ekspresi MnSOD dalam jaringan pada awal regenerasi diduga memiliki peranan yang berkaitan dengan netralisasi senyawa radikal dalam jaringan.

Kesimpulan: HIF 1 α ; HIF 2 α ; Cygb, PGC 1 α ; dan MnSOD masing-masing memiliki peran penting tersendiri dalam proses regenerasi jaringan.

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Background: In tissue regeneration there is an imbalance between oxygen demand and supply causes the tissue to experience relative hypoxia. Hypoxia is thought to have an important role in the tissue

regeneration. This research analyzed the expression of proteins and genes that play role in overcoming hypoxia (HIF-1 and HIF-2); the protein involved in oxygen supply (Cygb); the protein that stimulates mitochondrial biogenesis (PGC-1); and the enzyme counteract free radicals (MnSOD). The regeneration of house gecko's tail (*Hemidactylus platyurus*) was used as a model in this research, because it is the taxonomically closest animal to mammals that have a high capability in regeneration, compared to other vertebrates with the same ability. Hence, this study might become a model in studying tissue regeneration as an effort in developing a wound healing treatment.

Method: The research was performed in a descriptive experimental way, using 30 geckos, having undergone regeneration on day 1; 3; 5; 8; 10; 13; 17; 21; 25; and 30 after autotomy. The experiment used 3 repetitions for each observation. House geckos were obtained from the laboratory building of Zoology Research Center of Indonesian Institute of Sciences (LIPI) Cibinong and its surrounding area. The analysis for gene expression was performed using qRT-PCR method; the analysis for protein expression was undertaken using Western Blot method and immunohistochemistry. In addition to these, the structure analysis for the tissue histology was performed using Haematoxylin and Eosin (H&E) staining method. The study was conducted in the Laboratory of the Department of Molecular Biochemistry & Biology FKUI; Laboratory of the Department of Histology FKUI; laboratory of the Institute of Human Virology & Cancer Biology FKUI; and laboratory of Pathology Anatomy of Animal Medicine, Institute of Agriculture Bogor, in the year 2015-2018.

Results: The graph for the growth of the gecko tail tissue exhibits a slow growth pattern for the first 13 days, followed by a very swift growth between day 13 to 21, returning to slow growth afterwards until day 30. In the early growth stage, the expression of HIF-1 and HIF-2 were increased which showed the tissue was in hypoxia state. HIF protein regulates the contributing to the tissue regeneration process, leading to the increasing growth of tissue with the correlation values of $r = -0,853$ for HIF-1; $r = -0,75$ The substantial expression of Cygb observed throughout the process of tissue regeneration indicates its role in accommodating oxygen in the regeneration process. The expression of PGC-1 was observed to be high in the early stages of the process and remain so until the process ends. This indicates its function in maintaining that sufficient energy provided by mitochondrial biogenesis is available for the regeneration process. The high level of MnSOD expression in the tissue in the early stage of regeneration is thought to relate to its role in neutralizing radicals inside the tissue.

Conclusion: HIF 1, HIF 2, Cygb, PGC 1 and MnSOD have their own important roles in the tissue regeneration process.