Identifikasi copy number variation gen DOK5 pada pasien kanker kolorektal adenokarsinoma sporadik di Makassar Sulawesi Selatan : hubungannya dengan ekspresi gen DOK5 = Identification of dok5 gene copy number variation in sporadic colorectal cancer adenocarcinoma patient in Makassar South Sulawesi correlation with dok5 gene expression / Rinaldy Kusuma

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Abstrak

ABSTRAK
Latar Belakang: Kanker Kolorektal termasuk ke dalam lima besar kanker dengan tingkat insidensi dan mortalitas yang tinggi di Indonesia. Sebesar 20%-25% kejadian kanker kolorektal merupakan faktor keturunan melalui pewarisan mutasi gen-gen high penetrance yang berkontribusi signifikan terhadap pembentukan kanker kolorektal, sedangkan 80%-85% merupakan kanker kolorektal sporadik. Pada kanker kolorektal sporadik mutasi terjadi pada gen-gen low penetrance yang berisiko rendah terhadap pembentukan kanker kolorektal. Identifikasi mutasi pada gen-gen lain yang berpotensi memiliki kontribusi terhadap kanker kolorektal sporadik diperlukan. Prastudi sebelumnya menggunakan metode GWAS telah mengidentifikasi CNV di kromosom 7, 8, 18, dan 20 dari pasien kanker kolorektal sporadik. Penelitian ini bertujuan untuk mengidentifikasi gen dengan CNV di kromosom tersebut dan asosiasinya pada kanker kolorektal sporadik serta hubungan variasi genetik CNV terhadap tingkat ekspresi mRNA gen DOK5.

Metode: Identifikasi gen berdasarkan seleksi dari analisis data CNV dan SNP prastudi GWAS. Sebanyak 70 pasang sampel jaringan kanker kolorektal dan jaringan kolorektal sehat digunakan dalam penelitian ini dengan persetujuan komisi etik serta tujuh jenis sel lestari. Metode Real-Time PCR digunakan dalam analisis CNV gen DOK5 menggunakan DNA dari sampel jaringan maupun sampel sel lestari dan analisis ekspresi mRNA gen DOK5 dari sampel RNA sel lestari.

Hasil: Jumlah Copy Number (CN) gen DOK5 yang tinggi secara signifikan (P =0,01) terdapat pada kelompok usia ≥50 tahun (CN=1,58). Variabel CN gen DOK5 berasosiasi signifikan dengan variabel kelompok usia (P=0,028) dimana lebih dari 50% sampel ≥50 tahun memilik amplifikasi gen DOK5. Pada derajat diferensiasi histopatologi dan jenis kelamin tidak ada perbedaan dan asosiasi dengan CN gen DOK5 secara signifikan (P >0,05). CNV gen DOK5 berkorelasi positif kuat (R=0,890) secara signifikan (P=0,007) dengan tingkat ekspresi mRNA gen DOK5 di beberapa sampel sel lestari dari berbagai kanker.

Kesimpulan: Amplifikasi CNV gen DOK5 terkait dengan kanker kolorektal sporadik pada subyek berusia ≥50 tahun. CNV gen DOK5 yang teramplifikasi berpengaruh terhadap peningkatan ekspresi mRNA gen DOK5 di sel lestari.
<hr><i>ABSTRACT
Background: Colorectal Cancer (CRC) is among top five cancers that have high level of incidence and mortality in Indonesia. About 20%-25% of CRC were familial that inherited a mutation of high penetrance

genes with high predisposing risk of CRC, while the rest of 75%-80% were sporadic CRC. Most of predisposing mutation of genes in sporadic CRC were low penetrance genes with low risk of CRC. Identification of other predisposing genes that have significant and higher risk of sporadic CRC were needed. Our previous study using GWAS has identified a significant genetic variation in the form of CNV in chromosomes 7, 8, 18, dan 20 in sporadic CRC patient. This study aimed to identify the gene with CNV in those chromosomes and its association with sporadic CRC also the effect of CNV on expression level of the gene mRNA.

Method: The gene was identified by selection using analyzed data of CNV and SNP from GWAS study. Seven type of cell lines and 70 matched paired samples of cancerous and normal tissues were used in this study with approval from ethic commission. Real-Time PCR method was used to analyze both DOK5 gene CNV from DNA sample and DOK5 gene expression from RNA sample. DNA was extracted from tissues and cell lines, while RNA extracted from cell lines.

Results: Difference of DOK5 CN was significant in age group (P=0,01) with group ≥50 years old had higher CN of DOK5 gene (CN=1,58). DOK5 CN variable was associated with age group variable (P=0,028) where more than 50% samples of age ≥50 years old showed amplification of DOK5 CN. No significant difference (P >0,05) in CNV of DOK5 gene was detected in subjects if grouped based on their histopathology or gender. In cell lines, CN of DOK5 gene showed significant (P=0,007) and strong positive correlation (R=0,890) with mRNA expression level of DOK5 gene.

Conclusion: DOK5 gene was identified from GWAS CNV data. CNV amplification of DOK5 gene was associated with sporadic colorectal cancer in subject ≥50 years old. Amplified CNV of DOK5 gene affect the increased of DOK5 mRNA expression level in cell lines.</i>