



Figure S1 Marker methylation levels in purified leukocytes. Plotted are β -values of the identified markers in seven cell types isolated from blood samples of six healthy men Reinius et al. (2012). Abbreviated cell types: NE neutrophils, EO eosinophils, MO monocytes, CD4+ T cells, CD8+ T-cells, NK natural killer cells, B CD19+ B lymphocytes.

PubMed search terms:

```
(
  "Early Detection of Cancer"[MeSH Heading]
  AND "DNA Methylation"[MeSH Heading]
  AND "Biological Markers"[MeSH Heading]
)
OR (
  (
    "cancer"[Title/Abstract]
    OR "neoplasms"[Title/Abstract]
    OR "malignancies"[Title/Abstract]
    OR "carcinoma"[Title/Abstract]
  )
  AND (
    "blood"[Title/Abstract]
    OR "leukocytes"[Title/Abstract]
    OR "buffy coat"[Title/Abstract]
  )
  AND (
    (
      "methylation"[Title/Abstract]
      AND (
        "epigenetic"[Title/Abstract]
        OR "epigenome"[Title/Abstract]
      )
    )
    OR "CpG"[Title/Abstract]
    OR "450K"[Title/Abstract]
    OR "Infinium"[Title/Abstract]
  )
)
AND (
  "marker"[Title/Abstract]
  OR "biomarker"[Title/Abstract]
  OR "diagnosis"[Title/Abstract]
  OR "detection"[Title/Abstract]
  OR "screening"[Title/Abstract]
)
)
AND (("2011/01/01"[PDat] : "2016/06/31"[PDat]))
```

Table S1 Leukocyte DNA methylation markers in the literature.

Publication	Cancer	Markers
Wu (2016)	Hepatocellular carcinoma	cg12680131, cg22511877
Roos et al. (2016) ^a	Various	cg02444695, cg26079695, cg27094856, cg21046959
Konecny et al. (2016)	Lung cancer	SHOX2
Ho et al. (2015)	Colorectal adenomas	DNMT3B
Guo et al. (2016)	Esophageal squamous cell carcinoma	RASSF2
Joyce et al. (2015)	Various	IFNG, ICAM1, IL6
Cordero et al. (2015)	Colorectal and breast cancer	<u>No hits for CRC</u>
Dauksa et al. (2014)	Gastric cancer	APC, DAPK1, BCL2, CD44, TNFRSF10C, ACIN1
Yang et al. (2015) ^a	Breast cancer	HYAL2
Winham et al. (2014)	Ovarian cancer	cg04389426, cg05674150
Tahara et al. (2013)	Gastric cancer	SFRP1
You et al. (2013)	Nasopharyngeal carcinoma	CDK10
Hajikhan Mirzaei et al. (2012)	Breast cancer	RHOBTB2
Xu et al. (2013)	Breast cancer	cg17378989, cg10237911, cg22385477, cg08287471, cg13905388
Heyn et al. (2013)	Breast cancer	cg15652666
Langevin et al. (2012)	Head and neck cancer	FGD4, SERPINF1, CIAO1, IL27, HYAL2, PLEKHA6
Bosviel et al. (2012)	Breast cancer	BRCA1
You et al. (2012)	Esophageal squamous cell carcinoma	PTPRO
Marsit et al. (2011)	Bladder cancer	NLRP4, BDKRB1, ATG2B, COX7C, ZNF322B, HIGD2A, TBCA, BRD7, PSME2

^aPerformed adjustment for leukocyte composition

References (Supplement)

- Bosviel, R., Garcia, S., Lavediaux, G., Michard, E., Dravers, M., Kwiatkowski, F., ... Bernard-Gallon, D.J. (2012). BRCA1 Promoter Methylation in Peripheral Blood DNA Was Identified in Sporadic Breast Cancer and Controls. *Cancer Epidemiology*, *36*(3), e177–182. doi:10.1016/j.canep.2012.02.001
- Cordero, F., Ferrero, G., Polidoro, S., Fiorito, G., Campanella, G., Sacerdote, C., ... Naccarati, A. (2015). Differentially Methylated microRNAs in Prediagnostic Samples of Subjects Who Developed Breast Cancer in the European Prospective Investigation into Nutrition and Cancer (EPIC-Italy) Cohort. *Carcinogenesis*, *36*(10), 1144–1153. doi:10.1093/carcin/bgv102
- Dauksa, A., Gulbinas, A., Endzinas, Z., Oldenburg, J., & El-Maarri, O. (2014). DNA Methylation at Selected CpG Sites in Peripheral Blood Leukocytes Is Predictive of Gastric Cancer. *Anticancer Research*, *34*(10), 5381–5388.
- Guo, W., Dong, Z., Cui, J., Guo, Y., Shen, S., Guo, X., & Kuang, G. (2016). Aberrant Hypermethylation of RASSF2 in Tumors and Peripheral Blood DNA as a Biomarker for Malignant Progression and Poor Prognosis of Esophageal Squamous Cell Carcinoma. *Clinical & Experimental Metastasis*, *33*(1), 73–85. doi:10.1007/s10585-015-9759-5
- Hajikhan Mirzaei, Noruzinia, M., Karbassian, H., Shafeghati, Y., Keyhaneh, M., & Bidmeshki-Pour, A. (2012). Evaluation of Methylation Status in the 5'UTR Promoter Region of the DBC2 Gene as a Biomarker in Sporadic Breast Cancer. *Cell J*, *14*(1), 19–24.
- Heyn, H., Carmona, F.J., Gomez, A., Ferreira, H.J., Bell, J.T., Sayols, S., ... Esteller, M. (2013). DNA Methylation Profiling in Breast Cancer Discordant Identical Twins Identifies DOK7 as Novel Epigenetic Biomarker. *Carcinogenesis*, *34*(1), 102–108. doi:10.1093/carcin/bgs321
- Ho, V., Ashbury, J.E., Taylor, S., Vanner, S., & King, W.D. (2015). Gene-Specific DNA Methylation of DNMT3B and MTHFR and Colorectal Adenoma Risk. *Mutation Research/Fundamental and Molecular Mechanisms of Mutagenesis*, *782*, 1–6. doi:10.1016/j.mrfmmm.2015.09.005
- Joyce, B.T., Gao, T., Liu, L., Zheng, Y., Liu, S., Zhang, W., ... Hou, L. (2015). Longitudinal Study of DNA Methylation of Inflammatory Genes and Cancer Risk. *Cancer Epidemiology, Biomarkers & Prevention: A Publication of the American Association for Cancer Research, Cosponsored by the American*

Society of Preventive Oncology, 24(10), 1531–1538. doi:10.1158/1055-9965.EPI-15-0198

- Konecny, M., Markus, J., Waczulikova, I., Dolesova, L., Kozlova, R., Repiska, V., ... Majer, I. (2016). The Value of SHOX2 Methylation Test in Peripheral Blood Samples Used for the Differential Diagnosis of Lung Cancer and Other Lung Disorders. *Neoplasma*. Advanced online publication. doi:10.4149/210_150419N208
- Langevin, S.M., Koestler, D.C., Christensen, B.C., Butler, R.A., Wiencke, J.K., Nelson, H.H., ... Kelsey, K.T. (2012). Peripheral Blood DNA Methylation Profiles Are Indicative of Head and Neck Squamous Cell Carcinoma: An Epigenome-Wide Association Study. *Epigenetics*, 7(3), 291–299. doi:10.4161/epi.7.3.19134
- Marsit, C.J., Koestler, D.C., Christensen, B.C., Karagas, M.R., Houseman, E.A., & Kelsey, K.T. (2011). DNA Methylation Array Analysis Identifies Profiles of Blood-Derived DNA Methylation Associated With Bladder Cancer. *Journal of Clinical Oncology*, 29(9), 1133–1139. doi:10.1200/JCO.2010.31.3577
- Reinius, L.E., Acevedo, N., Joerink, M., Pershagen, G., Dahlén, S.-E., Greco, D., ... Kere, J. (2012). Differential DNA Methylation in Purified Human Blood Cells: Implications for Cell Lineage and Studies on Disease Susceptibility. *PloS One*, 7(7), e41361. doi:10.1371/journal.pone.0041361
- Roos, L., van Dongen, Jenny, Bell, C.G., Burri, A., Deloukas, P., Boomsma, D.I., ... Bell, J.T. (2016). Integrative DNA Methylation Analysis of Pan-Cancer Biomarkers in Cancer Discordant Monozygotic Twin-Pairs. *Clinical Epigenetics*, 8(1). doi:10.1186/s13148-016-0172-y
- Tahara, T., Maegawa, S., Chung, W., Garriga, J., Jelinek, J., Estécio, M.R.H., ... Issa, J.-P.J. (2013). Examination of Whole Blood DNA Methylation as a Potential Risk Marker for Gastric Cancer. *Cancer Prevention Research (Philadelphia, Pa.)*, 6(10), 1093–1100. doi:10.1158/1940-6207.CAPR-13-0034
- Winham, S.J., Armasu, S.M., Cicek, M.S., Larson, M.C., Cunningham, J.M., Kalli, K.R., ... Goode, E.L. (2014). Genome-Wide Investigation of Regional Blood-Based DNA Methylation Adjusted for Complete Blood Counts Implicates BNC2 in Ovarian Cancer: Ovarian Cancer DNA Methylation Study Implicates BNC2. *Genetic Epidemiology*, 38(5), 457–466. doi:10.1002/gepi.21815
- Wu, H.-C. (2016). Blood DNA Methylation Markers in Prospectively Identified Hepatocellular Carcinoma Cases and Controls from Taiwan. *World Journal of Hepatology*, 8(5), 301. doi:10.4254/wjh.v8.i5.301

- Xu, Z., Bolick, S.C.E., DeRoo, L.A., Weinberg, C.R., Sandler, D.P., & Taylor, J.A. (2013). Epigenome-Wide Association Study of Breast Cancer Using Prospectively Collected Sister Study Samples. *Journal of the National Cancer Institute*, *105*(10), 694–700. doi:10.1093/jnci/djt045
- Yang, R., Pfütze, K., Zucknick, M., Sutter, C., Wappenschmidt, B., Marme, F., ... Burwinkel, B. (2015). DNA Methylation Array Analyses Identified Breast Cancer-Associated HYAL2 Methylation in Peripheral Blood. *International Journal of Cancer*, *136*(8), 1845–1855. doi:10.1002/ijc.29205
- You, Y., Yang, W., Wang, Z., Zhu, H., Li, H., Lin, C., & Ran, Y. (2013). Promoter Hypermethylation Contributes to the Frequent Suppression of the CDK10 Gene in Human Nasopharyngeal Carcinomas. *Cellular Oncology (Dordrecht)*, *36*(4), 323–331. doi:10.1007/s13402-013-0137-5
- You, Y.-J., Chen, Y.-P., Zheng, X.-X., Meltzer, S.J., & Zhang, H. (2012). Aberrant Methylation of the PTPRO Gene in Peripheral Blood as a Potential Biomarker in Esophageal Squamous Cell Carcinoma Patients. *Cancer Letters*, *315*(2), 138–144. doi:10.1016/j.canlet.2011.08.032