Supplemental Table 1. Studies in humans that have examined the association between folate/folic acid intake or blood folate levels and DNA methylation

	Exposure Timing	Exposure and Duration/Sampling	Description	Subjects	Assay, Loci Included in Assay, Tissue Assayed for Methylation Status and Genotype Interaction	Result/Outcome	References
Clinical Trials							
Cancer Studies							
1.	Adults with adenomas and colorectal cancer	Folic acid supplementation trial-10mg/d 6 mo Samples: Baseline, 6mo and 3 mo after end of trial	A randomized placebo controlled trial of patients with colorectal cancer or adenoma given 6 mo of folic acid supplementation followed by a 3 mo washout.	N = 32 (age 49-82 y) N = 12 patients with adenoma (N = 5 given folic acid N = 7 placebo) N = 12 patients with colorectal cancer (N = 4 folic acid; N = 6 placebo) N= 8 patients getting colonoscopy without abnormality (not included in the folic acid supplementation)	Assay: [^{3}H] methyl acceptance Loci Included: Unmethylated CpG sites on double stranded DNA Tissue: colon Genotype Interaction: NA	Hypomethylation in cases normal appearing colorectal mucosa vs. controls ($p<0.005$), carcinoma vs. adenoma ($p<0.005$). DNA methylation increased after 6 mo of folic acid treatment in patient's colorectal mucosa ($p < 0.002$). Three months after the discontinuation of treatment DNA methylation decreased compared to 6 mo supplementation ($p < 0.05$). No significant changes were observed among those in the placebo group ($p = 0.4$)	Cravo et al. 1994 (30)

	Exposure Timing	Exposure and Duration/Sampling	Description	Subjects	Assay, Loci Included in Assay, Tissue Assayed for Methylation Status and Genotype Interaction	Result/Outcome	References
2.	Adults with colorectal adenomas	Folic acid supplementation trial of folate- 5mg/d, 3 mo Samples: Baseline, 3, 6 mo	Prospective crossover supplementation trial of patients with colorectal adenoma, given 5mg/d folic acid for three months then placebo for 3 mo (or vice versa).	N = 20 patients Two groups: Folate/placebo n=10 (age 58 +/- 11 y) Placebo/folate n=10 (age 56 +/- 11 y)	Assay: [^{3}H] methyl acceptance Loci Included: Unmethylated CpG sites on double stranded DNA Tissue: Colon Genotype Interaction: NA \	At baseline no association with folate intake and DNA methylation. Inverse association of caloric ($p = 0.03$) and fat ($p = 0.05$) intake with DNA methylation. No association of serum folate at baseline or 3 mo on DNA methylation. Increase in DNA methylation (decreased hypomethylation) only among those with single polyps after 3 mo of supplementation ($p < 0.05$).	Cravo et al. 1998 (118)
3.	Adults with colorectal adenoma	Controlled feeding trial of folic acid- 10w 400µg/d Samples: Baseline, 10 w	Double blind placebo controlled study of patients with a confirmed colorectal adenoma, randomized to folic acid treatment or placebo groups.	N=33 N= 17 folic acid treatment N=16 placebo (mean age 64 y)	Assay: [^{3}H] methyl acceptance Loci Included: Unmethylated CpG sites on double stranded DNA Tissue: Leukocytes and colorectal mucosa Genotype Interaction: NA	There was a 31% increase in DNA methylation in leukocytes ($p =0.05$) and 25% increase in colon ($p=0.09$).	Pufulete et al. 2005 (119)

	Exposure Timing	Exposure and Duration/Sampling	Description	Subjects	Assay, Loci Included in Assay, Tissue Assayed for Methylation Status and Genotype Interaction	Result/Outcome	References
4.	Adult with one previous colorectal adenoma	Controlled feeding trial of folate- 1 mg folic acid for 3 years. Samples at baseline, 3 y	Study of folic acid and/or aspirin for the prevention of colon cancer. Determination of the association of CpG island methylation with blood folate and other factors in normal colon.	N=389 patients N= 198 given 1 mg/d folic acid N=190 placebo (age 30-78 y)	Assay: Bisulfite Pyrosequencing Loci Included: *ERα* (3 CpG sites), *SFRP1* (5 CpG sites) Tissue: Normal colorectal mucosa Genotype Interaction: NA	There was no association with 1mg/d folic acid treatment for 3 years with either *ERα* or *SFRP1* methylation in either segment of the colon. In multivariate models RBC level was associated with an increasing trend of methylation at both *ERα* (Q1 to Q5 = 1% change in mean 10.03% to 11.3%, p=0.03) and *SFRP1* (Q1to Q5 1.8% change in mean 21.2% to 23.0% p=0.01). Plasma folate was not significantly associated. *ERα* and *SFRP1* methylation were both significantly associated with age and race. *ERα* was significantly associated with multivitamin use (supplements did not contain folic acid).	Wallace et al. 2010 (49)

	Exposure Timing	Exposure and Duration/Sampling	Description	Subjects	Assay, Loci Included in Assay, Tissue Assayed for Methylation Status and Genotype Interaction	Result/Outcome	References
Patients without cancer							
5.	Adults with hyperhomocystinemia and uremia	Controlled feeding trial of folate- 8 w 15 mg/d Samples: Baseline, after 2 month washout and after 8 wk folate treatment	Treatment of men with hyperhomocystinemia and uremia on dialysis with a 2 month folate washout followed by 15 mg oral methyltetrahydrofolate/d for 8 wk.	N=32 patients (Men mean age 61.3y, range 39-68 y) N =11 controls (Men mean age 58.7y range 42-65 y)	Assay: Southern blot probed with satellite 2, Digest Msp1 and Hpa II, cytosine extension and bisulfite sequencing of *SYBL1* promoter. Loci Included: 2 Global DNA methylation assays and bisulfite sequencing showing the methylation status of every CpG near the promoter of *SYBL1* (37 sites) Tissue: Isolated peripheral blood mononuclear cells Genotype Interaction: NA	Patients had less global methylation than controls by cytosine extension (p=0.0006) and by MspI/HpaII digest. Global hypomethylation correlated significantly with plasma homocysteine concentration (r=0.49 p=0.004). Some patients showed aberrant methylation and re-expression of *SYBL1* (3 of 32). *H19* in 3 patients with high homocysteine was aberrant methylation and bi-allelic expression return to a normal pattern after treatment. (Blood folate not reported)	Ingrosso et al. 2003 (140)

	Exposure Timing	Exposure and Duration/Sampling	Description	Subjects	Assay, Loci Included in Assay, Tissue Assayed for Methylation Status and Genotype Interaction	Result/Outcome	References
Healthy Adults							
6.	Healthy adults	Controlled feeding trial of folate Samples: Days 5, 41, 69 and 91	A controlled feeding trial of US based women. 91 day trial. Baseline-195 µg/d intake 5 day; day 6-41 of a low folate diet 56 µg/d folic acid, day 42-69 111 µg/d, day 70-80 286 µg/d, days 81-91 516 µg/d folic acid.	N=8 Postmenopausal women (49-63 y)	Assay: [^{3}H] methyl acceptance Loci Included: Unmethylated CpG sites on double stranded DNA Tissue: Lymphocytes Genotype Interaction: NA	Significant increases in DNA methylation were seen at day 69 after the 111 µg/d repletion ($p<0.001$). Folate depletion also increased the number of DNA breaks and uracil misincorporation.	Jacob et al.1998 (136)
7.	Healthy adults	Controlled feeding trial of folate- 7 wk repletion with 200 or 415 µg folate/d. Samples: Baseline, 7 and 14 weeks	A controlled feeding study of US based women aged 60-85 years. Consumed 118 µg folate/d for 7 wk, followed by a 7 wk repletion with 200 or 415 µg folate/d.	Elderly women (60-85 y) N=33	Assay: [^{3}H] methyl acceptance Loci Included: Unmethylated CpG sites on double stranded DNA Tissue: Leukocytes Genotype Interaction: NA	Folate depletion resulted in significantly decreased methylation (decreased methyl incorporation $p = 0.0025$) Repletion did not result in increased methylation. At baseline there was a correlation between homocysteine and DNA methylation ($p = 0.04$). At no other time was there a correlation between DNA methylation and serum or RBC folate or plasma homocysteine.	Rampersaud et al.2000 (135)

	Exposure Timing	Exposure and Duration/Sampling	Description	Subjects	Assay, Loci Included in Assay, Tissue Assayed for Methylation Status and Genotype Interaction	Result/Outcome	References
8.	Healthy adults	Controlled feeding trial of folate-7 weeks 400 µg DFE/d. Samples: Baseline, 7 and 14 weeks	Controlled feeding study of women placed on diets of 7 week 115 ug DFE/d; 7 weeks 400 ug DFE/d.	Women (20-30 y) *MTHFR* TT n= 19 *MTHFR* CC n=22	Assay: Liquid chromatography tandem mass spectrometry and [^{3}H] methyl acceptance Loci Included: All mC and unmethylated CpG sites Tissue: Leukocytes Genotype Interaction: *MTHFR*	During depletion there were decreases in DNA methylation ($p = 0.08$). During repletion there were increases in methylation only in the *MTFHR* TT group ($p < 0.05$). Homocysteine was associated with DNA methylation by [^{3}H] acceptance for both TT and CC groups $p < 0.001$)	Shelnutt et al. 2004 (137)
9.	Healthy adults	Controlled feeding trial of folate-7 w at 400 or 800µg/d folic acid supplementation. Samples: Baseline, 7 and 14 weeks	Women enrolled in a 14 week feeding trial. 7 week folate restricted (135 ug/d DFE) followed by 7 weeks at 400 or 800ug folic acid supplementation. All subjects with the MTHFR CC genotype.	Caucasian women n=14 African American women n=14 Aged 18-45 y	Assay: Cytosine extension Loci Included: Unmethylated CpG sites Tissue: Blood Genotype Interaction: *MTHFR*	Global DNA methylation levels did not significantly vary across the trial or by race.	Axume et al. 2007 (138)

	Exposure Timing	Exposure and Duration/Sampling	Description	Subjects	Assay, Loci Included in Assay, Tissue Assayed for Methylation Status and Genotype Interaction	Result/Outcome	References
Observational Studies **Cancer Studies**							
10.	Adults referred for colonoscopy with and without cancer	Folate Status Blood folates and a food frequency questionnaire in adults Samples: At the time of referral for colonoscopy	Hospital-based study of person referred for colonoscopy to examine the association of folate status, DNA hypomethylation, and *MTHFR* on the risk of colon cancer.	N=35 patients with adenoma N=28 patients with colon cancer N=76 controls (age 30-90y)	Assay: [^{3}H] methyl acceptance Loci Included: Unmethylated CpG sites Tissue: Leukocytes and colonic mucosa Genotype Interaction: *MTHFR, CBS, MS*	Decreased risk for cancer among those with higher folate status ($p = 0.01$). Cancer and adenoma patients had lower serum folate ($p=0.01$) and global hypomethylation ($p=0.08$). Adenoma patients had global hypomethylation ($p=0.009$). Global hypomethylation in the leukocytes and colon was associated with increased risk of adenoma ($p = 0.02$ and $p = 0.01$) and supplement use did not alter the trend.	Pufulete et al. 2003 (121)
11.	Women with cervical cancer Pre-and Post fortification	Pre-and Post fortification Samples: At the time of biopsy or curettage	Hospital-based study of global methylation levels in cervical cancer lesions pre-(1990-92) and post fortification (2000-02) at the Department of Pathology and the University of Alabama to determine if mandatory fortification was associated with changes in global DNA methylation in cervical cancer.	Pre-fortification N=152 Post fortification N=172	Assay: Immunohistochemical staining with monoclonal antibodies for 5 methyl cytosine (5-mc) Loci Included: 5-mc Tissue: Cervical cancer Genotype Interaction: NA	Global hypomethylation level did not change between pre- and post-fortification. Global hypomethylation was associated with severity of lesion (more hypomethylation in higher grade lesions $p < 0.001$).	Piyathilake et al. 2006 (122)

	Exposure Timing	Exposure and Duration/Sampling	Description	Subjects	Assay, Loci Included in Assay, Tissue Assayed for Methylation Status and Genotype Interaction	Result/Outcome	References
12.	Adults with and without head and neck squamous cell carcinoma (HNSCC)	Food frequency questionnaire Samples: After diagnosis	Population-based, matched case-control study of HNSCC in the Boston Metropolitan area to determine if DNA methylation in whole blood was a proxy for DNA methylation in a tumor tissue, associated with cancer risk and assess modification by other environmental risk factors.	Cases n= 278 (mean age 60.1y +/- 11.7 SD) Controls n= 526 (mean age 61.0y +/- 11.5 SD)	Assay: *LRE1* fluorescence assay Loci Included: *LRE1* element Tissue: Whole blood Genotype Interaction: *MTHFR*	Hypomethylation in the blood of the repetitive element *LRE1* was associated with an increased risk of HNSCC (aOR 1.6 95% CI 0.1-2.4). Among cases only, lowest tercile dietary folate (p=0.06) and *MTHFR* T allele (p=0.04) trended towards association with lower methylation levels.	Hsiung et al. 2007 (123)
13.	Adults with bladder cancer	Food frequency questionnaire Samples: After diagnosis before treatment	Hospital-based matched case-control study of bladder cancer in Spain to determine if DNA hypomethylation is associated with increased risk of bladder cancer.	N=775 cases (mean aged 68 y) N=397 controls (mean aged 65 y)	Assay: High-performance capillary electrophoresis and Hpa II digestion. Loci Included: Hpa II sites (proxy for global methylation) Tissue: Leukocytes Genotype Interaction: NA	Global methylation was not associated with reported folate intake. Global DNA methylation in leukocytes was lower in cases than controls (P<0.001). Global DNA methylation in controls was not associated with any of the patient characteristics. There was an interaction of smoking, global DNA methylation and cancer risk. No associations were found for genotypes that survived correction for multiple testing.	Moore et al. 2008 (124)

	Exposure Timing	Exposure and Duration/Sampling	Description	Subjects	Assay, Loci Included in Assay, Tissue Assayed for Methylation Status and Genotype Interaction	Result/Outcome	References
14.	Women with cervical cancer	Plasma folate	Prospective follow-up study of women diagnosed with abnormal cervical cells.	N = 376 women (age 19-50 y) N= 103 with ≥CIN 2 (cases) N = 273 ≤ CIN1 (non-cases)	Assay: Bisulfite pyrosequencing Loci Included: *LINE-1* methylation level Tissue: Peripheral blood mononuclear cells (PBMC) Genotype Interaction: *MTHFR, CBS, MTR, MTRR, SHMT1, CTH SLC19A1, TYMS, GSTM1*	The highest tertile LINE-1 methylation in PBMC were 56% less likely to be diagnosed with ≥CIN 2 (OR 0.44 95%CI 0.24-0.83 p = 0.011). High level "supraphysiological" plasma folate and sufficient B12 was associated with increased LINE-1 methylation in PBMC compared to women with lower folate and B12 among non-cases (OR 3.92 95% CI 1.06-14.52 p = 0.04).	Piyathilake et al. 2011 (125)
15.	Women with breast cancer	Folate Intake Samples: At diagnosis	Prospective cohort of breast cancer survival of HMO members (Kaiser Permanente). Analysis of primary breast tumors to determine if DNA methylation patterns are associated with patient characteristics or with outcome.	N= 162 Women with breast tumors (mean age 59.2y range 30-91 y)	Assay: Illumina bead array Loci Included: 1413 CpG sites in 773 genes Tissue: Primary breast tumors Genotype Interaction: NA	Among the tumors there were eight clusters of DNA methylation patterns as determined by unsupervised clustering. Dietary folate was associated with methylation class membership (Wald p<0.001) as were age, tumor size, and alcohol intake (p < 0.001).	Christensen et al. 2010 (131)

	Exposure Timing	Exposure and Duration/Sampling	Description	Subjects	Assay, Loci Included in Assay, Tissue Assayed for Methylation Status and Genotype Interaction	Result/Outcome	References
16.	Adults with colorectal cancer	Plasma folate and homocysteine measured Samples: At diagnosis	Hospital-based case-control study on newly diagnosed colorectal cancer to determine if folate related gene or if DNA methylation at specific genetic loci in blood cells are associated with colorectal cancer as a tool for non-invasive tumor detection.	N=67 colorectal cancer patients (mean age 62 +/-12) N=53 controls (hospital based) (age 59 +/-16)	Assay: Methylation specific PCR Loci Included: Fully methylated or unmethylated CpG covered by the primers for *p16^{in4A}, p73, hMLH1* Tissue: White blood cells Genotype Interaction: *MTHFR, MTR*	Individuals with the *MTHFR* TT genotype had reduced colorectal cancer risk compared to CC (OR 0.2; 95%CI 0.07- 0.6 p = 0.005) and lower plasma folate (p<0.05). High plasma folate among cases (≥6.7 ng/ml *vs.* <4.1 ng/ml) was associated with an increased methylation at *p73* (p = 0.023) but not *p16^{in4A}* or *hMLH1*.	Kim et al. 2011 (132)
Patients without cancer							
17.	Adults referred to cardiovascular surgery center.	RBC folate, plasma folate, B12 and homocysteine measured in blood sample. Samples: After referral	Study of person referred to Institute of Cardiovascular Surgery of Verona (Italy) for atherosclerosis or other reason to determine the effect of folate status on DNA methylation and the relationship between folate status and *MTHFR* CC or TT genotype	*MTHFR* N (CC) = 187 (mean age 60 y +/- SD 10 N (TT) = 105 (mean age 60 y +/- SD 11)	Assay: Liquid chromatography tandem mass spectrometry Loci Included: ng methylated cytosines per microgram DNA Tissue: Whole blood Genotype Interaction: *MTHFR*	Subjects with the *MTHFR* TT genotype had lower methylation compared to CC (p < 0.001). *MTHFR* TT subjects with lower than the mean folate (plasma folate <= 12 nmol/l or RBC <= 1.13 nmol/folate/g Hb) had lower levels of DNA methylation compared to higher plasma folate (both p < 0.05).	Friso et al. 2002 (139)

	Exposure Timing	Exposure and Duration/Sampling	Description	Subjects	Assay, Loci Included in Assay, Tissue Assayed for Methylation Status and Genotype Interaction	Result/Outcome	References
18.	Adults referred for colonoscopy	Serum and erythrocyte folates measured Samples: At the time of referral for colonoscopy	Hospital-based study of patients referred for colonoscopy without abnormality during colonoscopy to assess the effect of folate status on DNA methylation in the colon.	N=68 subjects (n=33 men, n=35 women) (aged 36-78 y)	Assay: [^{3}H] methyl acceptance Loci Included: Unmethylated CpG sites Tissue: Colon Genotype Interaction: *MTHFR, MS, CBS*	Adjusted analysis found a non-significant trend between lower methylation and lower serum folate (p=0.07, adjusted p=0.01 unadjusted) and erythrocyte folate (p=0.08 adjusted, p=0.03 unadjusted). MS genotype was significantly associated with DNA methylation (trend AA<AG<GG p=0.05). Increased hypomethylation was found for smokers (p = 0.05), increased age (p = 0.03) and increased homocysteine (p= 0.01).	Pufulete et al. 2005 (126)

	Exposure Timing	Exposure and Duration/Sampling	Description	Subjects	Assay, Loci Included in Assay, Tissue Assayed for Methylation Status and Genotype Interaction	Result/Outcome	References
Prenatal exposures							
19.	Periconceptional maternal folic acid use – 400µg/d in ~17 month old children	S-adenosylmethionine and S-adenosylhomocysteine in blood in mothers and children and self reported periconceptional folic acid use was reported at enrollment. Samples: At office visit.	Cross-sectional study of mothers with children between 12 and 18 months of age enrolled at Public Health centers in Rotterdam, Netherlands as healthy controls for another study (HAVEN).	Children (age 12-18 mo, mean 17 mo) N=86 (mothers periconceptional use of folic acid) N=34 (mothers did not take folic acid) (mothers' mean age 32 y SE 0.8).	Assay: Mass spectrometry Loci Included: *IGF2* DMR, 5 CpG sites within the *IGF2* DMR Tissue: Whole blood Genotype Interaction: NA	Maternal use of folic acid associated with a 4.5% increase in I*GF2* DMR methylation (49.5% vs. 47.4% p=0.014). I*GF2* DMR methylation in the child was also associated with maternal S-adenosylmethionine level and inversely with birth weight (-1.7% methylation per SD birth weight p = 0.034).	Steegers-Theunissen RP et. al 2009 (160)
20.	Prenatal vitamin use assessed in the third trimester	Cord blood serum folate, cord plasma homocysteine, maternal folic acid supplement use. Samples: At delivery	Hospital-based study in the UK examining the relationship between folic acid supplement use in pregnancy, blood folate and homocysteine, and repetitive element methylation in cord blood.	Women delivering term babies Mothers n=24 (mean age 29.4 y +/- 7.0 SD)	Assay: Pyrosequencing Loci Included: *LINE-1* elements Tissue: Cord blood Genotype Interaction: NA	No association of serum folate or folic acid intake with *LINE-1* methylation. Association of increasing maternal plasma homocysteine and decreasing percent methylation of *LINE-1* (r = -0.688 p=0.001). Plasma homocysteine was the only significant predictor in a multivariable stepwise regression model.	Fryer al. 2009 (162)

	Exposure Timing	Exposure and Duration/Sampling	Description	Subjects	Assay, Loci Included in Assay, Tissue Assayed for Methylation Status and Genotype Interaction	Result/Outcome	References
21.	Prenatal vitamin use assessed in the third trimester	Cord blood serum folate, cord plasma homocysteine, maternal folic acid supplement use Samples: At delivery	Hospital-based study in the UK examining the relationship between folic acid supplement use in pregnancy, blood folate and homocysteine, and DNA methylation patterns in cord blood. A subset of Fryer al. 2009 (162) selected for a range of *LINE-1* methylation levels.	Women delivering term babies Mothers n = 12 (age 19-40 y)	Assay: Illumina Bead Array 27,578 sites and Pyrosequencing Loci Included: Percent DNA methylation at those sites that met inclusion criteria (7,259 sites in 473 genes) and *LINE-1* elements Tissue: Cord blood Genotype Interaction: NA	CpG site within CpG islands showed hypomethylation. DNA methylation patterns formed two clusters. Significant correlation between methylation patterns, plasma homocysteine ($p = 0.038$), LINE-1 methylation ($p = 0.028$) and birth weight percentiles ($p = 0.019$). No significant associations between folic acid supplement use or serum folate and DNA methylation pattern cluster.	Fryer et al. 2011 (163)
22.	Adult women and cord blood	Maternal and cord blood serum folate Samples: At delivery	Hospital-based cross-sectional study of Chinese women delivering infants to determine the relationship between DNA methylation in maternal blood and cord blood and folate and B12.	99 newborns (52 boys, 47 girls) (mothers age 19-37y)	Assay: Real time Methylation Specific PCR Loci Included: *IGF2* promoter 2 and 3 Tissue: Cord blood Genotype Interaction: NA	No association of serum or cord blood folate levels with *IGF2* promoter methylation level. Maternal and cord blood B12 was associated with P3 methylation ($P<0.01$ and $P< 0.001$). P2 was also associated with maternal weight gain and exposure to smoking ($p=0.03$ and $p=0.02$).	Ba et al. 2011(164)

	Exposure Timing	Exposure and Duration/Sampling	Description	Subjects	Assay, Loci Included in Assay, Tissue Assayed for Methylation Status and Genotype Interaction	Result/Outcome	References
23.	Women's folate assessed within one week of pregnancy termination	Maternal serum folate Samples: Fetal tissue 18-28 weeks gestation Maternal serum folate ~1 week post-termination	Hospital based study in China examining the correlation of women's serum folate with fetal methylation comparing the 5mC in various tissues from fetus with NTDs to normal fetuses.	Terminated fetuses between 18-28 weeks gestation with NTD's and age and sex-matched controls NTDs n= 20 (18 spina bifida 2 anencephalus) Controls n=20	Assay: HPLC Loci Included: All mC Tissue: Fetal-heart, skin, brain, kidney, lung, and liver Genotype Interaction: NA	Hypomethylation in brain of fetuses with NTDs compared to controls ($p<0.01$). Lower mean serum folate in mother with NTD affected pregnancies ($p<0.01$). Correlation of maternal serum folate and 5mC in brain from NTD fetuses ($r=0.6$).	Chang et al. 2011(161)

	Exposure Timing	Exposure and Duration/Sampling	Description	Subjects	Assay, Loci Included in Assay, Tissue Assayed for Methylation Status and Genotype Interaction	Result/Outcome	References
24.	Pregnant women's prenatal vitamin use before and during pregnancy was assessed by self administered questionnaire between 19 -41 weeks gestation.	Maternal report of vitamin use before and during pregnancy Samples: At delivery	Hospital based study of pregnant women (NEST – Newborn Epigenetics Study) of women recuited from prenatal clinics to determine if periconeptional folic acid use is associated with DNA methylation in the cord blood of newborns.	Mothers at the time of delivery n = 438 (mean age 29, range 18-49 years)	Assay: Pyrosequencing Loci Included: *IGF2* (3 CpGs) and *H19* (4 CpGs) DMR Tissue: Cord blood Genotype Interaction: NA	No association of reported maternal folic acid/multivitamin use and DNA methylation level at *IGF2*. Folic acid intake before and during pregnancy decreased DNA methylation at *H19* DMR (2.8%, p =0.03 and 4.9% p = 0.04 respectively). Decreases in DNA methylation at *H19* were most pronounced in male infants (p=0.01).	Hoyo et al. 2011 (165)

	Exposure Timing	Exposure and Duration/Sampling	Description	Subjects	Assay, Loci Included in Assay, Tissue Assayed for Methylation Status and Genotype Interaction	Result/Outcome	References
25.	Embryonic exposure to starvation	Starvation Periconceptional exposure = conceived during the height of the famine. Late gestation exposure= born during the famine. Samples: At the time of recruitment.	Study of the *IGF2* DMR in individuals prenatally exposed to famine as part of the Dutch Hunger winter after WW II compared to their sex matched unexposed siblings to determine the association of starvation during pregnancy and DNA methylation levels 6 decades later.	Total subject 244 Periconceptionally exposed N= 60 and sex matched unexposed sibling controls (46.7% male; mean age 58.1 years at the time of blood draw) Late gestation exposed N= 62 and sex matched unexposed sibling controls (45.2% male mean age 58.8 years), N=122 Control mean age 57.1 y.	Assay: Mass spectrometry Loci Included: *IGF2* DMR (5 CpGs) Tissue: Whole blood Genotype Interaction: NA	In periconceptionally exposed persons DNA methylation level was lower at the *IGF2* DMR (average change -5.2% $p > 0.001$). There was no significant change in those exposed during late gestation.	Heijmans et al. 2008 (159)

	Exposure Timing	Exposure and Duration/Sampling	Description	Subjects	Assay, Loci Included in Assay, Tissue Assayed for Methylation Status and Genotype Interaction	Result/Outcome	References
26.	Embryonic exposure to starvation	Starvation Periconceptional exposure = conceived during the height of the famine. Late gestation exposure= born during the famine. Samples: At the time of recruitment.	Study of 15 loci in individuals prenatally exposed to famine as part of the Dutch Hunger winter after WW II compared to their sex matched unexposed siblings to determine the association of starvation during pregnancy and DNA methylation levels 6 decades later.	Total subject 244 Periconceptionally exposed N= 60 and sex matched unexposed sibling controls (28 male pairs and 32 female pairs, mean age 58.1 years at the time of blood draw) Late gestation exposed N= 62 and sex matched unexposed sibling controls (28 male pairs and 34 female pairs, mean age 58.4 years).	Assay: Mass spectrometry Loci Included: *IL10, GNASAS, INSIGF, LEP, MEG3, ABCA1, ABCA1 meth, KCNQ10T1, GRB10, GNASAB, APOC1, IGF2R, FTO, CRH, TNF, NR3C1* Tissue: Whole blood Genotype Interaction: NA	In periconceptionally exposure, DNA methylation level was lower at *INSIGF* and higher at *IL10, LEP, ABCA1, GNASAS* and *MEG3* compared to sex matched siblings (all P<0.001). An interaction with sex was found for *INSIGF, LEP* and *GNASAS*. Exposure to famine at later gestational ages resulted only in changes to *GNASAS* (P< 0.001). In men only, *LEP* was also significantly associated for later exposures.	Tobi et al. 2009 (153)

Abbreviations: NA- Not applicable, DFE- dietary folate equivalents; DMR- differentially methylated region; mC-methylated cytosine; CIN- Cervical intraepithelial neoplasia, *LINE*- long interspersed elements, *LREI*- a type of *LINE*.

Online Supporting Material