Supplementary Methods

Assessment of aspirin use

We have previously published a detailed description of the collection of information on aspirin use and the definition of regular aspirin use in these cohorts (1). In the Nurses' Health Study (NHS), aspirin use was first assessed in 1980 and every 2 years thereafter, except in 1986. The NHS participants were asked whether they took aspirin in most weeks, the number of tablets taken per week, and years of aspirin usage. In the NHS, regular aspirin users were defined as women who reported consumption of 2 or more aspirin tablets per week and nonusers as women who used fewer than 2 tablets per week or no aspirin. In the Health Professionals Follow-up Study (HPFS), in 1986 and every 2 years thereafter, participants were asked whether they used aspirin 2 or more times per week. In the HPFS, regular aspirin users were defined as men who reported consumption of aspirin at least 2 times per week and nonusers as men who consumed fewer than 2 times per week or no aspirin.

Statistical analysis to assess an interaction of tumor *MIR21* expression level and regular aspirin use after diagnosis in survival analysis

All statistical analyses were conducted using SAS (version 9.3, SAS Institute, Cary, NC) and all *P* values were two-sided. For a secondary analysis of the interactive associations of tumor *MIR21* expression level and regular aspirin use after diagnosis in colorectal cancer-specific survival and overall survival, we adjusted two-sided α level to 0.025 (= 0.05/2) by

simple Bonferroni correction for multiple hypothesis testing. To minimize ascertainment bias in aspirin use data collection after cancer diagnosis, we performed this secondary analysis limited to patients with stage I, II, or III disease. The statistical interaction was assessed by the Wald test on the cross-product term of tumor *MIR21* expression level (ordinal quartile categories [1 to 4]) and regular aspirin use after diagnosis (regular aspirin use vs. no aspirin use) variables in a Cox proportional hazards regression model.

To control for confounders, we used multivariable Cox proportional hazards regression models. In addition to the variables on tumor *MIR21* expression level and regular aspirin use after diagnosis, the multivariable model initially included sex, age at diagnosis (continuous), year of diagnosis (continuous), family history of colorectal cancer in a first-degree relative (present vs. absent), tumor location (proximal colon vs. distal colon vs. rectum), tumor differentiation (well/moderate vs. poor), MSI (high vs. MSI-low/MSS), CIMP (high vs. low/negative), *KRAS* (mutant vs. wild-type), *BRAF* (mutant vs. wild-type), *PIK3CA* (mutant vs. wild-type), and tumor LINE-1 methylation level (continuous). A single analysis model could estimate the effect of regular aspirin use after diagnosis in each stratum of tumor *MIR21* expression level, using a reparameterization of the interaction term (of tumor *MIR21* expression level and regular aspirin use after diagnosis) as previously described (2). A backward elimination was carried out with $P = 0.05$ as a threshold, to select variables for the final model. For cases with missing information on LINE-1 methylation level, we assigned a separate indicator variable. For cases with missing information in any of the categorical covariates, we included these cases in the majority category of a given covariate to minimize the number of variables in multivariable Cox models.

References

1. Chan AT, Ogino S, Fuchs CS. Aspirin and the risk of colorectal cancer in relation to the expression of COX-2. N Engl J Med 2007;356:2131-42.

2. Liao X, Lochhead P, Nishihara R, Morikawa T, Kuchiba A, Yamauchi M, et al. Aspirin use, tumor PIK3CA mutation, and colorectal-cancer survival. N Engl J Med 2012;367:1596-606.