Plasma ghrelin is probably not a useful biomarker for risk prediction or early detection of colorectal cancer

A recent study by Murphy et al reported an intriguing, time-dependent relationship between ghrelin, an appetite-stimulating hormone, and colorectal cancer (CRC). In the 10 years prior to diagnosis, but not earlier, low serum ghrelin concentrations were associated with a dramatic increase in CRC risk (OR=10.86). This raises the question of whether ghrelin might be a candidate biomarker for clinical use. However, whether ghrelin levels actually go down during the period approaching diagnosis is challenging to assess, requiring repeated prediagnostic blood samples from patients.

We conducted a validation of the study by Murphy et al using fasting plasma samples from the Västerbotten Intervention Programme, collected from CRC cases and matched controls within the 5 years preceding diagnosis of the cases. All participants had provided an additional blood sample 10 years earlier. This unique study design, described in detail elsewhere, allowed us to investigate potential changes in ghrelin levels over a long prediagnostic time period, from a generally tumour-free phase of 10–15 years prior to diagnosis to the window of opportunity for early detection.

Repeated plasma samples were available for 60 complete case sets, including 33 colon cancer and 27 rectal cancer cases, from our previous study. Plasma total ghrelin concentrations were analysed using sandwich ELISA (Merck, Germany). Our sample size provides 80% power to detect an OR of 3.3 for a median-split ghrelin variable at P<0.05 (and >99% power to detect an OR of 10). Characteristics of the participants are provided in table 1.

In our dataset, ghrelin was not associated with CRC risk (figure 1A). For samples collected within the 5 years preceding the CRC diagnosis of cases, we observed a multivariable OR and 95% CI of 0.98 (0.41 to 2.35) for levels below versus above the control–participant median of 496 pg/mL (figure 1A). Furthermore, ghrelin levels remained stable over time in both cases and controls (intra-class correlation coefficient=0.74, *P*interaction=0.81, figure 1B,C).

Our study is comparable to the report by Murphy et al in many respects. Both

### Table 1  Characteristics of colorectal cancer cases and matched control participants, all of whom provided repeated blood samples at 10-year intervals

| Time period prior to diagnosis of cases | >10 years | <5 years |
|----------------------------------------|----------|----------|
|                                       | Cases    | Controls | Cases    | Controls |
| N                                     | 60       | 60       | 60       | 60       |
| Males (%)                             | 34 (57)  | 34 (57)  | 34 (57)  | 34 (57)  |
| Age (years)                           | 50.0 (40.3–50.2) | 50.0 (40.2–50.2) | 59.8 (50.3–60.1) | 59.9 (50.2–60.1) |
| BMI                                   | 25.5 (23.6–27.0) | 24.6 (22.8–28.1) | 26.1 (24.4–28.9) | 25.5 (23.3–29.0) |
| Ever smoker, N (%)                    | 36 (60)  | 34 (57)  | 38 (63)  | 35 (58)  |
| Alcohol intake (g/day)                | 3.0 (0.9–6.3) | 3.4 (1.6–6.7) | 4.6 (1.7–8.1) | 3.2 (0.9–6.3) |
| No physical activity*                 | 28 (47)  | 23 (38)  | 27 (45)  | 25 (42)  |
| Ghrelin† (pg/mL)                      | 556 (323–772) | 494 (370–695) | 527 (338–758) | 504 (340–623) |
| <LOD, N (%)                           | 10 (17)  | 6 (10)   | 10 (17)  | 6 (10)   |

Summary statistics are presented as medians and interquartile ranges unless otherwise stated.

*A* Self-reported exercise frequency during leisure time on a scale from 1 to 5, where 1=none and 5=more than three times/week.

†Total fasting plasma ghrelin concentrations measured by sandwich ELISA (Merck, Germany).

BMI, body mass index in kg/m²; LOD, limit of detection.
were based on Scandinavian populations, had a prospective design with long follow-up, extensive data on potential confounders, fasting blood samples and low loss to follow-up, thanks to high-quality cancer registries. Notable differences include the lack of repeated measurements, male smoker inclusion criterion, the alpha-tocopherol, beta-carotene intervention and the radioimmunoassay analysis of serum ghrelin concentrations in Murphy et al’s study.4 Although we had limited power for subgroup analyses, we found nothing to suggest potential major heterogeneity in results based on sex, tumour site or tumour stage (data not shown). Plasma ghrelin concentrations were not associated with smoking in our study, which was in line with Murphy et al’s study.1 Use of ELISA was, if anything, an advantage in our study, as it may have less cross-reactivity, and thereby higher specificity, than radioimmunoassay.3 Ghrelin concentrations in plasma and serum are reportedly comparable,4 so the different media are also unlikely to explain the inconsistent findings.

In conclusion, we could not replicate the recently reported novel finding of a very strong association between lower circulating ghrelin concentrations and increased CRC risk in the years approaching diagnosis. Further examination of ghrelin in colorectal carcinogenesis may be warranted. However, our analyses, which were sufficiently powered for the purpose, suggest that ghrelin probably cannot be developed as a biomarker for risk prediction or early detection of CRC.

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Funding This study was funded by the Swedish Research Council (VR 2017-01737); the Lion’s Cancer Research Foundation, Umeå University (LP 17-2157); the Cancer Research Fund in Northern Sweden (CFF AMP 17-866); the Swedish Society of Medicine (SLS-594811); the Swedish Cancer Society (CANCAN 2014:780); Young Scientist and other research grants from the County Council of Västerbotten, Sweden, through the regional agreement between Umeå University and Västerbotten County Council on cooperation in the field of Medicine, Odontology and Health (VLL-547711); and the Faculty of Medicine at Umeå University, Umeå, Sweden.

Competing interests None declared.

Ethics approval The Regional Ethical Review Board at Umeå University.

Provenance and peer review Not commissioned; internally peer reviewed.

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To cite Sundkvist A, Myte R, Palmqvist R, et al. Gut 2019;68:373–374.

Received 26 January 2018
Revised 8 February 2018
Accepted 10 February 2018
Published Online First 28 February 2018

Gut 2019;68:373–374. doi:10.1136/gutjnl-2018-316110

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