

The Effect of Obesity on the Coccoid Form Transformation of *Helicobacter pylori* and Its Relationship with Gastric Inflammation Pattern: A Retrospective Comparative Study

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Abstract

Background/Aim:

Helicobacter pylori (*H. pylori*) can transform from its spiral form into a coccoid (viable but non-culturable; VBNC) state under adverse environmental conditions. The coccoid form is associated with antibiotic resistance and eradication failure. This study compared coccoid and spiral form proportions, *H. pylori* localization patterns (epithelial vs. crypt), and gastric inflammation parameters across three BMI-stratified groups.

Materials and Methods

This retrospective comparative study included 156 *H. pylori*-positive cases from the archives of Karamanoglu Mehmetbey University Faculty of Medicine (2016–2026). Groups: Group 1 (Severe obesity/Sleeve, BMI ≥ 35 ; sleeve gastrectomy, $n = 55$), Group 2 (Non-obese/Biopsy, BMI < 30 ; biopsy, $n = 68$), Group 3 (Obese/Biopsy, BMI 30–44; biopsy, $n = 33$). IHC with anti-*H. pylori* antibody was used for morphological assessment. Coccoid/spiral proportions, HP density, Sydney-criteria gastritis parameters, coccoid categorization ($< 5\%$, 5–19%, 20–39%, $\geq 40\%$), and epithelial/crypt HP localization were analyzed (SPSS 25.0; $p < 0.05$).

Results

Median corpus coccoid proportion was significantly higher in Group 1 (10%, IQR 5–30) vs. Group 2 (1%, IQR 1–1; $p < 0.001$), with strong BMI correlation ($r = 0.639$, $p < 0.001$). The $\geq 40\%$ coccoid category was exclusively observed in the severe obesity/sleeve group (23.6%; $p < 0.001$). Corpus spiral proportion decreased in a stepwise inverse gradient across categories (99%→90%→80%→40%; $H = 113.8$, $p < 0.001$). Corpus HP density (Sydney score) differed significantly across groups ($H = 13.8$, $p = 0.001$) and correlated positively with coccoid proportion ($r = 0.267$, $p = 0.003$). Corpus crypt *H. pylori* involvement was significantly more frequent in Group 1 (41.8%) vs. Group 3 (11.1%; $p = 0.045$). In antrum, crypt involvement was higher in non-obese (36.8%) vs. obese biopsy patients (9.7%; $p = 0.011$). Coccoid proportion showed no significant correlation with patient age or inflammation grade. Multivariable logistic regression identified BMI as the sole independent predictor of corpus coccoid $\geq 10\%$ (OR = 1.134 per 1 kg/m², 95% CI: 1.088–1.183, $p < 0.001$; C-statistic = 0.849), with age and sex non-significant after adjustment.

Conclusion

Obesity was strongly associated with coccoid-dominant *H. pylori* infection and increased corpus crypt colonization. The $\geq 40\%$ coccoid threshold is exclusive to the severe obesity/sleeve group and may

represent a histological signature of severe obesity. IHC-based morphological assessment and individualized eradication strategies are warranted in obese and severely obese HP-positive patients.

1. INTRODUCTION

Helicobacter pylori (*H. pylori*) is a Gram-negative microaerophilic bacterium colonizing the gastric mucosa of approximately 50% of the global population and is causally linked to chronic active gastritis, peptic ulcer disease, gastric adenocarcinoma, and mucosa-associated lymphoid tissue (MALT) lymphoma [1,2]. Despite available eradication regimens, with first-line eradication success rates declining to 70–80% in some regions [3]. While antibiotic resistance is the principal mechanism of failure, the bacterial capacity to adopt a “viable but non-culturable” (VBNC) state – the coccoid form – is an increasingly recognized contributor [4].

Under adverse environmental conditions including antibiotic exposure, extreme pH, elevated oxygen tension, and nutritional depletion, *H. pylori* transforms into spherical coccoid bodies through HdpA/Csd3-mediated progressive thinning of the lateral peptidoglycan (PGN) layer, leading to sacculus rupture and extrusion of the inner membrane-enclosed cytoplasm as a spheroplast-like structure [5]. Viable coccoid bacteria (subtype B) retain expression of major virulence genes (*ureA*, *ureB*, *vacA*, *cagA*, *cagE*, *BabA*), display low urease activity, and can revert to spiral forms when conditions normalize [6,7]. SEM studies confirm coccoid invasion of gastric epithelial cells and persistence within autophagosomes, creating a pharmacokinetic sanctuary from antibiotics [8].

Eradication failure linked to coccoid forms occurs via two mechanisms: (1) altered penicillin-binding protein profiles rendering β -lactams ineffective [9]; and (2) confinement within autophagosomes where drug concentrations may not reach bactericidal levels [10]. Clinical data corroborate this: eradication-failure strains exhibit higher invasiveness [11], and murine studies show re-colonization from intracellular/intravacuolar bacteria 8 weeks after standard triple therapy [10].

Obesity alters the gastric microenvironment through chronic low-grade systemic inflammation (elevated IL-1 β , TNF- α , IL-6, leptin), modified acid secretion, and changes in mucus composition and gastric microbiota. Mayer et al. [12] provided the first systematic clinical evidence that patients with severe obesity undergoing sleeve gastrectomy harbor a significantly higher median coccoid proportion (50%) than non-obese controls (10%; $p < 0.001$) and display predominantly chronic inactive rather than active gastritis. These findings raise the question of whether obesity-associated gastric alterations promote the VBNC transition and thereby contribute to lower eradication success in obese populations.

The present study extends this question by examining *H. pylori* morphological distributions across three BMI-stratified groups, quantifying BMI–coccoid correlations, introducing a four-tier coccoid categorization (< 5%, 5–19%, 20–39%, $\geq 40\%$), and for the first time systematically analyzing intragastric HP localization patterns (epithelial surface vs. glandular crypt involvement) across BMI groups.

2. MATERIALS AND METHODS

2.1. Study Design and Ethical Approval

This retrospective comparative study was conducted in accordance with the Declaration of Helsinki. Ethical approval was obtained from the Karamanoglu Mehmetbey University Faculty of Medicine Local Scientific Medical Research Ethics Committee (Application date: 26.02.2025). Patient data were anonymized prior to analysis; no new interventions were performed.

2.2. Study Population

The pathology archives were searched for *H. pylori*-positive gastric specimens processed between January 2016 and May 2026. A total of 156 cases fulfilling inclusion criteria were enrolled and stratified into three groups:

Group 1 (Severe obesity/Sleeve)

55 patients with BMI ≥ 35 kg/m² who underwent laparoscopic sleeve gastrectomy. Only corpus (oxyntic) mucosa was available.

Group 2 (Non-obese/Biopsy)

68 patients with BMI < 30 kg/m²; both antrum and corpus biopsies available.

Group 3 (Obese/Biopsy)

33 patients with BMI 30–44 kg/m²; both antrum and corpus biopsies available.

Exclusion criteria: age > 60 years, gastric malignancy or premalignant lesions, prior documented *H. pylori* eradication therapy, and insufficient tissue.

2.3. Histopathological and Immunohistochemical Evaluation

All FFPE tissue blocks were sectioned at 4 μ m and stained with H&E and Giemsa. IHC staining was performed with polyclonal anti-*H. pylori* antibody (CellMarque 215A-76, prediluted/ready-to-use format). Two experienced pathologists (B.S.Y. and S.A.) independently evaluated all slides while blinded to group assignment and clinical data. Each pathologist recorded all parameters separately without access to the other's assessments. Discordance was defined as a difference of ≥ 10 percentage points for continuous coccoid/spiral proportions or ≥ 1 grade for Sydney criteria ordinal scores. All discordant cases were re-examined jointly at a multihead microscope until consensus was reached. Recorded parameters included: (1) Coccoid and spiral form proportions (% summing to 100%) in corpus (all groups) and antrum (Groups 2–3); (2) HP density (Sydney 0–3 scale); (3) Epithelial surface HP% and glandular crypt HP% in both corpus and antrum; crypt involvement was defined as any HP% >0% in the glandular crypt

compartment; (4) Chronic inflammation grade (0–3); neutrophilic activation (0–3); intestinal metaplasia; glandular atrophy (all 0–3 per Sydney criteria) (Fig. 1).

(A–B) Corpus mucosa from a severe obesity/sleeve patient (Group 1) demonstrating dominant coccoid morphology ($\geq 40\%$ coccoid proportion). The majority of organisms display a spherical, non-flagellated appearance with near-complete loss of spiral forms. (A) $\times 200$; (B) $\times 400$.

(C–D) Corpus mucosa from an obese biopsy patient (Group 3) showing an intermediate mixed pattern with coexisting coccoid and spiral forms. (C) $\times 200$; (D) $\times 400$.

*(E–F) Corpus mucosa from a non-obese patient (Group 2) demonstrating predominantly spiral-form *H. pylori* along the epithelial surface, with minimal coccoid transformation. (E) $\times 200$; (F) $\times 400$.*

2.4. Corpus Coccoid Categorization

To identify clinically meaningful morphological thresholds, corpus coccoid proportions were stratified into four categories reflecting natural data breakpoints and clinical relevance: $<5\%$, $5\text{--}19\%$, $20\text{--}39\%$, and $\geq 40\%$.

2.5. Statistical Analysis

Statistical analyses were performed using IBM SPSS Statistics v25.0. Non-normal distribution of continuous variables was confirmed by Shapiro-Wilk test; results are presented as median (IQR) or mean $\pm$ SD. Group comparisons used Kruskal-Wallis with post-hoc Mann-Whitney U tests. Categorical variables were compared with chi-square or Fisher's exact tests. Spearman's rank correlation assessed BMI–coccoid and HP density–coccoid relationships. To assess BMI as an independent predictor of elevated coccoid burden, univariable and multivariable binary logistic regression analyses were performed with corpus coccoid proportion $\geq 10\%$ as the outcome variable. Predictors included BMI (continuous, per 1 kg/m²), age, and sex. Model discrimination was assessed by the C-statistic (area under the ROC curve) and calibration by the Hosmer-Lemeshow test. Variance inflation factors (VIF) were calculated to evaluate multicollinearity. Results are presented as odds ratios (OR) with 95% confidence intervals (CI). $P < 0.05$ was considered statistically significant.

3. RESULTS

3.1. Patient Demographics

A total of 156 *H. pylori*-positive patients were included: Group 1 ($n = 55$), Group 2 ($n = 68$), and Group 3 ($n = 33$). Mean BMI differed significantly across groups (Group 1: 47.4 ± 7.0 ; Group 2: 22.1 ± 2.7 ; Group 3: 42.8 ± 6.3 kg/m²; $p < 0.001$). Age distribution differed modestly ($H = 9.28$, $p = 0.010$). Female predominance was observed across all groups. Atrophy was absent in Group 1 and extremely rare in Groups 2–3 (≤ 2 cases each). Intestinal metaplasia was similarly rare in biopsy groups (Table 1).

Table 1
Patient demographics and baseline characteristics by group.

Variable	Group 1 Severe obesity/Sleeve (n = 55)	Group 2 Non-obese/Biopsy (n = 68)	Group 3 Obese/Biopsy (n = 33)	p
Mean BMI (kg/m ²)	47.4 ± 7.0	22.1 ± 2.7	42.8 ± 6.3	< 0.001
Mean age (years)	36.2 ± 10.7	39.5 ± 12.2	42.6 ± 9.4	0.010
Sex (F/M)	36/19	39/28	27/6	0.101
Specimen type	Sleeve gastrectomy (corpus only)	Endoscopic biopsy (antrum+corpus)	Endoscopic biopsy (antrum+corpus)	—
Atrophy (any)	0 (0%)	2 (2.9%)	1 (3.0%)	> 0.05
Intestinal metaplasia (any)	0 (0%)	5 (7.4%)	2 (6.1%)	> 0.05
<i>BMI: body mass index; F: female; M: male. Data presented as mean ± SD or n (%). Chi-square or Kruskal-Wallis tests used.</i>				

3.2. Coccoid and Spiral Form Proportions in Corpus Mucosa

Corpus coccoid proportions differed significantly across groups (Kruskal-Wallis H = 64.43, p < 0.001). The severe obesity/sleeve group (Group 1) had a median corpus coccoid proportion of 10% (IQR: 5–30%), substantially higher than the non-obese/biopsy group (Group 2: 1%, IQR: 1–1%; U = 2372, p < 0.001). The obese/biopsy group (Group 3) showed an intermediate median of 10% (IQR: 5–17.5%), significantly higher than Group 2 (U = 52, p < 0.001) but not significantly different from Group 1 (U = 608, p = 0.145). Results are detailed in Table 2.

Table 2
H. pylori morphological form proportions and density by group.

Parameter	Group 1 Severe obesity/Sleeve (n = 55)	Group 2 Non-obese/Biopsy (n = 68)	Group 3 Obese/Biopsy (n = 33)	p-value
Corpus coccoid % – median (IQR)	10 (5–30)	1 (1–1)	10 (5–17.5)	< 0.001*
Corpus spiral % – median (IQR)	90 (70–95)	99 (99–99)	90 (82.5–95)	< 0.001*
Antrum coccoid % – median (IQR)	N/A	1 (1–1)	10 (5–20)	< 0.001†
Antrum spiral % – median (IQR)	N/A	99 (99–99)	90 (80–95)	< 0.001†
Corpus HP density (Sydney) – mean (SD)	2.38 (0.71)	2.12 (0.82)	1.53 (0.80)	0.001*
Antrum HP density – mean (SD)	N/A	2.40 (0.65)	2.48 (0.57)	0.678†
N/A: Not applicable (antrum unavailable in sleeve specimens). *Kruskal-Wallis (3 groups). †Mann-Whitney U (2 groups). HP: Helicobacter pylori.				

3.3. Antrum Coccoid and Spiral Form Proportions

Antrum evaluation was restricted to Groups 2 and 3. The antrum coccoid proportion was significantly higher in Group 3 vs. Group 2: median 10% (IQR: 5–20%) vs. 1% (IQR: 1–1%; U = 112, p < 0.001). These findings corroborate the corpus results and reinforce a BMI-dependent shift in *H. pylori* morphology.

3.4. Correlation between BMI and Coccoid Form Proportions

Spearman’s rank correlation revealed a strong positive association between BMI and corpus coccoid proportion (r = 0.639, p < 0.001) and antrum coccoid proportion (r = 0.676, p < 0.001) across all 156 cases.

3.5. Categorized Analysis of Corpus Coccoid Proportions

Corpus-based categorical analysis included 121 cases with evaluable corpus coccoid data (Group 1: n = 55, Group 2: n = 48, Group 3: n = 18). Corpus coccoid proportions were stratified into four categories (Table 3; Chi²=84.6, p < 0.001). The most striking finding was the **≥ 40% category**: all 13 cases (23.6% of Group 1) were exclusively from the severe obesity/sleeve group; none of the 48 non-obese or 18 obese biopsy patients fell into this category within the present cohort (100% obesity-specificity). The 20–39% category also contained only severe obesity/sleeve and obese biopsy patients (0/48 non-obese). Conversely, the < 5% category was dominated by non-obese patients (47/48, 97.9%; Figs. 2 and 3).

Table 3

Distribution of corpus coccoid categories by group and associated morphological parameters (corpus-evaluable cases: N = 121; Group 1 n = 55, Group 2 n = 48, Group 3 n = 18).

Corpus Coccoid Category	n	Group 1 Severe obesity/Sleeve n (%) [*]	Group 2 Non-obese/Biopsy n (%) [*]	Group 3 Obese/Biopsy n (%) [*]	Median BMI (kg/m ²)	Corpus Spiral Median (%)
< 5%	61	12 (21.8)	47 (97.9)	2 (11.1)	24	99
5–19%	29	17 (30.9)	1 (2.1)	11 (61.1)	45	90
20–39%	18	13 (23.6)	0 (0.0)	5 (27.8)	45	80
≥ 40%	13	13 (23.6) ☒	0 (0.0)	0 (0.0)	45	40
p-value (Chi ² =84.6)	—	< 0.001	< 0.001	< 0.001	—	< 0.001‡
☒ All 13 cases with ≥ 40% coccoid are exclusively from the severe obesity/sleeve group (Group 1). [*] Column percentages. ‡Kruskal-Wallis H = 113.8; all pairwise p < 0.001.						

Strip and box plots of corpus coccoid proportions for Group 1 (Severe obesity/Sleeve, n = 55), Group 2 (Non-obese/Biopsy, n = 48 with evaluable corpus data), and Group 3 (Obese/Biopsy, n = 18 with evaluable corpus data). Total N = 121 (corpus-evaluable subset of 156 enrolled cases). Boxes represent IQR; horizontal bars = median; whiskers = range. Individual patients shown as open circles. ***p < 0.001

group.

Color intensity reflects the percentage of patients within each BMI group falling into that coccoid category (darker blue = higher proportion). ☒ The ≥ 40% coccoid category was exclusively observed in the severe obesity/sleeve group (23.6%; p < 0.001 vs. both other groups). Chi²=84.6, p < 0.001 (overall). BMI < 30 group: n = 48 of 68 enrolled with evaluable corpus coccoid data. BMI 30–44 group: n = 18 of 33 enrolled. Severe obesity/Sleeve group: all 55 enrolled had evaluable corpus data.

3.6. Corpus Spiral Form Gradient across Coccoid Categories

The corpus spiral proportion decreased in a precise stepwise gradient across increasing coccoid categories (Kruskal-Wallis H = 113.8, p < 0.001): <5% coccoid → spiral median 99% (IQR: 99–99%); 5–19% → 90% (IQR: 90–95%); 20–39% → 80% (IQR: 72–80%); ≥40% → 40% (IQR: 30–40%). All pairwise comparisons were statistically significant (all p < 0.001).

3.7. Corpus HP Density across Groups and Correlation with Coccoid Form

Corpus HP density assessed by Sydney criteria differed significantly across groups (Kruskal-Wallis H = 13.8, p = 0.001): Group 1 mean 2.38 ± 0.71 vs. Group 2 mean 2.12 ± 0.82 vs. Group 3 mean 1.53 ± 0.80.

Pairwise comparisons showed Group 1 and Group 2 did not differ significantly ($p = 0.094$), while Group 3 was significantly lower than both Group 1 ($p = 0.0003$) and Group 2 ($p = 0.010$). Across all 121 cases with corpus coccoid data, HP density correlated positively with corpus coccoid proportion (Spearman $r = 0.267$, $p = 0.003$). Stratified by HP density category, coccoid medians were: density 1 $\rightarrow$ 1% (IQR: 0–1%), density 2 $\rightarrow$ 1% (IQR: 1–10%), density 3 $\rightarrow$ 10% (IQR: 1–20%; Kruskal-Wallis $H = 9.6$, $p = 0.008$).

3.8. *H. pylori* Localization Pattern: Epithelial Surface vs. Glandular Crypt Involvement

Beyond morphological form, we analyzed the intragastric distribution of *H. pylori* between the surface epithelium and glandular crypts, as crypt involvement reflects deeper mucosal penetration and differs pathophysiologically from surface colonization. Results are summarized in Table 4.

In the corpus, crypt HP involvement (any HP% >0% in the crypt compartment) was significantly more frequent in Group 1 (23/55, 41.8%) compared to Group 2 (14/48, 29.2%) and Group 3 (2/18, 11.1%; $\chi^2=6.2$, $p = 0.045$). Continuous corpus crypt HP% did not differ significantly across groups in Kruskal-Wallis analysis ($H = 4.1$, $p = 0.126$), owing to the low absolute crypt proportions; however, the binary presence of crypt involvement showed a clear BMI-associated gradient. The corpus epithelial surface HP% was uniformly high across all three groups (median 99–100%; $p = 0.126$).

In the antrum (Groups 2 and 3 only), an inverse pattern was observed: antrum crypt involvement was significantly more frequent in non-obese patients (Group 2: 25/68, 36.8%) than in obese biopsy patients (Group 3: 3/33, 9.7%; $\chi^2=6.4$, $p = 0.011$). Correspondingly, antrum epithelial surface HP% was significantly higher in Group 3 than Group 2 (Mann-Whitney $p = 0.011$). This divergent pattern — deeper crypt involvement in non-obese antrum but predominantly surface epithelial localization in obese biopsy antrum — likely reflects the difference in gastritis type: Group 2 harbors active (neutrophilic) antral gastritis, which drives deeper HP infiltration, whereas the predominantly chronic inactive pattern in Group 3 is associated with more superficial colonization.

Table 4

H. pylori localization pattern: corpus and antrum epithelial surface vs. glandular crypt involvement.

Parameter	Group 1 Severe obesity/Sleeve (n = 55)	Group 2 Non-obese/Biopsy (n = 48)	Group 3 Obese/Biopsy (n = 18)	p-value
Corpus mucosa				
Epithelial surface HP% – median (IQR)	100 (99–100)	100 (98–100)	100 (100–100)	0.126*
Crypt HP involvement (> 0%) – n (%)	23 (41.8%)	14 (29.2%)	2 (11.1%)	0.045*
Antrum mucosa (Groups 2–3 only)				
Epithelial surface HP% – median (IQR)	N/A	100 (95–100)	100 (100–100)	0.011†
Crypt HP involvement (> 0%) – n (%)	N/A	25 (36.8%)	3 (9.7%)	0.011†
*Kruskal-Wallis (corpus, 3 groups). †Mann-Whitney U or Chi-square (antrum, 2 groups). N/A: Not applicable. Crypt involvement defined as any HP% >0% in the glandular crypt compartment. Highlighted cells indicate significant associations.				

3.9. HP Density Correlation with Coccoid Proportion and Absence of Age Effect

As noted in section 3.7, corpus HP density correlated positively with coccoid proportion across all groups ($r = 0.267$, $p = 0.003$), with the strongest density–coccoid association in Group 2 ($r = 0.426$, $p = 0.003$). Notably, coccoid proportion showed no significant correlation with patient age in any group or overall (corpus: $r = 0.008$, $p = 0.934$; antrum: $r = 0.151$, $p = 0.136$), indicating that the elevated coccoid burden in obese and obese biopsy patients reflects BMI-dependent rather than age-dependent gastric changes.

3.10. Coccoid Form and Gastric Inflammation Pattern

Corpus coccoid proportion showed no significant correlation with the degree of neutrophilic activation (Spearman $r = -0.119$, $p = 0.198$) or the grade of chronic inflammation ($r = 0.077$, $p = 0.408$) when analyzed across all cases. Comparison of patients with active (activation grade ≥ 1) vs. inactive (grade = 0) corpus gastritis revealed no significant difference in coccoid proportions (active: median 1%, IQR: 1–15% vs. inactive: median 5%, IQR: 1–15%; Mann-Whitney $p = 0.676$). This independence was also confirmed within Group 1 (active: median 20% vs. inactive: median 10%; $p = 0.631$). These findings indicate that coccoid burden does not track with the acute inflammatory response, consistent with the

known property of the VBNC state to suppress pro-inflammatory virulence factor expression. The gastritis pattern (chronic inactive vs. active) is thus a group-level phenomenon driven by BMI rather than a function of coccoid proportion per se.

3.11. Gastric Inflammation Parameters by Group

Gastritis parameters assessed by Sydney criteria are presented in Table 5. Corpus inflammation severity did not differ significantly across groups ($H = 2.27$, $p = 0.321$). Corpus activation differed significantly ($H = 7.48$, $p = 0.024$): Group 1 exhibited lower neutrophilic activity and lacked grade 3 activation (neutrophilic activation grade 0 in 40%, grade 1 in 36%, grade 2 in 24%; grade 3 absent), while Groups 2 and 3 showed higher activation including grade 3 in Group 2. Atrophy and intestinal metaplasia were absent in Group 1.

Table 5
Gastritis parameters (Sydney criteria) by group.

Parameter	Group 1 Severe obesity/Sleeve (n = 55)	Group 2 Non-obese/Biopsy (n = 68)	Group 3 Obese/Biopsy (n = 33)	p-value
Corpus inflammation grade (0/1/2/3) n	0/6/25/24	0/3/27/21	0/4/8/5	0.321*
Corpus activation grade (0/1/2/3) n	22/20/13/0	13/16/14/8	2/9/5/1	0.024*
Antrum inflammation grade (0/1/2/3) n	N/A	0/2/13/53	0/2/8/22	0.798†
Atrophy (any) n (%)	0 (0%)	2 (2.9%)	1 (3.0%)	> 0.05
Intestinal metaplasia (any) n (%)	0 (0%)	5 (7.4%)	2 (6.1%)	> 0.05
*Kruskal-Wallis (3 groups). †Mann-Whitney U (2 groups). N/A: Not applicable. Grades: 0 = absent, 1 = mild, 2 = moderate, 3 = severe.				

3.12. ROC Analysis: Optimal BMI Threshold for Predicting Corpus Coccoid ≥ 5%

ROC analysis identified BMI as a strong predictor of corpus coccoid proportion $\geq 5\%$ (AUC = 0.882, 95% CI: 0.814–0.943). The Youden-optimal BMI cut-off was ≥ 27 kg/m² (sensitivity 98.3%, specificity 77.0%, PPV 80.6%, NPV 97.9%, accuracy 87.5%, Youden index J = 0.754). Notably, a BMI ≥ 30 kg/m² (international BMI ≥ 30 threshold) yielded near-equivalent performance (Sens 96.6%, Spec 77.0%), and a BMI ≥ 35 kg/m² (obesity threshold) maintained high sensitivity (93.2%) with the same specificity. A secondary ROC analysis for corpus coccoid $\geq 10\%$ yielded an AUC of 0.846 (95% CI: 0.770–0.914), with BMI ≥ 27 kg/m² achieving sensitivity 98.0%, specificity 66.2%.

3.13. Multivariable Logistic Regression: BMI as an Independent Predictor of Corpus Coccoid ≥ 10%

To determine whether BMI independently predicts elevated coccoid burden after adjustment for potential confounders, univariable and multivariable binary logistic regression analyses were performed with corpus coccoid ≥ 10% as the outcome (N = 156; Table 6). In univariable analysis, BMI was the only significant predictor (OR = 1.125, 95% CI: 1.081–1.171, $p < 0.001$), while age (OR = 0.980, $p = 0.206$) and sex (OR = 1.298, $p = 0.466$) were not. In the multivariable model adjusting simultaneously for all three predictors, BMI remained the sole independent predictor of corpus coccoid ≥ 10% (OR = 1.134, 95% CI: 1.088–1.183, $p < 0.001$), indicating that each 1 kg/m² increase in BMI confers a 13.4% increase in the odds of reaching the ≥ 10% coccoid threshold. Age (OR = 0.968, $p = 0.107$) and sex (OR = 2.361, $p = 0.068$) were not independently significant, though a marginal trend was observed for male sex. The model demonstrated excellent discrimination (C-statistic = 0.849) and good calibration (Hosmer-Lemeshow $\chi^2=9.971$, $p = 0.267$), with Nagelkerke $R^2=0.438$. No multicollinearity was detected (all VIF < 1.02).

Table 6
Univariable and multivariable logistic regression for corpus coccoid ≥ 10% (N = 156).

Variable	Univariable OR (95% CI)	p	Multivariable OR (95% CI)	p
BMI (per 1 kg/m ²)	1.125 (1.081–1.171)	< 0.001	1.134 (1.088–1.183)	< 0.001
Age (per year)	0.980 (0.950–1.011)	0.206	0.968 (0.930–1.007)	0.107
Sex (male vs. female)	1.298 (0.643–2.618)	0.466	2.361 (0.938–5.944)	0.068†
Model fit statistics	N = 156; Nagelkerke $R^2=0.438$; C-statistic = 0.849; Hosmer-Lemeshow $\chi^2=9.971$, $p = 0.267$; all VIF < 1.02			

OR: odds ratio; CI: confidence interval; BMI: body mass index. †marginal trend ($p < 0.10$). Outcome: corpus coccoid proportion ≥ 10%. Bold: $p < 0.05$.

4. DISCUSSION

The principal findings of this study are: (1) patients in the severe obesity/sleeve group harbor a significantly higher proportion of *H. pylori* coccoid form in the gastric corpus compared to non-obese individuals; (2) BMI correlates strongly and positively with coccoid form proportion ($r = 0.639$, $p < 0.001$); (3) the ≥ 40% coccoid category is exclusively found in the severe obesity/sleeve group, representing a potential morphological signature of severe obesity; (4) corpus spiral proportions decrease in a precise stepwise gradient across coccoid categories; (5) corpus HP density (Sydney score) is significantly higher in the severe obesity/sleeve group and correlates positively with coccoid conversion; (6) corpus crypt *H. pylori* involvement is significantly more frequent in the severe obesity/sleeve group, while antrum crypt

involvement is paradoxically higher in non-obese patients; (7) coccoid burden is independent of patient age and does not correlate with inflammation grade.

Our results are consistent with and extend the findings of Mayer et al. [12], who reported a median coccoid percentage of 50% in patients with severe obesity undergoing sleeve gastrectomy versus 10% in non-obese controls ($p < 0.001$). Our three-group design with quantitative BMI data demonstrates that coccoid promotion is not a binary obese/non-obese phenomenon but occurs along a BMI gradient: obese biopsy patients (BMI 30–44) showed significantly higher coccoid proportions than non-obese patients ($p < 0.001$) but were statistically indistinguishable from the obesity/sleeve group ($p = 0.145$), suggesting the threshold for meaningful coccoid promotion may already begin at BMI ≥ 30 . Importantly, the observation that obese biopsy patients showed coccoid proportions comparable to those observed in the severe obesity/sleeve group argues against specimen size alone accounting for the observed association. This finding suggests that obesity-related biological factors, rather than differences in tissue sampling, contribute substantially to coccoid transformation.

The categorical analysis introduced in this study provides additional clinical granularity. The $\geq 40\%$ coccoid category was exclusively observed in the severe obesity/sleeve group, representing nearly one quarter of that cohort. In practical terms, a gastric specimen with $\geq 40\%$ coccoid *H. pylori* may be considered a potential histological indicator of obesity, pending validation in independent cohorts.

A novel contribution of this study is the analysis of intragastric *H. pylori* localization. Corpus crypt colonization was significantly more frequent in the severe obesity/sleeve group (41.8%) than in the obese biopsy group (11.1%; $p = 0.045$), suggesting that the obesity-associated gastric microenvironment facilitates deeper mucosal penetration by the pathogen. This deeper localization has clinical implications: bacteria residing in glandular crypts are more physically protected from luminal antibiotics and host immune surveillance, potentially contributing to eradication failure. Paradoxically, antrum crypt involvement was most frequent in non-obese patients (36.8% vs. 9.7% in obese biopsy patients; $p = 0.011$). This inverse antrum pattern aligns with the predominantly active (neutrophilic) antral gastritis in Group 2: neutrophilic activity drives *H. pylori* into deeper tissue compartments, a well-recognized histopathological observation [12,13]. In contrast, obese biopsy patients with their less active gastritis pattern show more superficial antrum colonization.

The positive correlation between corpus HP density and coccoid proportion ($r = 0.267$, $p = 0.003$) is consistent with a dose–response framework: a higher bacterial burden provides more organisms susceptible to VBNC-triggering environmental stress, amplifying coccoid conversion. Several mechanistic pathways may underlie obesity-associated coccoid promotion. The chronic low-grade inflammatory state (elevated IL-1 β , TNF- α , IL-6, leptin), altered gastric acid dynamics (including PPI use for reflux) [14], and adiposity-related changes in mucus composition collectively create a stressful intragastric microenvironment. From the ultrastructural perspective, Cheng et al. [5] demonstrated that the spheroplast-like coccoid structure is devoid of a complete PGN shell, rendering it inherently

insensitive to β -lactam antibiotics. The amplification of this process in obesity therefore directly worsens the antibiotic pharmacological access problem.

Multivariable logistic regression further corroborated the independence of BMI: after simultaneous adjustment for age and sex, BMI remained the sole significant predictor of corpus coccoid $\geq 10\%$ (OR = 1.134 per 1 kg/m², 95% CI: 1.088–1.183, $p < 0.001$), with the model demonstrating excellent discrimination (C-statistic = 0.849) and good calibration (Hosmer-Lemeshow $p = 0.267$). The observation that coccoid proportion is entirely independent of patient age (corpus: $r = 0.008$, $p = 0.934$; antrum: $r = 0.151$, $p = 0.136$) confirms that the obesity–coccoid association is not confounded by age-related mucosal changes. Similarly, the absence of a correlation between coccoid burden and inflammation grade, including within the severe obesity/sleeve group itself, indicates that the inactive gastritis phenotype seen in this group is a group-level property of the gastric microenvironment rather than being directly determined by coccoid proportion. This suggests that the metabolic and hormonal milieu of obesity – rather than the coccoid form per se – drives the immune suppression.

5. LIMITATIONS

Our study has several limitations. First, the retrospective single-center design precludes direct causal inference. Second, antrum evaluation was unavailable in sleeve gastrectomy specimens due to the surgical technique. Third, coccoid proportions were estimated visually rather than by quantitative image analysis. Fourth, prior antibiotic exposure, PPI use duration, and eradication history were not systematically available. Fifth, molecular VBNC confirmation (RT-PCR, flow cytometry) was not performed. Sixth, comparison between sleeve gastrectomy material and endoscopic biopsies poses an inherent methodological limitation: sleeve gastrectomy specimens provide a substantially larger mucosal surface area for assessment, potentially increasing the detection of minor HP populations compared to the limited surface area of pinch biopsies. We addressed this limitation by including a separate obese biopsy group (Group 3), whose comparison with the non-obese biopsy group (Group 2) provides a tissue-equivalent matched comparison unaffected by specimen size differences. The obese biopsy group thus serves both as an intermediate BMI category and as an internal methodological control. Seventh, formal interobserver agreement statistics (e.g., intraclass correlation coefficient or Cohen's κ) were not prospectively calculated, as only consensus-based final values were recorded; future studies should include pre-consensus independent ratings to quantify reproducibility. Finally, the proposed coccoid categorization cut-offs (particularly the $\geq 40\%$ obesity-specific threshold) require validation in independent prospective cohorts.

6. CONCLUSION

This study demonstrates that obesity is strongly associated with coccoid-dominant *H. pylori* infection (BMI–coccoid correlation $r = 0.639$, $p < 0.001$) and with deeper corpus crypt colonization (41.8% vs. 11.1% in obese biopsy patients; $p = 0.045$). The $\geq 40\%$ corpus coccoid threshold is exclusively observed in the severe obesity/sleeve group, constituting a potential severe-obesity-specific histological signature.

Corpus HP density correlates positively with coccoid proportion ($r = 0.267$, $p = 0.003$), and corpus spiral proportions decrease in a stepwise gradient (99%→90%→80%→40%). Coccoid burden is independent of patient age and gastric inflammation grade. Despite high HP density and elevated coccoid proportions, patients in the severe obesity/sleeve group exhibited lower neutrophilic activity and lacked severe (grade 3) activation. These findings support the selective use of IHC-based morphological assessment in obese H. pylori-positive patients, the inclusion of obese biopsy individuals in future eradication outcome studies, and the development of individualized eradication strategies informed by coccoid burden and BMI.

Abbreviations

H. pylori

Helicobacter pylori

BMI

body mass index

IHC

immunohistochemistry

VBNC

viable but non-culturable

HP

Helicobacter pylori

PGN

peptidoglycan

PPI

proton-pump inhibitor

IL-1 β

interleukin-1 beta

TNF- α

tumor necrosis factor alpha

IL-6

interleukin-6

ROC

receiver operating characteristic

AUC

area under the curve

OR

odds ratio

CI

confidence interval

VIF

variance inflation factor
RT-PCR
reverse transcription polymerase chain reaction
N/A
not applicable.

Declarations

Ethics approval and consent to participate:

This retrospective comparative study was conducted in accordance with the Declaration of Helsinki. Ethical approval was obtained from the Karamanoglu Mehmetbey University Faculty of Medicine Local Scientific Medical Research Ethics Committee (Decision No. 17-2025/20, dated 05.03.2025). Informed consent to participate was not obtained from individual patients, as this was a retrospective study based on archived, anonymized pathology specimens with no new interventions performed; the requirement for individual informed consent was therefore waived by the Karamanoglu Mehmetbey University Faculty of Medicine Local Scientific Medical Research Ethics Committee, in accordance with national regulations governing retrospective archival research.

Consent for publication:

Not applicable. As this study used retrospectively collected, fully anonymized archival pathology data with no identifiable patient information, images, or case details included, the requirement for consent for publication was waived by the ethics committee.

Competing Interests:

We have no conflicts of interest to declare.

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Author Contribution

B.S.Y. conceived and designed the study, performed histopathological and immunohistochemical evaluation of all slides, conducted the statistical analysis, interpreted the results, and wrote the main manuscript text. S.A. performed independent histopathological and immunohistochemical evaluation of

all slides, participated in consensus review of discordant cases, and contributed to data interpretation. All authors reviewed and approved the final manuscript.

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Not applicable.

Data Availability

All data generated or analyzed during this study are included in this published article and its supplementary information file (raw dataset provided as a Microsoft Excel file).

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Figures

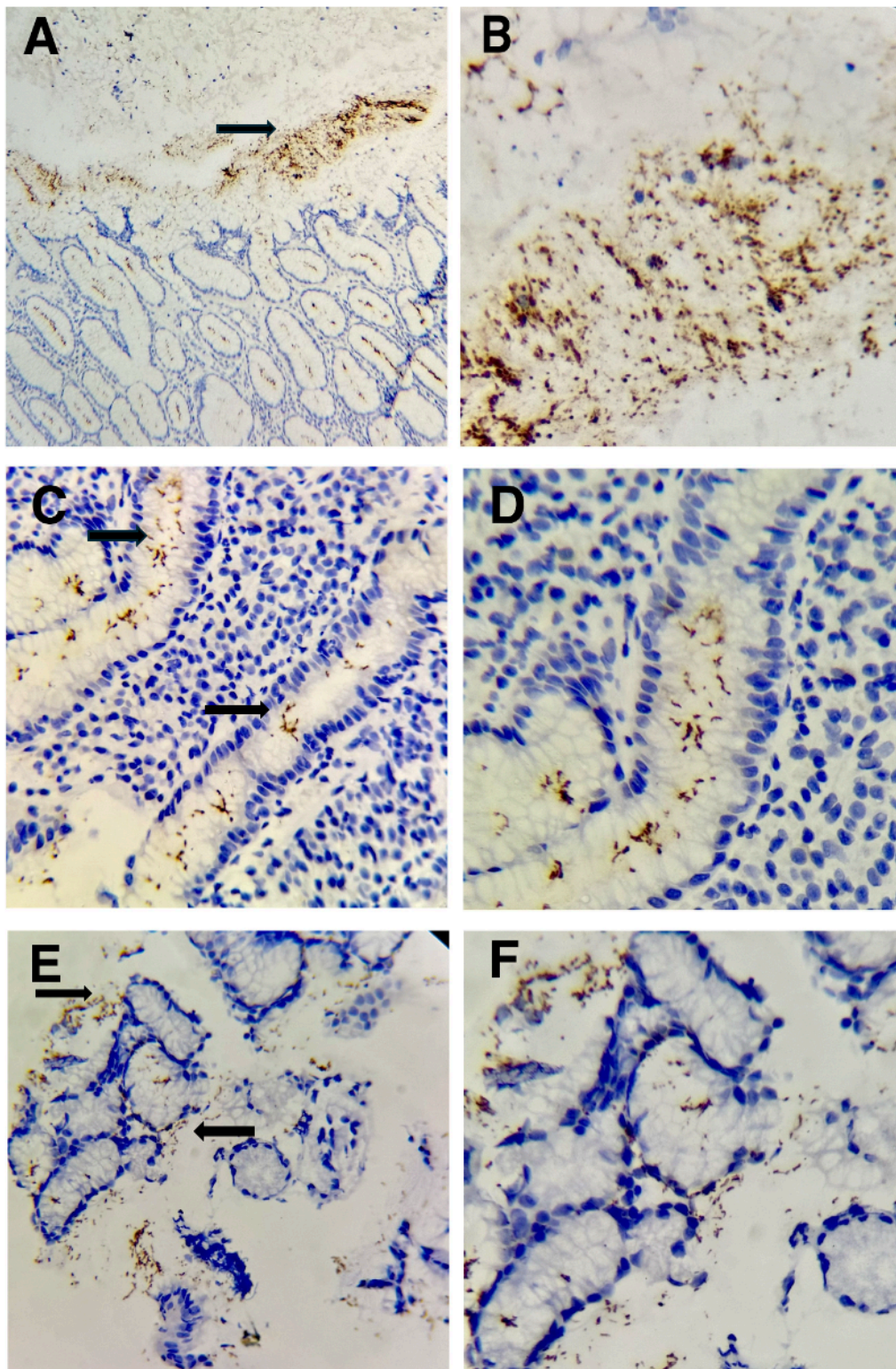


Figure 1

Immunohistochemical detection of *H. pylori* morphological forms across BMI groups (anti-*H. pylori* antibody, CellMarque 215A-76).

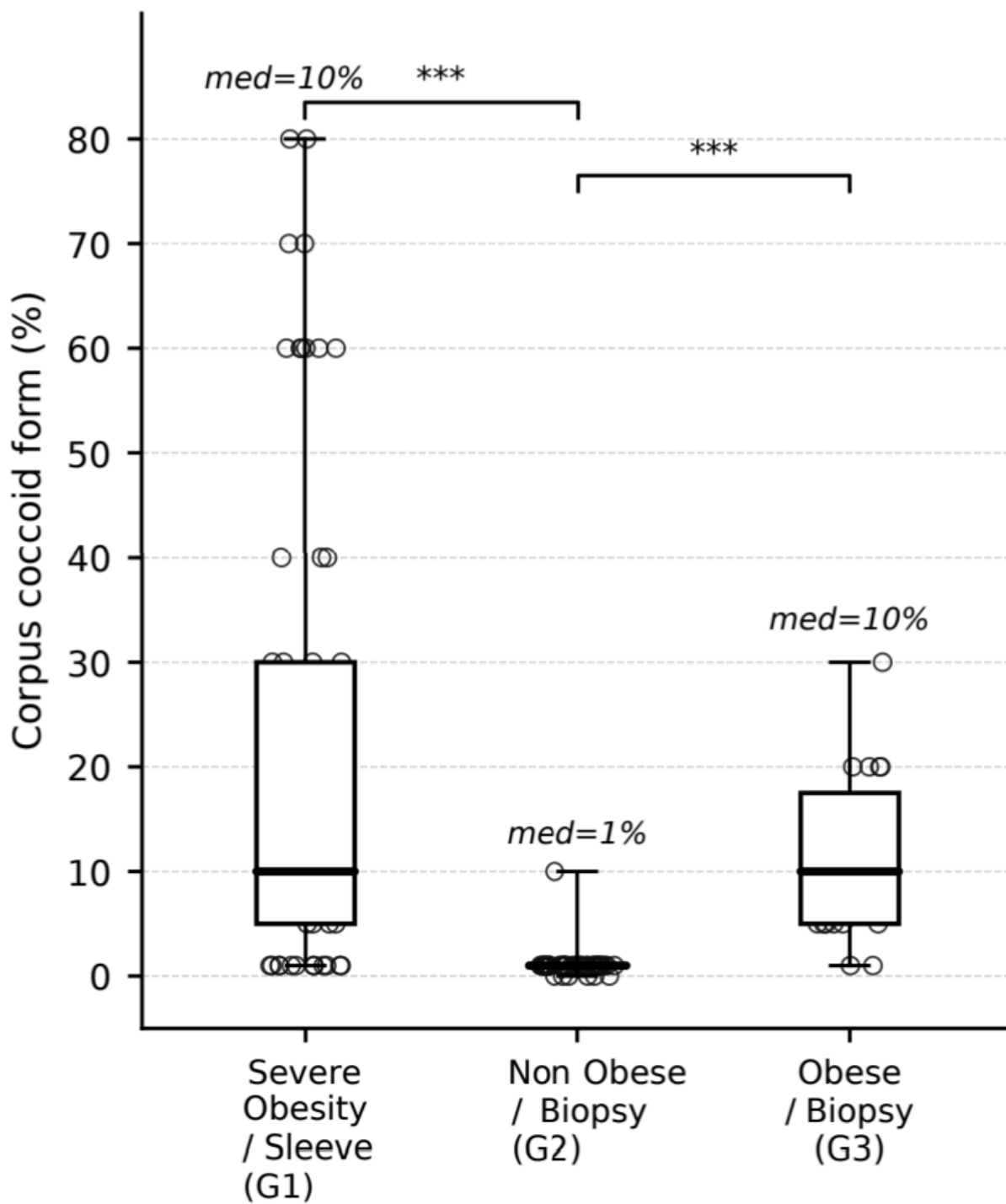


Figure 2

H. pylori coccoid form distribution across BMI groups and coccoid categories.

Strip and box plots of corpus coccoid proportions for Group 1 (Severe obesity/Sleeve, n=55), Group 2 (Non-obese/Biopsy, n=48 with evaluable corpus data), and Group 3 (Obese/Biopsy, n=18 with evaluable corpus

data). Total N=121 (corpus-evaluable subset of 156 enrolled cases). Boxes represent IQR; horizontal bars = median; whiskers = range. Individual patients shown as open circles. ***p<0.001

	<5%	5–19%	20–39%	≥40%
BMI <30 Non-obese/Biopsy (n=48)	97.9% n=47	2.1% n=1	0.0% n=0	0.0% n=0
BMI 30-44 Obese/Biopsy (n=18)	11.1% n=2	61.1% n=11	27.8% n=5	0.0% n=0
BMI ≥35 Severe obesity/Sleeve (n=55)	21.8% n=12	30.9% n=17	23.6% n=13	23.6% ★ n=13

Figure 3

BMI–coccoid form relationship: corpus coccoid category distribution by BMI group.

Color intensity reflects the percentage of patients within each BMI group falling into that coccoid category (darker blue = higher proportion). ☒ The ≥40% coccoid category was exclusively observed in the severe obesity/sleeve group (23.6%; p<0.001 vs. both other groups). Chi²=84.6, p<0.001 (overall). BMI <30 group: n=48 of 68 enrolled with evaluable corpus coccoid data. BMI 30–44 group: n=18 of 33 enrolled. Severe obesity/Sleeve group: all 55 enrolled had evaluable corpus data.

Supplementary Files

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- [Sleeveanonim.xlsx](#)