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GASTRITIS OLGA-STAGING & GASTRIC CANCER RISK:

A TWELVE YEAR CLINICO-PATHOLOGICAL FOLLOW-UP STUDY

MASSIMO RUGGE, MD,^{1,2} MICHELE DE BONI, MD,³ GIANMARIA PENNELLI, MD,¹ MANUELA DE BONA, MD,³
LUCIANO GIACOMELLI, BSc,⁴ MATTEO FASSAN, MD,¹ DANIELA BASSO, MD,^{1*} MARIO PLEBANI, MD,^{1*} &
DAVID Y GRAHAM, MD⁵

1 Department of Diagnostic Medical Sciences & Special Therapies (Pathology & Clinical Biochemistry* Units) University of
Padova – Italy

2 Istituto Oncologico del Veneto IOV-IRCCS - Padova, Italy

3 Department of Gastroenterology - Feltre Hospital- Feltre, Italy

4 Department of Pathology - Azienda Ospedaliera di Padova - Padova, Italy

5 Digestive Disease Division - Veterans Administration Hospital - Baylor College of Medicine - Houston, TX-USA

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CORRESPONDENCE TO:

Massimo RUGGE, M.D., F.A.C.G.
Cattedra di Anatomia Patologica
Università degli Studi di Padova
Istituto Oncologico del Veneto IOV-IRCCS
Via Aristide Gabelli, 61
35121 - Padova – Italia
Phone: (+39) 049 821 8990
Fax: (+39) 049 827 2277
e-mail: massimo.rugge@unipd.it

SUMMARY

Background. Intestinal-type gastric cancer (GC) still ranks among the high-incidence, highly lethal malignancies. Atrophic gastritis is the cancerization field in which GC develops. The current histological reporting formats for gastritis do not include any (atrophy-based) ranking of GC risk. *Aim.* This clinico-pathological long-term follow-up study tested the gastritis OLGA-staging in prognosticating neoplastic progression. *Methods.* Ninety-three Italian patients were followed up for more than 12 years (range=144-204 months). Clinical examinations, pepsinogen serology, endoscopy and histology (also assessing *H. pylori* status) were done both at enrolment (T1) and at the end of the follow-up (T2). *Results.* All invasive or intra-epithelial gastric neoplasia were consistently associated with high-risk (III/IV) OLGA stages. There was a significant inverse correlation between the mean pepsinogen ratio and the OLGA stage (test for trend; $p<0.001$). OLGA staging at T1 predicted both the OLGA stage (Kaplan Maier log rank test, $p=0.001$) and the neoplasia at T2 (Kaplan Maier log rank test, $p=0.001$). *Conclusions.* This long-term follow-up study provides the first evidence that gastritis OLGA-staging conveys relevant information on the clinico-pathological outcome of gastritis and, therefore, for patient management. According to OLGA-staging and *H. pylori*-status, gastritis patients could be confidently stratified and managed according to their different cancer risk.

INTRODUCTION

Gastritis is an inflammatory condition of the gastric mucosa with different etiologies, phenotypic expressions, and natural histories.^{1,2} Worldwide, *Helicobacter pylori* (*H. pylori*) is by far the most common etiological agent of non-self-limiting gastritis. Longstanding inflammation of the gastric mucosa is thought to trigger a cascade of genotypic and phenotypic derangements that may eventually result in intestinal-type (or epidemic) gastric cancer (GC).^{3,4}

Gastric mucosa atrophy is the soil in which gastric cancer most frequently develops and different patterns of atrophic-metaplastic gastritis are associated with different GC risks.⁴⁻⁹ A new system for gastritis histology reporting (the OLGA staging system) was recently established. Basically, exploiting experience gained by applying the Sydney System and other similar proposals,¹⁰⁻¹² OLGA-staging applies a vocabulary borrowed from oncology to rank the severity of the cancer risk due to atrophic gastritis. By simply combining atrophy scores relating to the antral mucosa with those obtained for the corpus mucosa (Figure 1), the stage should predict the gastritis-associated GC risk.^{13,14} Trials conducted in various epidemiological settings consistently associated GC with OLGA stages III/IV.¹⁵⁻¹⁷

Pepsinogens are proteolytic enzymes produced by gastrointestinal epithelia: pepsinogen I (PgI) is virtually found only in the gastric corpus, whereas pepsinogen II (PgII) is produced by both antral and oxyntic epithelia. Gastric mucosal atrophy results in a (progressive) decline in both serum PgI and the PgI/PgII ratio.¹⁸⁻²² Pepsinogen testing is widely used in Japan to assess GC risk.²³

The aim of the present long-term follow-up study was to test the efficiency of the OLGA staging system and gastric serology in prognosticating GC risk. With this aim, a consecutive series of dyspeptic patients was endoscopically, serologically and histologically

assessed at the beginning of the study, then the same patients were further tested at least 12 years later, using similar procedures.

MATERIALS AND METHODS

Patient recruitment

This long-term follow-up study involved the residents of a mountain region of Northern Italy (the Vanoi valley), which is known to be a high-risk area for gastric malignancies (270/100,000/5 years [years 1988-1992]).²⁴

The initial study population consisted of 100 consecutive dyspeptic subjects referred to the Gastroenterology Unit of the Regional Hospital in Feltre between April 1993 and April 1994 (T1). All the subjects considered underwent upper GI endoscopy with standardized gastric biopsy sampling under the personal supervision of one of the authors (MdB). A serum sample was also collected at the same time. No less than 12 years (T2) after their initial endoscopy, all the subjects were asked to return to the same Gastroenterology Unit for a clinical and endoscopic check-up: 7 failed to turn up. The remaining 93 subjects constituted the follow-up study population (mean follow-up= 149 months; range=144-204). In accordance with the 1975 Helsinki Declaration, informed consent was obtained from all the patients.

Clinical and endoscopic assessment (T1 & T2)

At both T1 and T2, the subjects underwent clinical examination, gastro-esophageal endoscopy, biopsy mucosa sampling and serum sampling. In all cases (at T1 and T2), at least 5 biopsy samples of the gastric mucosa were obtained (2 from the anterior and posterior walls of the antrum, 1 from the *angularis incisura* and 2 from the anterior and posterior walls of the oxyntic stomach). Any focal lesion was noted and sampled too. Any presence of peptic ulcers (active or scars, both duodenal and gastric) was recorded. All endoscopy procedures were performed or supervised by one of the authors (MdB).

Histology assessment (including *H. pylori* status)

At the end of the follow-up, all (T1 and T2) histology specimens (modified Giemsa stain for *H. pylori*, and Alcian blue periodic acid Schiff for intestinal metaplasia) were coded and jointly assessed by two pathologists (GP & MR), who were blinded to all clinical information (including the timing of the endoscopy).

The inter-observer consistency in assessing the OLGA stage between two pathologists involved in the study (MR & GP) was tested in 50 randomly-selected biopsy sets and interpreted in accordance with the Landis & Koch benchmarks (<0.4=poor agreement; 0.41-0.8=moderate/good agreement >0.8=excellent agreement).²⁵

At both T1 and T2, *H. pylori* status was assessed histologically (by modified Giemsa staining) and classified as following: a) persistent *H. pylori* infection, when the bacterium was detected histologically at both T1 and T2 (34 cases); b) native *Hp*-negative patients, when no *H. pylori* was detected at either T1 or T2 (11 cases); c) eradicated *Hp*-negative patients, who had revealed *H. pylori* at T1 but not at T2 (in 48 cases, eradication was confirmed by the patients' general practitioner [GB]).

At single biopsy level, mucosal atrophy was scored according to the OLGA Visual Analogue Scales; the stage of gastritis was assessed as detailed in a tutorial manuscript (Figure 1).^{14,16,17}

The histological assessment of neoplastic lesions was consistent with internationally validated criteria.^{14,16,26} Within the spectrum of gastric Intra-Epithelial Neoplasia (IEN), the categories considered were: (a) indefinite for IEN, (b) low-grade IEN (LG-IEN), and (c) high-grade IEN (HG-IEN). Gastric cancer was diagnosed in the presence of neoplastic epithelia infiltrating the lamina propria.^{27,28}

Serology

Blood was sampled immediately before endoscopy at T1 and T2. The serum was immediately separated and stored at -80°C until it was assayed for pepsinogens (PgI and PgII) (Biohit®, Helsinki, Finland) and the PgI/PgII ratio was calculated. All serology tests were performed under the supervision of one of the authors (DB).

Statistical analysis

Fisher's exact test, the Mann-Whitney test, the modified Kruskal Wallis non-parametric test for trend, and the Wilcoxon non-parametric test for matched pairs were used, as appropriate. The strength of the association between the variables considered and the OLGA stages was assessed by means of an ordinal logistic regression model (where an odds ratio [OR] of 1 means no association).

Sensitivity, specificity, and the positive (PPV) and negative (NPV) predictive values of the pepsinogen ratio at T2 were calculated for both high OLGA stages and neoplasia (at T2).

The inter-observer consistency in assessing the OLGA stage was ranked as "excellent" (K coefficient= 0.84).²⁵

The patients' clinico-pathological outcome (at T2) was correlated with the OLGA stage at T1 using the Kaplan-Meier method, comparing the groups using the log rank test. Significance was inferred at a p value of less than 0.05%. The statistical analysis was performed by one of the authors (LG) (STATA software; Texas-USA).

RESULTS

Patients at enrolment (T1)

The demographics and clinical data on the patients are summarized in Table 1.

Forty-eight patients were male (M/F= 1.07). The patients’ mean age at enrolment was 55 years (range: 22-73). Their *H. pylori* status at T1 is shown in Table 1 (none of the patients had been given anti-*H. pylori* therapy before recruitment). Among the 82 *Hp*-positive patients, the M/F ratio was 1.2 (mean age = 55 years; range 22-73). The M/F ratio among the 11 native *Hp*-negative subjects was 0.4 (mean age= 55 years; range 37-70).

The patients' distribution by OLGA stage and their mean age by stage are shown in Figure 2. In total, 83 out of 93 patients (89%) were staged as 0+I+II (*i.e.* low-risk stages); among them, no neoplastic lesions (either intra-epithelial or invasive) were encountered. Among the remaining 10 patients, 2 cases of low-grade IEN were seen (both associated with OLGA stage III).

The initial endoscopy detected 21 peptic ulcers (duodenal=19; gastric= 2) (Table 1). The duodenal ulcers (active= 7; scars= 12) showed no gender preference or significant association with the age brackets considered. All the duodenal ulcers identified at T1 occurred in *Hp*-positive patients, as did all the gastric ulcers (active= 0; scars= 2; M/F ratio= 2/0).

The pepsinogen ratios by stage (mean PgI/PgII and range) are given in Figure 2. A significant association emerged between mean PgI/PgII values and OLGA-stage (the lower the ratio, the higher the stage; by ordinal logistic regression: OR= 0.82; 95%CI= 0.72-0.94; p<0.006). The mean PgI/PgII ratio declined significantly as the OLGA-stage increased (test for trend; p<0.001). Significantly different mean PgI/PgII ratios were associated with low- versus high-risk stages (Stages 0+I+II: mean PgI/PgII= 6.80; SD= 4.63; Stages III+IV: mean PgI/PgII= 2.98; SD=1.7; Mann Whitney: p<0.002; Figure 3)

Gastritis stage at follow-up endoscopy (T2)

Figure 2 shows the patients' distribution by OLGA stage at T2. At the follow-up endoscopy, 6 patients had intra-epithelial or invasive neoplasia: 2 were indefinite for IEN, 1 was a LG-IEN (ascertained when the T1 histology specimens were reconsidered for this study, but overlooked at the initial diagnosis), 1 was a HG-IEN, and 2 were invasive carcinomas (one of them enrolled as LG-IEN). Both invasive cancers were located distally (1 case pTNM Stage II; 1 case pTNM Stage III).²⁹

Endoscopy at T2 also identified 9 ulcers or ulcer scars (duodenal= 7 [1 active]; gastric=2). All the duodenal ulcers occurred in Stage 0, *Hp*-eradicated patients. Both the gastric ulcers (1 in an *Hp*-eradicated patient and 1 in a case of persistent *H. pylori* infection, neither of them associated with NSAID therapy) were not documented at T1; they were both active and coexisted with GC.

The significant association between the PgI/PgII ratio and the OLGA stage was confirmed at T2 (ordinal logistic regression, OR= 0.71; 95%CI = 0.60-0.87; p<0.001). At T2, the mean PgI/PgII ratio was again significantly lower, the higher the stage of gastritis (test for trend; p<0.001). Taking all patients with OLGA stages 0+I+II together (low-risk stages), the mean PgI/PgII ratio (= 5.63, SD = 2.94) differed significantly from that of the cases with high-risk (III+IV) OLGA stages (= 2.94; SD =2.35) (Mann Whitney: p<0.001; Figure 3).

At T2, the median PgI/PgII ratio for cases with neoplasia (IEN+invasive adenocarcinoma) was lower (but not significantly so) than for non-neoplastic cases (median PgI/PgII ratio for neoplastic patients=2.10 *versus* median PgI/PgII ratio for non-neoplastic patients=5.07; Mann Whitney: p=0.09).

The power of the pepsinogen ratio at T2 to discriminate high-risk OLGA stages and neoplasia at T2 was assessed by assuming a PgI/PgII ratio cut-off ≤ 3.00 ²¹. In discriminating

high-risk *versus* low-risk OLGA stages, serology showed a 77% sensitivity (95%CI=54%-99%), 85% specificity (95%CI=77%-92%), 45% positive predictive value (PPV; 95%CI=24%-66%), and 96% negative predictive value (NPV; 95%CI=87%-100%). In discriminating cases with *versus* without neoplasia at T2, serology showed a 67% sensitivity (95%CI=29%-100%), 80% specificity (95%CI=70%-88%), 18% PPV (95%CI=2%-34%) and 97% NPV (95%CI=90%-100%).

Gastritis outcome (stages at T1 *versus* T2) and *Helicobacter pylori* status

The gastritis outcome (OLGA stage) vis-à-vis *H. pylori* status is given in Figure 4, which shows the OLGA stages at the baseline and at the end of the follow-up (the diagonal squares indicate the number of cases with the same stage at T1 and T2). Among the patients with persistent *H. pylori* infection (34 cases [Figure 4A]; mean follow-up = 12 years; range= 12-17), those whose stage of gastritis was worse at T2 significantly exceeded the number whose gastritis had improved (Figure 4A; Fisher’s exact test, $p<0.001$). Among the *H. pylori* eradicated patients (48 cases [Figure 4B]; mean follow-up= 12 years; range= 12-15), there was a significant prevalence of lower stages at T2 (Figure 4B; Fisher’s exact test, $p<0.006$). The difference in patients’ outcome according to their *H. pylori* status was statistically significant, i.e. 17/34 cases (50%) with persistent *H. pylori* infection had a higher OLGA stage, as opposed to 2/48 (4%) whose *H. pylori* had been eradicated, while the OLGA stage decreased in 3/34 cases (9%) with persistent *H. pylori* infection and in 8/48 cases (17%) after *H. pylori* eradication (Fisher’s exact test, $p<0.005$).

Based on the low cancer risk associated with stages 0, I, and II (as opposed to the high risk associated with stages III and IV), patients at T1 were merged into two groups (Stages 0+I+II= 83 *versus* Stage III+IV= 10). For these two cohorts, the OLGA stage at T1 significantly predicted patient outcome at T2. In fact, a high OLGA stage at enrolment predicted both a high OLGA stage [Kaplan Maier log rank test, $p= 0.001$; RR= 18.56,

95%CI= 5.54-62.14, $p= 0.001$], and a higher risk of neoplasia at T2. [Kaplan Maier log rank test, $p= 0.0001$; RR= 58.00, 95%CI= 5.67-592.53, $p= 0.001$]).

DISCUSSION

Despite its declining incidence, GC (especially the epidemic intestinal-type variant mainly associated with *H. pylori* infection) is still a highly lethal malignancy.³⁰

Primary GC prevention through *H. pylori* eradication is consistently recommended.³¹⁻³³ As for secondary prevention strategies, extensive gastric atrophy and (even more) IEN unequivocally identify patients at high risk of invasive adenocarcinoma. Unfortunately, both the histology profile and the clinical management of the patients with atrophy who warrant endoscopic follow-up are inconsistently defined.³⁴

The OLGA staging system was developed to deliver to clinicians a simple (prognostically useful) information on the gastritis-associated GC risk.¹³ Previous cross-sectional studies associated low-stage gastritis (0, I, II) with a low risk of GC.³⁵ In contrast, the consistent association of GC only with OLGA-stages III/IV identifies this (low-prevalence) population as an elective target for endoscopic surveillance.^{16,17}

In this long-term follow-up study, the prognostic impact of OLGA staging was tested in a consecutive series of dyspeptic outpatients. Both at enrolment (T1) and at the end of the follow-up (T2), the vast majority of the patients (89% and 86%, respectively) featured low-stage (0-I-II) gastritis. At both time points, all the neoplastic lesions encountered were clustered in the very low minority of patients featuring high-risk (III and IV) stages.

The long-term follow-up demonstrated that a high-stage gastritis at enrolment significantly predicted neoplasia at the end of the follow-up. Such a stage-related difference in

long-term outcome would represent a solid rationale for further exploring the impact of a stage-tailored follow-up in a larger population.^{16,17}

The clinico-biological consistency of the results achieved is further supported by the association between OLGA stages and peptic ulcer disease. Consistently with the inverse correlation between duodenal ulcer and GC risk, 18/19 cases of duodenal ulcer at T1 clustered in low OLGA stages; among these patients, *H. pylori*-positive gastritis significantly prevailed.^{1,2,4} Conversely, 3/4 gastric ulcers detected in this study (2 at T1, and 2 at T2) were associated with high-stage gastritis (both ulcers found at T2 coexisted with GC).^{1,2,4}

A significant correlation emerged between OLGA stages and pepsinogen serology.³⁶ The pepsinogen ratio dropped progressively with increasing gastritis stages, suggesting the wisdom of applying an integrated (non-invasive and invasive) strategy in secondary GC prevention.³⁷ In fact, by using a PgI/PgII ratio cut-off commonly used in the literature ($\text{PgI/PgII} \leq 3$), the high negative predictive value of serology in identifying both low-risk stages (NPV 96%) and non-neoplastic cases (NPV 97%) identifies patients who could be confidently excluded from any further endoscopic procedures.²³

Consistently with the notion that the PgI/PgII ratio gives an adequate picture of the severity of atrophy, but is not a biological marker of neoplastic transformation, the pepsinogen ratio failed to discriminate between neoplastic and non-neoplastic disease among cases with high-stage atrophic gastritis. As a consequence, serology tests (possibly including both the pepsinogen ratio and *H.pylori* status [see below]) only discriminated, from among the huge population of dyspeptic patients, the cases in whom second-line procedures (i.e. endoscopy and biopsy) can consistently assess a more “personalized” cancer risk. An additional finding of the present study is the divergent long-term outcome of gastritis depending on *H. pylori* status: even in the present (relatively small) cohort of patients,

etiological therapy resulted in an improvement in gastritis stage, whereas it worsened in association with persistent infection.³¹⁻³³

In conclusion, this long term follow-up study, shows that OLGA staging (and its functional serology counterpart) significantly prognosticated the gastritis-associated GC risk. On these bases, *H. pylori*-negative patients with low OLGA stages could be confidently excluded from any further invasive procedures (addressed to secondary GC prevention),³⁷ whereas patients with OLGA stages III-IV are definitely candidates for endoscopic surveillance.

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Table 1. Demographics and clinico-pathological data on patients at initial (T1) and final (T2) examination (mean follow-up= 149 months; range=144-204).

VARIABLE	T1	T2	STATISTICS
Gender: 48 males/45 females			
Mean age (range)	55 (22-73)	67 (34-85)	Wilcoxon's for matched pairs <i>p</i> = 0.0001
<i>H. pylori</i> status			
- positive	82	34	Fisher's exact test <i>p</i> =0.03
- negative	11	11	
- eradicated	0	48	
Peptic ulcer			
- duodenal (scars)	19 (12)	7 (6)	Fisher's exact test <i>p</i> = not significant
- gastric (scars)	2 (2)	2 (0)	
PgI/PgII ratio			
- mean	6.3	5.3	Wilcoxon's for matched pairs <i>p</i> =0.004
- range	0.5-27.1	0.7-18.0	
OLGA stage			
- 0	58	54	Fisher's exact test <i>p</i> =0.001
- I	16	16	
- II	9	10	
- III	6	7	
- IV	4	6	
Neoplasia			
- Absent	91	87	Fisher's exact test <i>p</i> =0.01
- Indefinite for IEN	0	2	
- Low-grade IEN	2	1	
- High-grade IEN	0	1	
- Invasive carcinoma	0	2	

Legends: Peptic ulcers= among brackets the number of scars; Pg=pepsinogen; IEN=Intra-Epithelia Neoplasia

CAPTIONS

Figure 1: The OLGA staging system: the stage of gastritis results by combining the atrophy score values as obtained in antral and oxyntic biopsy samples.

Figure 2: Patients' distribution by OLGA stage at T1 and T2. Pepsinogens ratio, prevalence of peptic ulcers GU: gastric ulcer; DU: duodenal ulcer) and neoplasia are also shown.

Figure 3: PgI/PgII ratio in low-risk (Stages o+ I+II [box A]) and high risk (Stages III+IV [box B]) OLGA-stages at the entry (T1) and at the end (T2) of the follow-up. The mean values of the ratio were consistently higher in low-risk than in high-risk stages (T1: Low-risk stages: mean PgI/PgII= 6.80; SD= 4.63; High-risk stages: mean PgI/PgII= 2.98; SD=1.7; Mann Whitney: $p<0.002$; T2: Low-risk stages: mean PgI/PgII= 5.63; SD= 2.94; High-risk stages: mean PgI/PgII= 2.94; SD= 2.35; Mann Whitney: $p<0.001$).

Figure 4: OLGA-stage (o, I, II, III, IV) at the entry (T1) and at the end of the follow-up (T2) in the subgroup of those 34 patients who retained the *H. pylori* infection (A), and of those 48 in whom *H. pylori* eradication was obtained (B). In the squares (diagonal) the patients who featured the same OLGA stage at both T1 and T2. Above the diagonal, cases in which the OLGA-stage at T2 was worse than that assessed at the entry (T1). Under the diagonal, cases in which the OLGA-stage at T2 was lower than that assessed at the entry (T1).

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Guarantor of the article: Dr Rugge had full access to all the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.

Specific author contributions:

MASSIMO RUGGE: Pathological assessment & clinico-pathological correlations

MICHELE DE BONI: Patient recruitment, clinical examination, and endoscopy assessment

GIANMARIA PENNELLI: Pathological assessment

MANUELA DE BONA: Patient recruitment

LUCIANO GIACOMELLI: Biostatistician

MATTEO FASSAN: Pathological assessment

DANIELA BASSO: Serology study

MARIO PLEBANI: Serology study

DAVID Y GRAHAM: Clinico-pathological correlations

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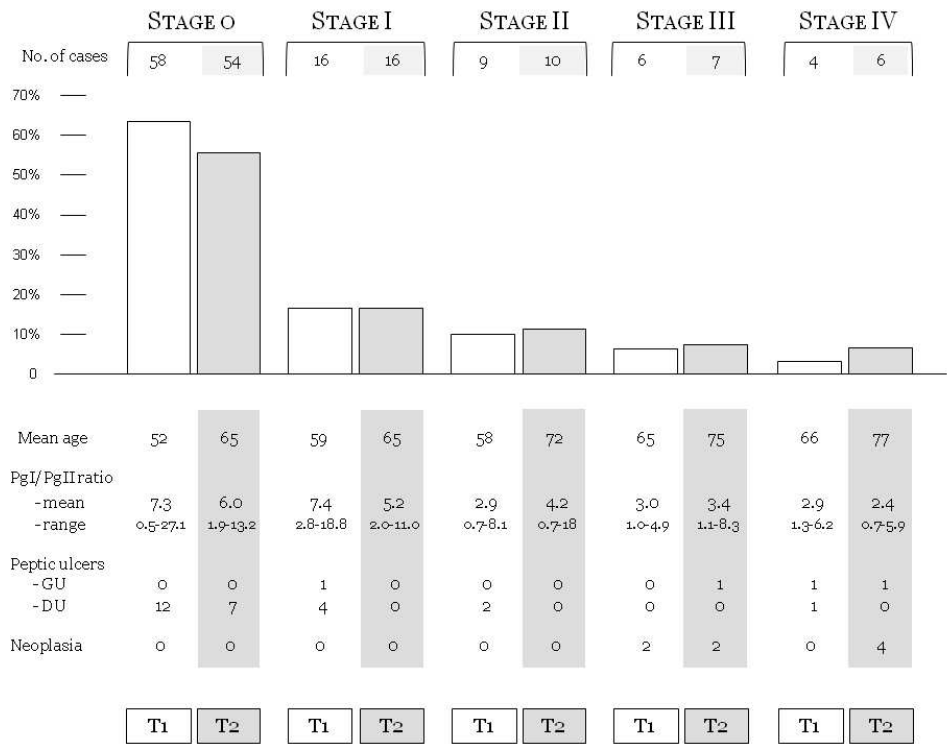
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Figure 1

		CORPUS			
ATROPHY SCORE		No atrophy (score 0)	Mild atrophy (score 1)	Moderate atrophy (score 2)	Severe atrophy (score 3)
A N T R U M	No atrophy (score 0) (including <i>incisura angularis</i>)	STAGE 0	STAGE I	STAGE II	STAGE II
	Mild atrophy (score 1) (including <i>incisura angularis</i>)	STAGE I	STAGE I	STAGE II	STAGE III
	Moderate atrophy (score 2) (including <i>incisura angularis</i>)	STAGE II	STAGE II	STAGE III	STAGE IV
	Severe atrophy (score 3) (including <i>incisura angularis</i>)	STAGE III	STAGE III	STAGE IV	STAGE IV

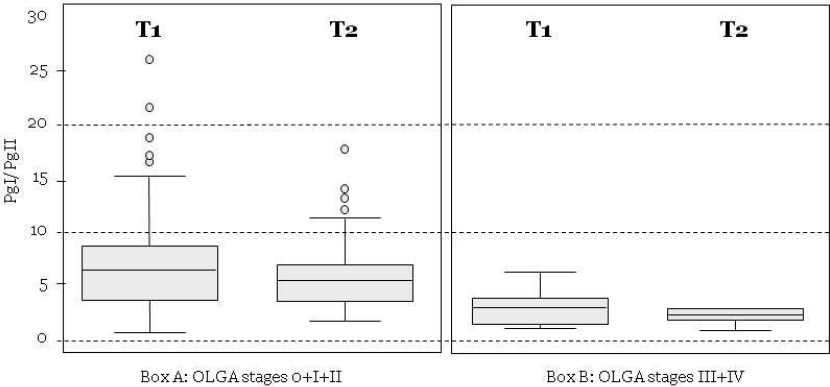
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Figure 2



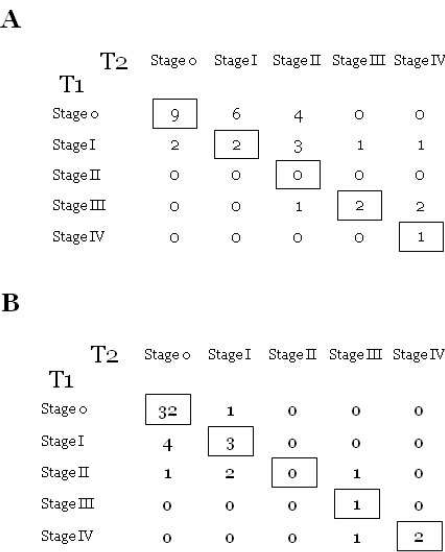
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Figure 3



254x300mm (96 x 96 DPI)

Figure 4 (A – B)



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