

Leading article

Prospects for intervention in gastric carcinogenesis: reversibility of gastric atrophy and intestinal metaplasia

For more than a quarter of a century it has been widely accepted that the majority of gastric cancers arise through a multistep process starting with chronic gastritis and progressing through atrophy, intestinal metaplasia (IM), and dysplasia to invasive carcinoma.¹ Corpus atrophy results in hypochlorhydria, which favours carcinogen formation, while atrophic gastritis and metaplasia are hyperproliferative states that favour mutagenesis. Thus atrophy and metaplasia are generally held to be premalignant conditions although the strength of the association is disputed. More recently, it has been established that infection with *Helicobacter pylori* is a major risk factor for gastric carcinogenesis and, in keeping with this, infection has been directly implicated in the development of atrophy and IM.²⁻⁴ This poses the question "Can eradication of *H pylori* reverse these premalignant conditions and interrupt the atrophy-carcinoma sequence?" In order to answer the question one first has to understand the biology and natural history of these conditions and then explore the possibilities for their reversal.

The nature of atrophy

Atrophy in the stomach is conventionally (and simply) defined as "loss of glands". Such loss may follow ulceration with destruction of the glandular layer or, more frequently, result from a prolonged inflammatory process in which multiple glandular units separately undergo destruction. However, atrophy can also be thought of as "a loss of specialised cells". Under this broader definition it is possible to include loss of parietal and chief (zymogenic) cells without glandular destruction. Such partial or "pre-atrophy" has been described in human autoimmune gastritis⁵ and is frequently encountered in animal models of both autoimmune gastritis⁶ and chronic *Helicobacter* infection.⁷ In these latter situations oxyntic cells are replaced within the intact glandular tubules by mucous neck cells (MNC). More interestingly, partial loss of parietal cells and replacement by MNC is a frequent but largely unrecognised finding in the inflamed corpus in *H pylori* gastritis.⁸ MNC were originally considered to be a transit cell population intermediate between stem cells and the fully differentiated parietal and chief cells. However, the repertoire of trefoil peptides and growth factors produced by MNC indicate that they are a distinct cell lineage which share the properties of other mucosally protective cell lineages in the gastrointestinal tract.⁹ Furthermore, it is apparent that proliferation of MNC explains the appearance of new glands formed in the wake of atrophy, so-called "pyloric metaplasia".

There are two principal routes to atrophy, one in which the stem cell compartment and/or glands are destroyed either by direct injury or as a consequence of the host inflammatory cell response, and the second when selective destruction of specialised epithelial cells occurs with preservation of stem cells. Both routes may apply in chronic *H pylori* infection; on the one hand bacterial toxins or, more

likely, proteases released by activated polymorphs could destroy glandular epithelium and stem cells¹⁰ while on the other autoantibodies are produced which react with epitopes on the proton pump in parietal cells.¹¹

The nature of intestinal metaplasia

Metaplasia represents a non-neoplastic change in cellular phenotype that usually arises in response to a sustained adverse environment. The altered phenotype is either a consequence of somatic mutation in stem cells or of epigenetic events which produce divergent differentiation in progeny cells. The subsequent emergence of the altered phenotype as the dominant population is a result of selection pressures exerted by the changed microenvironment.¹²⁻¹³ The pattern of gene expression determining cell phenotype is under the control of a complex hierarchy of transcription factors of which *homeodomain* proteins are important members. These proteins are themselves regulated by *homeobox* genes whose expression is pivotal to cellular differentiation and organogenesis.¹⁴ Thus the homeobox genes *Cdx-1* and *Cdx-2* are normally only expressed in the intestine but a corresponding transcription factor (CDX-1 protein) is present in the stomach.¹⁵ However, whether such expression results from a mutation in stem cells or epigenetic changes has not been determined. Certainly a wide range of genetic changes including telomere reduction, microsatellite instability, and mutations in p53, APC, and *k-ras* have been described in IM, even before the onset of dysplasia.¹⁶

IM can be divided into three subtypes which are likely to differ in their histogenesis and relationship to carcinogenesis—namely, type I (complete) which closely resembles small intestine, type II (incomplete—goblet cell metaplasia), and type III (incomplete—"colonic" type). Interestingly, it is the latter type that harbours most genetic changes¹⁷⁻¹⁸ and the only IM phenotype that carries a higher risk for gastric cancer.¹⁹

Is reversibility possible?

In any consideration of reversibility of atrophy a distinction has to be made between replacement of lost glands and regeneration of specialised cells within intact glands. In the latter situation the stem cell compartment is preserved and removal of an injurious factor could lead to regeneration of parietal and chief cells and full restoration of function. This has been clearly demonstrated in an animal model where withdrawal of a drug that induced selective loss of parietal cells was followed by complete restitution of the oxyntic mucosa.²⁰ Where glands and their associated stem cells have been completely destroyed, replacements would have to arise from adjacent intact pit gland units. In the developing mammalian stomach, multiplication of oxyntic glands occurs through a process of budding, duplication,

Abbreviations used in this paper: IM, intestinal metaplasia; MNC, mucous neck cell.

fission, and separation.²¹ Whether or not a similar process occurs in the adult is debatable. It seems clear that most regeneration following gland loss involves the MNC lineage but there is evidence, albeit from a small number of subjects, to indicate that full restoration of oxyntic glands can follow atrophy.²² When atrophy is combined with IM the scope for reversal is further limited. In IM the pit gland units are replaced by neocrypts in which the stem cell compartment is situated, as in the normal intestine, at the base of the crypt. While budding of intestinalised crypts is a distinct possibility it would be highly unlikely to give rise to normally arranged oxyntic glands, and would merely lead to expansion of the metaplastic foci.

IM is more likely to be reversible if it develops as an adaptation to an adverse factor that can be identified and removed. For instance, cytokines from chronic inflammatory cells and in particular Th2 helper lymphocytes may be responsible for “adaptive” IM in *H pylori* infection.²³ Certainly, intestinal cells cannot be colonised by the organism and will therefore enjoy a survival advantage. However, IM can have other causes (for example, bile reflux, high salt diet, and alcohol) some of which may be acting synergistically. This means that even in *H pylori* infected subjects, factors other than infection could be provoking or accelerating the metaplastic change. If IM is a consequence of stable somatic mutations in stem cells, changes in the immediate mucosal environment may not achieve reversal. While somatic mutation could be a factor in all types of IM it is interesting that the genetic lesions found in type III IM are similar to those found in gastric dysplasia (intraepithelial neoplasia).^{16–18, 24} These findings cast further doubt on the potential reversibility of this particular subtype of IM. Even when IM is a consequence of epigenetic changes, for example those resulting from methylation of genes involved in cell differentiation,²⁵ the scope for reversal may be strictly limited.

Does reversal of atrophy and IM occur?

There are several impediments to the proper assessment of reversibility, which include failings in histological interpretation, sampling errors, and “spontaneous regression”. When there is deep infiltration of the glandular layer by chronic inflammatory cells, separation of the glands can produce a spurious impression of atrophy. Resolution of inflammation subsequent to eradication of *H pylori* will lead, over several months, to a return to normal gland density. This is not reversal of atrophy but histological misinterpretation. Likewise, the assumption that a few small mucosal samples are representative of the whole is highly suspect when dealing with multifocal processes. Thus there are inherent difficulties in comparing atrophy and IM between baseline and follow up endoscopies. Finally, even in the absence of treatment, all follow up studies on chronic gastritis show apparent regression in a minority of subjects. In a large cohort follow up study in Colombia,²⁶ “regression” rates for the change from atrophy to normal (or superficial gastritis) and from IM to atrophy were 7.5 and 4.4/100 person years, respectively. While there was a net overall *progression*, the importance of sampling errors in giving rise to these results was emphasised.

Given that sampling errors will always confound histological interpretation it might be thought that indirect measures of atrophy would offer a more accurate estimate, but this approach is also flawed. It is now clearly established that hypochlorhydria and changes in serum pepsinogen I and II ratios can result from corpus inflammation alone.²⁷ Thus some patients with profound hypochlorhydria show a substantial increase in acid output after eradication of *H pylori* attributable to resolution of inflammation and not to regeneration of oxyntic glands.²⁸

Sampling errors are even more problematic in the assessment of IM. However, targeted biopsies from the margins of peptic ulcers and the subsequent “scar” clearly demonstrate that IM (usually of types I and II) frequently develops in regenerating epithelium and regresses with time.²⁹

What are the prospects for intervention?

With the realisation that *H pylori* plays a central role in gastric carcinogenesis the notion of intervention by eradication of infection has become increasingly attractive. Elimination of inflammation will have its own beneficial consequences but can atrophy and IM be expected to regress?

The published studies examining the reversibility of atrophy and IM following eradication of *H pylori* have yielded conflicting results. In general the longer the follow up the more likely the study will reveal regression of atrophy but the magnitude of change is small. The consensus of recent studies on IM status is that there is no discernible change. A more substantial benefit is claimed in the first reported randomised intervention trial of the effects of *H pylori* eradication and dietary supplementation with vitamin C or β -carotene.³⁰ This was carried out in a high incidence gastric cancer region (in Colombia) on 852 infected subjects who had atrophic gastritis and/or IM at entry and who were biopsied at 36 and 72 months following intervention treatment. The authors concluded that elimination of *H pylori* produced “a marked and statistically significant increase in the rate of regression of the precursor lesions”. But similar relative risks were obtained for the other treatments alone and, as Blot has pointed out,³¹ it is disturbing that anti-*H pylori* treatment was effective when given alone but conveyed no (added) benefit when given with vitamin C or β -carotene. Although the results are promising, Blot concluded that in view of the lack of consistency, the findings should be interpreted with caution. Perhaps further caution is called for when it is appreciated that a change from IM at baseline to multifocal atrophic gastritis at follow up qualifies as “regression” according to their histological protocol.

While true regression of atrophy and IM depend on the capacity of the mucosa to regenerate specialised glandular tissue, at least proper identification and elimination of the cause could be expected to prevent progression. However, it is clear that atrophy and IM have a number of causes—bile reflux, dietary irritants, and autoimmunity, as well as *H pylori* infection. Removal of infection may therefore be insufficient to produce reversal.

It seems most likely that regeneration of normal oxyntic glands following true glandular atrophy with replacement fibrosis will at least be limited. Likewise, restoration of normal differentiation in IM is improbable in the presence of stable mutations in stem cells. In effect, certain forms of atrophy and IM have passed a “point of no return” and reversal becomes impossible. In my view the hope that intervention by elimination of *H pylori* will of itself lead to substantial reversal of atrophy and IM is an unrealistic expectation. However, a more realistic outcome has been established already. *H pylori* eradication leads to resolution of inflammation, elimination of DNA damage by reactive oxygen species, a reduction in cell turnover, a rise in acid output in hypochlorhydric subjects, and a gradual return of ascorbic acid secretion into the gastric juice. These proven consequences could be much more important in the prevention of gastric cancer than the hoped for reversal of atrophy and IM.

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