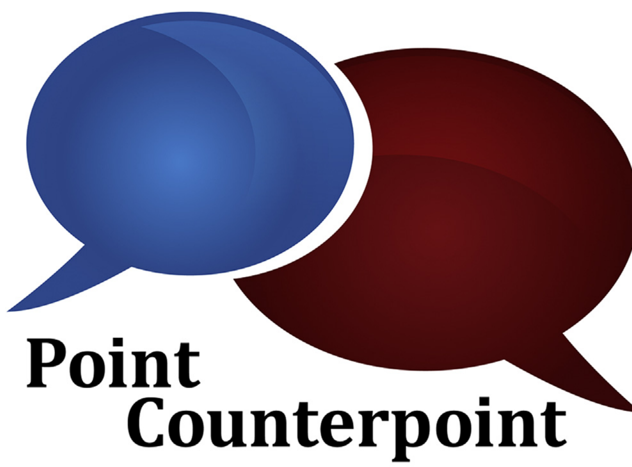
Isthmus Stem Cells Are the Origins of Metaplasia in the Gastric Corpus



The acquisition of genetic/epigenetic mutations in long-lived gastrointestinal stem cells leads to the development of cancer, as well as precancerous lesions such as metaplasia and dysplasia. In the proximal stomach corpus, this model of progression from stem cells has been supported by studies in mice and human beings, showing abundant proliferation in the isthmus and clonal expansion of mutated cells from the stem cell region. An alternative theory proposes that gastric metaplasia arises from mature differentiated chief cells. Despite reports of low levels of proliferation in chief cells in acute injury models, there is little evidence for reprogramming of chief cells into long-lived stem cells that continuously supply progeny over time. Critical flaws in the chief cell transdifferentiation theory include the definition of acute SPEM, the chief cell-damaging effect of chemical reagents, and the specificity of chief cell lineage tracing. In contrast, there is now strong evidence regarding the stem cell origins of gastric metaplasia that refutes the transdifferentiation theory. Here, we briefly review the history and definition of gastric metaplasia, and outline in detail the evidence that supports the stem cell origin of metaplasia. *(Cell Mol Gastroenterol Hepatol 2017;4:89–94; http://dx.doi.org/10.1016/j.jcmgh.2017.02.009)*

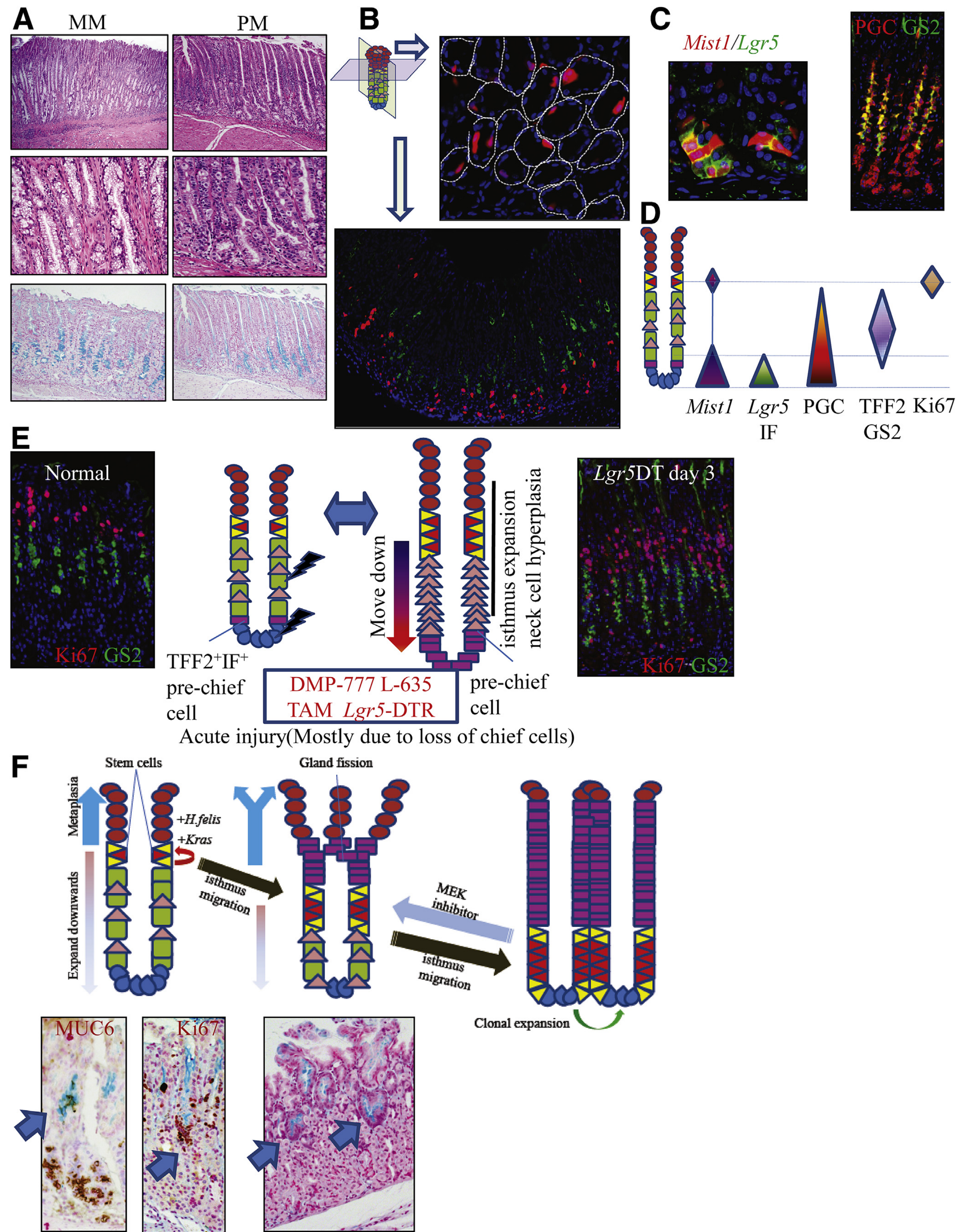
Metaplasia of the stomach gained increasing recognition when a link to gastric adenocarcinoma was noted and the Correa pathway was proposed. Although classic intestinal metaplasia (IM) with goblet cell differentiation initially received most of the attention, spasmolytic polypeptide-expressing metaplasia (SPEM) recently has attracted greater interest. SPEM was first characterized by "a marked expansion of an aberrant gastric mucous cell lineage that stained positive for spasmolytic polypeptide" in *Helicobacter felis*–infected mice (spasmolytic polypeptide was the original name given to trefoil factor family 2 [TFF2]).[1]

Helicobacter species induce a variety of histopathologic changes in mice, including oxyntic atrophy (loss of corpus chief and parietal cells), surface mucous pit-cell hyperplasia, mucous metaplasia (MM), and pseudopyloric metaplasia (PM).[2] In this process, chief cell disappearance precedes parietal cell loss and the development of SPEM.[2] Both MM and PM are classified as SPEM that expresses neck cell markers TFF2, gastric mucin-6 (MUC6), and Griffonia simplicifolia leaf lectin II (GS2), but they need to be distinguished. SPEM-MM is characterized by large, foamy TFF2^{+} cells that secrete neutral and acid mucins and replace lost parietal and chief cells (Figure 1*A*). In addition to morphologic differences from normal mucous neck cells, SPEM-MM expresses unique markers (CD44 and Sox9) that are absent in normal neck cells. Thus, MM is clearly a form of metaplasia and not simply neck cell hyperplasia. In contrast, SPEM-PM occurs later and includes less differentiated cell types that resemble the pyloric antrum. Dysplasia emerges after SPEM-PM is established.[2]

An entity similar to TFF2-expressing PM in mice was recognized in human gastric tissue in 1999 and the name SPEM formally was proposed to encompass TFF2-expressing metaplasia. SPEM development was linked to mucosal injury associated with parietal and chief cell loss, particularly in chronic *Helicobacter* species infection. Although initial studies pointed to SPEM as a preneoplastic lesion, knockout of the signature peptide, TFF2, in mice accelerated gastric inflammation and carcinogenesis, suggesting a possible role for TFF2 as a tumor suppressor.[2]

Observations in Patients Indicate a Stem Cell Link

Analysis of resected gastric specimens showed the frequent co-existence of SPEM and IM in the same compound glands. This raised the question of whether SPEM originated from tissue resident stem cells or from another source. The stability and durability of IM and SPEM suggests that they are maintained by a self-renewing stem cell. Although there was an implicit assumption that metaplasia arises from epigenetic changes in multipotent gastric stem cells, more recent studies[3] have shown that metaplastic gastric glands are clonal, maintained by multiple stem cells, and can form large patches that spread by glandular fission.


A
MM
PM
B
C
Mist1/Lgr5
PGC GS2
D
Mist1
Lgr5
IF
PGC
TFF2
GS2
Ki67
E
Normal
Ki67 GS2
Move down
isthmus expansion
neck cell hyperplasia
Lgr5DT day 3
Ki67 GS2
TFF2⁺IF⁺
pre-chief
cell
DMP-777 L-635
TAM Lgr5-DTR
pre-chief
cell
Acute injury(Mostly due to loss of chief cells)
F
Stem cells
Gland fission
Metaplasia
Expand downwards
+H.felis
+Kras
isthmus
migration
MEK
inhibitor
isthmus
migration
Clonal expansion
MUC6
Ki67

Thus, chronic inflammation leads to reconstruction and expansion of niche components with changes in the location of proliferation outside of the isthmus, which may cause the migration of isthmus stem/progenitor cells, rather than generation of new progenitors.

Development of Short-Term Models Mimicking Gastric Metaplasia

Although SPEM typically requires many months to develop, several acute chemically induced SPEM models have been described, including DMP-777, L-635, and high doses of tamoxifen. These models involve chemically induced injury with gastric atrophy and the development of SPEM-like lesions. Rodents treated with DMP-777 were reported to develop TFF2-expressing metaplasia after 7–10 days. Similarly, administration of high doses of tamoxifen caused rapid parietal and chief cell loss, and subsequently increased GS2^{+} cells near the base.[4] The most rapid SPEM-like model involved treatment with L-635, which lead to TFF2-expressing metaplasia in 1 week, accompanied by massive inflammation.[5]

In these models, a subset of metaplastic cells expressed chief cell markers and arose primarily in the lower third of the corpus glands. These correlated with other data suggesting a chief cell origin for these lesions. This led the researchers to formally propose that SPEM may be derived from mature chief cells through "transdifferentiation."[5]

To test this hypothesis, investigators performed lineage-tracing in *Mist1*-CreERT;*Rosa26*-LacZ mice to genetically mark *Mist1*$^{+}$ chief cells and their progeny during SPEM development.[5] After DMP-777 treatment, the number of TFF2-expressing cells in the midgland region increased quickly, but most of these cells were not marked by *Mist1*-CreERT lineage, suggesting that they arose from neck progenitors or other cells that did not express *Mist1*.[5] Indeed, there was actually a significant decrease in the number of *Mist1*-traced chief cells after DMP-777 treatment,[5] suggesting that DMP-777 ablates not only parietal cells but also chief cells, as we confirmed recently.[6] Nevertheless, a rare population of the remaining *Mist1*-traced chief cells expressed TFF2, indicating that a subset of *Mist1*$^{+}$ chief cells (most often at the top of the chief cell zone) could contribute in a limited fashion to SPEM-like lesions.

The investigators also showed full *Mist1*-lineage tracing of SPEM in both L-635 and *H felis* infection mouse models,[5] and, more recently, the group confirmed our previous findings[6] that expression of mutant *Kras*^{G12D} in the *Mist1* lineage leads to the development of SPEM, which they showed could be completely reversed by a mitogen-activated protein/extracellular signal-regulated kinase kinase (MEK) inhibitor.[7] From these models,[5,7] they concluded that the metaplasia originated primarily from chief cells.

Weaknesses in the Conclusion That Metaplasia Originates From Chief Cells

Although the conclusion of a chief cell origin is supported by expansion of double-positive cells (intrinsic factor (IF)$^{+}$TFF2^{+}) and *Mist1* tracing, the transdifferentiation

Figure 1. (***See previous page***). (*A*) *Left*: Histopathology of SPEM-MM. H&E staining of 9-month-old *H pylori* SS-1 strain–infected TFF2 knockout mice (*top* and *middle*) and Alcian blue staining of 4-month-old *H pylori* PMSS-1 strain–infected wild-type mice (*bottom*). *Right*: Histopathology of SPEM-PM. H&E (*top* and *middle*) and Alcian blue (*bottom*) staining of 6-month-old PMSS-1–infected wild-type mice. (*B*) *Mist1*$^{+}$ isthmus stem cells are present in almost every gland. Cross-section (*top*) and axial section (*bottom*) of *Mist1*-CreERT; *R26*-TdTomato mouse corpus 1 day after tamoxifen induction. *Bottom* panel is stained with GS2 (green). *Mist1* (red) is expressed abundantly in the basal chief cell region, but also consistently in isthmus stem cells. Cross-sectional analysis showed that an average of 1–2 *Mist1*$^{+}$ isthmus stem cells are present in each corpus gland, but the number of *Mist1*$^{+}$ chief cells is much greater than of *Mist*1^{+} stem cells. (*C*) *Left*: *Lgr5* marks chief cells. Gland base of *Mist1*-CreERT; *R26*-TdTomato; *Lgr5*-DTR-EGFP mouse corpus 1 day after tamoxifen. *Lgr5* expression (green) is restricted at the base and overlaps with the majority of *Mist1*$^{+}$ chief cells (red). *Right*: GS2 (green) and PGC (red) staining in wild-type mice. PGC is expressed in both chief and neck cells. (*D*) Schema of expression pattern in mouse corpus glands of *Mist1*, *Lgr5*, IF, PGC, TFF2/GS2, and Ki67. Neck cell markers such as TFF2 or GS2 are found between *Mist1*$^{+}$ chief cells and *Mist1*$^{+}$ isthmus cells. Proliferation (Ki67^{+}) is found solely within the isthmus region in normal state. Evidence suggests the presence of chief cell precursors (pre-chief cells) in the lower neck/top of the chief cell region, which express both chief and neck cell markers. (*E*) GS2 (green) and Ki67 (red) staining in wild-type (*left*) and *Lgr5*-DTR (*right*) mice 3 days after treatment with diphtheria toxin. *Lgr5*–diphtheria toxin treatment efficiently ablates chief cells, and there is a marked expansion of proliferation at the isthmus after chief cell ablation. Pit cells and GS2-expressing neck cells are increased and show hyperplasia. Because of the absence of basal chief cells, isthmus and neck cells migrate rapidly downward to the base, and therefore GS2^{+}IF^{+} pre-chief cells can be localized at the base. These cells occasionally proliferate (Ki67^{+}) and expand, reflecting their neck progenitor function, to compensate and regenerate ablated chief cells. Other acute injury models including DMP-777, L-635, and high dose of tamoxifen (TAM), cause a rapid loss of parietal cells, as well as a reduction in chief cells, therefore the increase of basal TFF2^{+} cells in these models is the result of a downward shift of TFF2^{+} neck/pre-chief cells that can proliferate to regenerate chief cells. During this process, immature IF^{+}TFF2^{+} cells are produced from upper isthmus stem and pre-chief cells, not from dedifferentiation of ablated mature chief cells. Loss of parietal cells alone is not sufficient to induce such IF^{+}TFF2^{+} cell hyperplasia, therefore acute SPEM lesions in injury models likely are caused by loss of damaged chief cells. (*F*) Diagram of changes in the gastric glands with *Kras*^{G12D} activation in the *Mist1* $^{+}$ cells. Mutant *Kras* expression is induced in both *Mist1*$^{+}$ isthmus stem cells and chief cells, but only isthmus stem cells can give rise to metaplasia, and they migrate toward the base in a manner that can be reversed with a MEK inhibitor. *Kras*-mutated *Mist1*$^{+}$ isthmus stem cells are proliferating rapidly. *Bottom*: MUC6 (*left*) staining and Ki67 (*middle*) staining with Alcian blue at 7 days after tamoxifen induction. Because of rapid turnover, chief cells are depleted at the base and MUC6^{+} neck cells occupy the basal region at this point, but importantly these MUC6^{+} cells do not show SPEM-like morphology, mucins, or proliferation. *Bottom right*: Alcian blue staining at 14 days after tamoxifen induction. Metaplastic *Mist1*$^{+}$ isthmus cell cluster expand toward the base as well as laterally through gland fission, eventually replacing the entire stomach with SPEM cells.

hypothesis remains flawed. First, very few actively cycling cells are detected at the gland base in chemical injury models; the majority of proliferating cells originate from the isthmus. The notion that chief cells interconvert into progenitor cells and migrate from the base toward the isthmus, against the flow from the main corpus stem cells, challenges numerous well-established paradigms. Although, a similar model was proposed in a study of *Troy-CreERT* knockin mice,[8] the chief cell behavior they proposed was proven to be perturbed by *Troy* gene haploinsufficiency later.[6]

Second, induction of metaplasia by the chemical agents is completely reversible within 2 weeks. Thus, these drugs do not cause permanent reprogramming at the stem cell level. *Helicobacter* species–induced metaplasia persists for >1 year and thus must be sustained by a long-lived stem cell. Thus, the nature of drug-induced metaplasia appears to be different from that of classic SPEM.

Third, DMP-777 damages chief cells and dramatically reduces the number of *Mist1*-traced chief cells,[5,6,9] which accounts for the extension of metaplasia to the base of the gastric glands; this is likely the case with the very similar compound L-635. One of the earliest changes in the *H felis* mouse model is rapid chief cell disappearance, an event that precedes parietal cell loss.[2] Indeed, the notion that part of the SPEM induction requires chief cell injury has been supported by a recent study showing that parietal cell ablation alone is insufficient to induce metaplasia. Therefore, the cellular changes observed at the gland base after drug treatment simply may reflect chief cell regeneration.

Indeed, although cells that express TFF2/GS2 and IF near the base often are categorized as SPEM, their histologic features are quite different from classic SPEM (either SPEM-MM or SPEM-PM) identified in *Helicobacter* species–infected mice. The acute SPEM-like cells show a different appearance with limited expansion and less mucin.[4,5] We conclude that the increase in TFF2$^+$IF$^+$ cells likely represents expansion and migration of progenitors that are known to be TFF2$^+$IF$^+$ from the neck region rather than true metaplasia. The definition of SPEM needs to be reconsidered carefully and more sharply limited, rather than loosely categorizing cells as metaplasia based simply on transient TFF2/gastric intrinsic factor (GIF) positivity.

Finally, the main problem with the transdifferentiation hypothesis is that it was based almost entirely on the assumption that *Mist1* is a specific marker of chief cells and does not mark any additional stem/progenitor cells, which is not the case.[6]

Mist1 Is Expressed in Quiescent Stem Cells in the Isthmus

We showed that *Mist1* is expressed not only in chief cells but also in quiescent, self-renewing stem cells within the corpus isthmus.[6] There is an average of 1–2 isthmus *Mist1*$^+$ cells per gland, much less than the number of *Mist1*$^+$ chief cells (average 9–10/gland) at the glandular base (Figure 1*B*), which explains why isthmus expression previously was overlooked. We found abundant long-term lineage tracing from *Mist1*$^+$ cells, but analyses of detailed time course and multicolor fluorescent reporter mice showed that *Mist1*-derived tracing arose from the isthmus, but not from the gland base.[6] Consistent with this, ablation of >95% of *Lgr5*$^+$ chief cells using *Lgr5*-DTR-EGFP mice that express diphtheria toxin receptor (DTR) and green fluorescent protein (GFP) in *Lgr5*$^+$ cells did not reduce the numbers of long-term *Mist1*-derived tracing events (Figure 1*A* and Figure 1*C*–*E*).[6] In

Table 1. Evidence for Cellular Origins of Metaplasia

	Evidence for chief cells	Evidence for *Mist1*$^+$ isthmus stem cells
Proliferation	None in normal state, and minimal (<5%) in injury	Normally slow proliferation, and marked expansion in injury
BrdU labeling	No uptake in normal state, with rare labeling after injury (eg, 5-FU), but no upward migration[8]	Labeled cells migrate bidirectionally from the isthmus upward toward the pits and downward to the base
Lineage tracing	*Troy*-derived long-term tracing in haploinsufficient *Troy* knockin models,[8] but not in *Troy* BAC models[6] Chief cells express *Lgr5*, but no tracing from *Lgr5*$^+$ chief cells in normal and injury states[9]; ablation of *Lgr5*$^+$ chief cells does not decrease *Mist1*-derived tracing[6] No evidence that chief cells expand clonally	*Mist1*-CreERT labels the isthmus stem cell as well as chief cells, and tracing arises from the isthmus stem cell[6] 5-FU–induced stem cell ablation eliminates *Mist1* tracing[6] Confetti mice show clonal expansion from the isthmus[6] eR1-CreERT mice show lineage tracing from the isthmus[10]
DMP-777	TFF2$^+$IF$^+$ cells present in lower third of glands after treatment, but morphology of these SPEM cells is atypical and changes are transient (<2 wk) DMP-777 reduces *Mist1*-traced chief cell number[5,6]	*Mist1*$^+$ isthmus stem cell–derived tracing is not altered after DMP-777[6] Human gastric metaplasia is stable and clonal with a field effect, indicating stem cell origins[3]
Mutant *Kras*	Proliferating GS2$^+$IF$^+$ cells at the base after 1 month[7] eR1-CreERT with *Kras* mutation leads to occasional GS2$^+$ metaplasia near the gland base[10] Most chief cells do not proliferate even after *Kras* induction at early time points, and are replaced rapidly by the migration of isthmus-derived SPEM cluster	Mucous-producing proliferating GS2/MUC6$^+$ SPEM that is Alcian blue$^+$ is present early on only in the isthmus Gland fission is present early only in the isthmus *Lgr5*-DTR–mediated ablation of chief cells does not reduce metaplasia, whereas suppression of the isthmus stem cell (with 5-FU) blocks metaplasia[6] Metaplasia in eR1-CreERT;LSL-*Kras* mice starts primarily in the isthmus[10]

BrdU, bromodeoxyuridine; 5-FU, 5-fluorouracil.

contrast, when isthmus stem cells ablated with a well-defined 5-fluorouracil regimen,[8] *Mist1*-derived tracing events rarely were seen.[6] Thus, the long-term, lineage-tracing events in *Mist1*-CreERT mice derive from isthmus stem cells, rather than chief cells. Therefore, lineage tracing from *Mist1*$^{+}$ cells cannot be used to support the concept of chief cell transdifferentiation. Long-term *Mist1*-traced lineage must be assumed to emanate from the isthmus stem cell.

Kras^{G12D} expression in the corpus isthmus (*Mist1*$^{+}$) stem cell results in the development of mucus-producing metaplasia within 1 month.[6] Within 5–7 days after *Kras* activation of the *Mist1*$^{+}$ cells, Ki67^{+} metaplastic foci were seen within the isthmus (Figure 1*F*). In contrast, few Ki67^{+} cells present at the base of the glands or associated with chief cells. At day 14, there was evidence for gland fission with Alcian blue^{+} mucous metaplasia, but only in the isthmus region (Figure 1*F*). These proliferating isthmal cell clusters then migrated toward the base and expanded laterally via gland fission(Figure 1*F*). Thus, temporal analysis showed a downward migration of the stem cell zone, consistent with our earlier findings in the *CK19-Kras* mice and *Helicobacter*-infected mouse models.[1] Moreover, depletion of *Lgr5*$^{+}$ chief cells did not impair metaplasia development, consistent with earlier findings[9] showing that SPEM was not derived from *Lgr5*$^{+}$ chief cells. Therefore, *Mist1*$^{+}$ isthmus cells, rather than *Mist1*$^{+}$ chief cells, are the clear origin of *Kras*-induced metaplasia.

Summary of the Evidence in Favor of a Stem Cell Origin of Metaplasia

The presence of *Mist1*$^{+}$ isthmus stem cells accounts for the origin of metaplasia in the models discussed earlier (Table 1), as well as the results from other models. Our findings regarding *Kras*-induced metaplasia development[6] were confirmed by Choi et al,[7] who showed that *Kras*-induced SPEM subsequently gives rise to IM and even dysplasia. *Kras*-induced metaplasia stably persists over time, similar to *H felis*–induced metaplasia, suggesting reprogramming of long-lived stem cells rather than transient transdifferentiation. Indeed, when MEK inhibitors reverse the *Kras*-induced metaplasia,[7] normalization of gland structure occurs in a gland-by-gland fashion with upward regression of the isthmus lesion but without any proliferation in chief cells. This finding strongly argues that metaplasia indeed is generated by *Kras*-activated stem cells, with normal gland components renewed from normalized stem cells after MEK inhibition.

An occasional criticism of the argument for the isthmus origin of metaplasia is that *Mist1*$^{+}$ corpus stem cells appear scarce and seem insufficient to account for all of the *Kras*-induced metaplasia, which replaces nearly the entire stomach within 1–2 months.[6] Indeed, in the initial report, *Mist1*$^{+}$ isthmus stem cells were not well represented in the 5-um–thickness axial sections analyzed.[6] In fact, each gland actually contains 1–2 *Mist1*$^{+}$ isthmus stem cells, which more clearly are shown in cross-sectional analysis (Figure 1*B*). Although *Mist1*$^{+}$ chief cells are more abundant, *Kras* mutant chief cells rarely proliferate. In contrast, the majority of *Kras*-mutated *Mist1*$^{+}$ isthmus cells become rapidly proliferative with expression of neck cell markers and Alcian blue^{+} mucins, suggesting a 50- to 100-fold greater proliferative potential in *Mist1*$^{+}$ stem cells compared with chief cells (Figure 1*F*). In addition, the notion that the numbers of *Mist1*$^{+}$ isthmus cells are insufficient ignores the fact that *Kras*$^{+}$ glands are known to outcompete wild-type glands with lateral expansion in the neutral drift model (Figure 1*F*). A recent study with eR1-CreERT;LSL-*Kras*^{G12D} mice has shown similar results,[10] with Alcian blue^{+} metaplasia also arising in the isthmus, supporting in our view an isthmal origin for metaplasia. Although an occasional gland base showed pepsinogen C (PGC)$^{+}$ metaplasia in these mice, it should be noted that PGC also is expressed in neck cells or above (Figure 1*C*), thus it does not support chief cell origin.

Although our detailed analyses confirm that isthmus stem cells are the main source of SPEM in acute injury and *Kras* models, we cannot exclude the possibility that neck or chief cell precursors, residing immediately above mature chief cells, may contribute to short-term regeneration and metaplasia. The presence of chief cell precursors in this region initially was suggested by Karam and Leblond,[11] and indeed approximately 15% of normal neck/chief lineage cells express both GS2/GIF, and they appear to show occasional limited proliferation after injury (Figure 1*E*). If some of these cells express *Mist1*, they might contribute to some of the short-term GS2/GIF lesions.

Conclusions

Gastric metaplasia can be fully generated by the isthmus stem cell. There is as yet no evidence for clonal expansion or upward migration of mature zymogenic cells from the gland base, even after injury. The role of the *Mist1*$^{+}$ isthmus stem cell remains to be fully elucidated in chemical injury models, but the definition of SPEM also needs further clarification. Although there may be some degree of plasticity for chief cells, they likely are unable to generate long-lived stem cells and the stable, clonal metaplasia found in patients and *H felis*–infected mice. Nevertheless, chief cells or their progenitors still may contribute in limited ways to early stages of gastric repair. More studies are needed to understand the factors involved in regulating the altered differentiation of the isthmus *Mist1*$^{+}$ stem cell that leads to the production of long-term SPEM.

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References

1. Wang TC, Goldenring JR, Dangler C, et al. Mice lacking secretory phospholipase A2 show altered apoptosis and differentiation with Helicobacter felis infection. Gastroenterology 1998;114:675–689.
2. Fox JG, Rogers AB, Whary MT, et al. Accelerated progression of gastritis to dysplasia in the pyloric antrum of TFF2 -/- C57BL6 x Sv129 Helicobacter pylori-infected mice. Am J Pathol 2007;171:1520–1528.
3. Gutierrez-Gonzalez L, Graham TA, Rodriguez-Justo M, et al. The clonal origins of dysplasia from intestinal metaplasia in the human stomach. Gastroenterology 2011;140:1251–1260 e1–6.
4. Huh WJ, Khurana SS, Geahlen JH, et al. Tamoxifen induces rapid, reversible atrophy, and metaplasia in mouse stomach. Gastroenterology 2012;142:21–24 e7.
5. Nam KT, Lee HJ, Sousa JF, et al. Mature chief cells are cryptic progenitors for metaplasia in the stomach. Gastroenterology 2010;139:2028–2037.e9.
6. Hayakawa Y, Ariyama H, Stancikova J, et al. Mist1 expressing gastric stem cells maintain the normal and neoplastic gastric epithelium and are supported by a perivascular stem cell niche. Cancer Cell 2015;28:800–814.
7. Choi E, Hendley AM, Bailey JM, et al. Expression of activated Ras in gastric chief cells of mice leads to the full spectrum of metaplastic lineage transitions. Gastroenterology 2016;150:918–930.
8. Stange DE, Koo BK, Huch M, et al. Differentiated troy(+) chief cells act as reserve stem cells to generate all lineages of the stomach epithelium. Cell 2013;155:357–368.
9. Nam KT, O'Neal RL, Coffey RJ, et al. Spasmolytic polypeptide-expressing metaplasia (SPEM) in the gastric oxyntic mucosa does not arise from Lgr5-expressing cells. Gut 2012;61:1678–1685.
10. Matsuo J, Kimura S, Yamamura A, et al. Identification of stem cells in the epithelium of the stomach corpus and antrum of mice. Gastroenterology 2017;152: 218–231.
11. Karam SM, Leblond CP. Dynamics of epithelial cells in the corpus of the mouse stomach. I. Identification of proliferative cell types and pinpointing of the stem cell. Anat Rec 1993;236:259–279.

Conflicts of interest
The authors disclose no conflicts.

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