Reviewer #1:

Change the title: it is not "new therapies" but GLP2 analogues

Answer: Thanks for the suggestion. We modified the title.

A table with all, the short (often only clinical case) and indirect, results of GLP2 analogues in Crohn's disease will be very useful for this article.

Answer: We added the Table I.

ref 81: is daily and not weekly for glepaglutide?

Answer: Indeed the cited Phase II study (now ref. 84) was performed with daily injections although the molecule is long-acting. The ongoing phase III trial (ref. 85) is comparing placebo, weekly and twice weekly dosing of glepaglutide (we corrected this).

Reviewer #2:

In the review article by Pizzoferrato et al. the authors the current clinical and pathophysiological knowledge about short bowel syndrome, focusing on Crohn's disease, moreover, they summarize the experiences from clinical studies about GLP2 analogues in SBS. The topic is of great clinical importance. The review is well structured, all the crucial data are mentioned and discussed.

Answer: We thank the reviewer for the comments.

Several recent articles about the topic is not referred (e.g.,J Invest Surg. 2018 Jun;31(3):243-252. doi: 10.1080/08941939.2017.1294217. or J Invest Surg. 2018 Jun;31(3):253-255. doi: 10.1080/08941939.2017.1300715.). I suggest to discuss the relation of GLP2 analogues to intestinal epithelial stem cell functions together with their possible future side effects.

Answer: A sentence on stem cells and the suggested reference has been added "The effect of teduglutide on intestinal epithelial stem cells is of particular relevance in the possible risk of the drug being tumorigenic [63]."

After minor revision I suggest accepting the manuscript for publication.

Reviewer #3: This is an excellent review on SBS, SBS-IF and GLP-2 pharmacotherapy, technically sound, and very well written. Definitely fun to read and very informative. The introduction and the positioning of the special, but common situation of CD-patients with SBS-IF, is well achieved, thoroughly reviewing the literature. The paragraph on the special focus is technically clear and well referenced, citing the current literature and describing the most recent and most important findings.

Answer: We thank the reviewer for the comments.

One little suggestion could be a paragraph or a statement on what further research is needed for this certain population. Effectiveness of GLP-2-analogues is mentioned, but would be expected. Maybe one could ask, if GLP-2 analogues are safe in CD-patients with clinically controlled, but subclinically active disease – some clinicians are worried of "potentiating" the inflammation, but maybe this is not the case, who knows….

Answer: We added to the last section GLP-2 ANALOGUES IN CROHN'S DISEASE the following sentence, after reporting the results of the RCT about use of teduglutide as anti-inflammatory therapy for CD: "AEs were not different between the placebo and treatment groups[97]. This last result is particularly relevant, because it came from a controlled study on a relevant number of patients with active CD, although not affected by SBS-IF. The good safety profile in this setting justifies the use of GLP-2 analogues in patients with SBS-IF and active CD, but more data on this specific population are needed.

Data on the use of the new GLP-2 analogues (apraglutide and glepaglutide) in CD are lacking."

We also stated in the conclusion: "More data are needed on the use of GLP-2 analogues in patients with active CD to clarify their safety and efficacy in this setting."

(Please, see also the answer to Reviewer #4, first point)

Some Minor suggestions:

Minor:

- Introduction: reference should be made that this review is focused on adult SBS – not pediatric SBS.

Answer: We added the specification to adult SBS in the first sentence of the Introduction.

- CD22.4 % - brackets are missing

Answer: Corrected

- The cited numbers in the last paragraph on the first page of the introduction -although from a very large cohort - may not necessarily reflect all IF-populations – compare with the intestinal transplant registry report from 2015 – the largest fraction in the US cohort were ischemia-SBS, this should be discussed or the numbers attenuated.

Answer: We added in the Introduction a sentence reporting what suggested " However, other reports, such as the US intestinal transplant (IT) registry, describe mesenteric ischaemia as

the first cause of IT (24%) and CD as the second most frequent cause (11%)[6] " *and the relative reference.*

- Ref [5] is not appropriate – a basic research study from mice is cited, but human SBS is referred to – Warner 2016 CMGH may be a better reference. - Ref [6] is not appropriate – relates to GLP-2-action, but endogenous adaptation is described – also Warner 2016 CMGH may be a better reference.

Answer: We modified the references as suggested.

- "possible severe dehydration" – this statement is not needed – the opposite may be the case in many situations due to ample volume resuscitation.

Answer: We deleted the statement.

- "Approximately 90% of the intestinal adaptation described above occurs in this phase." This is not referenced and not covered by data (90% of what?).

Answer: We modified to "Most of the intestinal adaptation…".

- "oral nutrition alone" – can already be introduced as "oral autonomy", because this term is an important technical term in the field and also used later.

Answer: We modified the sentence accordingly.

- "The most frequent and invalidating symptom is diarrhea due to…" – in most SBS-IF-patients, diarrhea is a problem, but not necessarily the "invalidating symptom".

Answer: We modified to "One frequent symptom is diarrhea…"

- Ref. [7] cites IBS-diarrhea, which is not applicable in the SBS-situation. Ref. [10] should be removed, because it is not applicable to the SBS-situation. A better citation for the bile acid problem could be "Hvistendahl et al. 2022 JPEN - Bile acid-farnesoid X receptor-fibroblast growth factor 19 axis in patients with SBS…". For the transit problem, another reference could be "Kunkel et al. 2011 Neurogastroenterol Motil.

Answer: We modified the ref [7, now 9] to Kunkel 2011; ref [10] has been removed , and we added ref [12] Hvistendahl 2022.

- Ref. [11] is cited at the statement that particularly patients with PN have high prevalence of gall stones – while it may true, it is not related to PN, this should be stated more clearly.

Answer: We rephrased the sentence accordingly "Asymptomatic gallbladder stones were reported in a population of SBS patients [13] ".

- The mechanism for oxalate stone formation is not completely deciphered, and it is not clear if increased passive diffusion is the only mechanism involved, this should be phrased a little

more cautiously. One reference for this could be "Saunders et al. 1975 Gut - Regional differences in oxalate absorption by rat intestine…" One important study has recently re-evaluated the prevalence and incidence of nephrolithiasis – "Yang et al. 2019 Ann Nutr Metab - Risk Factors for Nephrolithiasis in…", this study should be cited in this context.

Answer: We rephrased accordingly "Other mechanisms may be involved, such as regional differences in oxalate absorption [17]." and added the suggested references.

- "refractoriness" is not preferred terminology

Answer: We modified to resistance.

- "Patients with perianal disease have more bowel resections" cannot be maintained – it is probably a subgroup effect as described in the paragraph before – the p-phenotype is associated with higher risk of resection, because the disease phenotype is more severe. But phenotype also may exist independent of resections – In our IBD practice, we do have a very large cohort of perianal disease, but not many - if at all - bowel resections

Answer: This paragraph refers to the review of Limketkai, Inflamm Bowel Dis 2016 [26]. Having perianal disease phenotype at diagnosis is associated to a bad prognosis and a high risk of disabling disease course, including intestinal resections (Beaugerie, Gastroenterology 2006). We rephrased for better clarity :" Patients with perianal disease at diagnosis have higher risk of disabling disease course including bowel resections…"

- "greater age" – age is great, but should be termed "higher age"

Answer: We modified as required.

- "multidisciplinary team" – careful with the terminology here. If strictly only medical disciplines are meant, this would rather be termed "interdisciplinary", but then technically not involve a "nutritionist". If indeed "multidisciplinary" is meant, this would also involve special pharmacy, home care (infusion) nurse, stoma nurse, rehabilitation …

Answer: Indeed the required team is really multidisciplinary. We rephrased as follows: "A multidisciplinary team consisting of at least gastroenterologists, nutritionists, surgeons, radiologists, stoma therapists, care managers, pharmacists, and home care nurses is useful…."

- "dietary manipulation" – not really manipulation, but maybe rather intervention?

Answer: We modified as suggested.

- Ref. [29] pediatric study is linked – maybe rather an adult study?

Answer: We replaced the reference with Dibb Aliment Pharmacol Ther. 2013 (now Ref. 32).

- CRBSIs “the most feared complications” – I would fear IFALD/PNALD much more, although of course it is not as common as CRBSIs.

Answer: We modified “are among the most feared…”

- “CRSBI” “CRBSI”

Answer: We corrected accordingly.

- “Up to 32% of patients receiving HPN can develop microcytic or ….” This number is very much context-specific and should either be put in context of the cited ref. [52] or the statement on anemia should be phrased more broadly

Answer: We rephrased as follows: “Malabsorption-related anaemia may develop into microcytic anaemia due to iron deficiency or macrocytic anaemia due to malabsorption of vitamin B12 and folate, which lead to the need for iron and vitamin supplementation[3],[55].”

- “Intestinal absorption may also be increased by hormonal manipulation[56]–[58]. Refs [56] and [57] do not show this, ref. 56 shows the effect of IGF-1 on mucosal morphology – it could be cited with a statement on mucosal hypertrophy/proliferation. Ref. [57] discusses the anatomy of SBS-types, this could be cited in the introductory paragraph. A possible references for improved absorption due to GLP-2-therapy could be “Reiner et al. 2020 Dig Dis Sci - Teduglutide Promotes Epithelial Tight Junction Pore…”.

Answer: We deleted ref 56 and moved ref 57 to the introduction (now ref. 4). We added the suggested reference [61].

- “GLP2” -> “GLP-2”, should be always used.

Answer: We corrected accordingly.

- “One of the first open-label trials on teduglutide was carried out and published in 2005.” Why not “The first open label trial on teduglutide was published in 2005”. - “Sixty-three percent” -> 63%, or do not use at the beginning of the sentence.

Answer: We corrected accordingly.

- 20% -> missing)

Answer: The bracket is at the end of the sentence

- “Recently, new GLP2 analogue molecules have been studied”, references should be given, and references could cite elsiglutide and dapiglutide as well, although mostly preclinical data are available on these novel GLP-2 analogueues.

Answer: We added appropriate sentences on elsiglutide and dapiglutide and related references: "Other molecules are in the preclinical phase of their development. Elsiglutide is a GLP-2 analogue with a long half-life that reduced diarrhoea induced by lapatinib, a tyrosine kinase inhibitor, in a rat model[86]. Dapiglutide is a dual GLP-1 and GLP-2 agonist that showed beneficial effects in a rat model of SBS by improving body weight, promoting intestinal growth, increasing villous height and intestinal length, and reducing watery stool losses[87]."

- "apraglutide have also been studied in piglets. In this case, the two molecules showed similar results: the intestinal growth appeared to be a lasting outcome of treatment with long-acting GLP-2, persisting at least 7 days after the discontinuation; in contrast, mucosal hypertrophy appeared to regress 7 days after the end of treatment with both agents[77]." This finding must be interpreted with caution – I suggest not to spell this finding out so clearly here, because the experimental setup in the paper [77] is not 100% clear as to when the last dose before treatment discontinuation was given. Also, the discrepancy between longitudinal and horizontal growth is not entirely clear.

Answer: We added the word "apparently" to the description of the study results and we added this sentence: "However, these resultsmust be interpreted with caution and need further confirmation."

- Phase 2 should be spelled Phase II.

Answer: We corrected in the text, as the reviewer suggests. However, the original references published on The lancet gastroenterology and hepatology ad JPEN spell Phase 2 and we left this spelling in the references.

- Language/style issue as above: eighteen patients -> 18

Answer: Corrected.

- "before the beginning of therapy" -> "before the beginning of teduglutide therapy"

Answer: Corrected.

- "438.825 and 584.825 μm" -> avoid too many post decimals, maybe avoid the original numbers, the 33% may suffice.

Answer: Corrected.

- Ref [89] is an important and probably the most sophisticated study on the topic to date. It should also be discussed if the Crohn remission/response rates were due to reduced diarrhea as an item on the CDAI, which was used for the assessment clinical response/remission.

Answer: We added a sentence: "It is questionable whether the CDAI modifications described in this study were mostly due to the effect of teduglutide on diarrhoea because no significant modification of CRP was detected."

- Glepaglutide should also be mentioned under keywords.

Answer: We added glepaglutide to the keywords

Reviewer #4: This is an interesting review of the data concerning the treatment of patients with Crohn's disease who developed short bowel syndrome with intestinal failure due to surgical resection of the intestine, using Teduglutide and apraglutide.

Answer: We thank the reviewer for the comments.

My suggestions are the following:

• Is there literature information regarding the effect of the activity of the underlying Crohn's disease on the therapeutic effect resulting from the administration of the drugs?

Answer: There is no specific literature on this subject. The case series by Kochar (ref . 89) does not report data on Crohn's disease activity. A report of two cases by Al Draiiwesh (ref. 90) and one from Borghini (ref. 91) describe the efficacy of the drug in active Crohn' disesase patients, with improvement of both inflammatory activity and nutritional status. The most informative study is the one from Buchman (ref. 97) that is a study of teduglutide efficacy as therapy of active Crohn without SBS-IF. We added to the description of ref. 97 this comment for more clarity: "This last result is of particular relevance, coming from a controlled study on a relevant number of patients with active CD, although not affected by SBS-IF. The good safety profile that was demonstrated in this setting can justify the use of GLP-2 analogues in patients with SBS-IF and active CD, although more data on this specific population are needed." For these reasons we stated in the conclusion: "More data are needed on the use of GLP-2 analogues in patients with active CD to clarify their safety and efficacy in this setting."

• What is the effect of the use of biological agents, either as induction or as a maintenance treatment, on the therapeutic effect resulting from the administration of the drugs?

Answer: The reports described by Kochar (ref. 89) and Al Draiiwesh (ref. 90) described 8 and 2 Crohn's disease patients, respectively, successfully treated with both biologics (and/or immunosuppressants) and teduglutide.

Therefore we stated in the conclusion: "… signals from RCTs and real-life observations indicate that teduglutide is efficacious and well tolerated by CD patients, even if they are being treated with immunosuppressants and/or biological agents."

• What is the evolution of the patients after stopping the treatment? For how long can they be administered?

Answer: We added a paragraph to the section NEW THERAPIES: GLUCAGON-LIKE PEPTIDE-2 (GLP-2) ANALOGUES and a reference (88): "For the duration of therapy with GLP-2 analogues in patients with SBS-IF, the available data indicate that these drugs must be administered over a long life because reversal of the previous need for parenteral support occurs if teduglutide is discontinued[64]. A recent report described 13 patients (one with CD and one with ulcerative colitis) who discontinued teduglutide after a successful clinical outcome. The volume of PS remained stable in the first 4 years but later increased in 12/13 patients up to 9 years after withdrawal[88]. These data support further studies exploring the possibility of the periodic use of GLP-2 analogues for selected SBS-IF patients."

We also added this sentence and reference to the section GLP-2 ANALOGUES IN CROHN'S DISEASE: "Concerning teduglutide discontinuation, a report described 2 CD patients on chronic teduglutide treatment who were not able to tolerate even a few days withdrawal of the drug[95]."

• A more complete description of the pathophysiological actions of these drugs would probably be useful.

Answer: We added a sentence on this in the section NEW THERAPIES: "The mechanism of action of teduglutide is complex and involves direct and indirect effects of interaction with a GLP-2 receptor and includes the following most relevant factors: crypt cell proliferation, increase in bowel weight and villous growth, enhancement of intestinal barrier function, inhibition of motility of the gastrointestinal tract and gastric acid secretion, and increase of intestinal blood flow[60]. The effect of teduglutide on intestinal epithelial stem cells is of particular relevance in the possible risk of the drug being tumorigenic [63]."

• Finally, I wish for the sake of the readers that the authors would express more clearly their opinion as to the utility of these drugs in clinical practice.

Answer: We added a comment on that in the Conclusion: " The GLP-2 analogue teduglutide demonstrated a relevant clinical utility for SBS-IF patients, and it significantly reduced HPN volume and/or days of infusion, allowing oral autonomy in some patients. However, because this therapy is likely life-long or of long duration, more data are needed on the long-term safety and cost-effectiveness."

• A table listing details of existing clinical studies, e.g. drug dosage, clinical outcomes, laboratory data, and major side effects, would be helpful to readers.

Answer: We added the Table I, as also suggested by Reviewer #1.

• Regarding the cause(s) of the reduction in the surgical rates of patients with Crohn's disease (Conclusion part of the paper), I would suggest the authors mention the following (Dittrich et al Inflamm Bowel Dis 2020;19:1909-16): "…Although anti-TNF therapy seems to play a role, the decrease in surgical trends is likely multifactorial, owing to a decline in smoking trends, earlier diagnosis, earlier treatment, improved patient education, and changes in clinical practice…"

Answer: We added what suggested and the reference (24) in the section on SBS-IF IN CROHN'S DISEASE: "Surgery rates in CD declined in recent decades due to multifactorial reasons, including earlier diagnosis and treatment, the use of biological agents, a decline in smoking rates, and improved patient education[23], [24].