Modulation of inflammatory and disease biomarkers following 4 week treatment with AVX-470, an oral anti-TNF antibody, in ulcerative colitis patients

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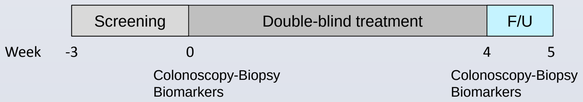
BACKGROUND & AIMS

AVX-470 is an oral, bovine-derived, polyclonal antibody designed to target tumor necrosis factor (TNF) locally in the gastrointestinal tract without significant systemic exposure. AB1101 was a double-blind, placebo-controlled first-in-human trial in patients with active ulcerative colitis (UC). AVX-470 administration was well tolerated, and was associated with dose-dependent increases in clinical and endoscopic remission (Fox et al, Am J Gastro 109: S2 (pp S494), data shown below).

Parameter	Placebo (n = 9)	0.2 g/d (n = 8)	1.6 g/d (n = 12)	3.5 g/d (n = 7)	Pooled Active (n = 27)
Clinical Response	1/9 (11.1)	3/8 (37.5)	2/12 (16.7)	2/7 (28.6)	7/25 (25.9)
Clinical Remission	0	0	0	1/7 (14.3)	1/27 (3.7)
Endoscopic Response	0	0	1/12 (8.3)	1/7 (14.3)	2/27 (7.4)
Endoscopic Remission	0	0	1/12 (8.3)	1/7 (14.3)	2/27 (7.4)

This study aimed to characterize AVX-470 modulation of TNF, MPO, and epithelial cell apoptosis in colon, and correlate change in inflammatory markers in tissue and serum with clinical and endoscopic response.

METHODS



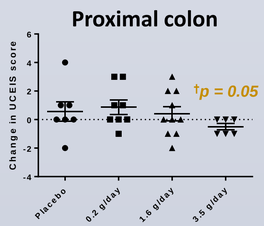
- 36 patients with active UC, 33 patients completed 4 week treatment
- 13 study centers located in US, Canada, Belgium and Hungary
- 3 ascending-dose cohorts, enteric coated capsule dosage form
  - AVX-470 was given orally at 0.2 g/d BID, 1.6 g/d BID, and 3.5 g/d TID in divided doses
  - Within each cohort, patients were randomized 3:1 to AVX-470 or placebo
- Colonoscopy with central reading at Baseline and Week 4
  - Biopsy tissue taken from cecum/ascending, transverse, descending, sigmoid, & rectum

IHC Staining Methods

Immunohistochemical (IHC) Methods: Standard IHC techniques were used to stain formalin-fixed, paraffin-embedded colon tissue biopsy sections with rabbit polyclonal antibodies to TNF (IHC World) and MPO (Abcam). TUNEL staining was performed using the TdT-FragEL DNA Fragmentation Detection Kit (Calbiochem). Digital image analysis was performed as follows: Levels of TNF and MPO were quantitated in the lamina propria and epithelial layers in 5 representative fields per sample, and geometric means were calculated. The number of cells positive for TUNEL staining were manually counted in the epithelial compartment (epithelial lumen + crypts), and means were calculated.

CLINICAL OUTCOME: Dose dependent improvement in endoscopy scores (UCEIS)

Graph shows mean ± SEM of change relative to baseline at Week 4. Dose dependent improvement in endoscopy scores was seen in proximal (right) but not distal colon (data not shown). †two-tailed, unpaired T-test, 0.2 vs 3.5 g/day group.



Proximal colon	Ascending and transverse regions
Distal colon	Descending and rectosigmoid regions

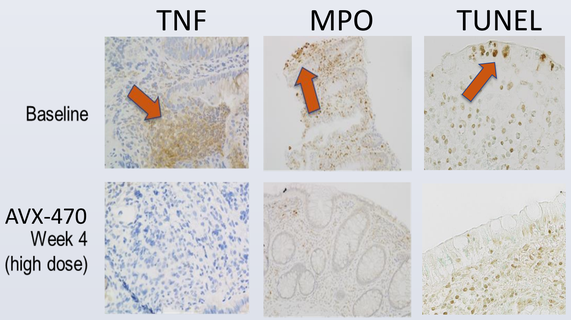
RESULTS

AB1101: First-In-Patient Clinical Trial in UC All Endpoints Met

Endpoint	Outcome
Safety	Safe and well-tolerated
Pharmacokinetics	Antibody in tissue and stool, not in serum
Immunogenicity	No induction of anti-drug antibodies
Pharmacodynamics	Inhibition of markers of local inflammation (TNF, MPO, TUNEL)
	Inhibition of systemic markers of disease (CRP, IL-6)
Clinical efficacy	Induction of clinical, histological and endoscopic response

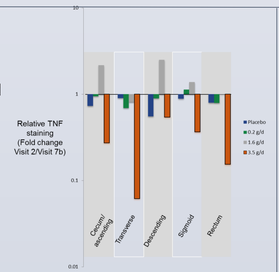
Biomarkers in Colon Biopsy Tissues

Representative images of IHC staining (brown color, arrows) in sigmoid colon biopsy samples in the high dose group. TNF & MPO staining in lamina propria and epithelial cell layers is dramatically reduced at Week 4. Apoptosis in the intestinal epithelial layer (IEC), indicated by TUNEL staining , is also reduced after AVX-470 treatment. Magnification 400x.



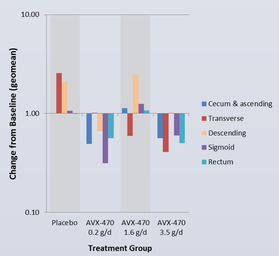
TNF

TNF levels are reduced in all 5 colon regions at Week 4 in the 3.5 g/d AVX-470 treatment group.



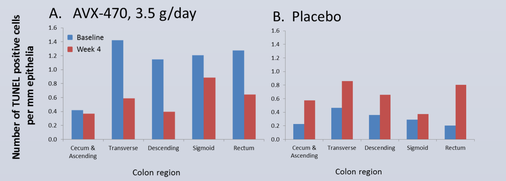
MPO

Decreased MPO levels seen in 4 of 5 colon regions in the AVX-470 0.2 and 3.5 g/day treatment groups.



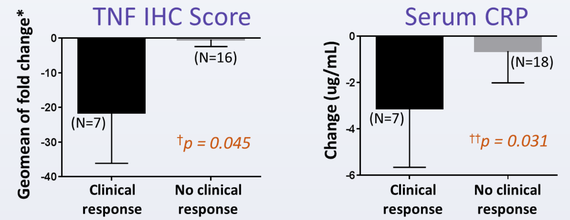
IEC TUNEL

Mean levels of IEC TUNEL staining were substantially reduced in patients from the AVX-470 treated group at Week 4, and in contrast, mean levels of IEC TUNEL staining were increased in the placebo group.



- Decrease in TNF and IEC TUNEL staining is seen predominantly in the proximal colon, consistent with reduction in endoscopy scores (left panel).
- MPO reduction is seen in both proximal & distal colon.

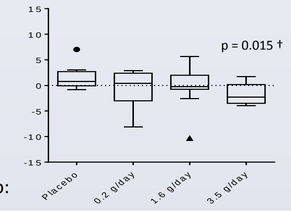
Reductions in levels of TNF in colon and serum CRP correlate with clinical response



Mean ± SEM of change relative to baseline at Week 4.
†two-tailed, unpaired T-test; †† Wilcoxon matched-pairs signed rank test.
*Across 5 colon regions (ascending, transverse, descending, sigmoid, rectum), AVX-470 treatment.

AVX-470 reduces serum IL-6 levels

†ANCOVA of raw change in IL6 levels as a function of treatment including placebo and AVX-470 3.5 g/d dose group, adjusting for baseline.

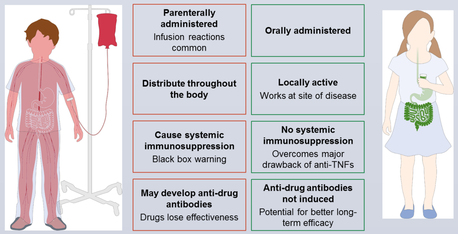


Data shown represent change following 4 weeks dosing. Serum IL-6 levels quantified by SIMOA ELISA analysis.

CONCLUSIONS

- AVX-470 mediated reduction in colon TNF levels and serum CRP levels was correlated with clinical response.
- Reduction in colon TNF and IEC TUNEL levels was proportionately greatest in the proximal colon, which correlated with changes in endoscopic activity. This suggests a linear gradient of response from the proximal to distal colon following oral dosing of AVX-470.
- These results show AVX-470 mediated reduction of inflammation, apoptosis of intestinal epithelial cells, and disease activity through local drug action in the colon.

This outcome advances the development of AVX-470, the first anti-TNF antibody that is orally delivered and stays in the digestive tract, with the potential for better safety. AVX-470 is especially well-suited for the treatment of pediatric IBD patients.



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