Results

The present study was conducted on thirteen male albino rats. They were divided into three groups: control group (ten rats), treated group with methotraxate (ten rats) and treated group with methotrexate plus vitamin A (ten rats).

(control group):

By light microscopic examination, the control rat jejunum was formed of mucosa, submucosa, muscularis mucosa and serosa. The mucosa is formed of villi and crypts. The villi appeared as tall finger like projections thrown into the lumen. The intestinal crypts (crypts of Lieberkuhn) appeared as tube like structures opened between the bases of the villi and extended deeply to the muscularis mucosa (fig. 5).

The surfaces of the villi were covered by absorptive cells which appeared tall columnar with basal oval nuclei .The goblet cells were scattered between the absorptive cells. The mucus content of these cells was often dissolved away and presented a foamy appearance. The lamina propria was formed of connective tissue surrounding the crypts and extending as central core in the villi. (figs 5&6). The covering surface of the epithelium showed intact brush border (fig. 6).

The lining epithelium in the upper half of the crypts was identical to that covering the villi. In the lower half of the crypts, the paneth cells appeared as pyramidal cells with basal rounded nuclei and apical eosinophilic secretory granules (figs. 7&8).

Sections stained with PAS showed neutral mucin – secreting goblet cells scattered along the villi and crypts. These appeared magenta red (figs. 8&9&10).

Brush border of the absorptive cells covering the villi appear pink line at the outer boundary of villi (figs. 10&11).

Sections stained with Alcian blue showed acid mucin – secreting goblet cells scattered along villi and crypts. These appeared turquoise- blue (figs. 12&13).

By electron microscopic examination, the rat jejunum showed that the absorptive cells were tall columnar cells with an oval pale basal nucleus which contain small rounded nucleolus. The cytoplasm of absorptive epithelial cells contained mitochondria with evident cristae and rough endoplasmic reticulum. The brush border was seen as layer of densely packed microvilli, each microvillus consisted of cell membrane enclosing a core of actin micro filaments (figs. 14&16). The core of actin microfilaments is associated with other cytoskeletal proteins. The cell junction between absorptive cells was evident and basal lamina appeared uniform, has trilaminar appearance, it composed of an outer dark membrane, inner dark membrane and middle light layer (figs. 14).

The goblet cell appeared among the absorptive cells, its apical part filled with secretory granules with golgi apparatus around these granules (figs. 14 &15).

Group B (Methotrexate(MTX)-treated group):

By light microscopic examination, the methotrexate treated rat jejunum showed loss of normal jejunal architecture varying from distortion in configuration of the villi in the form of broadening and fusion of some villi up to detachment and separation of the villi(figs. 17&18).

The lamina propria of the intestinal villi was infiltrated with inflammatory cells and extravasated by RBCs, with congested blood capillary and subepethelial vacuolation. Some absorptive cells are infiltrated by inflammatory cells (figs. 19&20).

There were sporadic areas of erosions (ulcerations) in surface epithelium of the villi (fig. 21).

Some absorptive cells became flattened with flat nucleus; other cells were cubical with rounded nucleus (fig. 22).

The lower part of intestinal crypts contained paneth cells; some appeared ballooned with flat nucleus and apparent increase in their apical eosinophilic granules, while other paneth cells were degenerated (fig. 23).

Some intestinal crypts are degenerated while other crypts are vacuolated and other intestinal crypts appear normal .The submucosa contain congested blood capillary (fig. 24).

The sections stained with PAS showed marked depletion of the neutral mucin – secreting goblet cells and their aggregations in the crypts (figs. 25&26). The surface the villi in some areas were covered by brush border (B), other areas are not covered by brush border (figs. 26).

Sections stained with Alcian blue showed marked depletion of the acid mucin – secreting goblet cells and their aggregations in the crypts (fig. 27).

By electron microscopic examination, the rat jejumum treated with MTX showed absorptive cells contained divided irregular nuclei with multiple nucleolui (fig. 28). Some absorptive cells became flat in shape with flat nucleus with few numbers of microvilli (fig 30).

The cytoplasm of the absorptive cells contained many groups of vacuoles and fat droplets (fig 28 &29 & 30). Also cytoplasm showed vacuolated mitochondria and lysosomes (fig 29).

Some areas of the luminal border of the absorptive cells show intact brush border and other showed areas of destruction (28&31).

Group C (Methotrexate and vitamin A -treated group):

By light microscopic examination, the rat jejunum treated with methotrexate plus vitamin A showed preservation of normal jejunal architecture (fig. 32).

The covering epithelium of the villi was intact in some areas; while in other area it was destructed (fig33).

The absorptive cells were tall columnar in shape with pale oval nucleus and intact brush border (fig. 34).

The intestinal crypts lined with normal absorptive cells and goblet cells. At its bottom, paneth cells appeared pyramidal in shape with basal rounded nucleus (fig. 35).

The sections stained with PAS showed the goblet cells that prouduce neutral mucin scattered along the villi and crypts (figs. 36&37). Brush border of the absorptive cells was intact (fig. 37).

The sections stained with Alcian blue showed the goblet cells that produce acid mucin scattered along villi and crypts (figs. 38).

By electron microscopic examination, the rat jejunum treated by methotrexate plus vitamin A showed that the absorptive cells were tall columnar with their basal oval nuclei and goblet cell filled with its secretory granules and small nucleus, the cell junction between the absorptive cells is evident (fig. 39). The cytoplasm shows mitochondria with evident cristae, vacuolated mitochondria, some swollen mitochondria, rough endoplasmic reticulum and golgi apparatus (figs. 40& 41).

Fig.(5): A light photomicrograph of T.S in rat jejunum (control group) showing: The lining mucosa was thrown into finger like projections called villi (v) which are covered by absorptive cells (A) and goblet cells (G) in between. The villi have core of connective tissue called lamina propria (L).At the bases of the villi there are intestinal crypts (C). The submucosa (Su), musculosa(M) and serosa(S) are seen.

(HX & E × 100)

Fig.(6): A light photomicrograph of T.S in rat jejunum (control group) showing: The intestinal villi which are covered by absorptive cells (A) and goblet cells (G) in between absorptive cells , the core of the villi is lamina propria (L). The absorptive cells have basal oval nucleus (N) and brush border (B).

(HX&E × 200)

Fig. (7): A light photomicrograph of T.S in rat jejunum (control group) showing: The intestinal crypt (C) which is lined by absorptive cells (A) and goblet cells in between (G), at the lower part of the crypt there are paneth cells(P) which are pyramidal shaped, contain acidophilic secretory granules in the apical cytoplasm (arrow) .

(HX & E $\times$ 200)

Fig. (8): A higher magnification of the previous section showing: The lower part of intestinal crypt (C) which is lined with absorptive cells (A) with oval basal nucleus (N) and paneth cell (P) with rounded basal nucleus (n) and its cytoplasm is filled with acidophilic secretory granules (sg).

(HX & E $\times$ 400)

Fig. (9): A light photomicrograph of T.S in rat jejunum (control group) showing: Neutral mucin - secreting goblet cells (G) scattered all over villi (V) and crypts (C).The submucosa (Su), musculosa (M) and serosa (S) are also seen.

(PAS × 40)

Fig. (10): A light photomicrograph of T.S in rat jejunum (control group) showing: Neutral mucin - secreting goblet cells (G) scattered all over the villi (V) .The brush border (B) cover the outer surface of the villi.

(PAS × 200)

Fig. (11) A higher magnification of the previous section showing: Neutral mucin - secreting goblet cells (G) cover the outer surface of the villi in between absorptive cells (A) which has brush border (arrow).

(PAS × 400)

Fig. (12): A light photomicrograph of T.S in rat jejunum (control group) showing: Acid mucin - secreting goblet cells (G) scattered all over the villi (V) and crypts (C). The submucosa (Su), musculosa (M) and serosa (S) are also seen.

(Alcain blue × 40)

Fig. (13): A light photomicrograph of T.S in rat jejunum (control group) showing: Acid mucin - secreting goblet cells (G) cover the outer surface of the villi (V) in between absorptive cells

(Alcain blue × 200)

Fig. (14): An electron photomicrograph of rat jejunum (control group) showing: Columnar absorptive cells (A) containing oval pale nucleus (N) with small rounded nucleolus (n). Cytoplasm contains mitochondria (m) and rough endoplasmic reticulum (rER). The luminal border of the absorptive cells shows microvilli (mv) thrown into the lumen (Lu). The cell junction between absorptive cells is evident (arrow). The apical part of goblet cell (G) appears filled with secretory granules (sg) and golgi apparatus (g). The basal lamina (BL) is uniform.

(E/M × 3000)

Fig. (15): An electron photomicrograph of rat jejunum (control group) showing: The apical part of mucus secreting goblet cells (G), golgi apparatus (g) surrounding the secretory granules. The luminal border of the cell shows microvilli thrown into the lumen (Lu) ,also goblet cell open into the lumen (arrow).
(E/M × 8000)

Fig. (16): An electron photomicrograph of rat jejunum (control group) showing: The upper part of absorptive epithelial cells, cytoplasm contains mitochondria (m) with evident cristae (c). The brush border is seen as layer of densely packed microvilli (mv), each microvillus consists of cell membrane (cm) enclosing a core of actin filaments (f), associated with other cytoskeletal proteins.

(E/M×15000)

Fig. (17): A light photomicrograph of T.S in rat jejunum treated with methotrexate (MTX) showing: Loss of normal jejunal architecture and inflammatory cell infiltration (I).The submucosa(Su), musculosa (M) and serosa (S) are seen.
(HX & E × 40)

Fig. (18): A light photomicrograph of T.S in rat jejunum treated with MTX showing: Loss of normal architecture with distortion in configuration of villi by broadening and fusion (circle) of some villi (V).The submucosa (Su), musculosa (M) and serosa(S) are also seen.

(HX & E × 100)

Fig. (19): A light photomicrograph of T.S in rat jejunum treated with MTX showing: The lamina propria (L) of the intestinal villi (V) are infiltrated with inflammatory cells (I) and extravasated by RBCs (R). Also, congested blood capillary (BC) and subepethelial vacuolation are seen in lamina propria.

(HX & E × 100)

Fig.(20): A higher magnification of the previous section showing: The lamina propria contains extravasation of RBCs (R) and congested blood capillary(BC). Some absorptive cells (A) are infiltrated by inflammatory cells (I).

(HX & E × 200)

Fig. (21): A light photomicrograph of T.S in rat jejunum treated with MTX showing: Erosions (arrow)s of some absorptive cells (A) in surface area of the villi (V). The lamina propria (L) shows vacuolations (Va).

(HX & E × 100)

Fig. (22): A light photomicrograph of T.S in rat jejunum treated with MTX showing: Some absorptive cells(A) become flattened with flat nucleus (f) , other cells are cubical with rounded nucleus (r). There is extravasation of RBCs (R) in the underlying lamina propria .

(HX & E × 400)

Fig. (23): A light photomicrograph of T.S in rat jejunum treated with MTX showing: The intestinal crypts(C) contain some ballooned paneth cell(BP) with flat nucleus, while others are degenerated (DP).

(HX & E × 200)

Fig. (24): A light photomicrograph of T.S in rat jejunum treated with MTX showing: Some intestinal crypts are degenerated (DC), while other crypts are vacuolated (Va) and other intestinal crypts appear normal (C).The submucosa contain congested blood capillary (BC).

(HX & E × 200)

Fig. (25): A light photomicrograph of T.S in rat jejunum treated with MTX showing: Depletion of neutral mucin - secreting goblet cells (G) in the villi (V) and their aggregations in the crypts (C).

(PAS × 40)

Fig.(26): A higher magnification of the previous section showing: depletion of neutral mucin - secreting goblet cells in villi(V) and their aggregations in the crypts(C). The surface the villi (V) in some areas is covered by brush border (B), other areas not covered by brush border (arrow).

(PAS × 100)

Fig. (27): A light photomicrograph of T.S in rat jejunum treated with MTX showing: Depletion acid mucin - secreting goblet cells in the villi (V) and their aggregations in the crypts (C). (Alcain blue × 40)

Fig. (28): An electron photomicrograph of rat jejunum treated with MTX showing: The absorptive cells containing divided irregular nuclei (N) which contain multiple nucleolui (n). The cytoplasm contains many vacuoles (Va).The luminal border shows intact micovilli are thrown into the lumen (Lu) in some area, other surface area show destruction of microvilli (arrow). The basal lamina (BL) of the cells is uniform. Notice: the cell junction between the cells is not evident.

(E/M × 3000)

Fig.(29): An electron photomicrograph of rat jejunum treated with MTX showing :The cytoplasm of the absorptive cell contains vaculations (Va) , lysosomes (Ly) , vacuolated mitochondria (vm) and nucleus(N).

(E/M× 8000)

Fig. (30): An electron micrograph of rat jejunum treated with MTX showing: Flattening of absorptive cells with flattening of its nucleus (N) with peripheral nucleolus. The luminal border contains few microvilli (mv). The cytoplasm is filled with fat droplets (FD). The basal lamina (BL) of the cell is uniform.

(E/M × 8000)

Fig. (31): An electron micrograph of rat jejunum treated with MTX showing: Brush border of the absorptive cells is destructed in some surface area (arrow) while other surface area contains intact micovilli (mv) with their actin microfilaments.The cytoplasm shows mitochondria (m) and vacuolation (va).

(E/M × 15000)

Fig. (32): A light photomicrograph of T.S in rat jejunum treated with MTX and vitamin A showing: Preservation of normal jejunal architecture. The mucosa thrown into villi(V) and crypts(C).The villi have core of lamina propria (L).The submucosa(Su), musculosa(M) and serosa(S) are also seen.

(HX & E × 40)

Fig. (33): A light photomicrograph of T.S in rat jejunum treated with MTX and vitamin A showing: The covering epithelium of the villi (V) is intact in some areas (arrow), while in other areas is destructed (d).

(HX & E × 100)

Fig. (34): A higher magnification of the previous section showing: Some areas of intestinal villi (V) covered with intact absorptive cells (A) and goblet cells (G).The absorptive cells have basal oval pale nuclei (N) and intact brush border (B). While other areas are not covered with absorptive cells (arrow).

(HX & E × 200)

Fig. (35): A light photomicrograph of T.S in rat jejunum treated with MTX and vitamin A showing: The intestinal crypt (C) is lined with absorptive cells (A) and goblet cells in between (G) and paneth cells in its bottom (P).
(HX & E $\times$ 400)

Fig. (36): A light photomicrograph of rat jejunum treated with MTX and vitamin A showing: Neutral mucin-secreting goblet cells (G) scattered all over the villi (V) and the crypts (C). (PAS × 40)

Fig. (37): A higher magnification of previous section showing: Neutral mucin-secreting goblet cells (G) scattered all over the villi(V) .The brush border (B) covers the outer surface of the villi.

(PAS × 200)

Fig. (38): A light photomicrograph of rat jejunum treated with MTX and vitamin A showing: Acid mucin-secreting goblet cells (G) scattered all over villi (V) and crypts (C).
(Alcain blue × 40)

Fig. (39): An electron micrograph of rat jejunum treated with MTX and vit. A showing: Absorptive cells (A) with its nucleus basal pale oval (N1) with small nucleolus (n1). The cytoplasm contain mitochondria and its luminal surface show microvilli (mv) thrown into the lumen (Lu). The cell junction between the absorptive cells is evident (arrow). The goblet cell (G) appears filled with secretory granules (sg). The cytoplasm of the goblet cell contains small nucleus (N2) and small rounded nucleolus (n2).

(E/M × 3000)

Fig. (40): An electron micrograph of rat jejunum treated with MTX and vit. A showing: Part of the cytoplasm of absorptive cell above nucleus (N) contains golgi apparatus (g), rough endoplasmic reticulum, mitochondria (m) with evident cristae (c) and some vacuolated mitochondria (vm) .

(E/M × 8000)

Fig. (41): An electron micrograph of rat jejunum treated with MTX and vit. A showing: Brush border of the absorptive cell shows intact microvilli (mv). The cytoplasm contains mitochondria (m) with evident cristae (c), swollen mitochondria (sm) and small areas of vacuolation (va).
(E/M × 15000)