



Influence of cigarette smoke on Follicle Associated Epithelium of murine Peyer's Patches

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Background

Recently, cigarette smoking has been associated with the development of several auto-immune diseases, including rheumatoid arthritis and Crohn's disease. The cellular and molecular mechanisms through which cigarette smoking predisposes to Crohn's disease are unknown. Cigarette smoke-induced apoptosis is described in several organs in *in vivo* and *in vitro* models of chronic smoke exposure, and might play a role in the pathogenesis of several smoke-associated diseases. The aim of this study was to quantify apoptosis in normal Follicle-Associated Epithelium (FAE) of murine Peyer's patches and compare this to apoptosis rates in the FAE of smoking mice.

Methods

C57BL/6 male mice were exposed to cigarette smoke for 24 weeks (chronic exposure) while a control group was exposed to air during the same period. After 24 weeks the mice were sacrificed and Peyer's patches were dissected for histology. Immunohistochemistry for active caspase-3 was performed on paraffin-embedded tissue sections of 11 Peyer's patches of smoking animals and 11 Peyer's patches of controls.

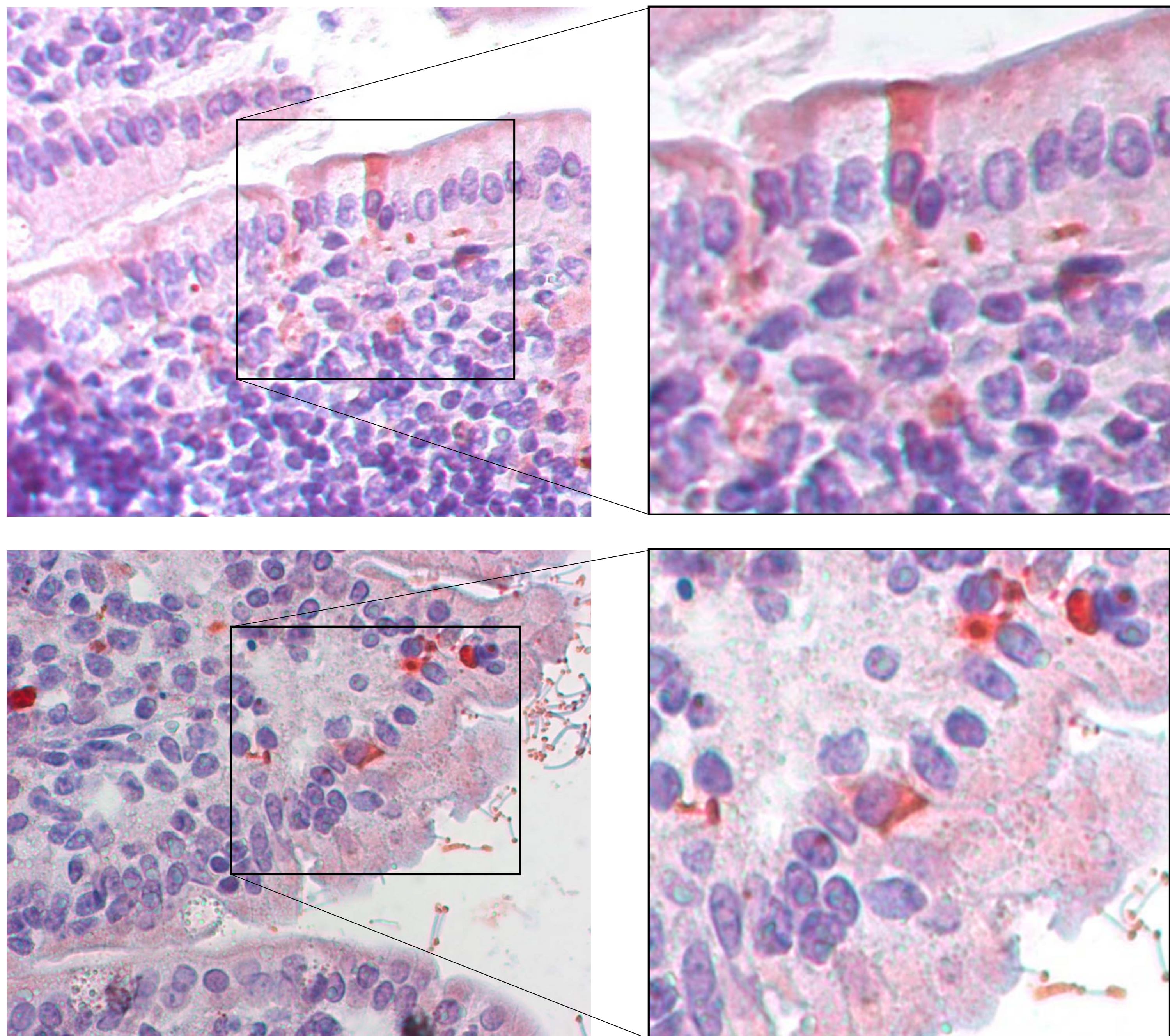


Figure 1: IHC for caspase-3 on murine Peyer's patches. 40x magnification

To compare apoptotic activities between smokers and controls, the apoptotic index (number of apoptotic cells per 100 cells) in the FAE was calculated. An unpaired student T-test was applied to evaluate statistical significance of the observed values.

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Results

A statistically significant increase in apoptosis of FAE cells was observed in smoking mice compared to air-exposed mice ($P=0.002$). In the FAE of smoking animals, the mean apoptotic index was 1.82 (95% CI 1.42-2.23), whereas the mean apoptotic index in the FAE of non-smoking animals was 0.92 (95% CI 0.64-1.12). In both groups, most apoptotic cells were seen at the apex of the FAE.

Non-smokers	Smokers
0,24	1,11
0,57	1,30
0,66	1,32
0,69	1,40
0,69	1,48
0,87	1,49
0,93	1,55
1,00	2,02
1,13	2,42
1,24	2,98
2,06	3,00

Table 1: Apoptotic index (%) for 11 Peyer's patches of smoking mice versus 11 Peyer's patches of non-smoking mice

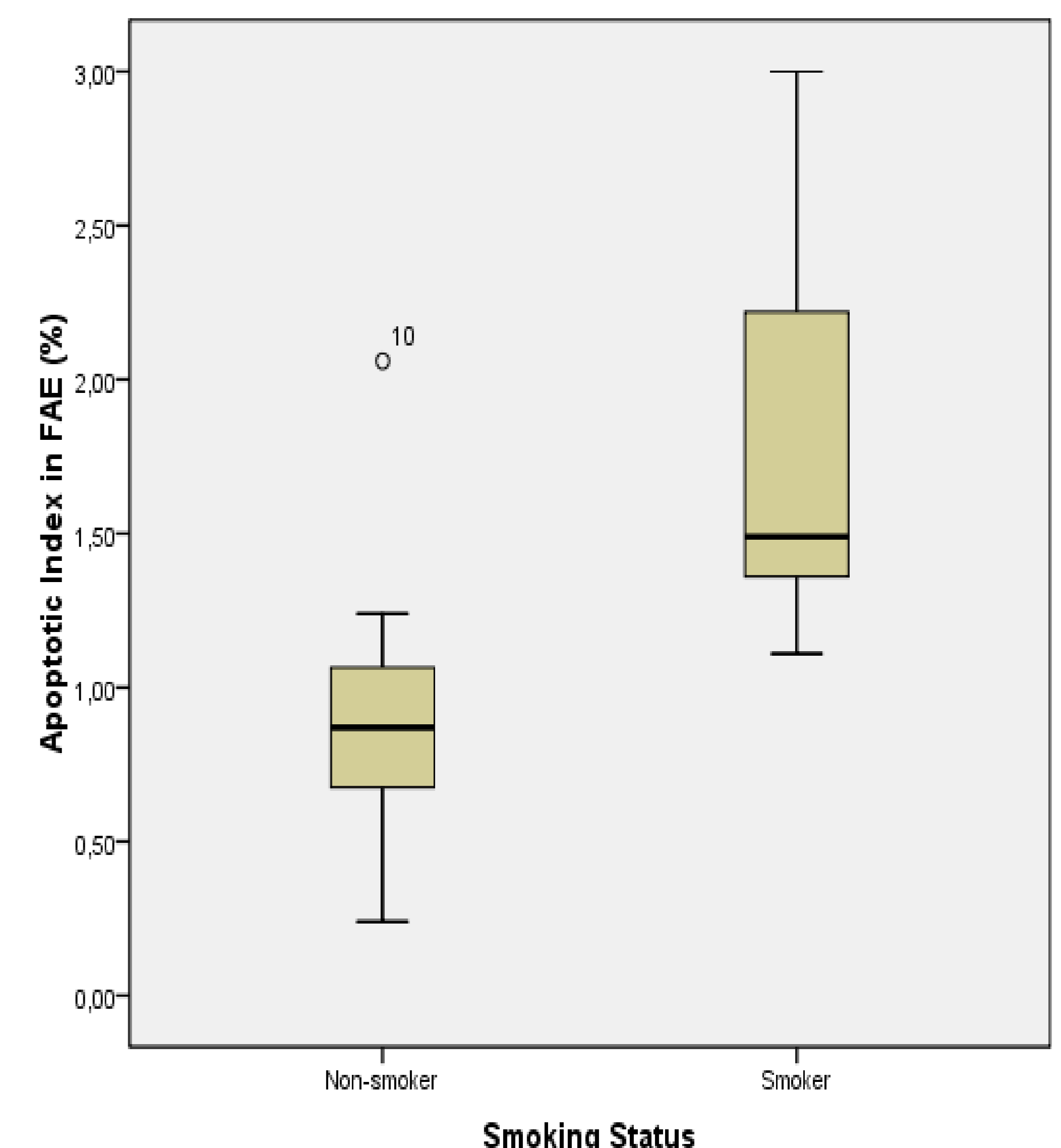


Figure 2: Boxplot demonstrating the difference in apoptotic index in smokers versus non-smokers

Conclusion

We quantified the rate of apoptosis in the FAE of Peyer's patches of smoking mice and their non-smoking siblings by caspase-3 immunohistochemical staining. An increased apoptotic index in the FAE of smoking animals was observed. Our results demonstrate that cigarette smoke induces a significant increase of apoptosis in the FAE of murine Peyer's patches and may point to a role for smoking in the pathogenesis of intestinal inflammation. Further investigation needs to clarify whether this increase in apoptosis influences normal function of the FAE.

References

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