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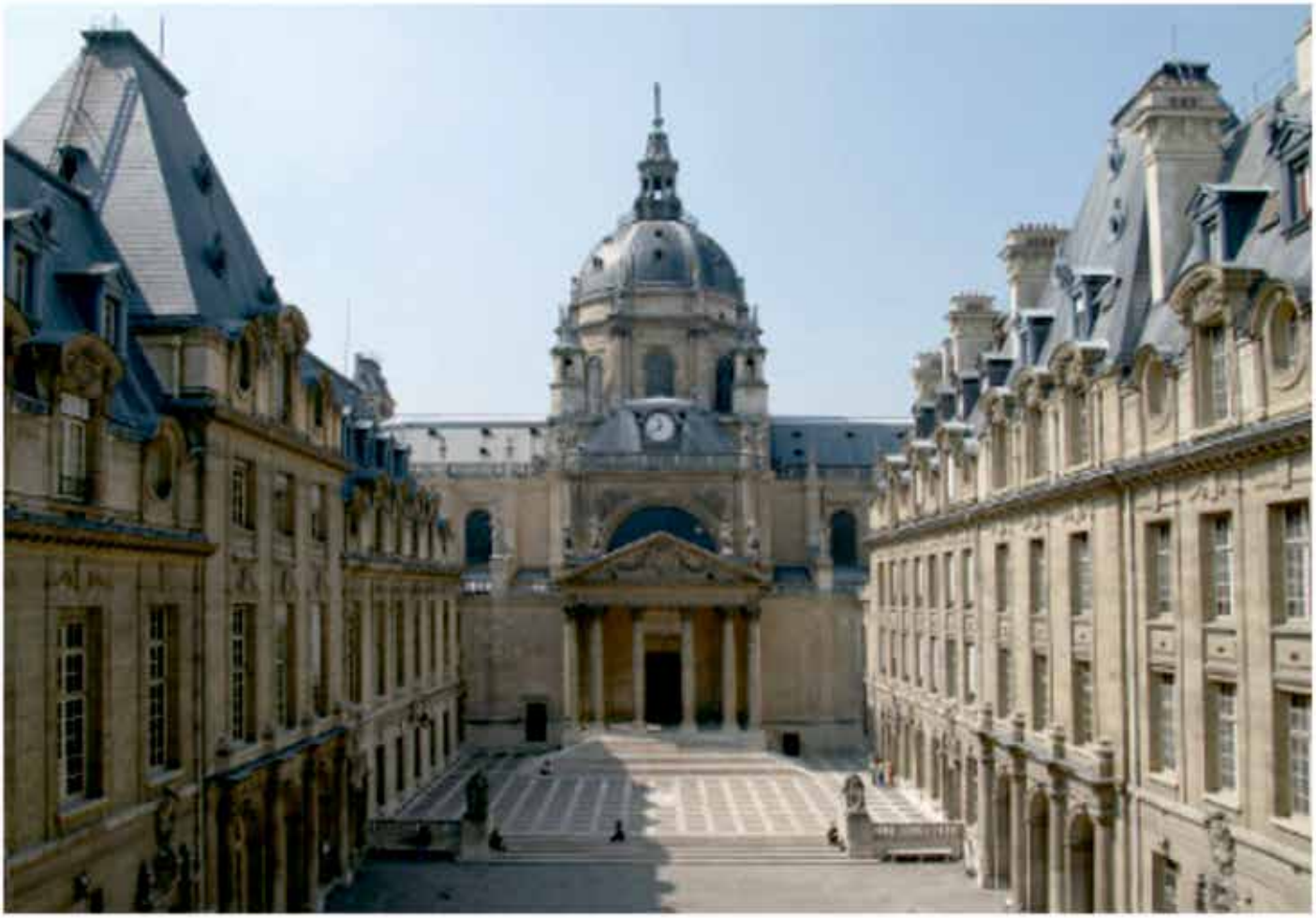

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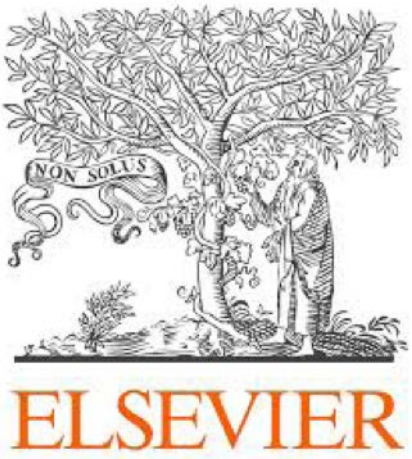

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Contents

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Morhometric characteristics of rat gingival mucosa in chronic ethanol intoxication

Abstract: The paper studies the effect of ethanol on the mucous membrane of the attached portion of gingivae. At the early stages of the experiment thickening of the total thickness of the epithelial lamina with follow-up sustainable thickening up to day 30 was noted. The total thickness of the proper lamina increases by 26.7% already on day 5 due to elevated amount of amorphous substance, persistent till the end of the observation. The minimum effect on the mean number of cell layers has been noted in the basal layer of the epithelial lamina. At the early stages of the observation lowering of the rate for acanthaceous and granular layers was found, whereas a significant increase in cell layers number was noted in the granular and corneous layers that is morphological manifestation of the epithelium defense reaction on the impact of ethanol.

Keywords: gingivae, ethanol, rats, morphometry.

Introduction

According to the WHO's investigations, the total rate for alcohol consumption per capita in Ukraine is twice higher the "dangerous" amount; alcohol abuse is responsible for more than 3.3 million deaths per year [13]. Currently rejuvenation and feminization of alcoholism is of special concern in the society [9]. The effect of ethanol on organs of nervous, uric, digestive and cardiovascular systems is fully

elucidated by the researchers [1.10-12]. However, the impact of chronic ethanol intoxication on oral mucosa remains to be understood [4].

Morphometric approach enables to detect reliable changes in organs' structural elements in stimulation, intoxication or change of condition of functioning [2].

Purpose

The paper was aimed at determining the features of metric indices of rat gingival mucosa in chronic ethanol intoxication.

Material and Methods

45 white outbred rats were involved into investigation. 5 animals were assigned into intact group who were ventricle administered with NaCl isotonic solution QID; 40 animals were assigned to experimental group who were ventricle administered with 12 mg/kg 40% ethanol (in recalculation for pure alcohol) [5].

The animals were killed under 25 mg/kg thiopental anesthesia overdose in compliance with the scheduled time periods (on days 5, 9, 12 and 30). Pieces of gingiva mucosa were put into epon-812 according to standard procedure [7]. Semi-thin sections were stained with polychrome stain [6].

The mean thickness of epithelial and proper laminae, number of layers in the basal, acanthaceous, granular and corneous layers was measured using the microscope with digital *Biorex 3* microphotohead with software, adapted to these studies. Statistical processing of morphometric data has been carried out using the Microsoft Excel [8].

Results and Discussion

It has been morphometrically found, that the mean thickness of epithelial lamina of the attached portion of gingivae of rat from the control group constituted (67,79±6,17) mcm, and (117,78±10,14) mcm for proper lamina (Table 1).

After administration of ethanol during 5 days the mean values of the epithelial lamina thickness was significantly reduced to (49,86 ± 6,71) mcm ($p < 0.05$) due to aggressive impact of ethanol on keratinocytes. Thickness rate of proper lamina of the attached portion of rat gingivae increased to (181,65 ± 5.15) mcm ($p < 0.05$) due to swelling and elevated amount of amorphous substances (Table 1).

On day 9 of the experiment, no significant increase in the mean thickness of the epithelial lamina was noted and constituted (56,34 ± 7.11) mcm. Thickness of proper lamina was tending to reduce (170,33 ± 8,64) mcm, but was significantly greater in $p<0,05$ over the control (Table 1).

Administration of ethanol during 12 days caused thickening of the gingival epithelium (79,58 ± 7.32) mcm, that was significantly greater than the value of the previous period of the experiment ($p < 0.05$), but differences were insignificant over the control value. The mean thickness of the proper lamina was tending to reduce and constituted (161,51 ± 7,42) mcm, but was greater than the value in a control group of rats (Table 1).

Up to day 30 of the observation epithelial lamina was thickening and the mean value was insignificant ($p < 0.05$) over the value on day 12 of the experiment, but significantly was 29.8% higher the control value. The mean thickness of the proper lamina was tending to reduce, but its thickness was 35% greater than the rate in the control group of rats (Table 1).

Table 1. The dynamics of metric values of epithelium and proper lamina of the attached portion of rat gingivae throughout the experiment (mcm)

Index	Control group (n=5)	Day 5 (n=10)	Day 9 (n=10)	Day 12 (n=10)	Day 30 (n=10)
Mean thickness of epithelium	67,79±6,17	49,86±6,71 *	56,34±7,11	79,58±7,32 **	88,0±2,44 *
Mean thickness of proper lamina	117,78±10,14	181, 65±5,15 *	170,33±8,64 *	161,51±7,42 *	159,01±3,36 *
Mean number of cell layers					
Basal layer	1,3±0,04	1,25±0,43	1,17±0,03 *	1,34±0,04 **	1,29±0,02
Acanthaceous layer	3,43±0,013	2,53±0,05 *	3,67±0,07 *, **	1,38±0,04 *, **	2,58±0,06 *, **
Granular layer	3,29±0,04	1,9±0,08 *	2,99±0,11 *, **	3,75±0,04 *, **	4,12±0,08 *, **
Corneous layer	2,89±0,13	3,32±0,04 *	2,16±0,07 *, **	4,42±0,16 *, **	4,91±0,05 *, **

*Notes: * - differences are significant as compared to the control (p<0,05); ** - differences are significant as compared to the previous time period of observation (p<0,05)*

While determining the mean number of cell layers in the epithelial lamina of the attached portion of gingivae of rat from the control group it was established that the value for the basal layer constituted 1.3 ± 0.04; 3.43 ± 0,013 for the

acanthaceous layer; 3.29 ± 0.04 for the granular layer; 2,89±0,13 for the corneous layer (Table 1).

It was morphometrically established that the ethanol effect during 5 days had the following outcomes: no changes in number of cell layers in the basal layer was noted; a significant reduce in the mean number of cell layers was found in the acanthaceous and granular layers from 3,43±0,013 to 2,53±0,05 and from 3,29±0,04 to 1,9±0,08, respectively, in $p<0,05$; the increase in mean number of cell layers by 14,8% ($p<0,05$) was found in the corneous layer (Table 1).

Up to day 9 of the observation the mean number of cell layers in the basal layer was reducing, indicating about the inhibition of proliferative activity of keratinocytes and significantly differed from the control (1,17±0,03 and 1,3±0,04, respectively ($p<0,05$). The mean number of cell layers in the acanthaceous layer significantly increased (by 45%) over the previous time period of the experiment and was 7% higher the control value ($p<0,05$). The value of the mean number of cell layers in the granular layer increased by 5% from 1,9±0,08 to 2,99±0,11 ($p<0,05$), but was 9% lower than the control value ($p<0,05$). A progressive reduce in mean number of cell layers from 3,32±0,04 to 2,16±0,07 and 2,89±0,13 ($p<0,05$) in the control group was noted in the corneous layer, which is the morphological proof of intensification of desquamative processes in epithelium of the attached portion of gingivae.

After 12 days from the beginning of the experiment the mean number of cell layers in the basal layer was 14,5% higher over the previous time period of the experiment, but no significant difference over the value from the control was noted (Table 1). The mean number of cell layers in the acanthaceous layer reduced and was by 60% different from the control values ($p<0,05$) and by 62% from the previous time period of the experiment ($p<0,05$). The number of cell layers in the granular layer was increasing and was 25% higher the value of the previous time period of the experiment ($p<0,05$) and 13,9% higher the control value (Table 1). A sudden increase in the number of cell layers (by 52,9%) was noted in the corneous layer, as compared with the previous time period of the experiment and was 104,6% higher the control value ($p<0,05$) (Table 1), indicating about the defense reaction of the epithelial lamina on the aggressive impact of ethanol.

On day 30 of the experiment the mean number of cell layers in the basal layer of the epithelial lamina of the attached portion of gingivae reduced, though no significant difference over the control value was noted. The values for acanthaceous

layer significantly increased by 37% ($p<0,05$), as compared with previous time period of the experiment, but was 25% less than the control value ($p<0,05$) (Table 1). While comparing with values from the control group of animals, the mean number of cell layers in the granular layer significantly increased by 25% ($p<0,05$) and was 9,8% ($p<0,05$) higher the value on previous time period of the experiment. A significant increase in the mean number of cell layers (by 11,1% ($p<0,05$) was noted in the corneous layer, as compared with previous time period of the experiment and was 69,9% ($p<0,05$) higher the control values (Table 1).

Conclusions

Administration of ethanol to rats leads to structural changes in the mucous membrane of the attached portion of gingivae, which become apparent at the early stages of the experiment by reducing of the total thickness of the epithelial lamina with the following sustainable thickening up to day 30 of the experiment. The total thickness of the proper lamina increases by 26.7% already on day 5 due to elevated amount of amorphous substances with no recovery till the end of the observation. The minimum effect on the mean number of cell layers has been noted in the basal layer of the epithelial lamina. It has been established that at the early stages of the observation lowering of the rate for acanthaceous and granular layers was found, whereas a significant increase in cell layers number was noted in granular and corneous layers that is the morphological manifestation of the epithelium defense reaction on the impact of ethanol.

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HPLC method for the determination of 3-chlorophenol as azoderivates

Abstract: Studied azoderivation reaction of 2-chlorophenoles with 4-nitrophenildiazonium cation. Investigated the optimum conditions Designed and tested method of determining the 3-chlorophenol in soil by HPLC.

Keywords: 3-chlorophenole, azoderivation reaction, method for determination, HPLC.

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ВЭЖХ метод определения 3-хлорофенола в виде азодеривата

Аннотация: Исследованы условия азодеривации 2-хлорофенола с 4-нитрофенилдиазоний катионом. Установлены оптимальные условия, диапазон определяемых концентраций.

Ключевые слова: 2-хлорфенол, реакция азодеривации, методика определения, ВЭЖХ.

В обычных условиях, 3-хлорфенол (ХФ), бесцветные игольчатые кристаллы с запахом, $t_{пл.}$ = 35°C, $t_{кип}$ = 214°C, плотность 1,258 г/см^3 (20°C) [1]. Растворим в бензоле, воде, диэтиловом эфире, лигроине этаноле.

Опасен при ингаляции, проглатывании и попадании на кожу. При ингаляции першение в горле, кашель, сонливость, головная боль; судороги, потеря сознания. При попадании внутрь – боли по ходу пищевода и в желудке. При попадании на кожу – краснота, боль, отек.

Используется для промышленного синтеза. ХФ в природной среде образуется в процессе метаболизма водных организмов, при биохимическом