

## MORPHOFUNCTIONAL ALTERATIONS OF THE ESOPHAGUS INDUCED BY CARBON MONOXIDE EXPOSURE AND POTENTIAL APPROACHES FOR THEIR CORRECTION

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**Objective:** To investigate structural and immunohistochemical alterations in the esophagus caused by prolonged carbon monoxide (CO) exposure and to assess the therapeutic potential of phytochemicals derived from *Silybum marianum* (milk thistle) and *Carthamus tinctorius* (safflower)

**Materials and Methods:** A total of 200 outbred white rats (200–250 g) were included in the experiment and randomly assigned to four groups: control; chronic CO exposure; CO + *Silybum marianum*; CO + *Carthamus tinctorius*.

Chronic CO intoxication was simulated by exposing animals to concentrations of 200–300 mg/m<sup>3</sup> (0.02–0.03%) for 1–2 hours daily, 5–6 times per week, over a period of 1–3 months to induce sustained hypoxic conditions.

After completion of the exposure period, the animals were sacrificed, and esophageal tissue samples were collected. Specimens were fixed in 10% neutral formalin, dehydrated, and embedded in paraffin. Sections (5–7 μm) were stained with hematoxylin–eosin and according to Van Gieson.

Immunohistochemical staining was performed using the streptavidin–biotin–peroxidase technique with Ki-67 as a marker of cell proliferation and Bcl-2 as an indicator of anti-apoptotic activity. Quantitative assessment of immunopositive cells was carried out per 1 mm<sup>2</sup> using morphometric analysis.

**Results:** Prolonged CO exposure resulted in marked structural deterioration of the esophageal mucosa. Epithelial thickness significantly decreased (from 45 ± 2.3 to 26 ± 1.2 nm), while edema (from 10 ± 0.6 to 20 ± 1.1 nm), inflammatory cell infiltration (from 35 ± 3 to 140 ± 9 cells/mm<sup>2</sup>), and overall inflammatory activity (from 0.5 ± 0.1 to 4.0 ± 0.2 points) were notably increased.

Administration of phytochemicals mitigated these adverse changes. In the *Carthamus tinctorius* group, epithelial thickness reached 42 ± 2.0 nm, inflammation decreased to 1.5–2.0 points, and infiltration to 60–70 cells/mm<sup>2</sup>. In the *Silybum marianum* group, epithelial thickness was 43 ± 2.1 nm, inflammation 1.3 ± 0.1 points, and infiltration 55 ± 4 cells/mm<sup>2</sup>.

**Conclusion:** Long-term exposure to carbon monoxide leads to significant morphofunctional impairment of the esophagus. Treatment with phytochemicals

based on *Silybum marianum* and *Carthamus tinctorius* effectively attenuates inflammatory and degenerative alterations. These findings highlight the potential role of plant-based therapies in protecting esophageal tissue under chronic hypoxic conditions.