Effect of *Ononis angustissima* extract on the Aluminium alterations in intestinal histology

Efeito do extrato de *Ononis angustissima* nas alterações induzidas por alumínio na histologia intestinal

Efecto del extracto de *Ononis angustissima* sobre las alteraciones inducidas por aluminio en la histología intestinal

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ABSTRACT
Ononis angustissima (Oa) is an endemic desert plant with medicinal qualities. Nevertheless, aluminum (Al) is a poisonous metal found in large quantities in nature. This investigation seeks to assess the impact of an aqueous Oa extract on adult rats intoxicated with AlCl3 at a dose of 100 mg/kg. The findings show that Al significantly reduced organ weight ($p < 0.001$), perturbed biochemical parameters, and caused histological changes in the intestine, including lymphoid tissue hyperplasia, villi erosion, and an increase in the number of goblet cells. However, treatment with the aqueous Oa extract exhibited anti-inflammatory effects, epithelial regeneration, and control of goblet cells. It is concluded that Al causes structural and metabolic alterations in intestinal tissue, but Oa has limited this damage.

Keywords: *Ononis angustíssima*, AlCl3, rats, biochemical test, intestinal histology.

RESUMO
Ononis angustissima (Oa) é uma planta endêmica de regiões áridas com propriedades medicinais. No entanto, o alumínio (Al) é um metal tóxico amplamente encontrado na natureza. Este estudo visa avaliar o impacto do extrato aquoso de Oa em ratos adultos intoxicados com $AlCl_3$ na dose de 100 mg/kg. Os resultados mostram que o Al reduziu significativamente o peso dos órgãos ($p < 0,001$), alterou parâmetros bioquímicos e causou mudanças histológicas no intestino, incluindo hiperplasia do tecido linfoide, erosões das vilosidades e aumento no número de células caliciformes. Contudo, o tratamento com o extrato aquoso de Oa demonstrou efeitos anti-inflamatórios, regeneração epitelial e controle das células caliciformes. Concluiu-se que o Al provoca alterações estruturais e metabólicas no tecido intestinal, mas o Oa limitou esses danos.

Palavras-chave: *Ononis angustíssima*, $AlCl_3$, ratos, testes bioquímicos, histologia intestinal.

RESUMEN
Ononis angustissima (Oa) es una planta endémica de zonas áridas con propiedades medicinales. Sin embargo, el aluminio (Al) es un metal tóxico que se encuentra en grandes cantidades en la naturaleza. Este estudio tiene como objetivo evaluar el impacto del extracto acuoso de Oa en ratas adultas intoxicadas con $AlCl_3$ a una dosis de 100 mg/kg. Los resultados muestran que el Al redujo significativamente el peso de los órganos ($p < 0,001$), alteró parámetros bioquímicos y causó cambios histológicos en el intestino, incluyendo hiperplasia del tejido linfoide, erosión de las vellosidades y aumento en el número de células caliciformes. Sin embargo, el tratamiento con el extracto acuoso de Oa demostró efectos antiinflamatorios, regeneración epitelial y control de las

células caliciformes. Se concluyó que el Al provoca alteraciones estructurales y metabólicas en el tejido intestinal, pero el Oa limitó estos daños.

Palabras clave: *Ononis angustissima*, $AlCl_3$, ratas, pruebas bioquímicas, histología intestinal.

1 INTRODUCTION

In recent decades, the effects of industrialism have disrupted the balance between nature and humankind, while the pollution of the environment by heavy metals, including aluminum (Al), has gained importance due to its health risks, which are still poorly assessed (Hasni, 2015). In terms of abundance, aluminum (Al) is found after oxygen and silicon in the lithosphere. It exists in soils, rocks, aerosols, and water, and its use has been described on a large scale in the construction, sanitation, and cosmetic sectors, as well as in food manufacturing, due to its excellent physicochemical properties (Sonthonnax, 2014; Esquerre, 2016; Niu, 2018).

Its bioaccessibility becomes extremely intense, especially through multiple sources and routes of exposure (Esquerre, 2016; Inan-Eroglu and Ayaz, 2018). As a result, this metal affects several organs, with the major impacts observed in neurological impairments, cardiovascular and kidney ailments, gastrointestinal disorders, and cancers (Shaw and Tomljenovic, 2013; Tair, 2016). The intestinal tract is an essential barrier that plays a crucial role in nutrient absorption, immunity, and detoxification of xenobiotics along with the liver. This makes it a target for toxic agents (Vignal et al., 2016; Jeong et al., 2020). Aluminum ingestion tends to accumulate in the intestinal mucosa, damaging gut homeostasis, regulation, permeability, and even the microflora. Its involvement in chronic inflammatory bowel diseases has also been confirmed (Vignal et al., 2016; Yu et al., 2019; Jeong et al., 2020).

Natural resources are full of beneficial virtues, whose importance has increased due to their use in agro-alimentary and pharmacological applications. These resources have always served as remedies for several illnesses, providing alternatives to synthetic compounds (Djeridane et al., 2006; Dibong et al., 2011; Rebbas et al., 2012; Ramdane, 2018; Berroukeche et al., 2024). The geographical position of Algeria offers a wide range of bioclimatic stages, supporting a rich and diverse vegetation (Emberger, 1971; Burits and Bucar, 2000; Wollenweber et al., 2003). Among these, the Fabaceae (Leguminosae) family holds significant importance as it constitutes the backbone of the food system along with cereals (Klouche, 2014). This family includes many medicinal plants, such as the genus *Ononis*, which comprises 75 species. It is traditionally known for its curative properties against skin cancer, gout, diarrhea, rheumatism, jaundice, urinary tract inflammation, kidney stones, and for its antiseptic, antimicrobial properties, or as a treatment for

wounds and eczema (Benabderahmane et al., 2014; Youcef et al., 2014; Ghribi et al., 2016; Guettaf et al., 2016).

Ononis angustissima is a Saharan endemic plant with yellow flowers, found in regions such as Guardaia, Bechar, Biskra, Boussaada, and Beni Abbas (Wollenweber et al., 2003; Bouheroum et al., 2009; Benabderahmane et al., 2014; Guettaf et al., 2016; Yerlikaya et al., 2017). Previous studies have reported its biological activities and its role as a powerful supplier of secondary metabolites (Guettaf et al., 2016; Yerlikaya et al., 2017).

In light of these data, our research investigates the effect of *Ononis angustissima* extract *in vivo* against aluminum toxicity in Wistar rats.

2 THEORETICAL FRAMEWORK

Aluminum is a known as environmental toxin that causes oxidative stress (Jeong et al., 2020), leading to significant damage on tissues, which including alterations in intestinal histology (Abu-Almaaty et al., 2019), while the intestine is the primary site of nutrient absorption, is particularly susceptible to such toxic effects. However, Ononis angustissima, as medicinal plant traditionally used for its antioxidant and anti-inflammatory properties (Yerlikaya et al., 2017)., it may alleviate these harmful effects.

3 MATERIALS AND METHODS

3.1 EXTRACTION

Ononis angustissima, the selected species, was collected in autumn 2019 during the end of the flowering stage and the beginning of the fruiting period in the region of "Chott El Chergui," located between the wilayas of Saïda and El-Bayadh (Algeria). It was identified by a taxonomist expert (Pr. Hasnaoui).
The sample was stored, and the voucher specimen, labeled P-201997, was placed in the herbarium of the Biology Department, Faculty of Sciences, University of Saïda, Algeria.
Ten grams of plant material were mixed with 100 ml of distilled water in a flask with a condenser on top, heated for 30 minutes, filtered, and the residue recovered. The volume was readjusted to 100 ml (Rebaya et al., 2014).

3.2 THE ANIMALS, HANDLING, AND HUSBANDRY CONDITIONS

The experiments were carried out on albino rats of the Wistar strain, housed in the local animal facility of the University Dr. Moulay Tahar, Saïda, Biology Department. The animals were accommodated in conventional cages with access to standard food (pellets) and water *ad libitum*.

They were maintained at a temperature of 22–23°C and a constant light cycle (12 hours of darkness and 12 hours of illumination).

3.3 EXPERIMENTATION

The animals in the experiment were categorized into the following groups:

- **Control (TE):** Rats with no treatment.
- **Intoxicated (INT):** Rats receiving aluminum chloride (AlCl3) (100 mg/kg body weight) dissolved in drinking water for 12 weeks.
- **Treated (TR):** Rats gavaged with *Ononis angustissima* extract (Eq Oa) at a dosage of 20 mg/kg body weight.
- **Intoxicated-Treated (INTT):** Rats intoxicated with AlCl3 (100 mg/kg body weight) and treated with aqueous extract (Eq Oa) at a bolus of 20 mg/kg body weight.

The rodents were weighed weekly. At the end of the experiment, the animals were anesthetized and sacrificed, and their intestines were collected for further study.

3.4 BIOCHEMICAL TESTS

Biochemical analyses of the intestinal mucosa aimed to evaluate the impact of long-term Al exposure on intestinal parameters in adult rats and verify the effect of plant therapy.

3.4.1 Total protein determination

The Bradford technique (1976) was used to determine the total protein levels of the supernatant fractions. The protein content was determined using a calibration curve established with bovine serum albumin (BSA) (0–10 µg/ml). Each assay was performed in a spectrophotometer as follows: 100 µl of a solution containing 1–10 µg of protein was mixed with 5 ml of Coomassie blue reagent, homogenized by inversion, and incubated for 5 minutes (allowing the staining to stabilize). Optical density (OD) was measured at 595 nm, with a blank tube prepared using distilled water for the standard range.

3.4.2 Total sugar determination

The total sugar content of the supernatant fractions was determined using the technique of Dubois et al. (1956). This method allows for the determination of all monosaccharides in a simple, fast, and reliable way, though it is not specific for hexoses.

In test tubes, 1 ml of the sample containing 10–70 µg of total sugars was used, followed by the addition of 1 ml of phenol solution. The tubes were carefully shaken, and 5 ml of concentrated

sulfuric acid was added in 5 seconds with a buret, dropped rapidly onto the surface of the liquid. The mixture's temperature rose to approximately 110°C. After quick stirring, the tubes were maintained at 100°C for 5 minutes.

After 30 minutes in the dark, absorbance was measured at 490 nm. The coloration remained stable for 3–4 hours in the light.

3.4.3 Alkaline Phosphatase (ALP) activity

ALP activity was measured using the Chronolab colorimetric technique. Under alkaline conditions and in the presence of ALP, p-nitrophenyl phosphate is hydrolyzed into p-nitrophenol and phosphate. The absorbance at 405 nm correlates with the ALP activity.

3.5 HISTOLOGICAL STUDY

Intestinal samples were fixed in a 10% neutral buffered formalin solution, dehydrated in graded ethanol (70–100%), clarified using xylene, and embedded in paraffin. Sections 5 µm thick were prepared and stained with hematoxylin and eosin (H&E) for microscopic observation (Suvarna et al., 2013).

3.6 STATISTICAL STUDY

The results are presented as the mean (M) of individual values, accompanied by the standard error of the mean (SEM). Comparisons were carried out using one-way ANOVA with the intoxication factor ($AlCl_3$, T).

Time-factor analysis was performed using repeated-measure ANOVA, and the statistical analysis software SigmaStat was used. Statistical significance was defined as a probability of $p < 0.05$ (SPSS Inc., Chicago, IL, USA).

4 RESULTS AND DISCUSSIONS

4.1 RESULTS

- THE YIELD EXTRACTION

The yield of the extraction was approximately 14.40%.

- EFFECT OF ALUMINUM ON BODY WEIGHT

The findings indicate that animals exposed to AlCl3 during the 12 weeks of experimentation experienced a significant decrease ($p < 0.05$) in body weight compared to the control group. In contrast, the intoxicated-treated group showed a significant increase ($p < 0.05$) (Table 01).

Table 1. Evaluation of body weight of the control, intoxicated and intoxicated-treated with (*EOa*) groups.

	TE	INT	INTT	TH
Body weight (g)	110.5 ± 3,783	97.75 ± 3,837*	106.75 ± 4,150*	111.5 ± 3,617

Values are expressed as mean ± SEM (*: p <0.05) (Source: Elaborated by the authors 2024).

- ORGAN WEIGHT

Evaluation of intestinal weights revealed significant decreases ($p < 0.001$) in the relative weight of the intestines of intoxicated rats compared to the control group (Table 02), demonstrating that aluminum (Al) has a harmful effect on organ growth. However, treatment with EOa resulted in a slight correction of this parameter.

Table. 2. Evaluation of the intestine weight of the experienced lots.

Lots	TE	INT	INTT	TH
The intestine weight (g)	10.88 ± 2,424	7.66 ± 1,607***	8.99 ± 0.189***	10.85 ± 0.431

Values are expressed as mean ± SEM (***: p <0.001) (Source: Elaborated by the authors 2024).

- BIOCHEMICAL ANALYSIS OF INTESTINAL MUCUS
  - ➢ TOTAL PROTEIN ASSAY

Statistical analysis of the results revealed a significant decrease ($p < 0.001$) in total protein content across different intestinal parts (duodenum, jejunum, ileum, colon) in the intoxicated group (Table 03). This indicates that aluminum (Al) induces a loss of protein composition. However, an increase in protein levels was observed in animals receiving EOa-based treatment.

Table 3. Intestinal mucus protein assay (µg/ml).

	Duodenum	Jejunum	Ileum	Colon
T	3.80 ± 0.31 ***	2.61 ± 1.85 ***	4.07 ± 0.97***	1.75 ± 0.23 ***
TH	3.79 ± 0.34	2.37 ± 0.60	4.32 ± 0.52	1.89 ± 0.78
INT	1.94 ± 1.20 ***	1.76 ± 0.72 ***	1.86 ± 1.47***	0.23 ± 0.78 ***
INTT	1.97 ± 0.58	1.98 ± 0.91	1.99 ± 1.02	0.87 ± 0.75

Values are expressed as mean ± SEM (***: p <0.001) (Source: Elaborated by the authors 2024).

  - ➢ TOTAL SUGARS ASSAY

The determination of total sugars in the intestinal mucus of the different groups (Table 04) statistically indicates a significant decrease ($p < 0.01$) in the total sugar content across various intestinal fragments in the groups of rats exposed to AlCl3 compared to the other groups.

Table 4. Total sugar levels in intestinal mucus (µg/ml).

	Duodenum	Jejunum	Ileum	Colon
T	123.4 ± 0.67**	67.8 ± 0.32**	177.01 ± 0.03***	45.7 ± 0.22*
TH	125.3 ± 0.13	68.17 ± 0.11	178.10 ± 0.09	44.17 ± 0.59
INT	8.52 ± 0.43**	48.68 ± 0.55 **	19.89 ± 0.32***	27.41 ± 0.21*
INTT	37.28 ± 0.74	60.29 ± 0.49	128.72 ± 0.42	57.18 ± 0.71

Values are expressed as mean ± SEM (***: p <0.01, **: p <0.01, *: p <0.05) (Source: Elaborated by the authors 2024).

➢ ALKALINE PHOSPHATASE (AP) ASSAY

The statistical results of the intestinal alkaline phosphatase assay (duodenum, jejunum, ileum, colon) (Table 05) reveal that aluminum disrupts the balance of intestinal AP by causing a decrease ($p < 0.01$) in the duodenum and jejunum and a significant increase ($p < 0.05$) in the ileum and colon in AlCl3-intoxicated rats compared to the control group. However, therapy with EOa regulated the intestinal AP levels.

Table 5. Intestinal mucus alkaline phosphatase assays

	Duodenum	Jejunum	Ileum	Colon
T	38 ± 0.07*	26 ± 0.04**	102 ± 0.11*	10 ± 1.04*
TH	36 ± 0.06	29 ± 0.05	101 ± 0.17	11 ± 0.58
INT	14 ± 0.08*	11 ± 0.05**	424 ± 0.01*	20 ± 0.06*
INTT	28 ± 0.29	23 ± 0.71	200 ± 1.07	17 ± 0.22

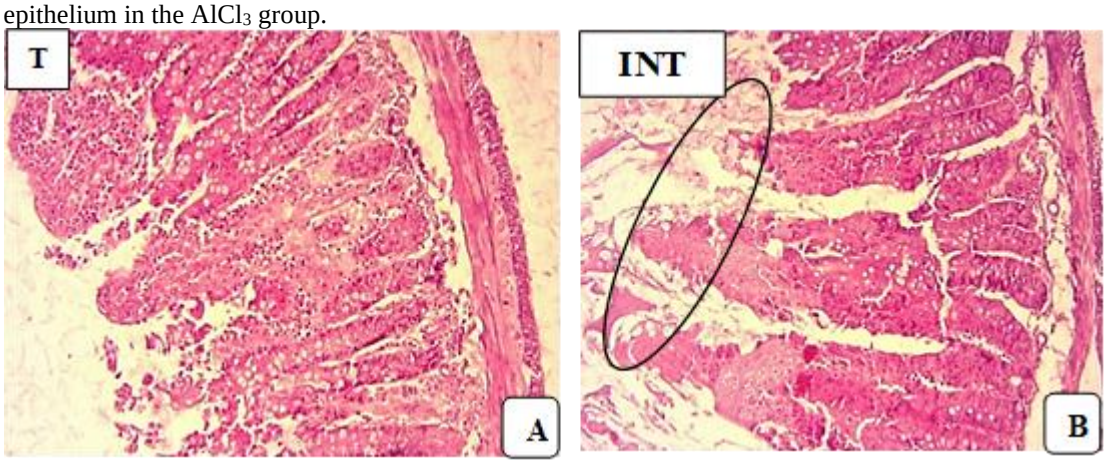
Values are expressed as mean ± SEM (**: p <0.01, *: p <0.05) (Source: Elaborated by the authors 2024).

➢ THE EFFECT OF ALCL3 ON GUT HISTOLOGY

Histological analysis of the intestine from control rats revealed a preservation of normal cellular architecture in T rats (Figure 1A), including the four cell layers: mucosa, submucosa, muscularis mucosa, and serosa, with ordinary cellular appearance (nuclei and cytoplasm). However, microscopic observation of the sections from Al-intoxicated rats revealed intense ulceration and epithelium erosion, accompanied by necrosis of enterocytes at the apex of the villi (Figure 1B), hyperplasia of goblet cells, and infiltration of inflammatory cells (Figure 2A).

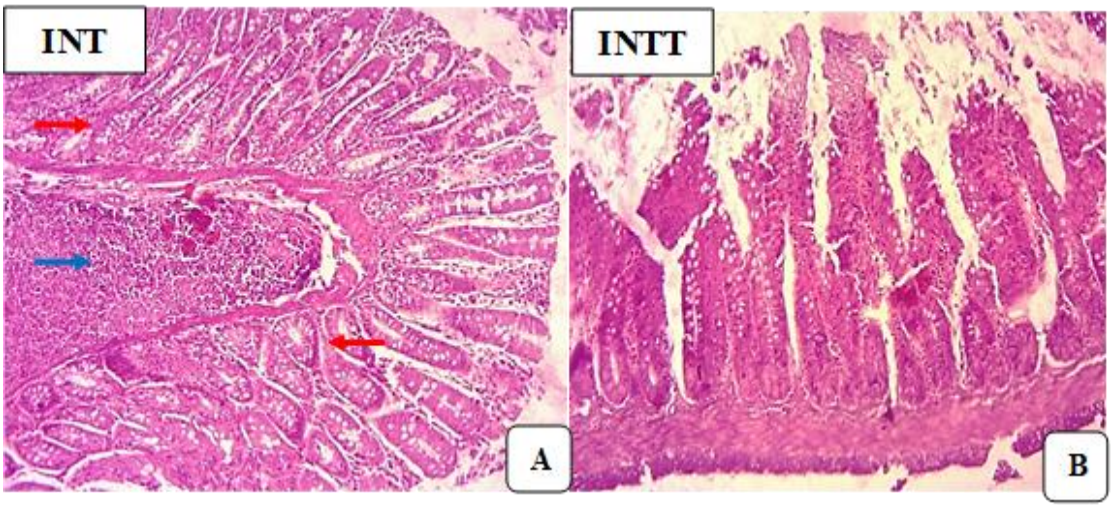
Treatment with the aqueous *Ononis angustissima* extract (INTT sections) showed an anti-inflammatory effect, regeneration of the epithelium, and a reduction in goblet cells (Figure 2B).

Figure 1. A microphotograph of the intestinal sections; A: normal cytoarchitecture in control group B: degeneration of the gut epithelium in the $AlCl_3$ group.



(Source: Elaborated by the authors 2024).

Figure 2. A microphotograph of the intestinal sections; A: hyperplasia of goblet cells (red arrow) and the inflammatory ducts of lamina propria (blue arrow) in the INT lot, B: the therapeutic effect of *EOa* at the histological level.



(Source: Elaborated by the authors 2024).

4.2 DISCUSSIONS

Metals are always present in ecosystems in various forms due to natural or industrial processes. They are constituent elements of the Earth's crust, and their dispersion in the environment is considered toxic (Elder, 1998; Gandois, 2009).

Aluminum is known for its adverse impacts on organs and their various functions due to its ubiquity (Ozkaya et al., 2010). Furthermore, a correlation has been highlighted between high doses of aluminum and impairments or pathologies, including Alzheimer's disease, Parkinson's disease, and anemia (Bekaria et al., 2002; Yokel, 2002).

Many studies have illustrated its damage to the brain, liver, and bones. Alongside this, particular attention has been paid to its toxic effects on the gut (Yu et al., 2016; Jeong et al., 2020).

This study focuses on the species *Ononis angustissima*, a Saharan endemic plant with significant therapeutic properties. It represents an excellent source of secondary metabolites (Djeridane et al., 2010; Ghribi et al., 2015; Guettaf et al., 2016).

The results of this study highlighted the extraction yield for an aqueous decoction of the aerial parts of *Ononis angustissima*, which was 14.40% in dry weight. However, these results differ from those found by Ghettaf et al. (2016), who reported 9.75%. An important extraction rate was recorded in another species from the same Fabaceae family (*Genista sahareae*), around 10.33% (Ghettaf et al., 2016). These fluctuations in yield are attributed to several factors, including pedoclimatic conditions such as temperature, relative humidity, and soil type. Additionally, variations may arise from the period and location of harvest, extraction techniques, drying duration, grind granulometry, time, temperature, solvent nature and polarity, and the solvent-to-mass ratio (Boukhatem et al., 2010; Koné et al., 2017).

The effect of aluminum on body weight was detected through recorded results, which indicate that animals exposed to aluminum (100 mg/kg, orally) showed a significant decrease in body weight compared to controls. Moreover, aluminum administration reduced the weight of various intestinal parts. This aligns with the findings of Yeh et al. (2009) and Mohammad et al. (2015), who also recorded weight reductions in the liver, kidneys, and testes of aluminum-exposed rats.

Several studies have reported reduced body weight gain caused by aluminum exposure (Abu-Almaaty et al., 2019; Balgoon, 2019; Hosseini et al., 2020). Notably, studies by Kaizer et al. (2005) attribute the decline in weight and relative organ mass to aluminum accumulation in these tissues (Anand et al., 2012; Hosseini et al., 2020).

The results of Abu-Almaaty et al. (2019) demonstrated that aluminum exposure disrupts gastrointestinal homeostasis and absorption, causing organ damage, which subsequently reduces protein synthesis and leads to weight loss.

According to Bekhedda et al. (2020), this decrease is probably due to the anorectic effect of aluminum through a deficiency in serotonin and dopamine production. These neurotransmitters are directly involved in appetite regulation and satiety control.

This study also indicates that aluminum exposure causes an imbalance in intestinal mucosa constituents, including a decrease in total proteins and sugars and a disturbance in alkaline phosphatase (ALP) levels. This suggests that chronic exposure to aluminum induces intestinal lesions and depletes the biochemical composition.

Yu et al. (2016) and Jeong et al. (2020) reported that aluminum generates oxidative stress and inflammation, leading to intestinal epithelium dysfunction and decreased tight junction protein expression. This increases intestinal permeability (Yu et al., 2016), causing a loss of cellular content due to tissue destruction and degeneration. The leakage of biomolecules is evidenced by high plasma levels of total proteins and sugars (Al-Hashem, 2009; Hadi et al., 2009; Al-Hazmi et al., 2021; Atanu and Okpe, 2021; Marza Hamza et al., 2021).

Intestinal alkaline phosphatase (ALP) plays a crucial role in maintaining intestinal homeostasis by regulating pH and absorbing calcium and phosphorus. It also has immune functions, including detoxifying xenobiotics and infectious agents, attenuating inflammation, and preserving the microbiota (Chen et al., 2010; Fawley and Gourlay, 2016; Lallès, 2019; Liu et al., 2021). In this study, aluminum toxicity impaired ALP activity, likely due to its inhibitory effects (Marza Hamza et al., 2021). Furthermore, ALP, a sensitive biomarker for metal exposure, reflects tissue damage and alterations (Hadi et al., 2009). Aluminum also causes zinc (Zn) deficiency (Mohammad

et al., 2015; Mahmoud et al., 2020). As ALP activity depends on zinc, its depletion may further contribute to ALP dysfunction (Charuel, 1971; Ensinger et al., 1978).

Histopathological analysis revealed that aluminum exposure significantly disrupted intestinal homeostasis, compromised enterocyte viability, and caused inflammation. Similar anomalies were reported by Buraimoh et al. (2012), Pineton de Chambrun et al. (2014), Eltahawy et al. (2016), Vignal et al. (2016), and Yu et al. (2019).

Cunat et al. (2000) and Gad El-Hak et al. (2020) found that aluminum accumulates in the intestine. Buraimoh et al. (2012) observed that aluminum chloride alters the small intestine's histology in Wistar rats, causing mucosal degeneration. Eltahawy et al. (2016) highlighted that Paneth cells, which synthesize antimicrobial peptides, are particularly sensitive to aluminum toxicity. Aluminum exposure also changes intestinal cytoarchitecture, leading to crypt and lamina propria hyperplasia, increased goblet cell activity, and villi erosion.

Hao et al. (2022) noted that aluminum disrupts tight junctions, promotes lymphoid tissue hyperplasia, and alters enzymatic activity. Its accumulation causes dysbiosis. Pineton de Chambrun et al. (2014) concluded that aluminum damages the intestinal epithelium and induces inflammation by stimulating inflammatory cytokines and granuloma formation (Eisenbarth et al., 2008).

Studies by Esquerre (2016) revealed aluminum's procarcinogenic effects, increased intestinal permeability, and altered microbiota. These changes disrupt the cell cycle, promote epithelial cell necrosis, and induce apoptosis and inflammation (Yu et al., 2019). Jeong et al. (2020) and Sieg et al. (2022) also linked oxidative stress to aluminum's intestinal toxicity.

The genus *Ononis* (Fabaceae) has gained attention for its antipyretic, antimicrobial, anti-inflammatory, antiseptic, cytotoxic, and hypoglycemic properties (Ghribi et al., 2016; Guettaf et al., 2016; Sayari et al., 2016; Dénes et al., 2017; Mezrag et al., 2017; Yerlikaya et al., 2017; Stojković et al., 2020).

Administering *Ononis angustissima* aqueous extract led to body weight recovery, restored biochemical parameters, and repaired aluminum-induced intestinal lesions. This restorative effect is attributed to the plant's richness in polyphenols, flavonoids, and terpenes (Bouheroum et al., 2009; Mezrag et al., 2013; Ghribi et al., 2015; Laoufi et al., 2017; Benmerzoug et al., 2020).

Flavonoids, terpenes, and alkaloids are known for their antioxidant and anti-inflammatory properties. These compounds modulate inflammatory cascades, regulate enteroendocrine cells, and maintain intestinal barrier integrity (Vezza et al., 2016; Li and Weigmann, 2022). Isoflavones such as Formononetin-7-O-β-D-glucoside exert anti-inflammatory effects by reducing inflammatory mediator synthesis (Zhang et al., 2020).

Polyphenols alleviate inflammation by inhibiting nitric oxide production, suppressing prostaglandins and cytokines, and promoting beneficial microbiota (Li and Christman, 2020; Da Fonseca Machado et al., 2021). As metal chelators, they protect biomolecules, stimulate antioxidant enzyme expression, and neutralize free radicals (Basra et al., 2020; Mahfuz et al., 2021).

5 CONCLUSION

This study demonstrates that aluminum chloride ($AlCl_3$) exposure induces significant structural and biochemical damage in the intestine, including weight loss, disruption of intestinal homeostasis, decreased levels of total proteins, sugars, and alkaline phosphatase activity, and also severe histological alterations such as villi erosion, enterocyte necrosis, and inflammatory responses, for instanve, these findings highlight the profound toxic effects of aluminum on intestinal homeostasis.

However, the administration of *Ononis angustissima* (Oa) extract showed promising protective effects, while the extract improved body weight, restored biochemical parameters, and mitigated histological damage by promoting epithelial regeneration, reducing inflammation, and regulating goblet cell activity. These effects are attributed to the plant's richness in polyphenols, flavonoids, and terpenes, which are known for their antioxidant, anti-inflammatory, and cytoprotective properties.

Our research emphasizes the potential of *Ononis angustissima* as a natural therapeutic agent against aluminum-induced intestinal damage, it may pave the way for developing plant-based interventions to combat heavy metal toxicity, particularly in regions with significant environmental aluminum exposure, and future studies could explore the detailed mechanisms underlying Oa's protective effects and its application in other models of heavy metal toxicity.

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