



## **Comparative Study of Some Histological Features in the Pancreas of Some Mammals Having Different Feeding Habits**

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### **Abstract**

**Background:** Mammals' endocrine and exocrine systems depend heavily on the pancreas, which aids in functions including digestion and blood sugar management. The pancreas may have differences in histological structure that reflect adaptations to various dietary habits, even though its functional responsibilities are comparable across species. These differences can shed light on the functional and evolutionary connections between pancreatic shape and nutrition. **Objective:** This study aims to compare the histological features of the pancreas, body weight, and blood glucose level in some mammals with different feeding habits. **Methodology:** Ten female pangolins, rabbits, and rats each were studied. Body weight and fasting blood glucose levels were measured. The pancreas was harvested post-sacrifice and analysed histologically using Hematoxylin and Eosin, Masson's Trichrome, Von Gieson, and Gomori aldehyde fuchsin stains to evaluate endocrine and exocrine structures. **Results:** Rabbits had the highest body (1975±66.2) and blood glucose levels (662.9±6.153), followed by pangolins (1327±48.01)(116.6±6.806) and rats (215.3±6.648) (121.1±6.873). Pangolins had the heaviest pancreas in absolute terms (1.930±0.068), while rats showed the highest relative pancreatic weight (0.0050±0.0) p value < 0.01. Histologically, pangolins exhibited compact acinar architecture with indistinct islets of Langerhans. Rabbits showed loosely organised acini with poorly defined islets, while rats had clearly delineated islets and well-structured lobules. **Conclusion:** The observed structural and physiological differences in the pancreas across species reflect dietary specialisation and metabolic demands. Pangolins exhibit a predominantly exocrine pancreas adapted to a protein-rich insectivorous diet, while rats and rabbits demonstrate mixed pancreatic features aligned with their respective feeding habits.

**Keywords:** pancreas, islet cells, pangolin, acinar cells, comparative histology, endocrine pancreas

### **Introduction**

The pancreas is a glandular organ found in vertebrates' endocrine and digestive systems. It is situated behind the stomach in the human abdominal cavity. (Mihoc et al., 2024). This endocrine gland produces a number of vital hormones that are released into the bloodstream, such as insulin, glucagon, somatostatin, and pancreatic polypeptide. (Al-Suhaimi et al.,

2022). The pancreas is also a digestive organ, secreting pancreatic juice containing digestive enzymes that assist in the digestion and absorption of nutrients in the small intestine. These enzymes help to further break down the carbohydrates, proteins, and lipids in the chyme (Moini & Ferdowsi, 2024). The evolution of the mammalian pancreas is closely tied to dietary specialization. Insectivores such

as pangolins exhibit exocrine-dominant pancreases suited for protein-rich, carbohydrate-poor diets, with indistinct islets of Langerhans (Makanya et al., 2015; Lemelin et al., 2008). Herbivores display enhanced enzymatic diversity and endocrine prominence to regulate postprandial glucose surges (Cheeke, 1987; Mohammed, 2019). Omnivores like rats show balanced exocrine and endocrine components, reflecting dietary versatility (Kumar & Prasad, 2021). Across vertebrates, compact islet organization in mammals represents an evolutionary refinement associated with homeothermy and increased metabolic demands (Slack, 1995; Da Silva, 2018).

Among insectivorous mammals, the pangolin (*Phataginus spp.*) offers a particularly intriguing model. Pangolins feed almost exclusively on ants and termites, a diet rich in proteins but low in carbohydrates. Their narrow dietary range suggests a highly specialized digestive system with distinct structural features, particularly within the pancreas. Studies have indicated that insectivores tend to possess compact, enzyme-rich exocrine tissues but may show reduced development of endocrine structures due to minimal dietary glucose intake (Makanya et al., 2015). Yet, detailed histological data on the pangolin pancreas remain limited due to conservation concerns and restricted access to specimens (Archer, 2024).

Herbivores like the domestic rabbit (*Oryctolagus cuniculus*) rely heavily on fermentable carbohydrates and fibrous plant material. Their digestion depends not only on mechanical and microbial breakdown in the hindgut but also on pancreatic enzymes that assist in polysaccharide and protein degradation (Cheeke, 1987; Lambert et al., 2004). Consequently, rabbits are expected to possess well-developed endocrine components for managing glucose influx, although histological studies suggest that islets may be less distinct without immunohistochemical labeling

(Mohammed, 2019; Ateia et al., 2024). Their pancreatic architecture typically displays loosely packed acinar cells with visible ductal structures and collagenous septa separating lobules (Hale & Dayan, 2010).

Rats (*Rattus norvegicus*), commonly used in laboratory settings, serve as a reference species for both metabolic and anatomical studies. As omnivores, they consume a diet containing both animal and plant matter, necessitating flexible digestive and endocrine responses. Histological analyses consistently show prominent islets of Langerhans and well-defined lobular exocrine organization in rat pancreas—features that facilitate dynamic regulation of blood glucose and digestive enzyme output (Kumar & Prasad, 2021; Kierszenbaum & Tres, 2020). These attributes make rats an ideal baseline for comparative analyses involving species with more specialized feeding patterns.

Structural variation in the pancreas is not limited to the balance of acinar and islet cells; connective tissue organization also reflects functional adaptations. Collagen distribution, ductal architecture, and lobular integrity influence pancreatic resilience, secretory efficiency, and disease susceptibility. Histochemical stains such as Hematoxylin and Eosin (H&E), Masson's Trichrome, Von Gieson, and Gomori aldehyde fuchsin provide critical insights into these structural differences (Jones et al., 2008; Abunasef, Amin, & Abdel-Hamid, 2014). For instance, Masson's trichrome highlights collagen-rich fibrous septa, while Gomori staining distinctly reveals the endocrine islets, enabling comprehensive evaluation of both exocrine and endocrine components across species.

Despite the known general functions of the pancreas across mammals, few comparative studies integrate morphometric, biochemical, and histological parameters to elucidate species-specific adaptations systematically. Particularly, the pangolin remains underrepresented in comparative anatomy literature, even as its conservation status draws increasing attention

(Ampitan *et al.*, 2021; Schaake, 2022). A better understanding of pangolin biology, including organ structure, can inform veterinary care, conservation biology, and potentially even zoonotic disease research, given the species' ecological interactions and human-wildlife conflict dynamics (Yan *et al.*, 2022).

Therefore, the present study aimed to compare the pancreatic architecture, body weight, and blood glucose levels of pangolins, rabbits, and rats—representing insectivorous, herbivorous, and omnivorous dietary niches, respectively. We hypothesized that differences in feeding strategy would be reflected in the structural complexity and distribution of exocrine and endocrine tissues within the pancreas. By integrating quantitative analysis and detailed histological evaluation, this work contributes to our understanding of functional morphology. It offers a foundation for future investigations into pancreatic adaptation, evolution, and physiology in mammals.

## **Materials and Methods**

### **Animals**

Ten female pangolins were bought from the Asejire Forest Reserve, and ten female rabbits and rats were also purchased from the animal house of the College of Health Sciences, Osun State University. The care and handling of the animals conform to the rules and guidelines of the animal rights committee of Osun State University, Oshogbo, Nigeria. They were all housed in the animal house for 24 hours before being taken to the animal house laboratory unit for sacrifice. The rats and rabbits were sacrificed using cervical dislocation, while the pangolin was sacrificed using a stun gun.

### **Sample Selection**

The study utilized a purposive sampling approach to include three mammalian species with distinct dietary habits—insectivorous pangolins (*Phataginus spp.*), herbivorous rabbits (*Oryctolagus cuniculus*), and omnivorous rats (*Rattus norvegicus*). A total of thirty healthy adult females (ten per species)

were selected to minimize variability due to sex- and age-related differences in pancreatic structure and metabolism.

### **Body Weight Measurement**

The body weight of each animal was recorded prior to sacrifice using a calibrated digital electronic balance (Mettler Toledo, Switzerland; sensitivity  $\pm 0.1$  g). Each animal was weighed individually after an overnight fast to minimize variations due to gut content, and results were expressed in grams (g) (Guyton & Hall, 2021; Gonzalez *et al.*, 2020).

### **Harvesting of the organs**

This was done in the animal house laboratory unit, after which the animals were sacrificed and blood samples of each animal were collected for blood glucose and other biochemical analysis. The pancreas was weighed, washed in normal saline, and fixed in formalin for tissue processing.

### **Blood Glucose Measurement**

Following humane sacrifice—via cervical dislocation for rabbits and rats and stun gun for pangolins—fasting blood samples were collected via cardiac puncture into heparinized tubes. Fasting blood glucose levels were measured immediately using a glucometer (Accu-Chek® Active, Roche Diagnostics), which employs an enzymatic electrochemical detection principle based on glucose dehydrogenase-mediated oxidation of glucose, with the resulting electron transfer quantified as an electrical signal proportional to glucose concentration with compatible glucose test strips, following the manufacturer's protocol. All animals had been fasted overnight (~12 hours) before measurement to ensure baseline glucose levels (Benninger & Kravets, 2022; Kawasaki *et al.*, 2002).

### **Pancreatic Weight Determination**

Immediately after sacrifice, a midline abdominal incision was made, and the pancreas was carefully dissected from surrounding

tissues. The gland was blotted dry using filter paper to remove excess moisture and weighed using a high-precision analytical balance (sensitivity  $\pm 0.001$  g). Pancreatic weight was recorded in grams and documented for each specimen (Makanya *et al.*, 2015; Kierszenbaum & Tres, 2020).

### Relative Pancreatic Weight

Relative pancreatic weight (RPW) was calculated to normalize pancreatic mass relative to total body mass, allowing for interspecies comparison. The RPW was expressed as a percentage using the formula:

$$\text{The relative weight of pancreas} = \frac{\text{MWP (mgs)}}{\text{MFWA (gms)}} \times 100 \text{ --- (eqn 1)}$$

MWP = Mean weight of pancreas (mg),

MFWA = Mean Final weight of the animal (g)

This method is widely employed in comparative anatomical studies to account for allometric scaling of organ size to body mass (Slack, 1995; Kumar & Prasad, 2021).

### Histological procedures

The pancreases were harvested from the sacrificed rats after dissection and were weighed and washed with saline. The specimens were stretched on filter paper and fixed in 10% buffered formalin (pH 7.4). The fixed specimens were sliced, processed, and embedded in paraffin blocks. The blocks were cut into 4  $\mu\text{m}$  paraffin sections by a rotator microtome. The sections were stained with Hematoxylin and Eosin (H&E), Masson trichrome, Gomori Aldehyde, and Von Gieson (Abunasef *et al.*, 2014).

## Results

### Body weight

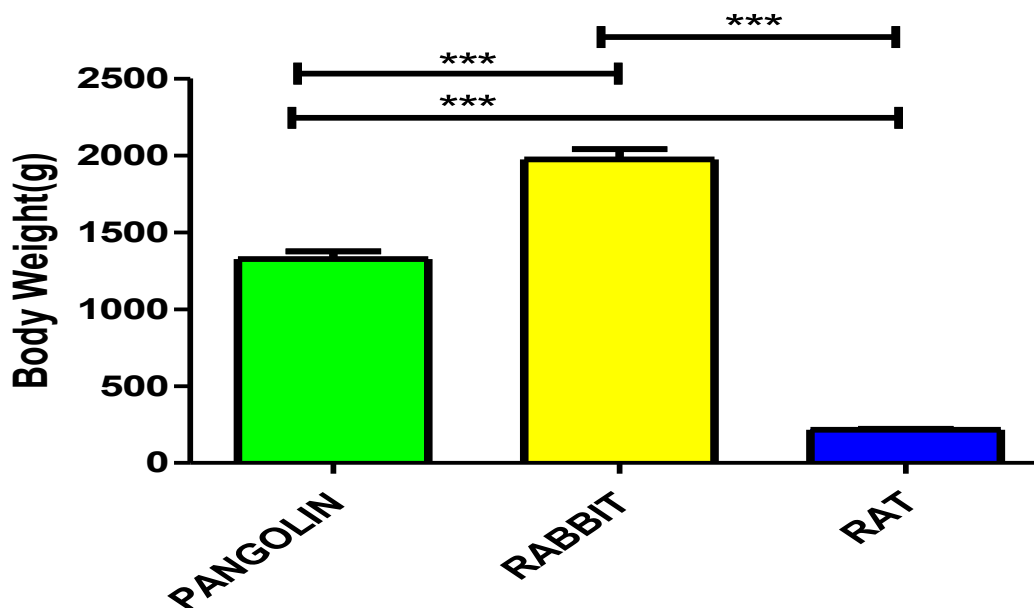
It was visually evident that rabbits had the highest average body weight, followed by pangolins, while rats had the lowest. To confirm the significance of these observed differences, a one-way analysis of variance (ANOVA) was performed. The results showed a highly significant difference in mean body weights

among the groups, with a P value of less than 0.0001. The F-statistic was 352.9, indicating a strong effect of species on body weight. The coefficient of determination ( $R^2$ ) was 0.9632, suggesting that over 96% of the variability in body weight was attributable to differences between the species rather than random variation. Bartlett's test for homogeneity of variances returned a corrected statistic of 27.71 with a P value also less than 0.0001, indicating that the assumption of equal variances among groups was violated. Despite this, Bonferroni's multiple comparison test was employed to determine which pairs of species had significant differences in body weight. The pairwise comparisons revealed that all three species differed significantly from each other. Specifically, rabbits were significantly heavier than pangolins (mean difference = -648.0 g;  $t = 9.671$ ), and pangolins were significantly heavier than rats (mean difference = 1112 g;  $t = 16.59$ ). The greatest difference was observed between rabbits and rats, with a mean difference of 1760 g and a t-value of 26.26. All comparisons had confidence intervals that did not cross zero, confirming statistical significance at the 0.05 level.

From the visual data, it is evident that rabbits had the highest blood glucose levels, while pangolins and rats had comparatively lower and similar values. Statistical analysis using one-way ANOVA confirmed that the differences among the groups were highly significant, with a P value less than 0.0001. The F value was 14.88, and the R-squared value was 0.5244, indicating that over 52% of the variation in blood glucose levels was due to differences between the species. Bartlett's test for equal variances produced a non-significant result ( $P = 0.9391$ ), suggesting that the assumption of homogeneity of variance was met and validating the use of ANOVA for the comparison. Post hoc analysis using Bonferroni's multiple comparison test revealed a significant difference between the pangolin and rabbit groups, with rabbits having markedly higher glucose levels

(mean difference = -46.30 mg/dL,  $P < 0.001$ ). Similarly, a significant difference was found between rabbits and rats, with rabbits again showing higher values (mean difference = 41.80

mg/dL,  $P < 0.001$ ). However, no significant difference was observed between pangolins and rats (mean difference = -4.50 mg/dL,  $P > 0.05$ ).



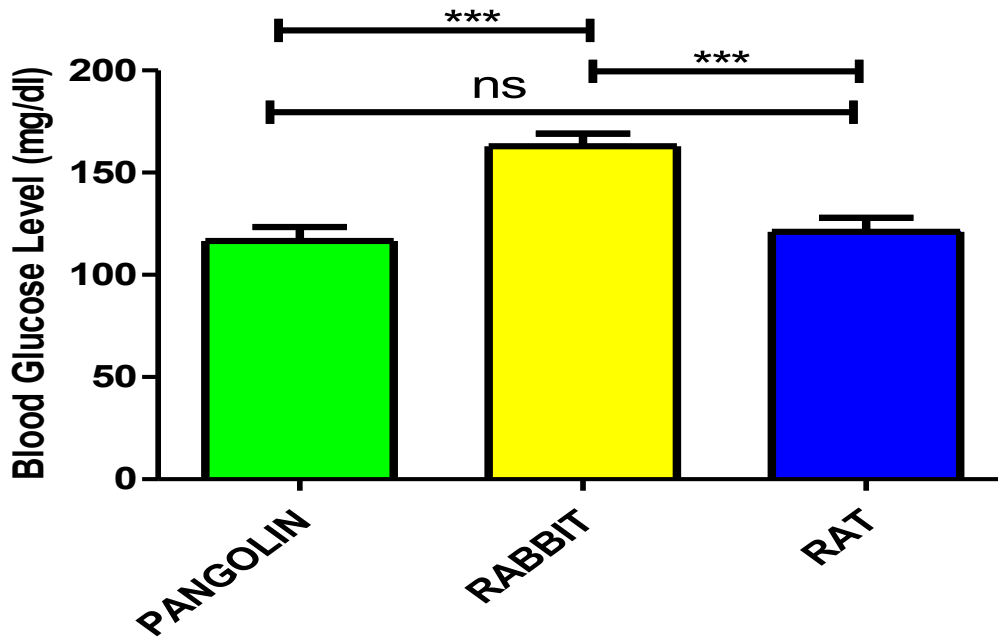
**Figure 1:** Bar graph showing the mean fasting blood glucose levels ( $\pm$  SEM) in Pangolin, Rabbit, and Rat. Rabbits exhibited significantly higher glucose levels compared to Pangolins and Rats ( $***P < 0.001$ ), while no significant difference was observed between Pangolin and Rat (ns).

### Blood Sugar Level

Bar graph showing the mean fasting blood glucose levels ( $\pm$  SEM) in pangolin, rabbit, and rat. Rabbits exhibited significantly higher glucose levels compared to pangolins and rats ( $***P < 0.001$ ), while no significant difference was observed between pangolins and rats (ns). Bar chart showing the mean body weights (in grams) of pangolins, rabbits, and rats. Rabbits had the highest average weight, followed by pangolins, with rats having the lowest. Error bars represent standard deviation. Significant differences were observed between all groups ( $***P < 0.001$ , ANOVA with Bonferroni post hoc test). The bar graph illustrated that pangolins had the highest average pancreatic weight, followed by rabbits, while rats had the lowest. To assess the significance of these differences, a one-way analysis of variance (ANOVA) was conducted. The results showed a highly significant difference in pancreatic weights among the three groups, with a P value less than

0.0001. The F value was 72.83, and the  $R^2$  value was 0.8436, indicating that approximately 84% of the variability in pancreatic weight was due to differences between species rather than within-group variation. Bartlett's test for equal variances was also performed to check the assumption of homogeneity of variance, which is important for ANOVA. The test returned a P value of 0.1096, indicating no significant difference in variances across the groups. Therefore, the assumption of equal variances was upheld, validating the use of ANOVA in this case. Further analysis using Bonferroni's multiple comparison test revealed that all pairwise differences between species were statistically significant. Pangolins had significantly heavier pancreases than rabbits, with a mean difference of 0.55 grams, while the difference between pangolins and rats was even more pronounced at 0.852 grams. Rabbits also had significantly heavier pancreases than rats, with a mean difference of 0.302 grams. All these differences

had confidence intervals that did not include zero, reinforcing their statistical significance.



**Figure 2:** Mean fasting blood glucose levels ( $\pm$  SEM) in pangolins, rabbits, and rats. Rabbits showed significantly higher levels than pangolins and rats ( $***P < 0.001$ ), with no difference between pangolins and rats (ns).

The bar graph illustrated that pangolins had the highest average pancreatic weight, followed by rabbits, while rats had the lowest. To assess the significance of these differences, a one-way analysis of variance (ANOVA) was conducted. The results showed a highly significant difference in pancreatic weights among the three groups, with a P value less than 0.0001. The F value was 72.83, and the R<sup>2</sup> value was 0.8436, indicating that approximately 84% of the variability in pancreatic weight was due to differences between species rather than within-group variation. Bartlett's test for equal variances was also performed to check the assumption of homogeneity of variance, which is important for ANOVA. The test returned a P value of 0.1096, indicating no significant difference in variances across the groups. Therefore, the assumption of equal variances was upheld, validating the use of ANOVA in this case. Further analysis using Bonferroni's multiple comparison test revealed that all pairwise differences between species were statistically significant. Pangolins had significantly heavier pancreases than rabbits, with a mean difference of 0.55 grams, while the difference

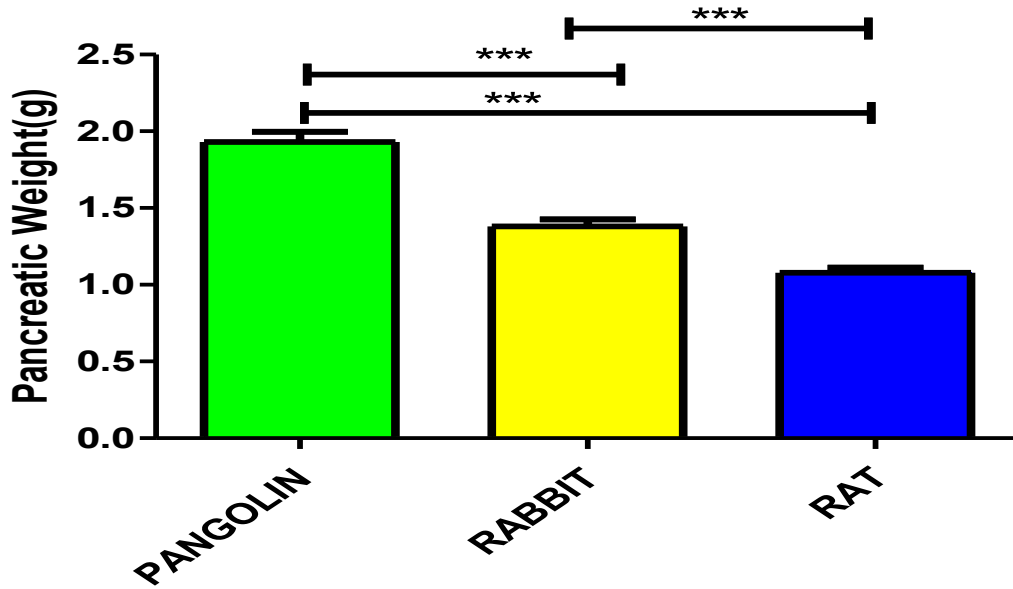
between pangolins and rats was even more pronounced at 0.852 grams. Rabbits also had significantly heavier pancreases than rats, with a mean difference of 0.302 grams. All these differences had confidence intervals that did not include zero, reinforcing their statistical significance.

#### Relative Pancreatic Weight

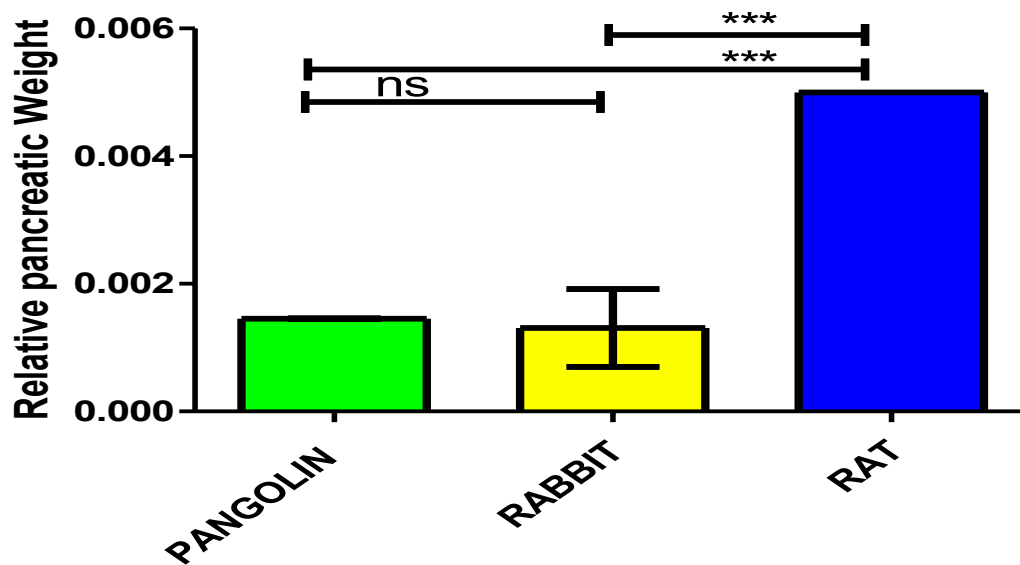
The bar chart illustrates that rats exhibited the highest relative pancreatic weight, followed by rabbits and pangolins, which showed similar values. The statistical analysis confirmed these observations. A one-way analysis of variance (ANOVA) revealed a highly significant difference among the groups, with a P value less than 0.0001 and an F value of 35.23. The R-squared value of 0.7230 indicates that approximately 72.3% of the variation in pancreatic weight is attributable to differences between the species. To validate the assumption of homogeneity of variances, Bartlett's test was performed and returned a non-significant result, indicating that the variances among the groups were statistically equal. Further pairwise comparisons were carried out using

Bonferroni's multiple comparison test. There was no significant difference in relative pancreatic weight between pangolins and rabbits ( $P > 0.05$ ). However, rats had significantly higher pancreatic weights compared to both pangolins and rabbits ( $P < 0.001$  for

both comparisons). Specifically, the mean difference between pangolins and rats was  $-0.003548$ , and between rabbits and rats was  $-0.003691$ , with confidence intervals that excluded zero, confirming the significance of these differences.



**Figure 3:** Bar chart showing the mean pancreatic weights (in grams) of pangolins, rabbits, and rats. Pangolins had the highest pancreatic weight, followed by rabbits and rats. Error bars represent standard deviation. All group differences were statistically significant ( $***P < 0.001$ , one-way ANOVA with Bonferroni post hoc test).



**Figure 4:** Graph showing the relative pancreatic weight (mean  $\pm$  SEM) in pangolin, rabbit, and rat. Rats exhibited significantly higher pancreatic weight compared to pangolins and rabbits (\*\*P < 0.001), while no significant difference was observed between pangolins and rabbits (ns).

### Histology

**Gomori staining** was employed to differentiate the endocrine and exocrine components of the pancreas, particularly highlighting the islets of Langerhans and surrounding acinar tissue across pangolin, rabbit, and rat specimens. In the **pangolin**, the islets of Langerhans were not clearly identifiable. They stained with similar intensity to the surrounding acinar tissue, offering minimal or no contrast. The exocrine acinar cells displayed intense staining and were densely and tightly packed, reflecting a compact and organized glandular structure. In the **rabbit**, the islets of Langerhans were present but less distinct than those observed in the rat. They showed moderate to light staining, with borders that were poorly demarcated from the surrounding tissue. The acinar cells in the rabbit pancreas were more loosely arranged, resulting in a less compact architecture. This gave the tissue a more open appearance compared to the denser structure seen in the pangolin. The **rat** pancreas exhibited the most prominent islets of Langerhans. These islets were large, clearly defined, and stained densely at the center, indicating high cellularity. The separation between endocrine and exocrine tissue was distinct. The exocrine component was well organized into distinct lobules with visible ductal structures, reflecting a highly structured and functional pancreatic architecture.

Histological examination of the pancreas from the pangolin, rabbit, and rat using hematoxylin and eosin (H&E) staining revealed distinct structural features across the three species. In the **pangolin**, the pancreas displayed a clearly defined lobular arrangement, with fine connective tissue septa demarcating individual lobules. The acinar cells were densely packed and contained basophilic nuclei, indicative of active protein synthesis. Although the islets of Langerhans seem present, they were not distinctly separated from the surrounding exocrine tissue. Overall, the pangolin pancreas appeared compact and highly cellular, characterized by well-preserved acini reflecting a dense and functional exocrine component. The **rabbit** pancreas exhibited a highly organized lobular structure, with lobules arranged in distinct circular patterns. The acinar cells

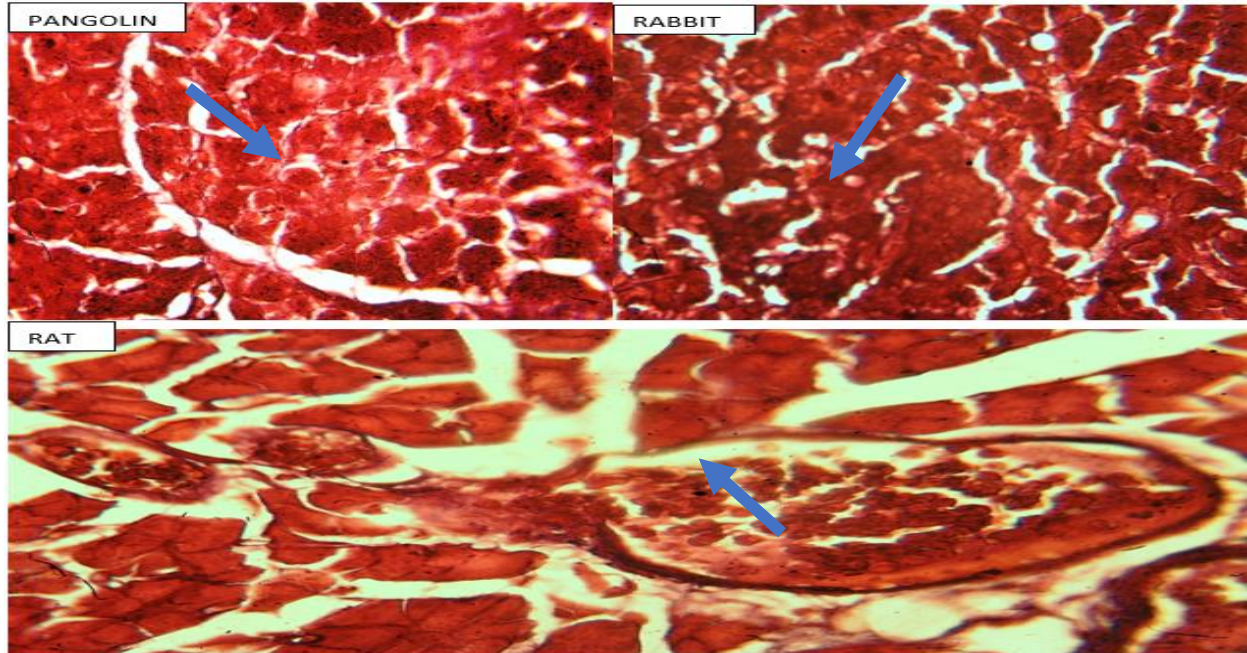
were uniform in appearance and often displayed central duct-like structures. The islets of Langerhans were not prominently visible, likely embedded within the eosinophilic exocrine matrix, making them less distinguishable under standard magnification. The circular clustering of acini and the pronounced lobular architecture suggested a unique structural organization compared to the pangolin. In the **rat**, the pancreas showed a very clear lobular architecture, with broader connective tissue septa distinctly separating the lobules. Acinar cells were well organized, featuring prominently stained basophilic nuclei and lightly eosinophilic cytoplasm. Although the islets of Langerhans were not easily identifiable at the observed magnification, they are expected to be located between the lobules. The rat pancreas demonstrated a well-structured exocrine component with evident cellular detail, suggesting high secretory activity.

Masson's trichrome staining was utilized to assess the structural organization of the pancreas in pangolin, rabbit, and rat, with particular focus on the delineation of lobules, acinar tissue composition, and the distribution of connective tissue. In the **pangolin**, the pancreatic tissue displayed prominently defined lobules, separated by thin fibrous septa stained green, indicative of collagen presence. The acinar tissue stained intensely red, reflecting a well-developed and compact exocrine component. Moderate amounts of collagen were observed within the interlobular spaces and around blood vessels, suggesting a balanced connective tissue framework that supports glandular architecture.

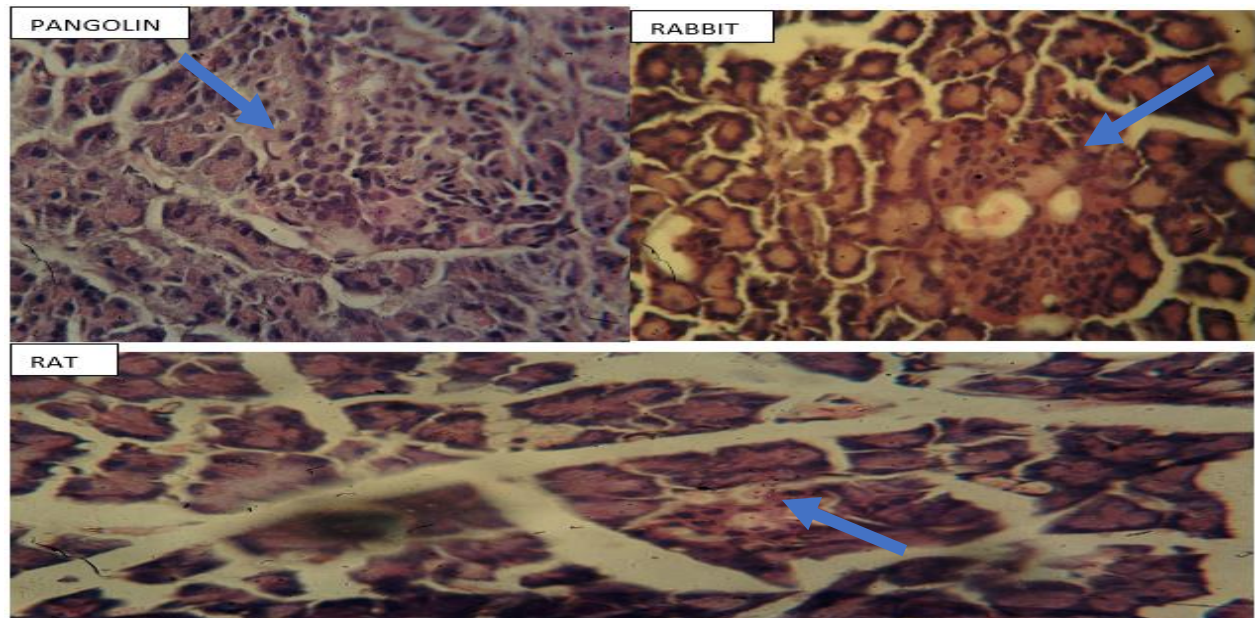
The **rabbit** pancreas exhibited more loosely arranged lobules, although fibrous septa clearly demarcated these. The acinar tissue showed dense eosinophilic staining (appearing red to pink), but with greater separation between lobules compared to the pangolin. Notably, collagen deposition was more pronounced, with abundant green-stained fibers particularly concentrated around ducts and blood vessels, suggesting a more fibrous or connective tissue-rich environment. In the **rat**, the pancreas demonstrated a highly organized structure, characterized by numerous small lobules distinctly separated by broad fibrous septa. The acinar cells

were uniform and densely packed, exhibiting strong red cytoplasmic staining. Extensive collagen deposition was evident, especially surrounding the

vasculature and ductal systems, as indicated by the intense green staining of connective tissue.

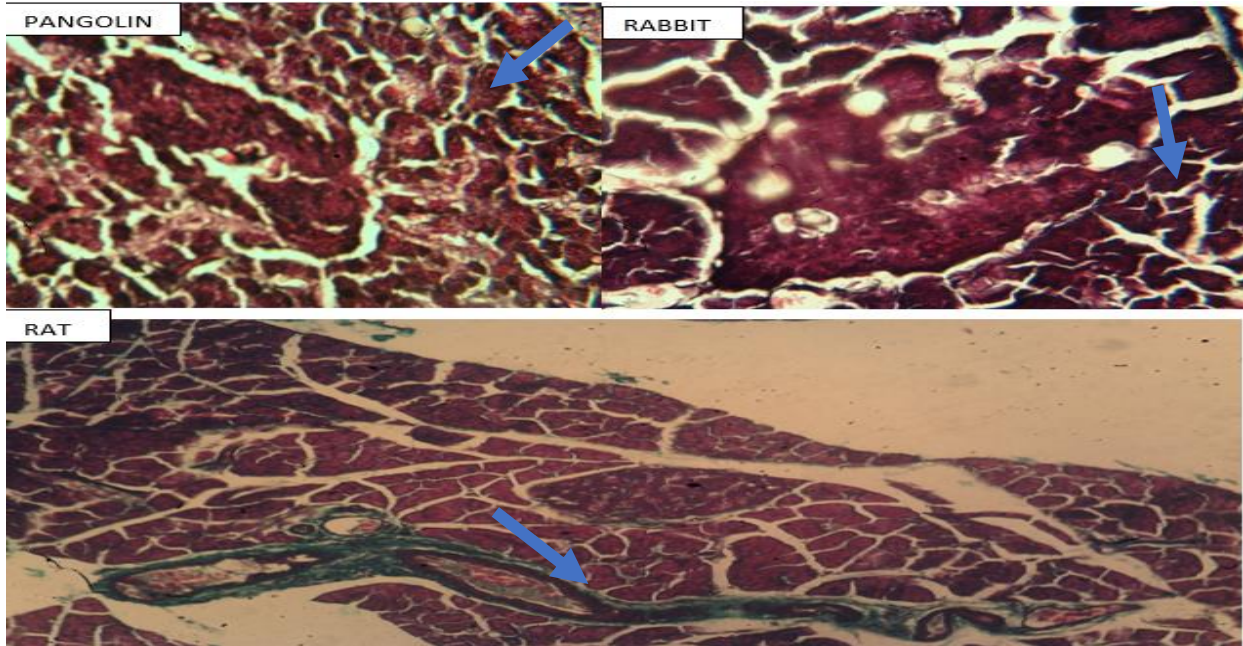


**Plate 1:** Gomori-stained pancreatic sections showing lightly stained islets and dense acini in pangolin, faint islets with loose acini in rabbit, and prominent, well-defined islets with organized exocrine lobules in rat. (X 400)

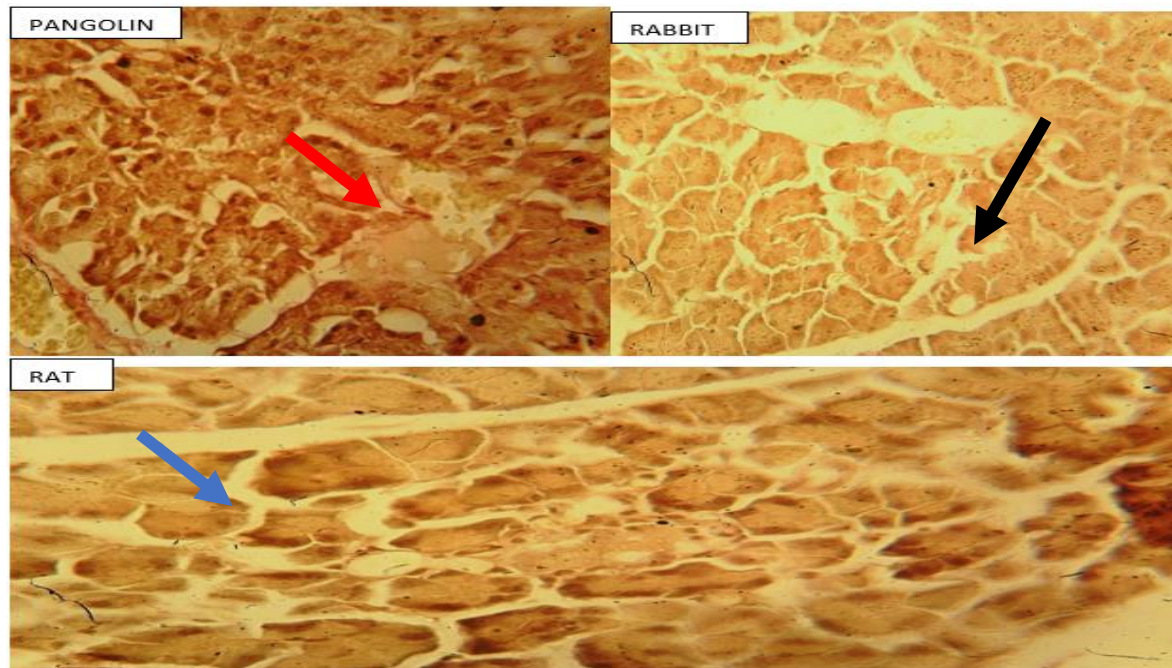


**Plate 2:** Histological sections of the pancreas from pangolin, rabbit, and rat (H&E stain) (x 400) show species-specific architecture. The pangolin pancreas appears compact with dense acinar cells and indistinct

islets. The rabbit shows circular lobular patterns with uniform acini and obscured islets. The rat displays well-defined lobules with broad septa and organized acinar cells, though islets are not clearly visible.



**Plate 3:** Masson's trichrome-stained sections of the pancreas from pangolin, rabbit, and rat reveal species-specific connective tissue organization. In pangolins, lobules are compact with moderate collagen (green) in septa and around vessels. Rabbit pancreas shows more fibrous septa and abundant collagen around ducts. The rat pancreas exhibits broad septa with extensive collagen deposition and densely packed acinar tissue (reddish), indicating a highly structured exocrine architecture (x 400).



**Plate 4:** Von Gieson staining of pangolin, rabbit, and rat pancreas shows species-specific structure. Pangolin has compact lobules with prominent collagen (red). Rabbit shows loosely arranged lobules with sparse collagen. The rat displays well-defined lobules, moderate collagen, and dense acinar cells (yellow)(x 400).

Von Gieson staining was applied to pancreatic tissue sections from pangolin, rabbit, and rat to highlight differences in lobular organization, collagen distribution, and acinar cell architecture. In the **pangolin**, the pancreas exhibited well-organized lobules, clearly separated by distinct red-stained connective tissue septa. The presence of collagen fibers was prominent, particularly in the interlobular spaces, as evidenced by the deep red staining. The exocrine acinar cells were densely packed and stained a vibrant yellow, indicating well-preserved cytoplasmic detail and a tightly organized glandular structure. In the **rabbit**, the lobular architecture appeared less defined compared to the pangolin. Lobules were larger, more loosely arranged, and separated by thinner connective tissue. The red staining in interlobular areas was sparse, suggesting lower collagen content. The acinar cells stained pale yellow and were loosely packed with increased spacing between them, giving the tissue a less compact appearance. The **rat** pancreas displayed highly developed lobular architecture, with numerous small lobules clearly delineated by fibrous septa. The connective tissue showed moderate to strong red staining, indicating a well-developed collagen framework surrounding the lobules. Acinar cells stained yellow and were densely packed within the lobules.

### **Discussion**

This comparative study of the pancreas across pangolin, rabbit, and rat species reveals significant interspecies variation in morphological, histological, and physiological parameters. The observed differences in body weight, pancreatic mass, and histoarchitecture underscore the evolutionary and functional adaptations of each species to its ecological niche, diet, and metabolic needs.

Body weight differed significantly among species, with rabbits exhibiting the highest mean body weight, followed by pangolins and then rats. The high F-value and  $R^2$  ( $F = 352.9$ ;  $R^2 = 0.9632$ ) indicate that species identity accounts for the majority of variability in body mass, consistent with their known physiological differences and life strategies (Kumar & Prasad, 2021). Interestingly, while pangolins had

a lower overall body weight than rabbits, they possessed the highest absolute pancreatic weight, suggesting an organ size disproportionate to body mass—a finding that could be attributed to dietary specialization. Contrastingly, rats had the highest relative pancreatic weight, suggesting a higher pancreatic investment relative to their body mass. This aligns with their omnivorous diet and rapid metabolic rate, which demand both robust exocrine enzyme production and endocrine regulation for energy metabolism (Gonzalez *et al.*, 2020). The significant difference in blood glucose levels, with rabbits showing the highest values, reflects variation in glucose regulation. Rabbits' herbivorous diets, high in fermentable fibers and complex carbohydrates, may stimulate greater glucose absorption and insulin response, contributing to this observation (Cheeke, 1987; Lambert *et al.*, 2004).

Histological assessment using H&E staining revealed clear lobular organization in all species, but varying degrees of acinar cell density and islet visibility. The pangolin pancreas demonstrated a dense exocrine structure, with acini tightly packed and lobules separated by delicate connective septa. Although islets of Langerhans were indistinct under both H&E and Gomori staining, this may reflect either poor histochemical contrast or inherently small and sparse islets, consistent with reports in other insectivores (Makanya *et al.*, 2015). This compact architecture and high exocrine cellularity may be an adaptation to a protein-rich, low-carbohydrate diet consisting mainly of ants and termites, necessitating potent enzymatic activity (Lemelin *et al.*, 2008). In rabbits, the pancreas exhibited loosely packed acini and more prominent interlobular spaces, especially under Masson's trichrome and Von Gieson staining. The relative invisibility of islets under routine staining might suggest their integration within a diffuse exocrine matrix or a smaller islet-to-acinar ratio. However, the higher blood glucose levels suggest functionally competent endocrine activity, which may not be histologically prominent without immunolabeling (Gonzalez *et al.*, 2020). Rats demonstrated the most distinct pancreatic organization. Islets of Langerhans were large and clearly visible, especially under Gomori staining,

suggesting a well-developed endocrine function. This is consistent with their need for tight glucose regulation due to their high metabolic turnover and frequent feeding behavior. Furthermore, the acinar tissue was well-organized into distinct lobules with visible ductal structures, indicating a highly structured exocrine component likely capable of adjusting enzyme output to variable dietary inputs (Kierszenbaum & Tres, 2020).

Masson's trichrome and Von Gieson stains highlighted interspecies differences in the collagen framework. Pangolins showed moderate but distinct collagen distribution, mainly around blood vessels and ducts, supporting the compact glandular structure. Rabbits had looser lobular integrity and more widespread collagen deposition, particularly around ducts, possibly reflecting connective tissue remodeling or age-related changes (Hale & Dayan, 2010). In contrast, the rat pancreas exhibited thick fibrous septa with moderate collagen deposition, indicative of structural reinforcement for a functionally active organ (Sleight & Potter, 2015). These differences in connective tissue composition might influence both pancreatic resilience and susceptibility to fibrotic or inflammatory conditions. For instance, excessive collagen around ducts in rabbits could impair exocrine outflow in pathological states, while dense septa in rats could isolate damage but also limit regenerative capacity (Slack, 1995).

The histological and morphometric findings suggest evolutionary adaptations shaped by dietary and metabolic demands. Pangolins' specialized myrmecophagous diet likely necessitates a pancreas geared toward high-efficiency protein digestion with limited reliance on insulin-mediated glucose regulation—explaining their indistinct islets and large exocrine mass. Rabbits, as hindgut fermenters, require carbohydrate-digesting enzymes and a stable glucose-insulin axis to manage fermentable substrates, even though their pancreatic structure appears less compact. Rats, omnivorous and opportunistic feeders, display a versatile pancreatic design that balances both endocrine and exocrine functions efficiently. These interspecies differences reinforce the importance of considering both ecological context and evolutionary history in comparative anatomical studies. The findings also emphasise the limitations of relying solely on conventional histology to assess endocrine function—future studies employing immunohistochemistry (e.g., insulin, glucagon, or

somatostatin labelling) or gene expression profiling could provide more precise characterisation (Kawasaki *et al.*, 2002; Heller *et al.*, 2005).

### **Conclusion**

The present study highlights species-specific adaptations in pancreatic structure and function among pangolins, rabbits, and rats. These differences, evident in both gross and microscopic features, reflect evolutionary pressures related to diet, metabolism, and ecological niche. While pangolins exhibit a predominantly exocrine pancreas suited for protein digestion, rabbits and rats show varying balances between endocrine and exocrine components. This comparative insight provides a basis for using these species in translational models and emphasizes the necessity of integrating histological, biochemical, and molecular tools for a holistic understanding of organ function.

### **Recommendations**

Future studies should apply molecular profiling and enzyme assays to clarify functional differences. Integrating histology with dietary and metabolic data will deepen understanding of pancreatic adaptations across mammals.

### **Professional Implications of the Study**

This study enhances understanding of species-specific pancreatic adaptations, offering valuable reference data for veterinary pathologists, wildlife biologists, and comparative anatomists. The findings support improved diagnostic interpretation, diet formulation, and health monitoring in both domestic and wild mammals, while informing conservation strategies for endangered species such as the pangolin.

### **Limitations of the Study**

The study was limited by a lack of molecular and enzymatic analyses, reducing the functional correlation of histological findings. Additionally, the cross-sectional design did not capture age-related or seasonal variations in pancreatic structure and function.

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