Guidance Document on Amphibian Thyroid Histology Part 2: Approach to reading studies, diagnostic criteria, severity grading, and atlas

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Table of Contents

List of Tables 3
List of Figures 4
I. General guidelines for reading studies 5
IA. Pathologist qualifications 5
IB. Approach to reading studies 6
1C. Selection of Histologic Sections for Evaluation 6
II. Diagnostic Criteria and Severity Grading 6
IIA. Diagnostic Criteria 6
Core Criteria 7
Additional Qualitative Criteria 7
IIB. Severity Grading 8
General severity grading scale 8
III. Reference Atlases 12
IIIA. General normal appearance of *X. laevis* thyroid microanatomy 13
IIIB. Developmental series of normal *X. laevis* thyroid microanatomy – H&E 14
IIIC. Reference atlas of diagnostic criteria and severity grades 15
IIIC1. Thyroid gland hypertrophy – general range of changes 15
IIIC2. Thyroid gland atrophy - general range of changes 20
Grade 0 – Thyroid gland atrophy 21
Grade 1 – Thyroid gland atrophy 22
Grade 2 – Thyroid gland atrophy 23
Grade 3 – Thyroid gland atrophy 24
IIIC3. Follicular cell hypertrophy – general range of changes 25
Grade 0 - Follicular cell hypertrophy 26
Grade 1 - Follicular cell hypertrophy 27
Grade 2 - Follicular cell hypertrophy 28
Grade 3 - Follicular cell hypertrophy 29
IIIC4. Follicular cell hyperplasia 30
Grade 0 – Follicular cell hyperplasia 30
Grade 1 – Follicular cell hyperplasia 31
Grade 2 – Follicular cell hyperplasia 32
Grade 3 – Follicular cell hyperplasia 33
IIIC5. Colloid quality 34
Homogenous 34
Foamy appearance 35
Granular colloid 36
Tinctorial quality of colloid 37
IIIC6. Miscellaneous findings 38
Eosinophilic droplets: 38
Karyomegaly 38
Midline thyroid follicles 39
Interfollicular pigment 40
IIIC7. Artifacts 41
Colloid fall-out 41
IV. Data Reporting 42
Reference List 43
Appendix A: Histopathology Reporting Tables 44
Core Criteria 44
Additional Criteria 45
Narrative Descriptions 46

List of Tables

Table 1. Severity grading scheme for thyroid gland hypertrophy. 10
Table 2. Severity grading scheme for thyroid gland atrophy..... 10
Table 3. Severity grading scheme for follicular cell hypertrophy. 11
Table 4. Severity grading scheme for follicular cell hyperplasia. 11

List of Figures

Figure 1. Normal *X. laevis* thyroid glands…. 13
Figures 2-5. Developmental series of normal *X. laevis* thyroid microanatomy, 48 hours post-stage 54, 96 hours post-stage 54, 144 hours post-stage 54, 192 hours post-stage 54. 14
Figures 6-9. General ranges of thyroid gland hypertorphy 16
Figure 10. Grade 0 – Thyroid gland hypertrophy 16
Figure 11. Grade 1 – Thyroid gland hypertrophy 17
Figure 12. Grade 2 – Thyroid gland hypertrophy 18
Figure 13. Grade 3 – Thyroid gland hypertrophy. 19
Figures 14-17. General ranges of thyroid gland atrophy 21
Figure 18. Grade 0 – Thyroid gland atrophy. 21
Figure 19. Grade 1 – Thyroid gland atrophy. 22
Figure 20. Grade 2 – Thyroid gland atrophy 23
Figure 21. Grade 3 – Thyroid gland atrophy s. 24
Figures 22-25. General ranges of follicular cell hypertrophy 25
Figure 26. Grade 0 – Follicular cell hypertrophy.. 26
Figure 27. Grade 1 – Follicular cell hypertrophy. 27
Figure 28. Grade 2 – Follicular cell hypertrophy. 28
Figure 29. Grade 3 – Follicular cell hypertrophy. 29
Figure 30. Grade 0 – Follicular cell hyperplasia. 30
Figure 31. Grade 1 – Follicular cell hyperplasia 31
Figure 32. Grade 2 – Follicular cell hyperplasia. 32
Figure 33. Grade 3 – Follicular cell hyperplasia. 33
Figure 34. Grade 3 – Follicular cell hyperplasia – high magnification 33
Figure 35. Colloid quality - homogenous 34
Figure 36. Colloid quality - foamy. 35
Figure 37. Colloid quality - granular. 36
Figure 38. Colloid quality – heterogenous tinctorial quality 37
Figure 39. Miscellaneous findings – eosinophilic droplets. 38
Figure 40. Miscellaneous findings – karyomegaly. 38
Figure 41. Miscellaneous findings – midline thyroid follicles 39
Figure 42. Miscellaneous findings – interfollicular pigment. 40
Figure 43. Artifacts – colloid fall-out. 41

Objective:

The goal of this document is to facilitate the standardization of the histopathologic analysis of thyroid tissues from *Xenopus laevis* to fulfill the needs of the Amphibian Metamorphosis Assay. Standardization of reading practices, diagnostic criteria, severity grading and data reporting and compilation will be addressed in an effort to maximize comparability between pathologists and to reduce bias.

This document draws from the guidance provided for the "Histopathology guidelines for Phase 1B of the OECD Fish Screening Assay for EDC's" and the guidance given in the general protocol for the Phase 2 Amphibian Metamorphosis Assay, from pathologists experienced reading toxicologic pathology studies in amphibians, the scientific literature, and from the OECD Amphibian Metamorphosis Assay thyroid histopathology consultation held in Washington, DC 2006.

This guidance document is divided into four sections:

I. General guidelines for study reading practices

II. Diagnostic criteria and severity grading

III. Reference atlas of normal microanatomy of *X. laevis* thyroid glands and an atlas of core diagnostic criteria with examples of severity grades

IV. Data recording and compilation.

I. General guidelines for reading studies

The purpose of this section is to provide general guidance for the light microscopic evaluation of thyroid gland tissue sections to improve diagnostic consistency among pathologists.

IA. Pathologist qualifications

Studies are to be read by individuals experienced in reading toxicologic pathology studies, and who are familiar with normal *X. laevis* thyroid histology, with thyroid gland physiology, and with general responses of the thyroid gland to agonists or antagonists. Pathologists may be board certified (e.g. American College of Veterinary Pathologists, The European Centre of Toxicologic Pathology, or other certifying organizations), however certification is not a requirement as long as the pathologist has obtained sufficient experience with, and knowledge of, amphibian thyroid histology and toxicologic pathology. Technicians should not be used to conduct readings due to the subtle nature of some changes and the need for subjective judgments based on past experience.

It is recognized that there is a limited pool of pathologists with the necessary training and experience that are available to read the thyroid histopathology for the amphibian metamorphosis assay. If an individual has toxicological pathology experience and is familiar with thyroid histology in other species, he/she may be trained to read the

amphibian assay. If pathologists with little experience are used to conduct the histopathological analysis, informal peer review may be necessary.

IB. Approach to reading studies

Pathologists are to read the studies non-blinded (i.e. with knowledge of the treatment group status of individual tadpoles). However, it is expected that any potential compound-related findings will be re-evaluated by the pathologist in a blinded manner prior to reporting such findings, when appropriate. Certain diagnostic criteria, such as thyroid gland hypertrophy or atrophy, cannot be read in a blinded manner due to the diagnostic dependence on control thyroid glands. As a rule, treatment groups should be evaluated in the following order: Control, High-dose, Intermediate-dose, and Low-dose.

It is suggested that the pathologists be provided with all available information related to the study prior to conducting their readings. Information regarding developmental stage at sacrifice, gross morphologic abnormalities, mortality rates, and general test population performance and health are useful for pathologists to provide comprehensive reports and to aid in the interpretation of findings. For a more comprehensive discussion of standard reading approaches for toxicologic pathology studies, please refer to the Society of Toxicologic Pathology Best Practices for reading toxicologic histopathology studies [1].

1C. Selection of Histologic Sections for Evaluation

Examination of two sections is sufficient for evaluation of the thyroid lesions that may present in the amphibian metamorphosis assay. These two sections should be selected from a minimum of five sections, they should be selected for maximum cross sectional amount of representative thyroid tissue present, the absence of artifacts in the tissue section, and they should be at least two step sections apart, if possible. If, for some reason, the five sections initially evaluated have prominent, though not consistent changes, up to three sections should be evaluated.

II. Diagnostic Criteria and Severity Grading

The purposes of this section are to provide common technical “language” and to describe an approach to severity grading of lesions.

IIA. Diagnostic Criteria

Histopathology is a descriptive and interpretive science, and therefore somewhat subjective. However, histopathologic evaluations of the same study by any qualified pathologist should identify the same treatment-related findings [1]. Therefore, we aim to define the diagnostic criteria that will likely be encountered during the histopathologic analysis of the amphibian metamorphosis assay.

A consolidated set of diagnostic criteria follow. These criteria are based on pathologists’ experience with the amphibian metamorphosis assay and known changes of the thyroid gland in response to chemical exposure, however novel findings that are

exposure-related shall also be reported. These criteria were not necessarily chosen based on biological significance and it is not implied that other lesions that may be found in the thyroid gland are biologically less significant.

The criteria below have been divided into two sections: 1. Core criteria, and 2. Additional criteria. The core criteria are severity graded on a numerical scale. The additional criteria are either graded on a numerical scale, or are qualitatively described. The additional criteria need only be addressed if they represent exposure-related findings.

Core Criteria:

1. **Thyroid gland hypertrophy/atrophy**: Increases (hypertrophy) or decreases (atrophy) in the overall size of the thyroid gland are consequent of changes in follicular cell size and number. The severity of either hypertrophic or atrophic observations is to be graded on an overall, general appearance of the thyroid gland. Because the diagnosis of hypertrophy or atrophy is dependant on a comparison to controls, it is necessary to establish the normal variability of thyroid gland sizes in control tadpoles prior to making determinations on thyroid gland size in dose groups.

2. **Follicular cell hypertrophy**: Hypertrophic follicular cells, defined as tall columnar cells, are to be graded based on the percentage of the cells exhibiting this feature. It is recognized that follicular cell hypertrophy may present as a generalized lesion and interpreted thus. Because normal amphibian thyroid glands show heterogeneity in follicular cell shape, ranging from squamous to tall columnar, severity is determined by the change in percentage of cells exhibiting tall columnar structure.

3. **Follicular cell hyperplasia**: Follicular cell hyperplasia is diagnosed when there is follicular cell crowding, stratification (multiple layers), or papillary infolding of single or multiple layers of follicular cells. The severity grading scheme for follicular cell hyperplasia is based on the percentage of follicles that exhibit hyperplasia, and/or the percentage of tissue that is affected.

Additional Qualitative Criteria:

1. **Follicular lumen area (previously colloid area)**: Lumenal area can be reduced or increased. Severity of effects on lumenal area is to be graded based on the generic grading scheme. Colloid quantity shall also be considered.

2. **Colloid quality**: Colloid quality is generally considered in association with colloid content decreases. However, changes in colloid quality can exist independent of changes in follicular lumen area. Typical descriptions include homogeneous, heterogeneous, lacy or granular. If present, these findings are to be reported in a narrative format.

3. **Follicular cell height/shape**: Follicular cell height/shape can range from squamous, to cuboidal, to low columnar to high columnar. Increased epithelial cell height (progression from cuboidal to columnar) can lead to glandular hypertrophy. A narrative description of the predominant cell shape (squamous, cuboidal, low columnar, tall columnar) can be provided if necessary to reflect a chemical-dependent change.

IIB. Severity Grading

Severity grading is a method by which a range of variation is assigned to ordinal classes, generally being listed as minimal, mild, moderate and severe [2] and is semi-quantitative. The purpose of severity grading is to provide an efficient, semi-objective mechanism for comparing changes (compound-related effects) among animals, treatment groups, and studies.

The current approach for grading severity incorporates a four score range, including: 0 – non-remarkable, 1-mild, 2-moderate, and 3-severe. This severity grading method has been evaluated and determined to be the best method for discriminating subtle pathologic changes in the thyroid gland.

For the frog metamorphosis studies, quantitative scoring may not be suitable for all criteria. The core criteria (i.e., atrophy/hypertrophy, follicular cell height/shape, and follicular hyperplasia) will most effectively be measured using the severity scoring system. In addition to the severity grade, pathologists can document qualitative changes associated with the lesions. The additional criteria will be documented using descriptive terms only, except for follicular lumen area, which will be documented using the generalized grading scheme. Follicular lumen area need only be documented if the finding is exposure-related.

For thyroid gland hypertrophy and glandular atrophy, the pathologist is to score compound-exposed animals relative to the control animals using the atlas provided and the descriptions below. For follicular cell hypertrophy and hyperplasia, the pathologist is to base severity grades on the percentage of cells affected, and/or the percentage of the tissue which is affected.

Severity grading shall employ the following system:

Grade 0 (not remarkable)
Grade 1 (mild)
Grade 2 (moderate)
Grade 3 (severe)

General severity grading scale:

- **Grade 0:** Non-remarkable to minimal. Ranging from inconspicuous to barely noticeable but so minor, small, or infrequent as to warrant no more than the least assignable grade. For multifocal or diffusely-distributed alterations, this grade is used for processes where less than 20% of the tissue in the section is involved.

- **Grade 1: Mild.** A noticeable feature of the tissue. For multifocal or diffusely-distributed alterations, this grade is used for processes where 30-50% of the tissue in the section is involved.

- **Grade 2: Moderate.** A dominant feature of the tissue. For multifocal or diffusely-distributed alterations, this grade is used for processes where 60-80% of the tissue in the section is involved.

- **Grade 3: Severe.** An overwhelming feature of the tissue. For multifocal or diffusely-distributed alterations, this grade is used for processes where greater than 80% of the tissue in the section is involved.

Because the grading system to be used for the amphibian metamorphosis assay is to promote consistency in the histological analysis, proposed descriptions for scoring the various criteria are presented below. These descriptions are to support the photomicrographic examples of each criterion with severity grades, which follow. In addition examples of normal histomorphology of the amphibian thyroid gland during metamorphosis, and examples of common artifacts are presented.

Core Criteria:

Table 1. Severity grading scheme for thyroid gland hypertrophy.

Grade	Descriptor	Criteria
0	Non-remarkable	Less than 20% enlargement of glands in comparison to controls.
1	Mild	Diffuse enlargement of glands that exceeds control glands size by 30-50%.
3	Moderate	Diffuse enlargement of glands that exceeds control glands size by 60-80%.
4	Severe	Diffuse enlargement of glands that exceeds control glands size by over 80%. There is contact of both glands at the midline and they exceed normal boundaries into surrounding tissue space.

Table 2. Severity grading scheme for thyroid gland atrophy.

Grade	Descriptor	Criteria
0	Non-remarkable	Less than a 20% reduction in size in comparison to controls.
1	Mild	Gland size is 30-50% reduced from the size of control glands.
2	Moderate	Gland size is 60-80% reduced from the size of control glands.
3	Severe	Gland size is over 80% reduced from the size of control glands.

Table 3. Severity grading scheme for follicular cell hypertrophy.

Grade	Descriptor	Criteria
0	Non-remarkable	Fewer than 20% of the cells exhibit hypertrophy.
1	Mild	30-50% of follicular cells exhibit hypertrophy.
2	Moderate	60-80% of follicular cells exhibit hypertrophy.
3	Severe	Over 80% of follicular cells exhibit hypertrophy.

Table 4. Severity grading scheme for follicular cell hyperplasia.

Grade	Descriptor	Criteria
0	Non-remarkable	Focal or diffuse crowding of follicular cells affecting less than 20% of the tissue.
1	Mild	Focal or diffuse crowding of follicular cells affecting 30-50% of the tissue, and/or single or multiple papillary infoldings of follicular cell layer.
2	Moderate	60-80% of the follicles exhibit focal hyperplasia characterized by pseudostratified or stratified follicular epithelium – papillary infolding may be present.
3	Severe	Over 80% of follicles exhibit extensive hyperplasia with stratification 2-3 cell layers thick – papillary infolding may be present.

Additional Criteria:

Follicular lumenal area increase or decrease: Use the generic grading scheme presented above to document exposure related changes.

Colloid Quality: Document changes in colloid quality using narrative descriptions. Examples include homogeneous, heterogeneous, lacy or granular. Pathologists may also comment on the tinctorial quality of the colloid.

III. Reference Atlases

Below are photographic examples of normal thyroid histomorphology of *X. laevis* during metamorphosis, examples of the core criteria and assigned severity grades for each lesion, examples of colloid quality changes, some miscellaneous changes that might be encountered while performing histological analyses on the test subjects, and common artifacts that might contribute to interpretive difficulty. Several caveats to these images are as follows:

1. The examples are not all-inclusive, nor complete, and may not be the most representative example of each lesion.
2. Sections are either transverse or frontal, however it is expected that the thyroid changes presented can be accurately applied to multiple sectioning planes.
3. Examples have been obtained from tadpoles in different developmental stages.
4. Some photographs contain several different lesions that are to be scored independently.

IIIA. General normal appearance of *X. laevis* thyroid microanatomy

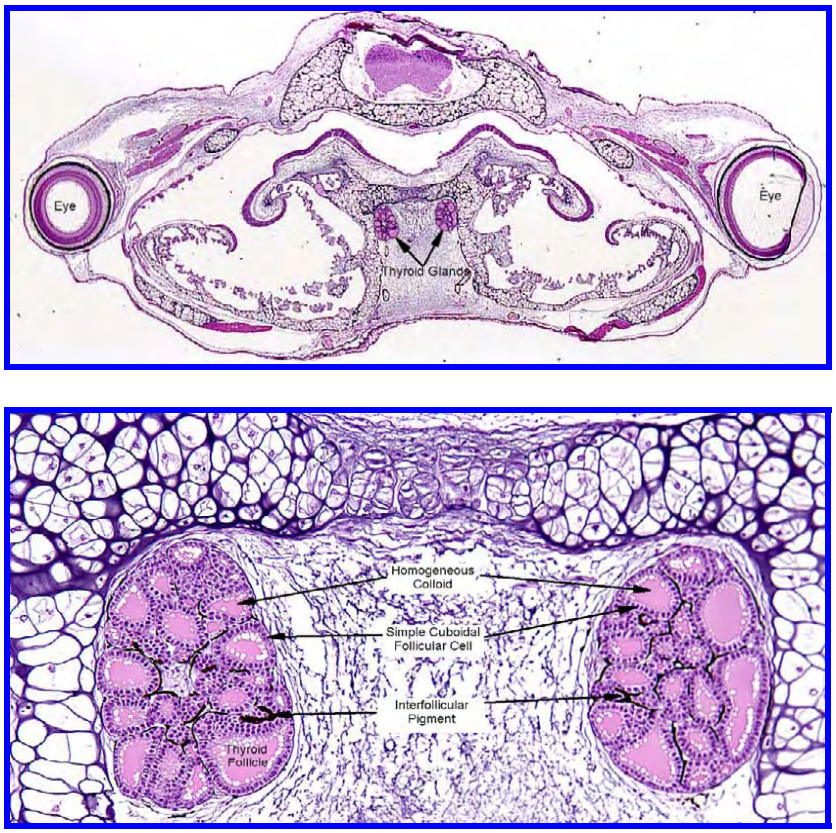


Figure 1. Low (1.25x) and high (10x) magnification photomicrographs of transversally sectioned thyroid glands. The colloid is homogeneous and light eosinophilic, and the follicles are lined by simple cuboidal epithelium.

IIIB. Developmental series of normal *X. laevis* thyroid microanatomy – H&E

Figure 2. 48 hours post-stage 54

Figure 3. 96 hours post-stage 54

Figure 4. 144 hours post-stage 54.

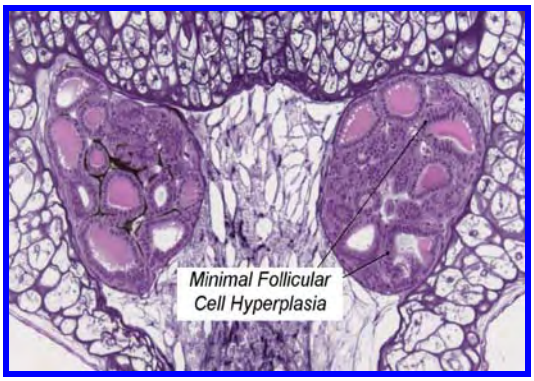


Figure 5. 192 hours post-stage 54

In an effort to establish histologic references for *X. laevis* thyroid glands, a time course experiment was executed. Beginning at stage 54, tadpoles were sequentially sacrificed at 48-, 96-, 144-, and 192-hours post stage 54, as represented above. These images were obtained to demonstrate the normal changes of the amphibian thyroid gland during metamorphosis.

IIIC. Reference atlas of diagnostic criteria and severity grades

IIIC1. Thyroid gland hypertrophy – general range of changes

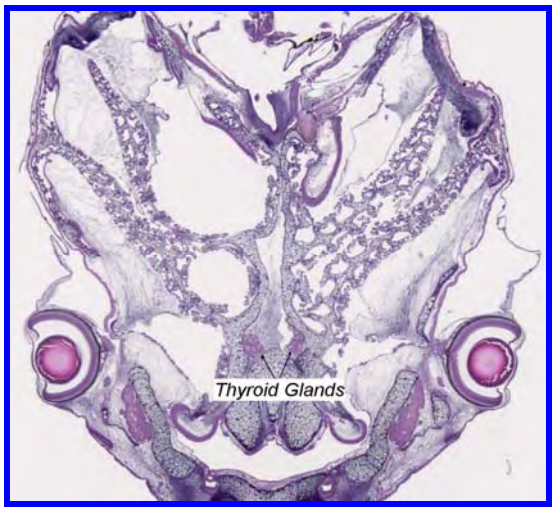


Figure 6. Thyroid hypertrophy - Grade 0

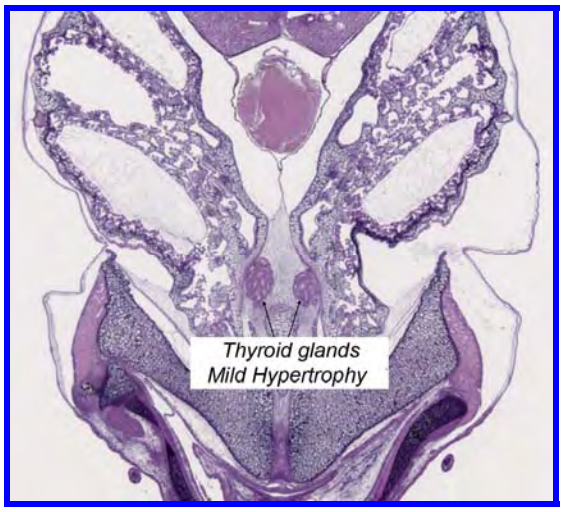


Figure 7. Thyroid hypertrophy – Grade 1

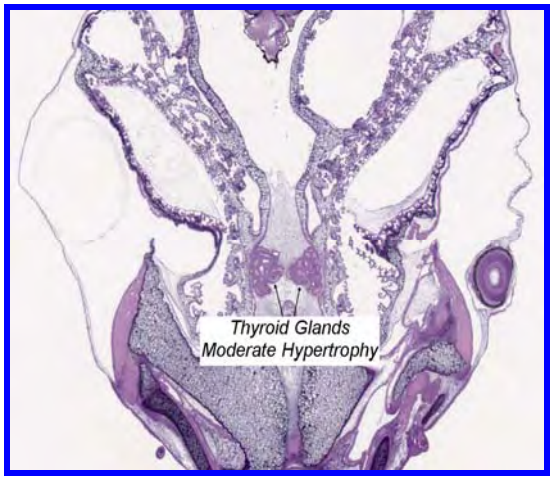


Figure 8. Thyroid hypertrophy – Grade 2

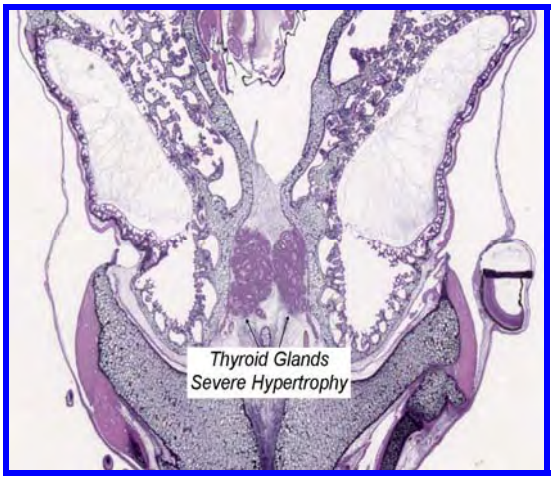
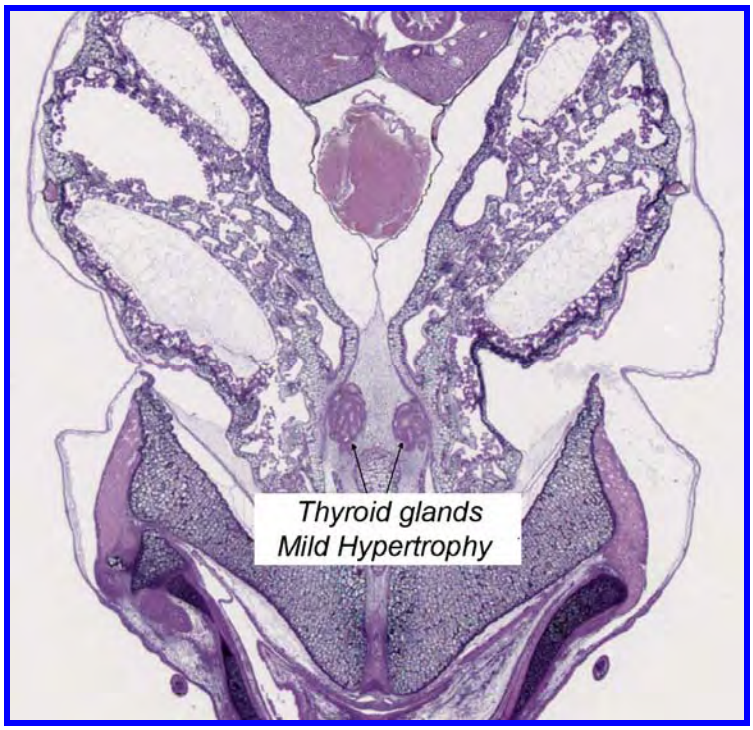


Figure 9. Thyroid hypertrophy – Grade 3

Grade 0 – Thyroid gland hypertrophy:

Figure 10. Low (1.25x) and medium (10x) magnification photomicrographs of thyroid glands from a *X. laevis* that was a control animal. The thyroid follicles are colloid-filled and lined by simple cuboidal follicular cells. There is minimal follicular cell hyperplasia.

Grade 1 (Mild) – Thyroid gland hypertrophy:



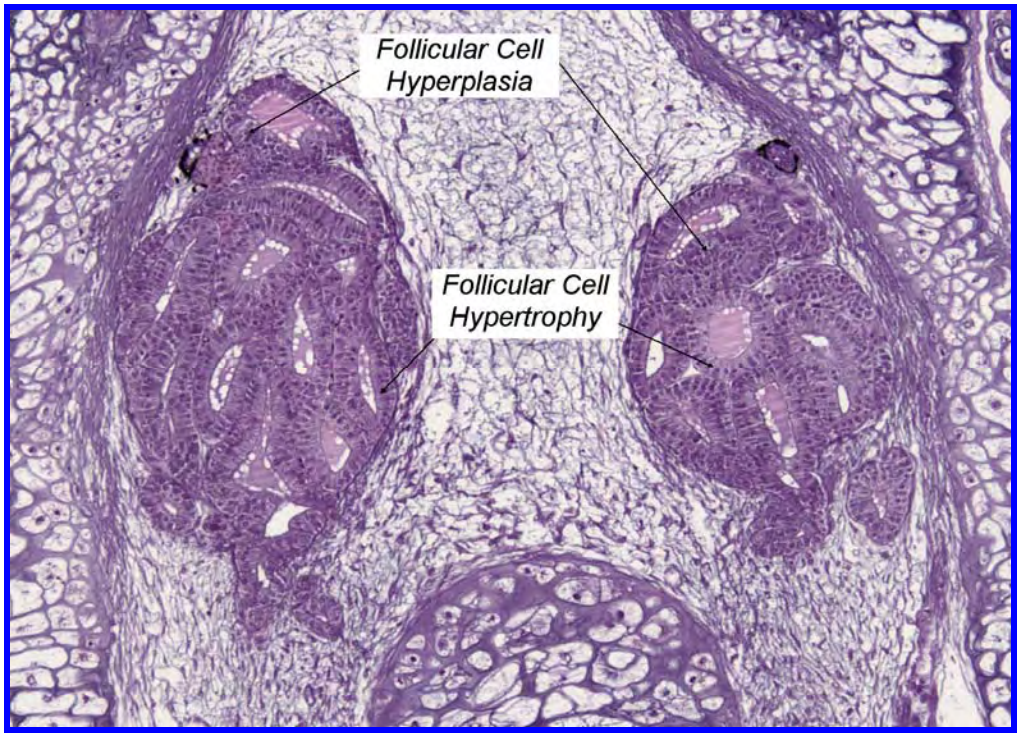


Figure 11. Low (1.25x) and medium (10x) magnification photomicrographs of a *X. laevis* exposed to 62.5 µg/L perchlorate for 21 days. There is moderate hypertrophy of the follicular cells and slight/mild follicular cell hyperplasia. There is mild glandular hypertrophy and there is slight/mild decreased colloid in the thyroid follicles.

Grade 2 (Moderate) – Thyroid gland hypertrophy:

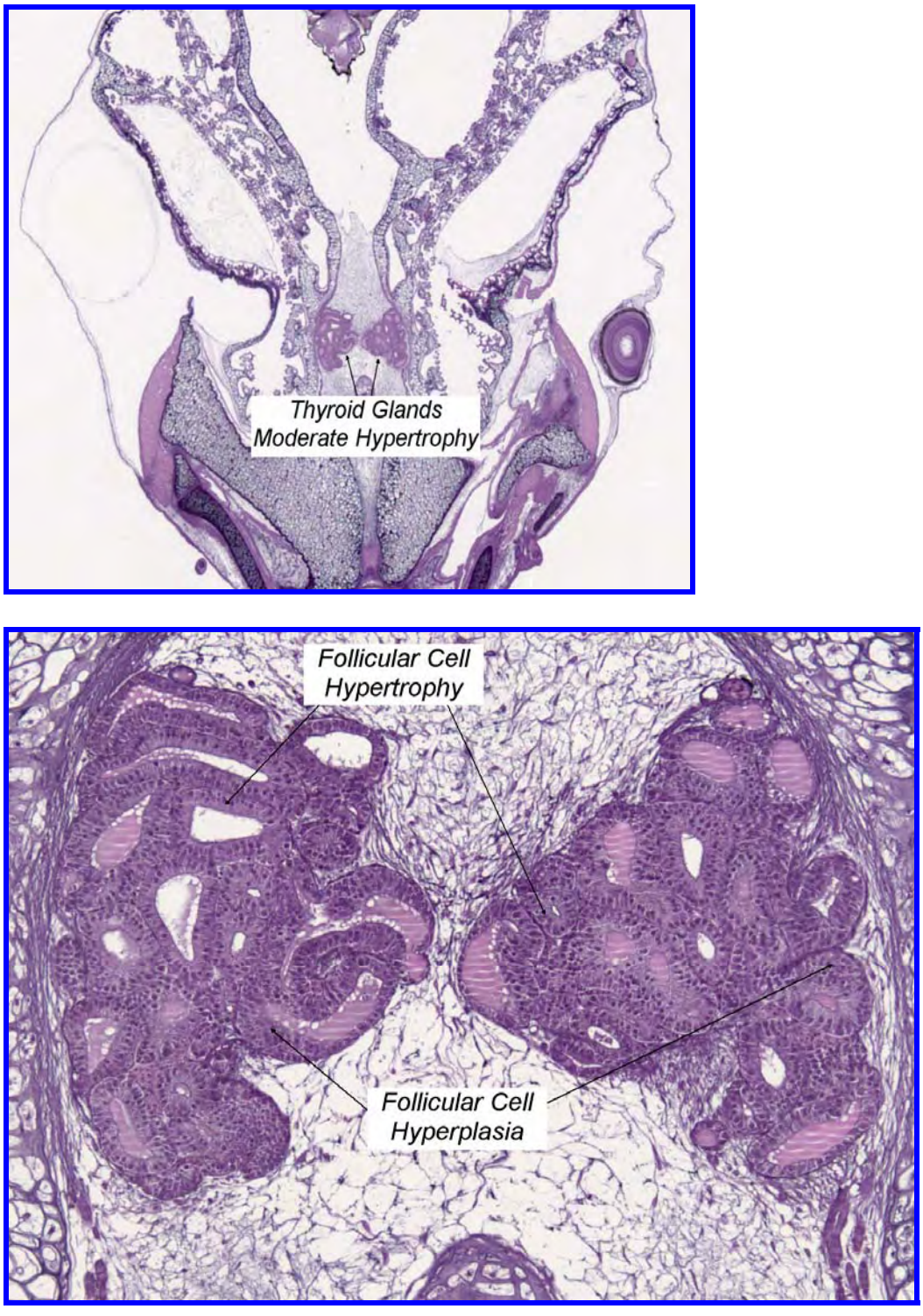


Figure 12. Low (1.25x) and medium (10x) magnification photomicrographs of a *X. laevis* exposed to 62.5 µg/L perchlorate for 21 days. Moderate follicular cell hypertrophy and moderate follicular cell hyperplasia are present. The thyroid glands are moderately enlarged overall and there is slight/mild decreased colloid in the thyroid follicles.

Grade 3 (Severe) – Thyroid gland hypertrophy:

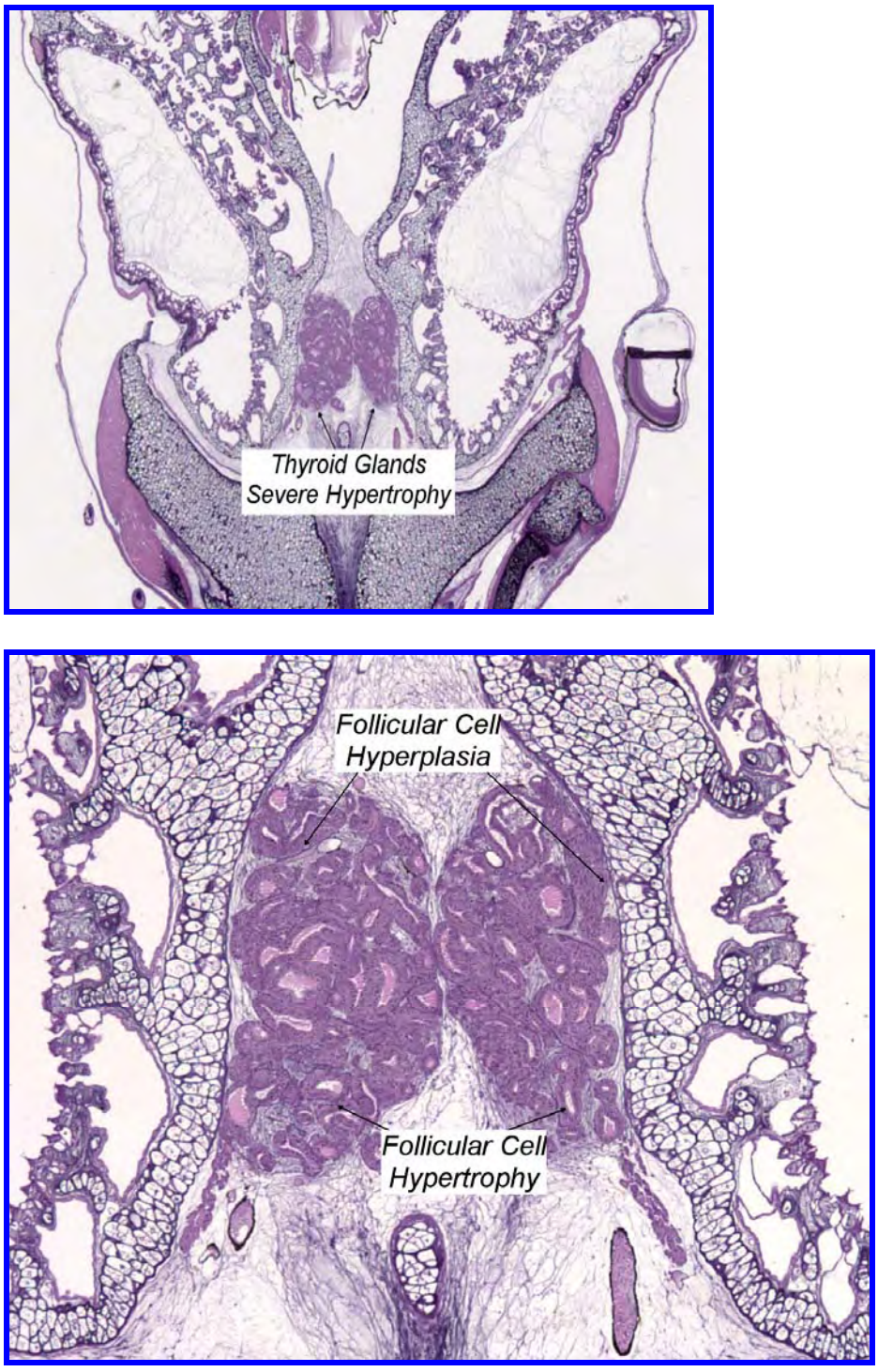


Figure 13. Low (1.25x) and medium (4x) magnification photomicrographs of a *X. laevis* exposed to 250 µg/L perchlorate for 21 days. There is severe hypertrophy of the thyroid glands with contact at the midline. There is moderate follicular cell hypertrophy and there is moderately severe follicular cell hyperplasia. There is slight/mild decreased colloid in the thyroid follicles.

IIIC2. Thyroid gland atrophy - general range of changes

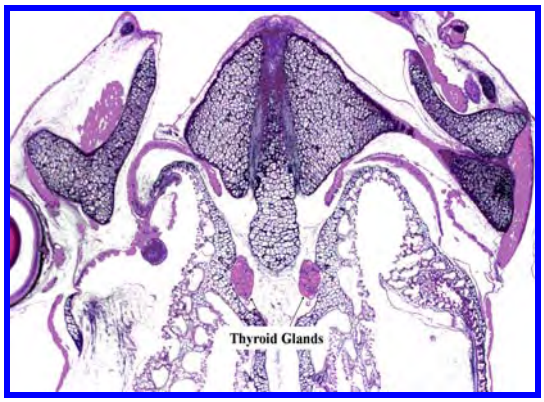


Figure 14. Thyroid atrophy – Grade 0

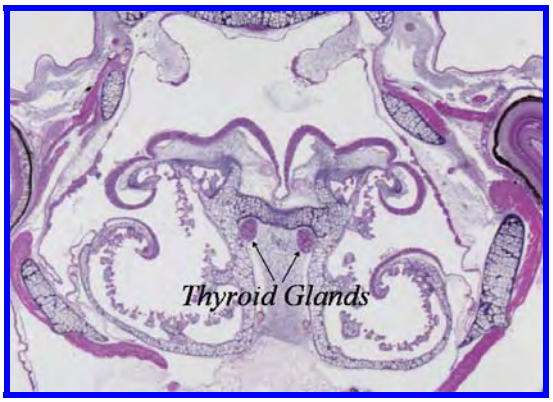


Figure 15. Thyroid atrophy – Grade 1

Figure 16. Thyroid atrophy - Grade 2

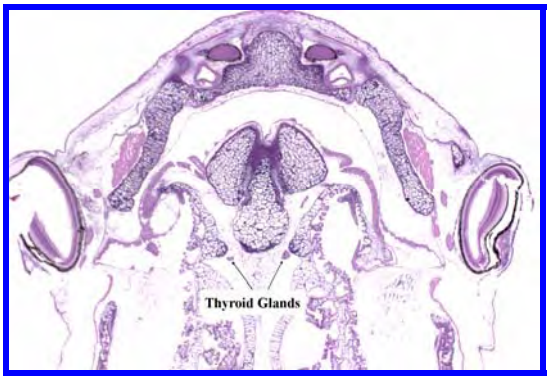


Figure 17. Thyroid atrophy - Grade 3

Grade 0 – Thyroid gland atrophy

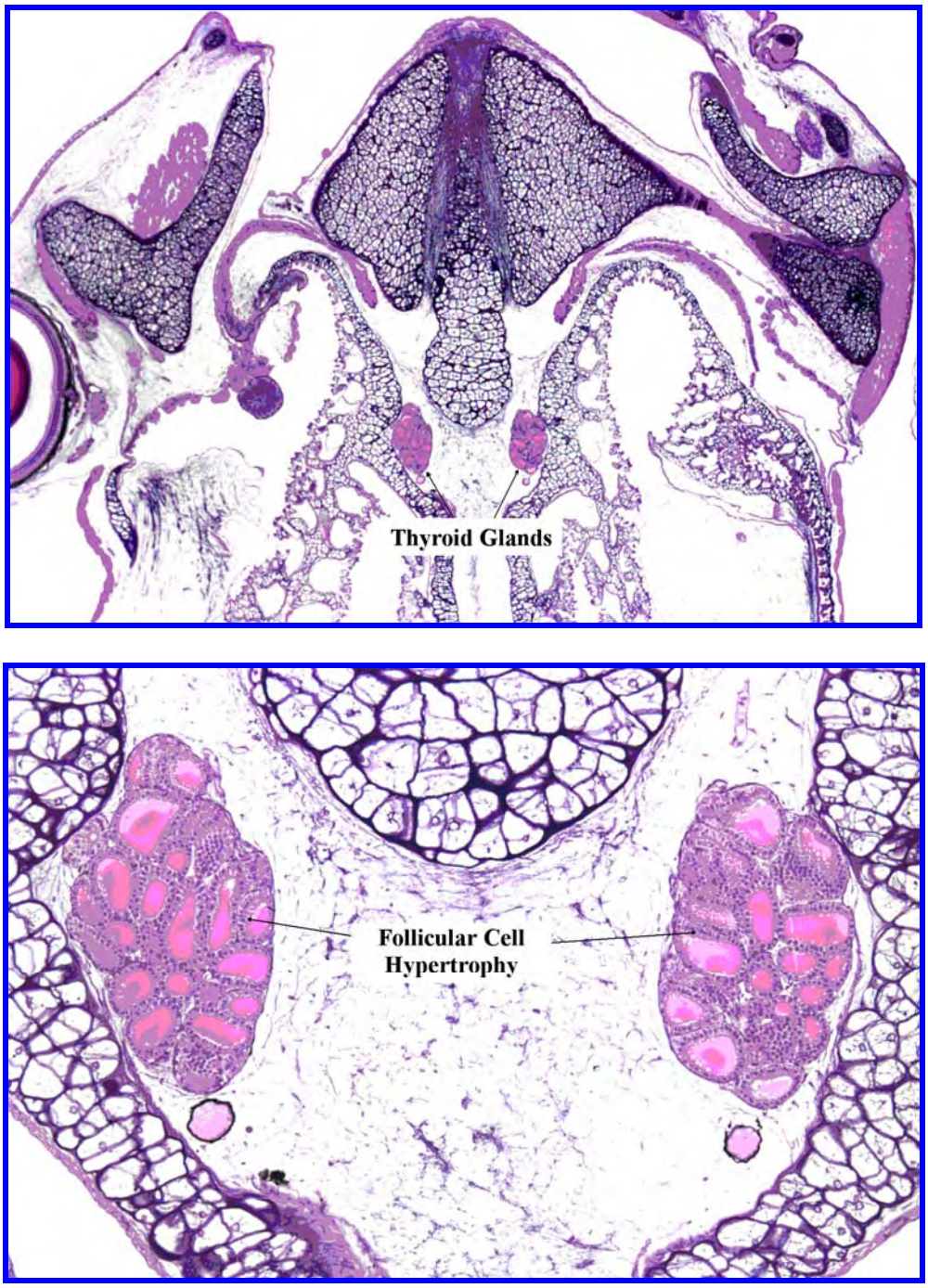


Figure 18. Low (1.25x) and medium (10x) magnification photomicrographs of thyroid glands from *X. laevis* that was a control animal. The thyroid follicles contain eosinophilic homogeneous colloid and are lined by simple cuboidal follicular cells. There is minimal follicular cell hyperplasia and hypertrophy which may be present in normal thyroid glands. The thyroid follicles are variable in size.

Grade 1 – Thyroid gland atrophy:

Figure 19. Low (1.25x) and medium (10x) magnification photomicrographs of thyroid glands from *X. laevis* exposed to 1 µg/L thyroxine. There is minimal decreased quantity of colloid and a minimal decrease in the size of thyroid follicles as compared to control frogs. There is mild atrophy of the thyroid glands.

Grade 2 – Thyroid gland atrophy:

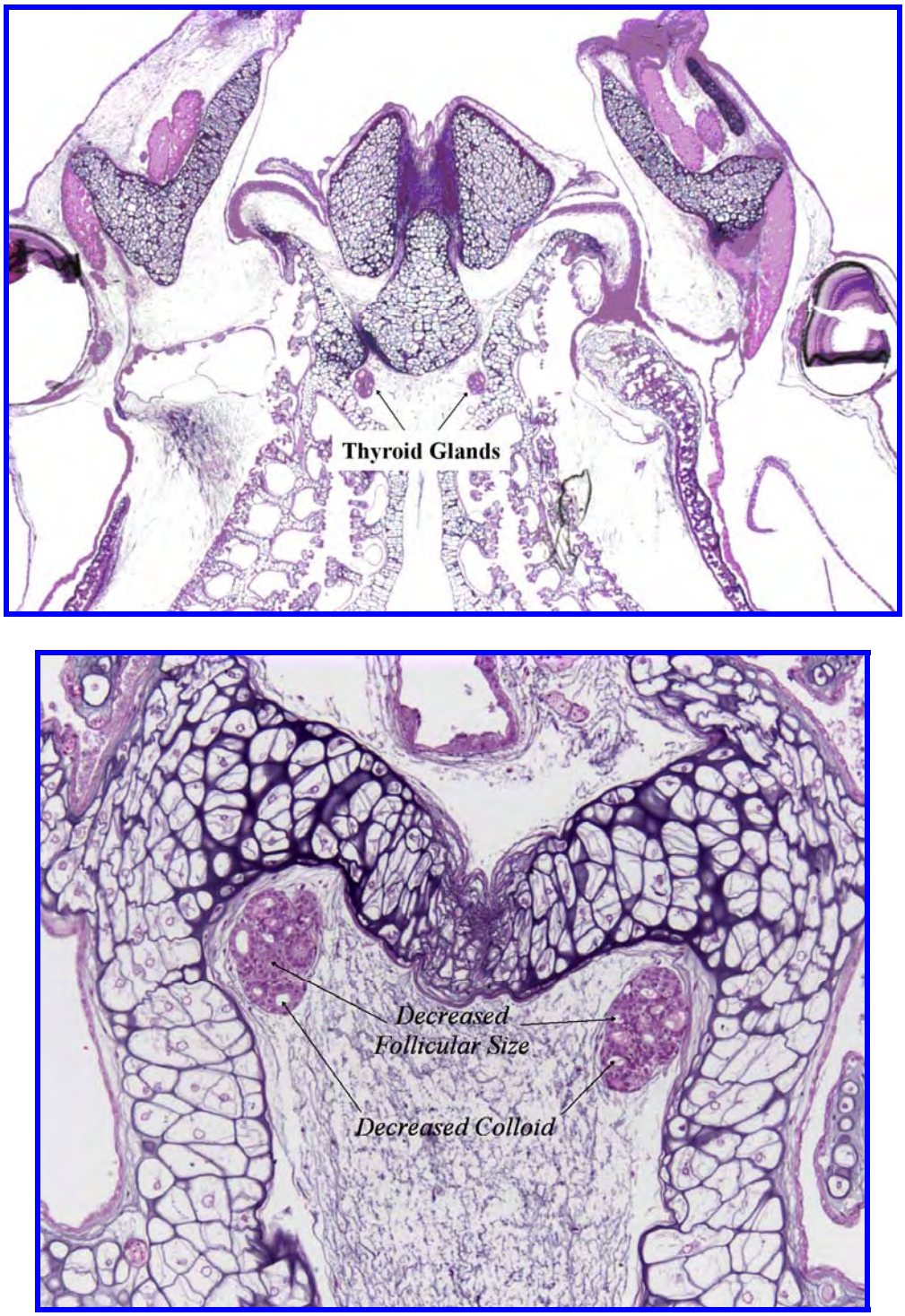


Figure 20. Low (1.25x) and medium (10x) magnification photomicrographs of thyroid glands from thyroid glands of *X. laevis*. There is a moderate decrease in the size of the thyroid follicles and moderately decreased colloid. The glands are moderately atrophied.

Grade 3 – Thyroid gland atrophy:

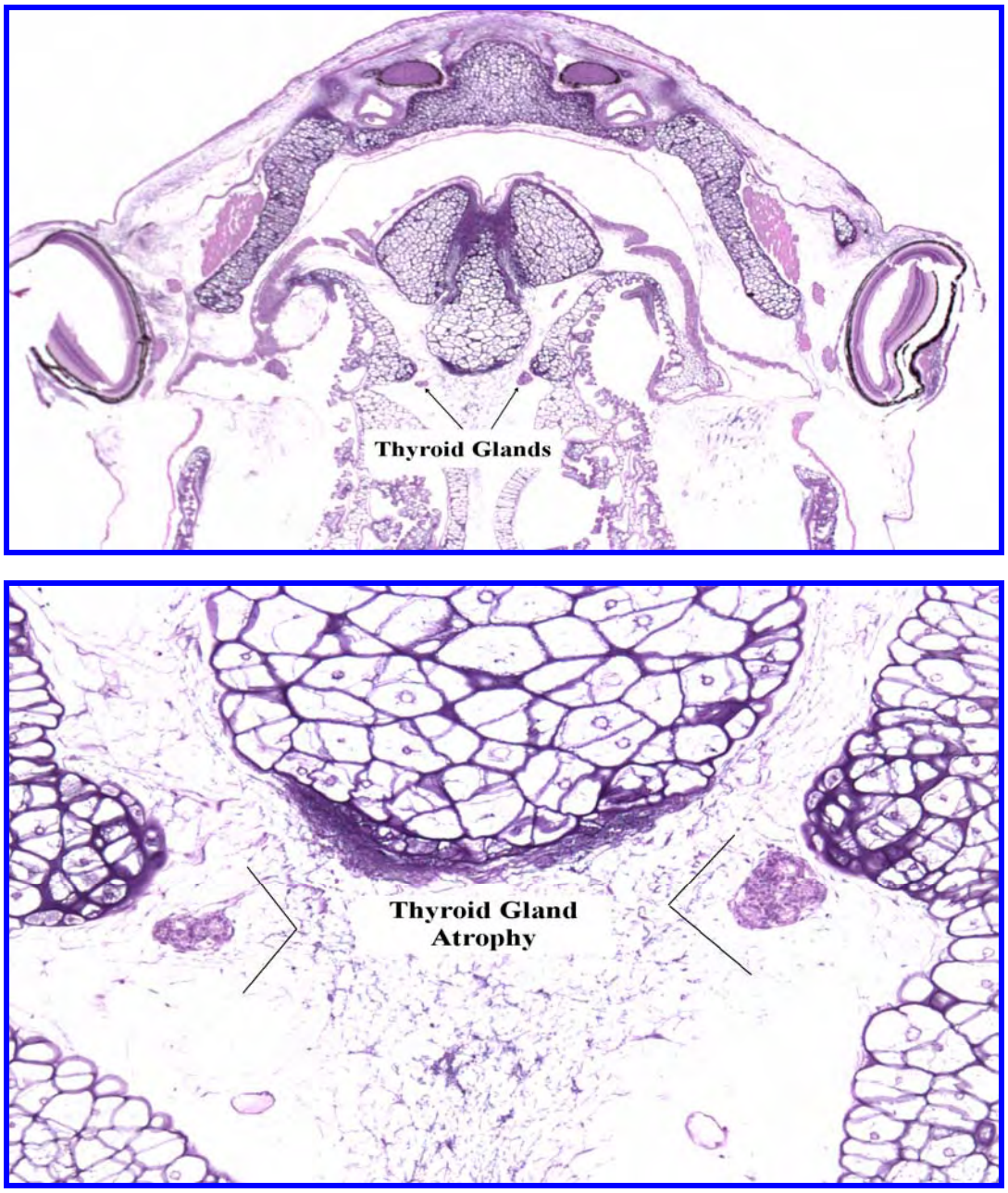


Figure 21. Low (2x) and medium (10x) magnification photomicrographs of thyroid glands from *X. laevis*. There is severe thyroid gland atrophy and the thyroid follicles are uniformly small. There is a moderately severe decease in amount of colloid, but what little colloid is present is eosinophilic and homogeneous.

IIIC3. Follicular cell hypertrophy – general range of changes

Figure 22. Follicular cell hypertrophy - Grade 0

Figure 23. Follicular cell hypertrophy – Grade 1

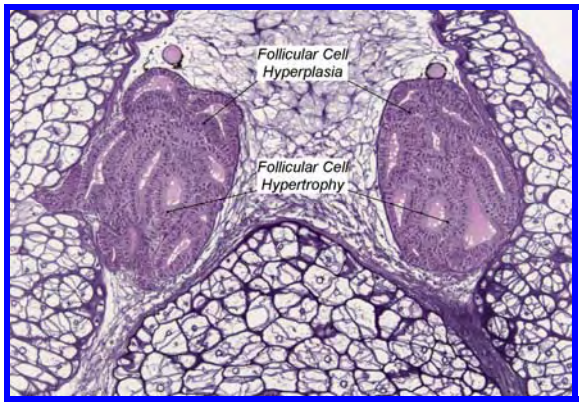


Figure 24. Follicular cell hypertrophy – Grade 2

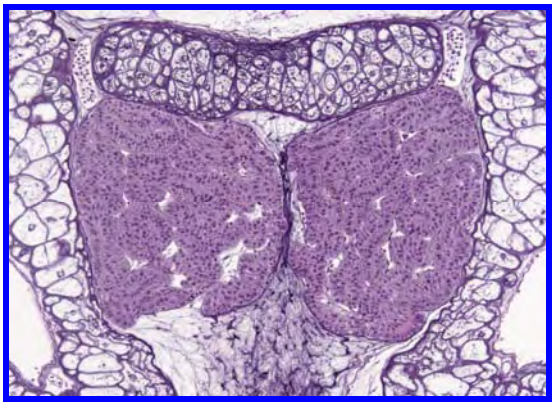


Figure 25. Follicular cell hypertrophy – Grade 3

Grade 0 - Follicular cell hypertrophy:

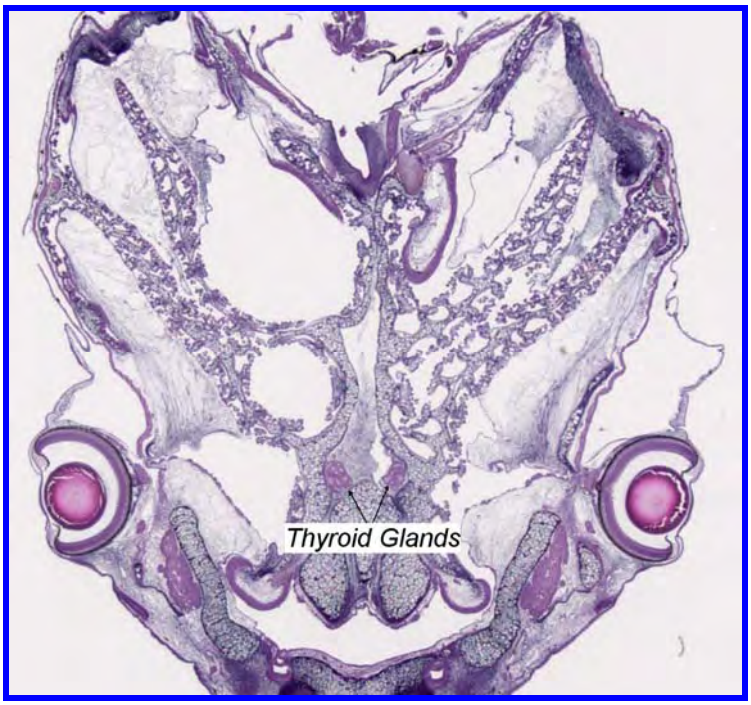


Figure 26. Low (1.25x) and medium (10x) magnification photomicrographs of thyroid glands from *X. laevis* that was a control animal. The thyroid follicles are colloid-filled and lined by simple cuboidal follicular cells. There is minimal follicular cell hyperplasia.

Grade 1 - Follicular cell hypertrophy:

Figure 27. Thyroid glands from a Stage 54 *X. laevis* exposed to methimazole for 96 hours. There is mild follicular cell hypertrophy characterized by cells that are plump and columnar as compared to the usual cuboidal cells. There is also mild follicular cell hyperplasia. A lower magnification image is not available.

Grade 2 - Follicular cell hypertrophy:

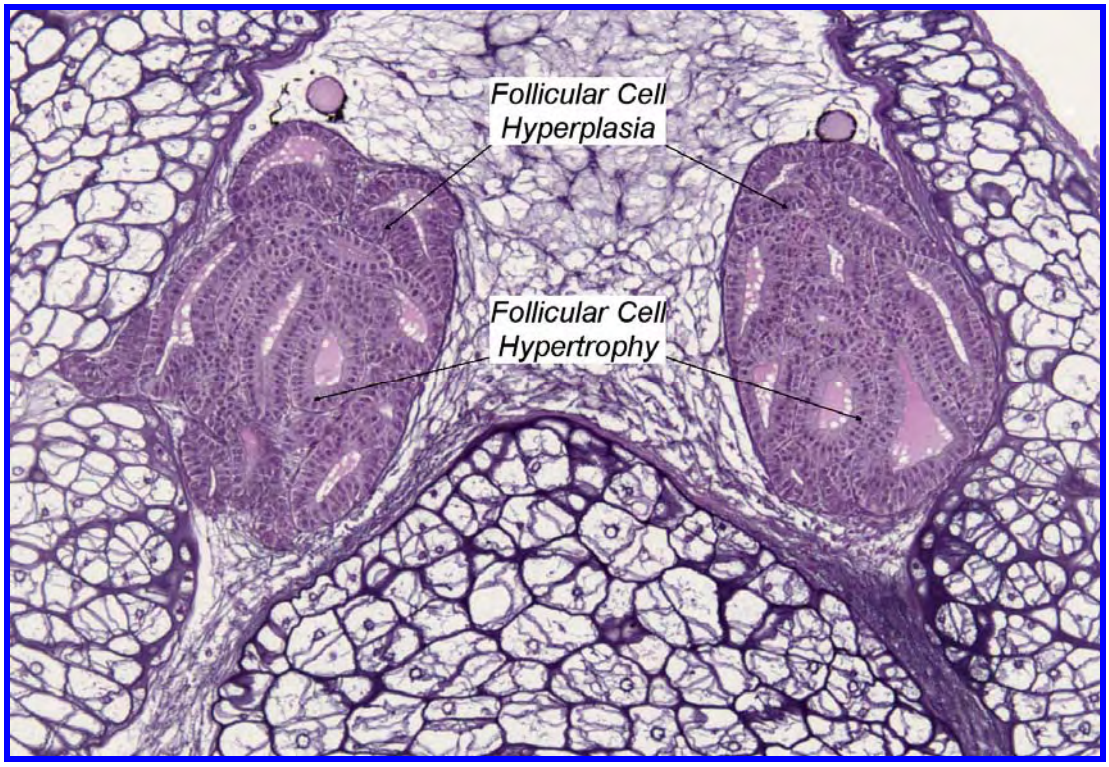


Figure 28. Low (1.25x) and medium (10x) magnification photomicrographs of a Stage 51 *X. laevis* exposed to 125 µg/L perchlorate for 21 days. There is slight/mild follicular cell hyperplasia and there is moderate follicular cell hypertrophy. There is slight/mild decreased colloid in the thyroid follicles.

Grade 3 - Follicular cell hypertrophy:

Figure 29. Thyroid glands from a Stage 54 X. laevis exposed to methimazole for 192 hours. There is moderately severe hypertrophy of the thyroid glands. The amount of colloid in the follicles is severely reduced and there is severe follicular cell hypertrophy and moderately severe hyperplasia. A lower magnification image is not available.

IIIC4. Follicular cell hyperplasia

Grade 0 – Follicular cell hyperplasia:

Figure 30. High (20x) magnification photomicrograph of thyroid gland from *X. laevis*. Minimal thyroid follicular cell hyperplasia is characterized by crowding of columnar cells. This follicular cell hyperplasia is representative of the upper limit of grade 0 because it is focal and affects less than 20% of the tissue.

Grade 1 – Follicular cell hyperplasia:

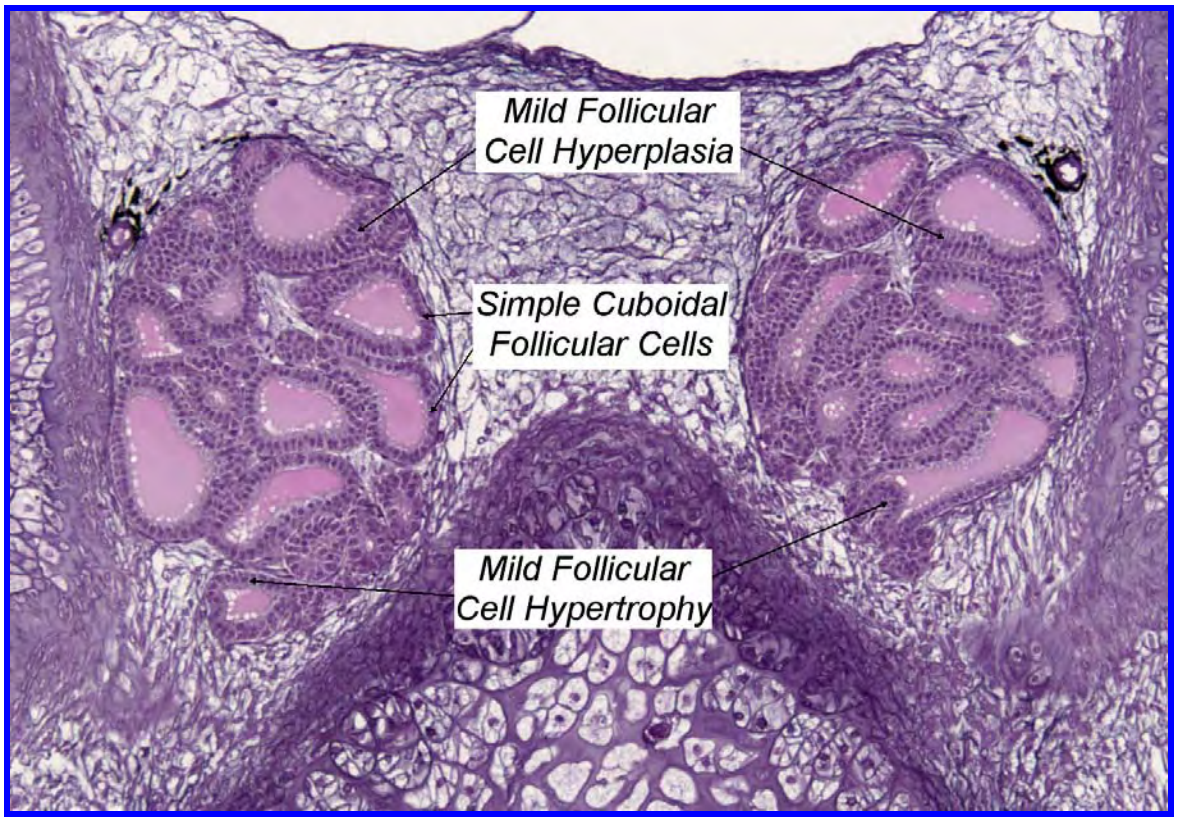


Figure 31. Medium (10x) magnification photomicrograph of thyroid glands from *X. laevis* that was a control animal. The thyroid follicles are colloid-filled and most are lined by simple cuboidal follicular cells. Slight/mild follicular cell hypertrophy and mild follicular cell hyperplasia are present demonstrated by papillary infolding and crowding of follicular cells.

Grade 2 – Follicular cell hyperplasia:

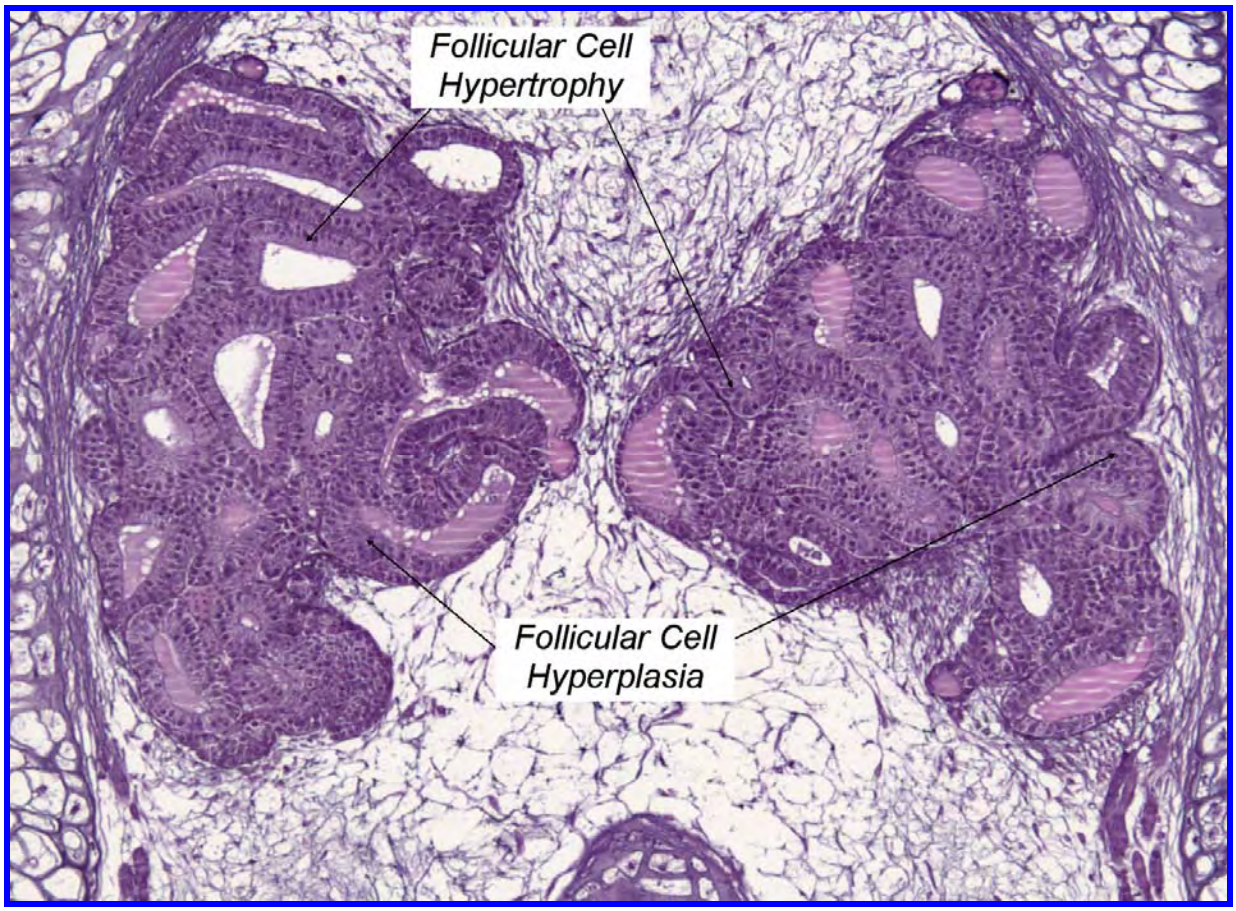


Figure 32. Medium (10x) magnification photomicrographs of a Stage 51 *X. laevis* exposed to 62.5 µg/L perchlorate for 21 days. Moderate follicular cell hypertrophy and moderate follicular cell hyperplasia are present. The thyroid glands are moderately enlarged overall and there is slight/mild decreased colloid in the thyroid follicles. The hyperplasia affects 30-50% of the tissue.

Grade 3 – Follicular cell hyperplasia:

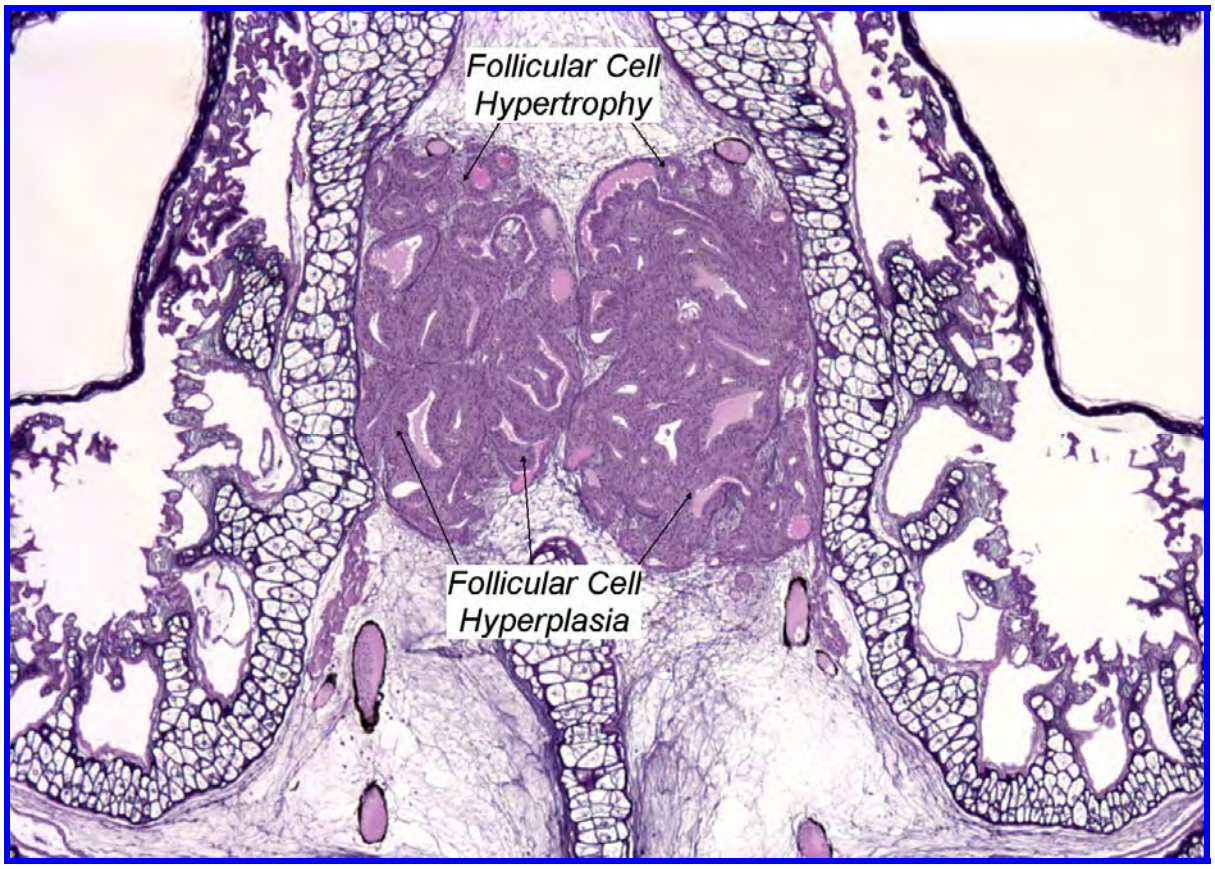


Figure 33. Medium (4x) magnification photomicrographs of a Stage 51 *X. laevis* exposed to 500 µg/L perchlorate for 21 days. There is severe hypertrophy of the thyroid glands and the colloid is slightly/mildly decreased in the thyroid follicles. There is severe follicular cell hyperplasia, affecting over 80% of the tissue, and there is moderate follicular cell hypertrophy.

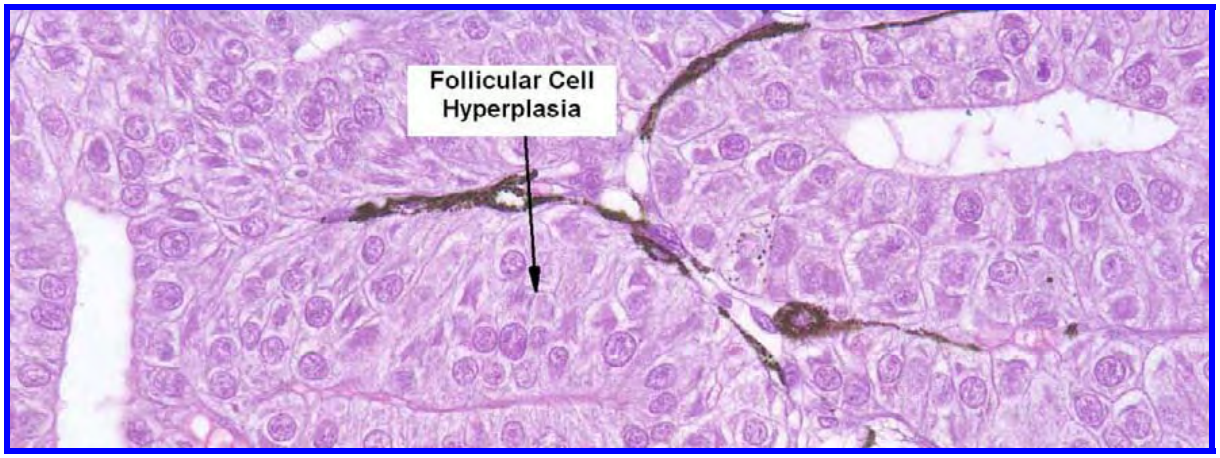


Figure 34. High (40x) magnification photomicrograph of thyroid gland from *X. laevis*. There is severe follicular cell hyperplasia demonstrated by the stratification of the follicular cells.

IIIC5. Colloid quality

Homogenous:

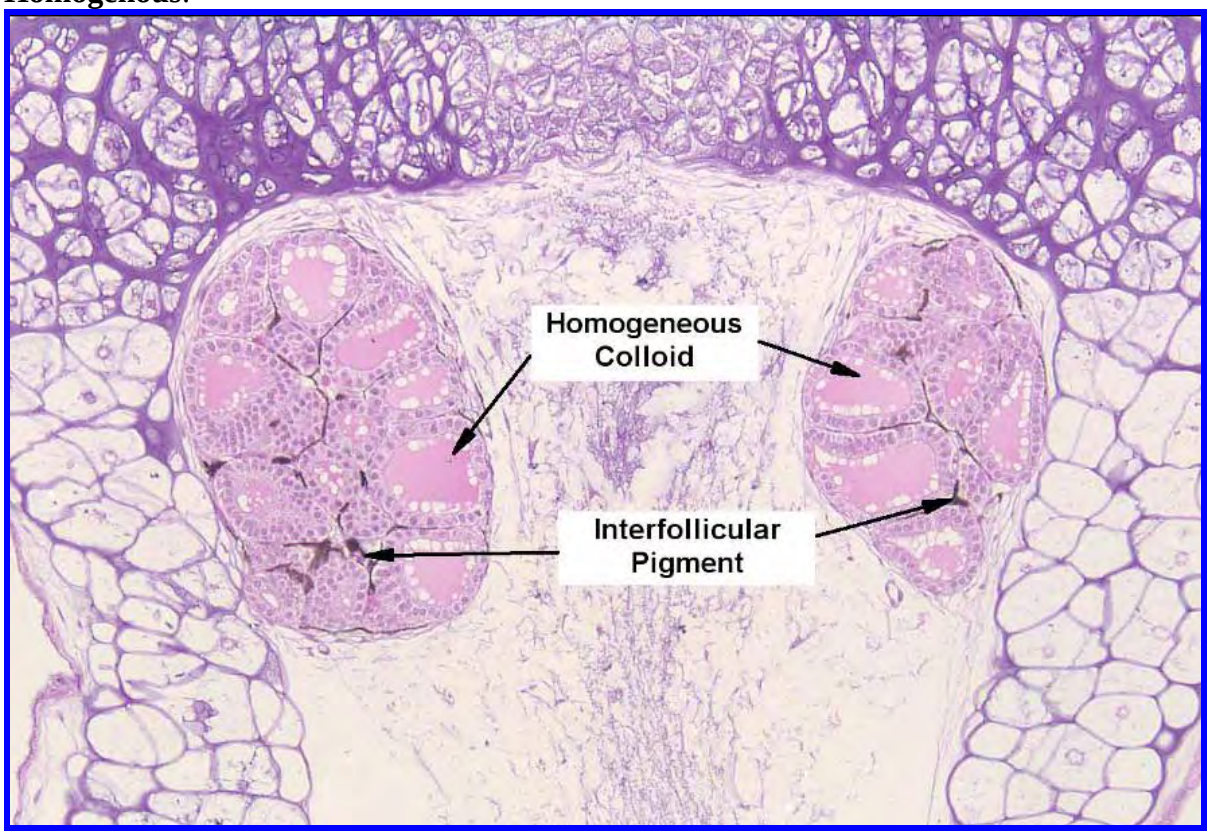


Figure 35. Medium (10x) magnification photomicrographs of thyroid glands from *X. laevis*. The colloid is homogeneous and light eosinophilic and the follicles are lined by simple cuboidal epithelium.

Foamy appearance:

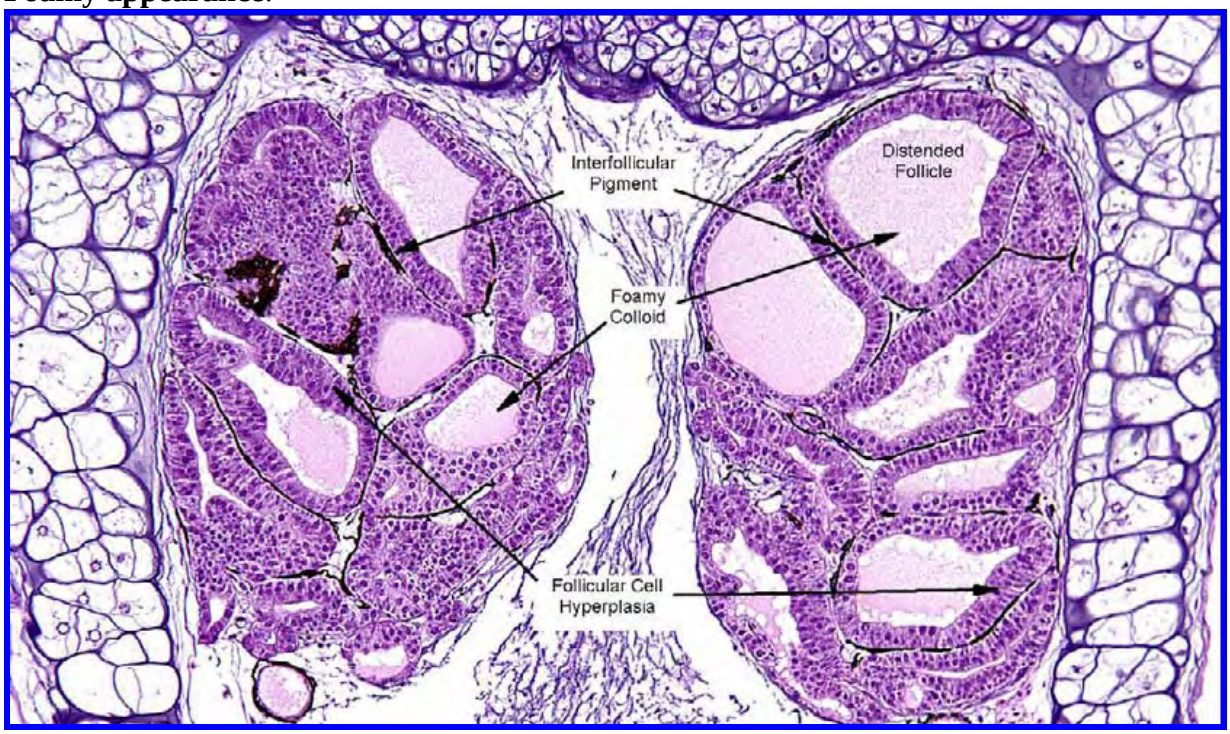


Figure 36. High (10x) magnification photomicrographs of thyroid glands from *X. laevis*. The colloid has a foamy appearance.

Granular colloid:

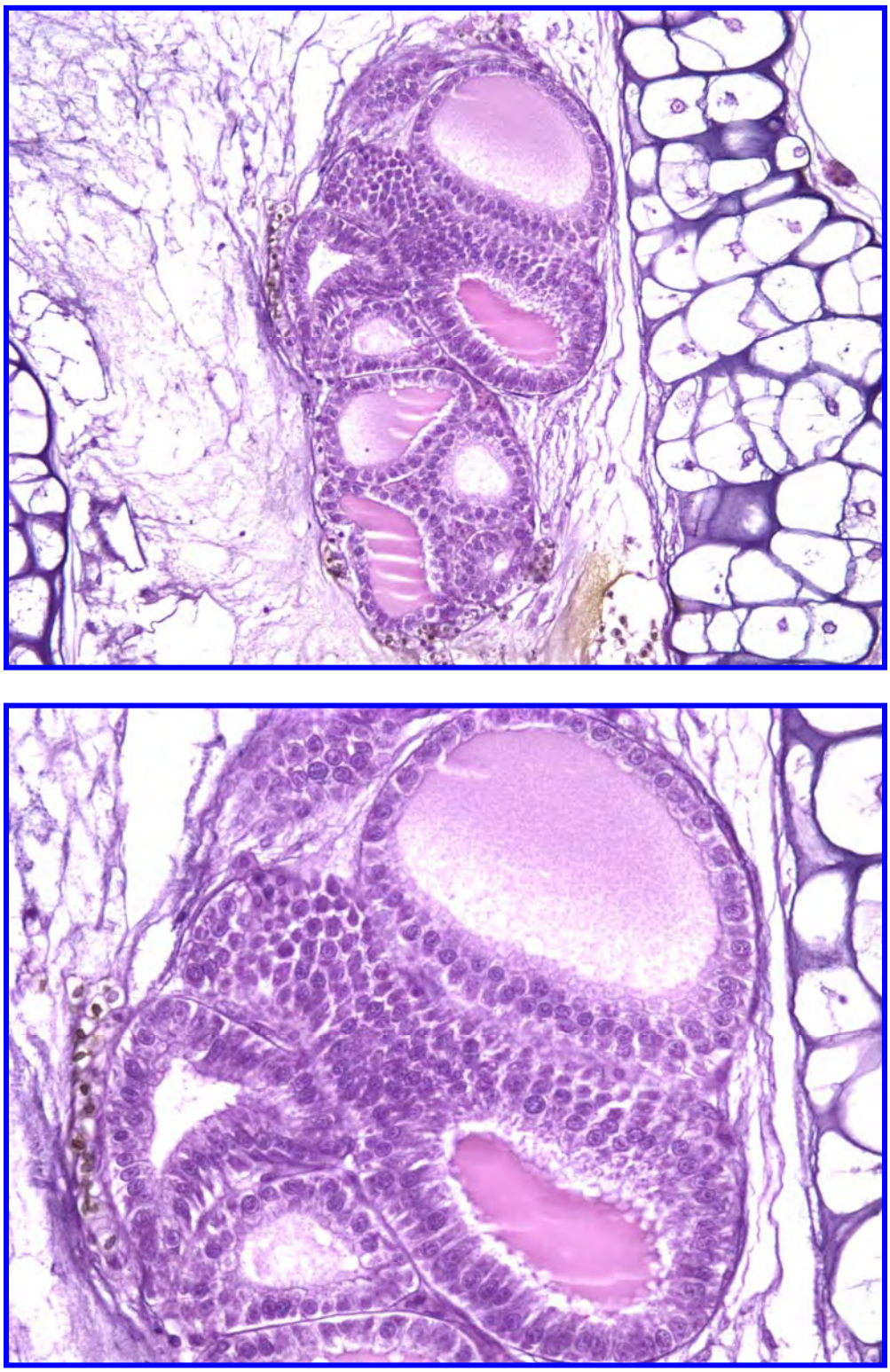


Figure 37. Medium (20x) and high (40x) magnification of an *X. laevis* thyroid gland. The colloid in the upper-right follicle is granular.

Tinctorial quality of colloid:

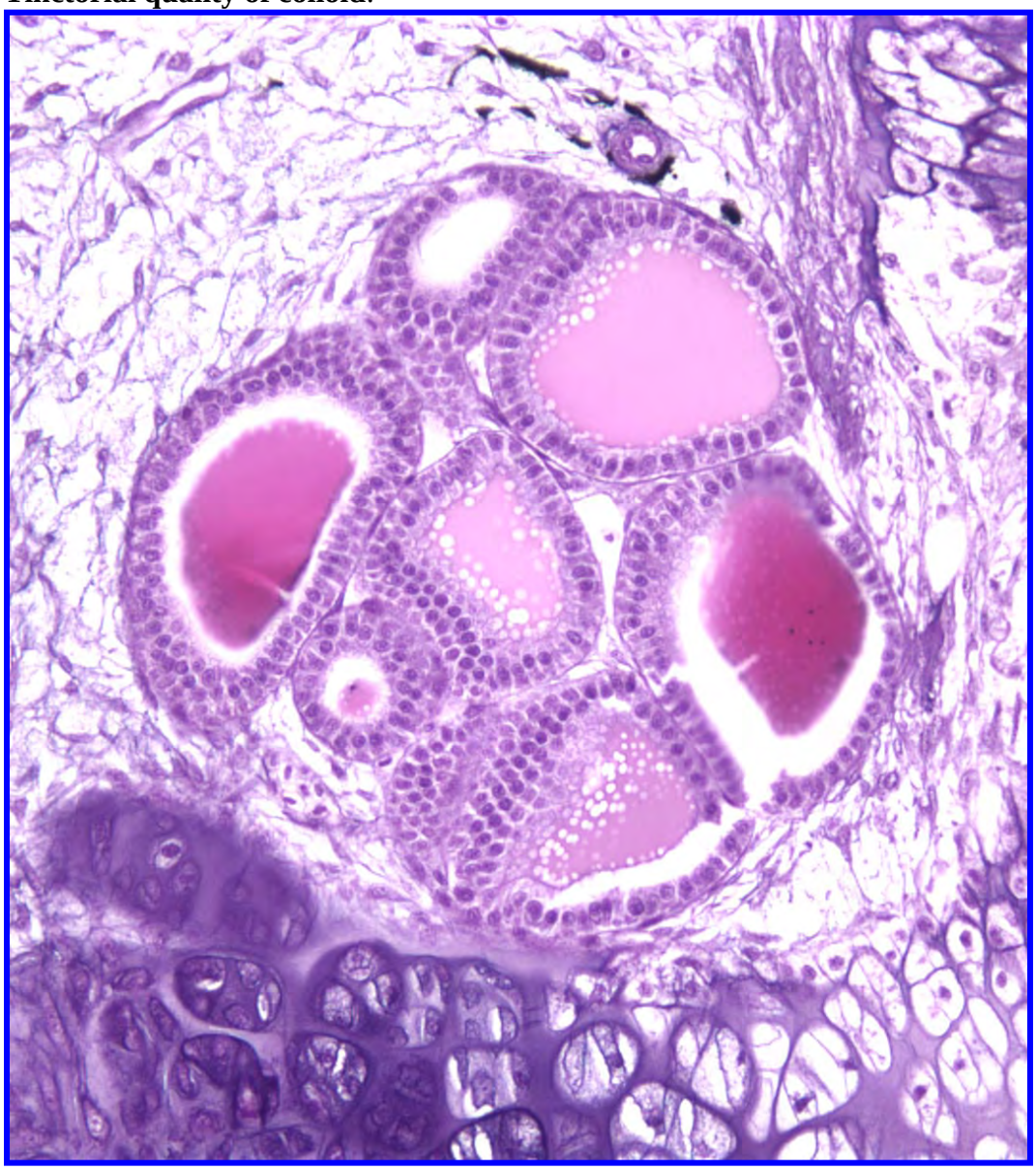


Figure 38. Thyroid gland of a stage 61 *X. laevis* tadpole. The tinctorial quality of the colloid is heterogenous in this example. Tinctorial quality may be affected by the thickness of the section, the batch of stain used, and variation of staining within the same gland.

IIIC6. Miscellaneous findings

Eosinophilic droplets:

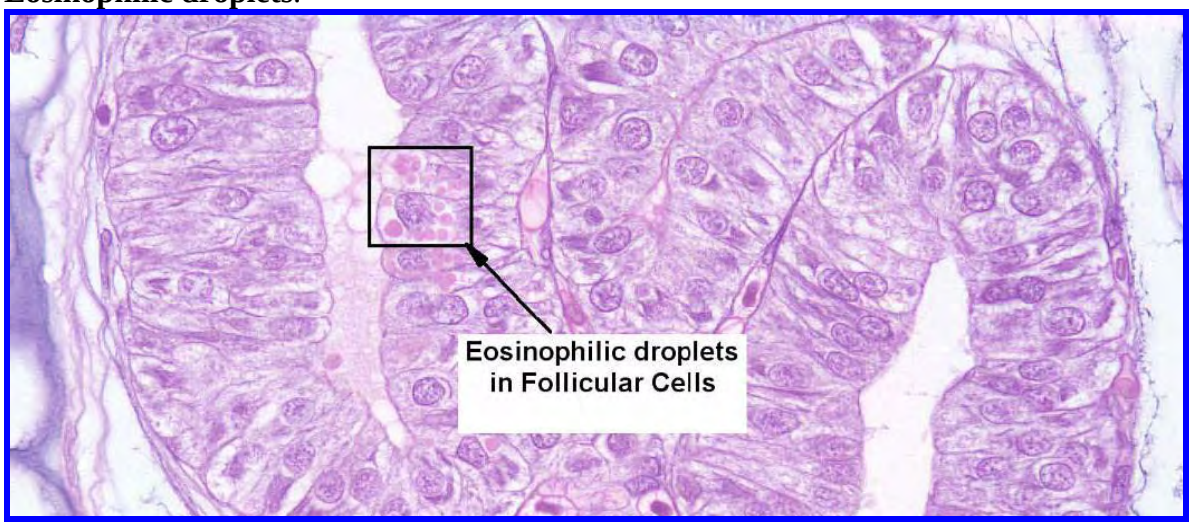


Figure 39. High (40x) magnification photomicrograph of thyroid gland from *X. laevis*. Note the eosinophilic droplets in follicular cells.

Karyomegaly:

Figure 40. High (20x) magnification photomicrographs of thyroid glands from *X. laevis*. Minimal karyomegaly is characterized by occasional enlarged nuclei.

Midline thyroid follicles:

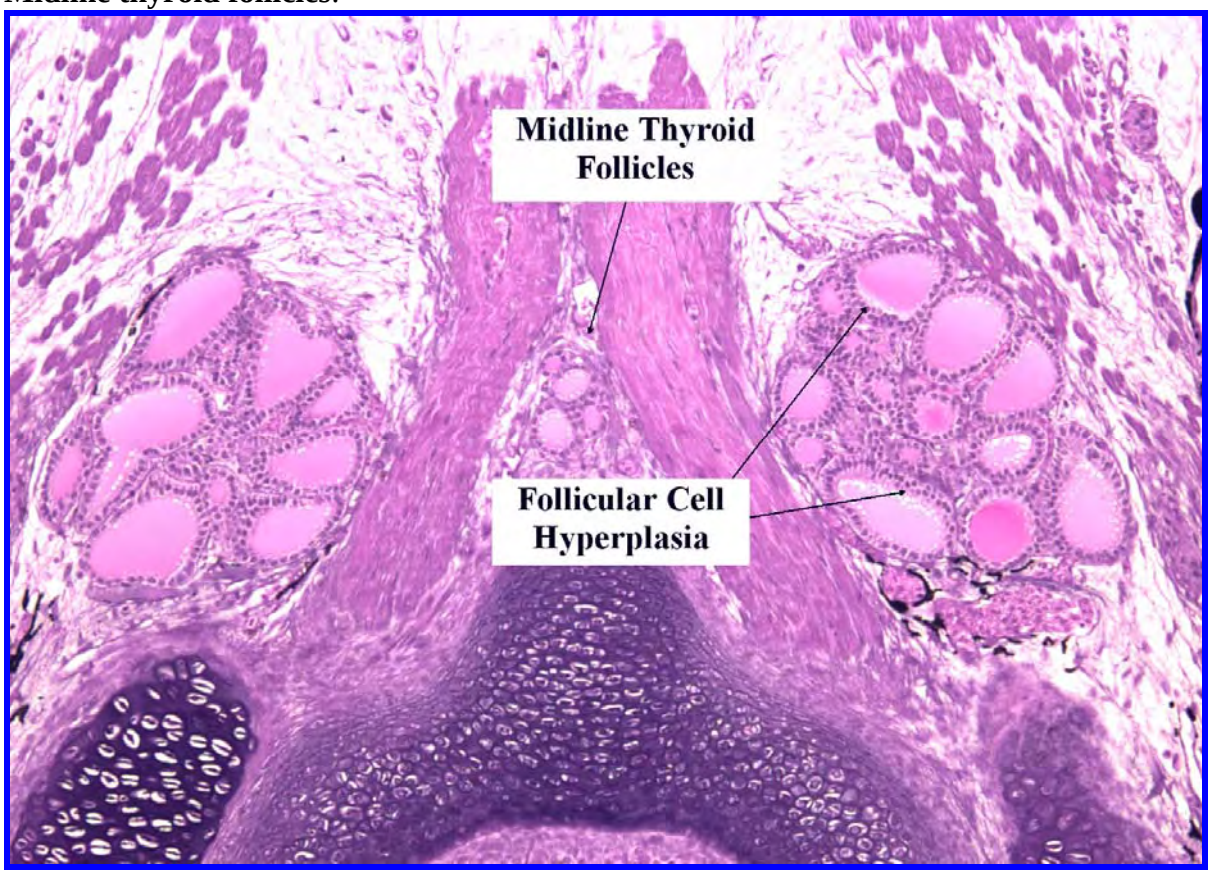


Figure 41. Medium (10x) magnification photomicrographs of thyroid glands from *X. laevis* (frontal section). The thyroid follicles contain eosinophilic homogeneous colloid and are lined by simple cuboidal follicular cells. There is minimal follicular cell hyperplasia. Thyroid follicles are present also on the midline in this frog.

Interfollicular pigment:

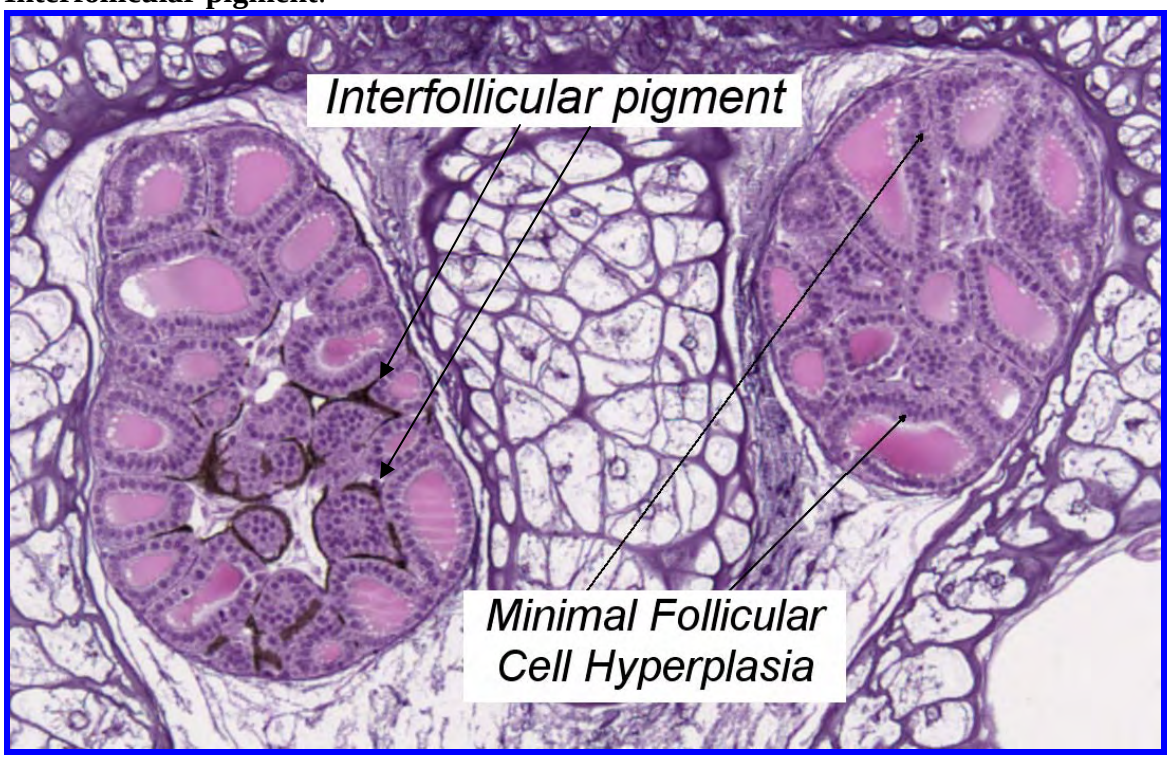


Figure 42. Medium (10x) magnification photomicrographs of thyroid glands from *X. laevis* (frontal section). Pigment surrounding follicles is present in the gland on the left. This pigment is inconsistently present in normal thyroid glands. It may occur unilaterally or bilaterally in the glands.

IIIC7. Artifacts

Colloid fall-out:

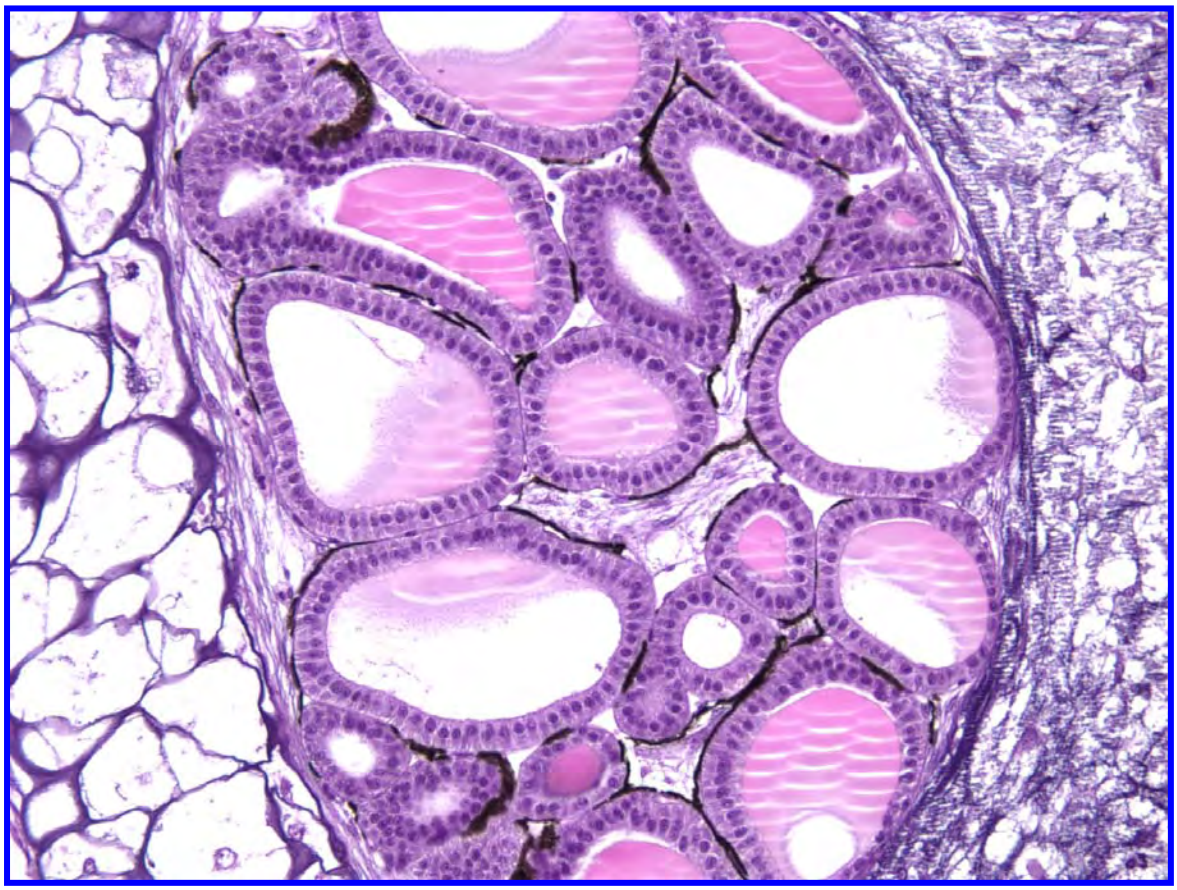


Figure 43. Thyroid gland from a stage 60 *X. laevis* tadpole. Some follicles are completely or partially devoid of colloid. This is an artifact that sometimes occurs during sectioning and is not to be mistaken with decreased colloid content. The equally distributed lines within the colloid are also a result of the sectioning process.

IV. Data Reporting

An Excel worksheet form has been created to facilitate histopathology data collection (Appendix A) and which will be provided electronically. In this worksheet, each data entry cell represents an individual tadpole. Space is also provided for comments. For each tadpole, the pathologist records the severity grade in the corresponding cell to the diagnostic criterion. If there is no reasonably appropriate diagnostic criterion for a particular finding, the pathologist can create a term that can be assigned to an unused portion of the table. If there are no findings for a tadpole, this should be recorded as ”0” (not remarkable). If insufficient tissue is available for diagnosis, this should be recorded as **IT** (insufficient tissue).

Adding a modifier to a diagnosis may help to further describe or categorize a finding in terms of chronicity, spatial distribution, color, etc. In many instances, modifiers are superfluous or redundant (e.g., fibrosis is always chronic); therefore, the use of modifiers should be kept to a minimum. An occasionally important modifier for evaluating paired thyroid glands is *unilateral* (**UNI**); unless specified in this manner, all thyroid diagnoses are assumed to be bilateral. Other modifier codes can be created as needed by the pathologist.

Reference List

1. Crissman,J. *et al.* Best Practices Guideline: Toxicologic Pathology. *Toxicologic Pathology* **32**, 126-131 (2004). http://www.toxpath.org/ToxHistopath.pdf

2. Holland,T. A survey of discriminant methods used in toxicological histopathology. *Toxicol Pathol* **29**, 269-273 (2001).

Appendix A: Histopathology Reporting Tables

Core Criteria:

Date: Chemical: Pathologist:

		Thyroid gland hypertrophy	Thyroid gland atrophy	Follicular cell hypertrophy	Follicular cell hyperplasia
Control Animal ID - replicate 1					
Control Animal ID - replicate 2					
Total:					

		Thyroid gland hypertrophy	Thyroid gland atrophy	Follicular cell hypertrophy	Follicular cell hyperplasia
___ Dose Animal ID - replicate 1					
___ Dose Animal ID - replicate 2					
Total:					

		Thyroid gland hypertrophy	Thyroid gland atrophy	Follicular cell hypertrophy	Follicular cell hyperplasia
___ Dose Animal ID - replicate 1					
___ Dose Animal ID - replicate 2					
Total:					

		Thyroid gland hypertrophy	Thyroid gland atrophy	Follicular cell hypertrophy	Follicular cell hyperplasia
___ Dose Animal ID - replicate 1					
___ Dose Animal ID - replicate 2					
Total:					

Additional Criteria:

Date: Chemical: Pathologist:

		Follicular lumen area increase	Follicular lumen area decrease
Control Animal ID - replicate 1			
Control Animal ID - replicate 2			
Total:			

		Follicular lumen area increase	Follicular lumen area decrease
____ Dose Animal ID - replicate 1			
____ Dose Animal ID - replicate 2			
Total:			

		Follicular lumen area increase	Follicular lumen area decrease
____ Dose Animal ID - replicate 1			
____ Dose Animal ID - replicate 2			
Total:			

		Follicular lumen area increase	Follicular lumen area decrease
____ Dose Animal ID - replicate 1			
____ Dose Animal ID - replicate 2			
Total:			

Narrative Descriptions:

Date:
Chemical:
Pathologist:

		Narrative description
Control Animal ID - replicate 1		
Control Animal ID - replicate 2		
Dose ____ Animal ID - replicate 1		
Dose ____ Animal ID - replicate 2		
Dose ____ Animal ID - replicate 1		

____ Dose ____ Animal ID - replicate 2		

____ Dose ____ Animal ID - replicate 1		
____ Dose ____ Animal ID - replicate 2		