

Investigation of a novel modified fixed dose determination protocol for
radioiodine treatment of feline hyperthyroidism

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ABSTRACT

Background: Radioiodine is the treatment of choice for feline hyperthyroidism. The ideal method of dose determination of ^{131}I remains controversial.

Objective: To compare a method of radioiodine dose determination that utilized thyroid scintigraphy with a standard fixed dose for treatment of feline hyperthyroidism.

Methods: Fifty-seven and 23 cats were in the novel and fixed dose groups, respectively. Cats with a percent dose uptake as determined using $^{99\text{m}}\text{TcO}_4^-$ uptake on thyroid scintigraphy <5%, 5-10%, and >10% were designated to receive 3 mCi, 3.5 mCi, or 4.5 mCi of ^{131}I , respectively, administered subcutaneously. Radioiodine dose was adjusted by thyroid size, determined by evaluating the thyroid:salivary size ratio (T:S) and categorized as <5, 5-10, and >10. If the thyroid size fell into a higher dosing category than percent uptake, the dose was increased accordingly. Cats in the fixed dose group received 4.5 mCi of ^{131}I . Six months after treatment, cats were determined to be euthyroid, hypothyroid, or hyperthyroid based on serum T4 concentrations relative to an established reference interval. Univariate analysis using Chi-square was used to determine associations between treatment and outcome.

Results: There was no difference in outcome between the novel and fixed dose treatments. Euthyroidism, hypothyroidism, and hyperthyroidism developed in 61, 30, and 9% of cats in the fixed dose group, respectively compared to 58, 26, and 16% in the novel dose group.

Conclusions: A modified fixed dose method of radioiodine based upon thyroid size and percent dose uptake was ineffective in improving outcomes over a standard fixed dose method.

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GENERAL ABSTRACT

Feline hyperthyroidism is the most common endocrinopathy in the cat. Radioiodine is considered the treatment of choice. Despite being the treatment of choice, dosing of radioiodine is controversial. No one method of radioiodine dosing has been able to consistently successfully treat hyperthyroidism in the cat.

The goal of this study was to utilize thyroid scintigraphy to determine a radioiodine dose to improve outcomes. Fifty-seven cats were dosed with 3.0, 3.5, or 4.5 mCi of radioiodine based upon thyroid size and percent dose uptake of technetium. Cats were evaluated at 1, 3, and 6 months to determine if they were successfully treated. These results were compared to a group of 23 cats dosed with a standard dose of 4.5 mCi.

There was no difference in outcome between the two groups of cats. A radioiodine dose method based upon thyroid size and percent dose uptake was ineffective in improving outcome over a standard 4.5 mCi dose method.

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ABBREVIATIONS

NIS	sodium iodide symporter
ROI	region of interest
T3	3,5,3' triiodothyronine
T4	thyroxine
T:B	thyroid to background ratio
T:S	thyroid to salivary ratio
TG	thyroglobulin
TPO	thyroid peroxidase
TRH	thyroid releasing hormone
TSH	thyroid stimulating hormone

Chapter 1: Literature Review

A. Background of feline hyperthyroidism

Feline hyperthyroidism was first described in 1979¹ and later in 1980.² Prior to this, rare instances of thyroid enlargement and nodules seen on histopathology were noted on feline necropsies. Gross abnormalities such as cardiac enlargement, thin body condition, and metastatic calcium deposition that have been associated with hyperthyroidism were found in a few cats, however the majority of cats did not display clinical signs of hyperthyroidism.^{3,4} Since being first described, there has been a dramatic increase in the number of cases of feline hyperthyroidism reported, making it the most common endocrine disorder in cats and one of the most commonly diagnosed disorders in small animal practice.⁵ Actual prevalence rates vary widely according to geographic location. The overall prevalence of feline hyperthyroidism in England is 2.38-3%^{6,7} with a prevalence in cats over nine years of age of 7.8%.⁷ In Warsaw, Poland, the overall prevalence of hyperthyroidism in cats over 7 years of age was reported to be 20%.⁸ Most cats diagnosed with hyperthyroidism are middle-aged to older.⁹

B. Thyroid physiology/potential factors in development of disease

The feline thyroid gland consists of two lobes that lie adjacent to the left and right side of the trachea in the mid-cervical region, and in the non-diseased state are not palpable. The functional unit of the thyroid gland is the follicle which contains colloid that stores thyroglobulin (Tg) that serves as a reservoir for thyroid hormones.¹⁰ Each molecule of (Tg) contains approximately 70 tyrosine molecules, from which thyroid hormones are derived.¹¹ Since thyroid hormones are iodinated, iodide is the rate limiting substance for their formation.¹² Most dietary iodine is reduced to iodide before absorption in the small intestine. After intestinal absorption,

iodide is transported in plasma to the thyroid gland where it is concentrated in the thyroid gland, eventually forming 3, 5, 3'-triiodothyronine (T3) and thyroxine (T4). Uptake of iodide by the thyroid gland, called iodide trapping, is dependent on availability, with the percentage uptake increased in states of iodine deficiency.¹³ Iodine uptake by the thyroid gland is increased in hyperthyroidism, with a rate of uptake of ¹³¹I at 24 hours of 2.02 +/- 0.79% and 22.0 +/- 13.4% in euthyroid and hyperthyroid cats, respectively.¹⁴

Synthesis of thyroid hormones is regulated by thyroid stimulating hormone (TSH), secreted by the pituitary gland. TSH binds and activates the TSH receptor on the thyroid follicular cells, which results in increased iodide uptake via the sodium iodide symporter (NIS). Iodide is transported into thyroid follicular cells by the NIS located at the basolateral membrane of the thyrocyte. Iodide is then oxidized by thyroid peroxidase (TPO) which subsequently catalyzes iodination of tyrosine molecules on Tg in the presence of hydrogen peroxide. This process, known as organification, results in the formation of monoiodotyrosine and diiodotyrosine. Thyroxine (T4) is formed when two diiodotyrosine residues couple with one another, and triiodothyronine (T3) is formed when one molecule of monoiodotyrosine couples with one molecule of diiodotyrosine.¹⁵ T3 and T4 are cleaved from the thyroglobulin molecule by pinocytosis and the digestion of Tg molecules by multiple proteases, which release T3 and T4 in the free form. Tg is recycled back into the follicular cell and T3 and T4 then diffuse through the base of the thyroid cell into the surrounding capillaries, resulting in thyroid hormones being released into the bloodstream.¹¹ Control of plasma thyroid hormone concentrations is via negative feedback of circulating T3 and T4 suppressing TRH secretion from the hypothalamus and TSH secretion from the pituitary gland. In primary hyperthyroidism, excessive production of T4 and T3 leads to suppression of both TRH and TSH.¹⁶ Although T4 is the major product

secreted from the thyroid gland, it is thought to primarily be a prohormone for T3 which is 3-5 times more potent than T4.⁵

The most common pathology seen with hyperthyroidism in the cat is functional thyroid adenomatous hyperplasia or a benign thyroid adenoma. It is similar pathologically and clinically to toxic nodular goiter (Plummer's disease) in humans.¹⁷ Normally, there is a subpopulation of follicular cells in the feline thyroid gland that have a high growth potential. The multinodular nature of the disease is likely associated with this subpopulation of thyrocytes that replicate in an autonomous way, independent of TSH.¹⁸ Disease may affect one or both thyroid lobes, with one recent study reporting unilateral disease in 32%, bilateral disease in 63%, and suspected carcinoma in 1.7% of cats diagnosed with hyperthyroidism confirmed by thyroid scintigraphy.¹⁹ It was originally thought that the pathogenesis of malignant tumors differed from that of the benign thyroid changes more commonly seen.¹⁸ A more recent study suggests that the prevalence of thyroid carcinoma increases with disease duration, suggesting the development of thyroid carcinoma is a result of progression of hyperthyroid disease.¹⁹ It is unclear over what time period clinical disease develops. Clinical signs develop after the adenomatous thyroid tissue begins to concentrate iodide and secretes excessive amounts of T4 and T3. Thyroid follicular cells replicate and the thyroid hormones are secreted in an autonomous fashion, independent of any negative feedback from the hypothalamic-pituitary-thyroid axis.¹⁸

The cause of thyroid hyperplasia and autonomous function is unknown, though several theories exist regarding genetic, nutritional, and environmental factors. It is believed that mutations of the TSH receptor gene or its associated G proteins may play a role in the development of disease. G proteins can either be stimulatory or inhibitory to the activity of the adenylate cyclase enzyme which stimulates hormone production. A reduction in inhibitory G

proteins may lead to increased hormone production and hyperthyroidism. One study has shown a decreased amount of the inhibitory G protein subtype $G_{i\alpha 2}$ in feline thyroid adenomas.²⁰ Another study identified nine somatic mutations in the TSH receptor gene of hyperthyroid cats, five of which had been previously identified in human cases of hyperthyroidism.²¹ Burmese, Persian, Siamese, Himalayan and other purebred cats have decreased risk of developing hyperthyroidism, possibly indicating a genetic protective component against hyperthyroidism in these breeds.^{7,22}

Commercial cat food diets have been implicated due to widely variable amounts of iodine between foods. As iodine is an essential component of thyroid hormone production, it is hypothesized that an iodine deficient diet initially results in decreased thyroid hormone production. Persistently decreased thyroid hormone production then results in chronic TSH release and overstimulation resulting in hyperplasia and the development of autonomous nodules within the thyroid gland.²³ When iodine intake increases, production of thyroid hormones subsequently increases. Toxic nodular goiter in humans can occur when a diet is changed from an iodine deficient diet to an iodine replete diet, and this might happen in cats as well.^{15, 24} Other nutritional factors implicated in the development of feline hyperthyroidism include dietary selenium and soy flavonoids that are known to be endocrine disrupters.¹⁵

Environmental factors that have been implicated in the development of feline hyperthyroidism include Bisphenol A (BPA) used in liners of canned cat food,²⁵ and polybrominated diphenyl ethers (PBDEs) used in flame retardants.²⁶ BPAs and PBDEs are known endocrine disrupters that because of their structural similarity to thyroid hormone interfere with the hypothalamic-pituitary-thyroid gland axis.^{26,27} Both flea control products,¹⁸ and cat litter²⁸ have also been implicated as potential causes. Though several risk factors have

been identified, the definitive underlying cause for the development of hyperthyroidism in the cat remains unknown, and may be multifactorial.

C. Clinical signs and complications

Thyroid hormones affect many body functions and play an important role in the regulation of many enzymes, regulation of hormones, and the metabolism of vitamins, minerals, lipids, proteins, and carbohydrates.²⁹ They also have chronotropic and inotropic effects on the heart, are necessary for normal respiratory function, and stimulate both the formation and resorption of bone.¹⁰ There is virtually no body system that is not regulated in some manner by thyroid hormones.

The clinical signs of hyperthyroidism are directly attributable to excess thyroid hormones. Presenting history typically includes weight loss despite having a normal or increased appetite, often accompanied by gastrointestinal signs, hyperactivity and other behavioral changes, polyuria, and polydipsia. Some cats will have the apathetic form of hyperthyroidism and will be inappetent or anorexic. Physical exam findings usually include a palpable goiter, tachycardia with or without a heart murmur, and decreased body condition from weight loss. Thoracic radiographs may show left-sided cardiomegaly, echocardiography may show left ventricular hypertrophy with left atrial and ventricular dilation, and a small number (2-3%) will have signs of heart failure.³⁰ Hypertension may be present, but in many cases it is difficult to determine if it is related to chronic renal disease that exists concurrently in many geriatric cats or as a direct effect of hyperthyroidism. Some cats may have ocular abnormalities such as tortuous retinal vessels secondary to hypertension. Studies have shown 54-86% of cats to have cardiovascular manifestations at time of diagnosis.^{9,31,32} Most cardiovascular effects are reversible with appropriate treatment of hyperthyroidism, but hypertension may persist.

The effects of hyperthyroidism on renal function are well documented. Thyroid hormone acts directly on vascular smooth muscle cells, causing them to relax, resulting in decreased peripheral vascular resistance.³³ Subsequent activation of the renin-angiotensin aldosterone system (RAAS) occurs and has been documented in cats with hyperthyroidism.³⁴ Increased plasma volume results in an increase in cardiac preload and combined with decreased peripheral vascular resistance ultimately increases cardiac output.³⁵ In addition, hyperthyroidism may result in upregulation of beta-adrenergic receptors within cardiac tissue causing increased heart rate.³⁶ As a result of decreased vascular resistance, activation of RAAS, and increased cardiac output, subsequent increases in renal blood flow and glomerular filtration rate (GFR) occur. Renal proteinuria, which is a risk factor for development or progression of azotemia, is a common finding in cats with hyperthyroidism.^{37,38} Decreases in GFR post treatment have been documented, with previous studies showing 15.3-49% of hyperthyroid cats developing azotemia after treatment regardless of treatment method.^{32,39-43} It has also been shown that most cats treated with radioactive iodine have a significant drop in GFR one month post treatment, with no further decline six months after treatment. This same study showed that cats with subnormal T4 had GFR increase markedly once T4 levels normalized.⁴⁴ One study evaluating the long-term survival of hyperthyroid cats treated with I¹³¹ found that 41% had renal abnormalities just before death.³² It is unclear whether hyperthyroidism results in damage to the kidneys or if renal injury is a consequence of the high prevalence of renal failure in the geriatric population.

Other concurrent diseases have been reported to occur frequently in cats diagnosed with hyperthyroidism. One study showed 18% of cats to have concurrent diseases, with the two most common being alimentary lymphoma and chronic enteropathy.⁴⁵ Other studies have shown

urinary tract infections in 12%⁴⁶ and diabetes mellitus in up to 5.8%⁴⁷ of hyperthyroid cats.

D. Diagnosis

The diagnosis of hyperthyroidism in the cat is usually not difficult, but cats presented with mild hyperthyroidism or concurrent non-thyroidal disease can be problematic. Diagnosis is based upon compatible physical exam findings (i.e. palpable thyroid nodule, tachycardia, thin body condition) and routine laboratory testing, with confirmation using thyroid function tests. Serum T4 is the preferred screening test for hyperthyroidism, as over 90% of hyperthyroid cats have a serum T4 concentration that is above the reference interval.^{9,48,49} Approximately 10% of all hyperthyroid cats and over 30% of cats with early or mild hyperthyroidism have serum T4 concentrations within reference range.^{9,48-50} It has been reported that 2-4% of euthyroid cats have a serum T4 concentration above the reference range.⁵⁰ A diagnosis of hyperthyroidism should never be based upon just an elevated T4 without other evidence of hyperthyroidism. If an elevated serum T4 is found on routine screening in an otherwise asymptomatic cat, one should perform a thorough review of history and physical exam findings to look for concurrent disease, and repeat T4 measurement to verify results prior to treatment.⁴⁹ Fluctuations of T4 in and out of the reference range, and suppression of T4 into the reference range because of concurrent non-thyroidal illness are possible explanations of why a T4 concentration can be within the reference range in a hyperthyroid cat.^{48,49,51} Most cats with early hyperthyroidism that have T4 concentrations within the reference range will subsequently have elevated serum T4 concentrations upon retesting a few weeks to a few months later.¹⁷

Serum T3 concentrations are highly correlated with serum T4 in hyperthyroid cats. Measurement of serum T4 is preferred over T3 for diagnosis of hyperthyroidism because of its considerably higher sensitivity. Most hyperthyroid cats with normal T3 concentrations and

mildly elevated T4 concentrations have early disease. As with T4, if the hyperthyroid state is allowed to progress untreated, T3 would eventually rise above reference range. Measurement of serum T3 is not recommended as a diagnostic test for hyperthyroidism, and if used, should always be used concurrently with a serum T4 concentration.^{9,48}

In cats suspected to have hyperthyroidism with a normal serum total T4 concentration, free T4 concentrations can help in obtaining a diagnosis.^{49,50,52} A variety of methods are used to measure free T4. The equilibrium dialysis technique is believed to be a more accurate technique with the non-equilibrium dialysis technique having little benefit over serum T4 alone.⁴⁹ Even though free T4 is more sensitive than T4 for diagnosing hyperthyroidism in the cat, the specificity of the test is low, with up to 20% of sick euthyroid cats having falsely elevated free T4 results.⁴⁹ To better obtain an accurate diagnosis, free T4 should always be used in conjunction with clinical signs and serum T4. The specificity of the test for hyperthyroidism increases if serum T4 concentrations are in the upper end of the reference range.⁴⁸⁻⁵⁰

Measurement of serum TSH may be of benefit in diagnosing hyperthyroidism in the cat, especially in early or subclinical stages. A feline-specific assay has not been developed, however a canine TSH assay is widely available and believed to provide useful diagnostic information. Feline TSH is 94-96% homologous to canine TSH.⁵³ Canine TSH assays however cannot distinguish between low normal and subnormal concentrations of TSH in cats.^{49,50} One study showed that euthyroid cats with undetectable TSH at baseline were significantly more likely to be diagnosed with hyperthyroidism in the future.⁵⁴ Another study showed TSH to be suppressed below the limit of detection of the assay in 98% of hyperthyroid cats, but found that concentrations were measurable in a few cats with mild to moderate hyperthyroidism. In the same study, approximately 30% of older euthyroid cats were also found to have undetectable

TSH concentrations.⁵⁰ Because of the poor specificity of serum TSH concentrations, it should never be used as a solo test. However, when measured in conjunction with T4 and fT4, serum TSH enhances the specificity of the tests.

Dynamic function tests such as T3 suppression testing and TRH stimulation may be considered in rare situations where clinical signs of hyperthyroidism are not supported by an increase in serial serum T4 concentration or free T4 concentrations. Disadvantages of these tests include cost and client compliance. TSH stimulation testing is not recommended as a diagnostic test for hyperthyroidism in the cat due to the high cost to perform and lack of validity.⁴⁸

Thyroid scintigraphy is used as an aid in estimating thyroid size and to quantify function by measuring uptake of a radionuclide (either pertechnetate or radioiodine). Thyroid scintigraphy is considered the gold standard method in diagnosing feline hyperthyroidism in cases of mild or occult disease.⁴⁹ A study using thyroid scintigraphy to determine the thyroid-to-salivary ratio as a diagnostic test for feline hyperthyroidism had a 98.7% sensitivity compared to a sensitivity of 90.9% for serum T4 concentration.⁵⁵ Scintigraphy has the benefit of being able to distinguish between bilateral and unilateral disease, assesses thyroid size as well as physiologic activity, and identifies ectopic and metastatic thyroid tissue.^{55,56} Thyroid scintigraphy is very sensitive and has the ability to diagnose hyperthyroidism prior to thyroid hormone tests being abnormal. It can also exclude the diagnosis of hyperthyroidism in euthyroid cats with falsely elevated T4 or free T4.⁴⁹ Potential disadvantages of scintigraphy is that interpretation can be affected by observer variability,⁵⁷ concurrent iohexol administration,⁵⁸ rhTSH administration,⁵⁹ varying anesthetic protocols,⁶⁰ restricted iodine diet,⁶¹ and methimazole administration.^{62,63}

Additional imaging modalities used to aid in the diagnosis of hyperthyroidism include

ultrasonography and computed tomography (CT). Ultrasonography is operator specific and can be technically challenging. Despite this, it has been used to determine the volume and dimensions of thyroid glands in both euthyroid and hyperthyroid cats. One study showed 85.7% agreement with scintigraphy in defining normal and abnormal lobes.^{52,64} A recent study comparing CT to scintigraphy found that size estimates of thyroid lobes were comparable with each modality, however CT was poor at identifying unilateral vs. bilateral disease.⁶⁵ Treatment with methimazole has been shown to result in a decrease in attenuation and heterogeneity of thyroid lobes, but not thyroid size.⁶⁶

E. Treatment

Treatment options for feline hyperthyroidism include administration of antithyroid drugs, surgical thyroidectomy, dietary iodine restriction, or the administration of radioiodine (¹³¹I). Regardless of the treatment modality chosen, the goal is to restore euthyroidism, avoid the development of hypothyroidism, and to have minimal side effects. It is recommended that all hyperthyroid cats, even those with concurrent disease including renal failure, be treated.⁶⁷ The most commonly used antithyroid drug in North America is methimazole. Methimazole's mechanism of action is inhibition of thyroid peroxidase, the enzyme responsible for formation of T4 and T3 through the rate limiting oxidation of iodide as discussed previously. It does not block the release of preformed thyroid hormones, so there is a delay after onset of treatment until a decrease in thyroid hormone is appreciated.⁶⁸ The medication is available in oral and transdermal form. Most hyperthyroid cats treated with methimazole have successful control of their disease.⁶⁹ Poor client compliance, especially with the oral administration is a major drawback of methimazole treatment, and can lead to treatment failure. The transdermal form, though effective, has been shown to require higher doses with prolonged treatment.⁷⁰

Methimazole administration does not affect growth of the adenoma, so thyroid tumors continue to grow and potentially undergo malignant transformation.¹⁹ Side effects of methimazole include vomiting, anorexia, blood dyscrasias, facial pruritus and excoriation, hepatotoxicity, coagulation problems, and acquired myasthenia gravis.⁶⁸ Carbimazole, a pro-drug that is converted to methimazole, is an alternative medication that is not currently available in the United States. Propylthiouracil is another drug not routinely used due to its higher prevalence of side effects compared to methimazole.

Surgical thyroidectomy is a treatment option that requires no special equipment and many veterinarians are capable of performing. Studies have shown a success rate after thyroidectomy of anywhere from 89-95%.^{71,72} Recurrence of disease post thyroidectomy has been reported as little as 3 months and as long as 59 months post thyroidectomy.⁷² Another study documented an asymptomatic elevation in T4 63 months post thyroidectomy.⁷¹ Surgery is not recommended without scintigraphy because atypical or ectopic location of hyperplastic thyroid tissue increases chance of recurrence.⁷² Complications of thyroidectomy include hypocalcemia, hypothyroidism, hemorrhage, and recurrent laryngeal nerve damage.⁷³ Hypothyroidism often occurs at least transiently in animals undergoing bilateral thyroidectomy, but permanent hypothyroidism is thought to be rare.⁷¹ Others prefer to supplement levothyroxine indefinitely, monitoring thyroid levels every 6 months, adjusting the dose accordingly.⁷²

As previously mentioned, dietary iodine is essential for thyroid hormone production. It has been shown that restricting dietary iodine intake can decrease or normalize serum T4 concentrations in hyperthyroid cats.⁷⁴ One study showed that strictly feeding a commercial iodine restricted diet (Hill's y/d) resulted in a reduction of T4 concentration to within the reference range in 64% and 75% of cats after 4 and 8 weeks, respectively.⁷⁵ Another study

showed 42% and 83% of cats at 60 and 180 days of feeding the iodine restricted diet, respectively, had normal serum T4 concentrations. Despite normalization of serum T4 concentrations, body weight of cats did not improve at 180 days.⁷⁶ Clinical signs of hypothyroidism were not documented in either study despite several cats having T4 concentrations below the reference range. Long term effects of feeding an iodine restricted diet have not been studied and are not well understood. Humans eating an iodine deficient diet chronically can develop a nodular goiter that results from increased secretion of TSH. Hypertrophy, hyperplasia, and autonomous nodules may result.¹⁵ Like methimazole, growth of hyper-functioning thyroid tissue can continue to occur on an iodine restricted diet, with possible malignant transformation over time.¹⁹ Another disadvantage is that cats must be strictly fed this food, as even a small amount of another diet renders this method of treatment ineffective. This can cause difficulty with compliance in multi-cat households and with indoor-outdoor cats.

Radioiodine (¹³¹I) is considered to be the treatment of choice for most hyperthyroid cats, as it eliminates the necessity of daily medication and potential side effects, eliminates diet restrictions, and avoids surgical and anesthetic risks. Thyroid cells do not differentiate between stable and radioactive iodine, therefore radioiodine is concentrated like iodine by the thyroid gland.¹⁴ ¹³¹I emits both β and γ radiation. β particles, which are locally destructive, travel 1-2 mm in thyroid tissue, sparing adjacent tissues, and cause the majority of tissue damage by their ionizing effects.^{77,78} Radioiodine is primarily concentrated in hyperactive thyroid tissue directly destroying hyperplastic cells by its irradiation effects. Normal tissue has limited damage as iodine uptake is suppressed because pituitary TSH secretion is decreased resulting from negative feedback of thyroid hormones. However, large doses of radioiodine may damage even atrophied thyroid tissue.^{48,79} Disadvantages of treatment are that a limited number of facilities can

perform radioiodine therapy due to the necessity of a radioactive materials license and isolation requirements to prevent human exposure requires prolonged hospitalization.⁶⁷ At least 75% of the injected dose is excreted in the feces and urine, which can be a significant health hazard to caretakers.⁸⁰ Additionally, treatment failures have been well documented. Studies have shown 1.0-9% of cats to be persistently hyperthyroid.⁸¹⁻⁸⁷ Factors associated with persistent hyperthyroidism include a pretreatment T4 >250 nmol/L, greatly enlarged thyroid glands, thyroid carcinoma, and receiving anti-thyroid drugs within one month prior to treatment.^{81,86,88} The development of hypothyroidism with or without clinical signs requiring levothyroxine supplementation has been shown to occur in 2.1-30% of cats after treatment^{83-86,89} most often in those with bilateral disease.⁸⁹ The development of hypothyroidism can have serious clinical implications as iatrogenic hypothyroidism can contribute to the development of azotemia and reduced survival time.⁹⁰ Also, many owners are unable to administer oral medication, which is often a principal reason for choosing radioiodine treatment. Despite being the treatment of choice, radioiodine dose determination is controversial. No single method of dose determination is widely accepted. Thyroid scintigraphy is often used to evaluate disease and to help determine a radioiodine dose.

F. Assessing feline hyperthyroidism using thyroid scintigraphy

The principle behind thyroid scintigraphy is the selective uptake of a radionuclide by thyroid tissue that is able to be imaged by a gamma camera.^{56,91} The radionuclide of choice is Technetium-99m in the form of sodium pertechnetate ($\text{Na}^+ \text{}^{99\text{m}}\text{TcO}_4^-$). Technetium is a transition metal and in the form of sodium pertechnetate acts in a manner similar to iodine due to size and valence. It is actively trapped and concentrated in the thyroid gland. However, it does not undergo organification and incorporation into thyroid hormones. Unlike iodine, it is neither

bound to thyroglobulin nor stored in the thyroid gland.⁹¹ Pertechnetate is preferred over ¹²³I (a radioisotope of iodine with γ -ray emissions) due to availability and cost, and ¹³¹I (a radioisotope of iodine with β and γ -ray emissions) because of a shorter half-life and lack of beta emission, reducing exposure of personnel to radiation.^{91,92} Since the NIS is also present in salivary tissue, radionuclide uptake is also visualized in the salivary glands. The amount of radionuclide concentrated within the thyroid gland is determined using a gamma camera and an imaging computer. Low-energy-all-purpose (LEAP) collimators are used most often in veterinary medicine and are best for image analysis allowing quantifying multiple regions-of-interest without concerns of geometry and depth of targeted tissues. Pin-hole collimators can be used to magnify the thyroid gland and provide better spatial resolution. Usually imaging is performed 20 minutes after intravenous injection of pertechnetate, though differences in uptake between 20 and 60 minutes have not been shown to be significant.⁶²

The thyroid scintigraphy image is evaluated both qualitatively and quantitatively. A normal thyroid scintigraphy scan will show uniform distribution of radioactivity throughout both thyroid lobes that are symmetrical in size and shape.⁵⁶ Occasionally some euthyroid cats will have thyroid gland asymmetry, so caution must be taken when they are evaluated visually. Subjectively, the radionuclide uptake should appear similar in the thyroid lobes and salivary glands.^{92,93} Bilateral disease may be symmetric or asymmetric, with one or both having greater intensity than the salivary glands.⁵⁵ In cases of unilateral hyperthyroidism, the contralateral thyroid gland will not be visualized because of suppression of TSH rendering the contralateral gland inactive and atrophied. Ectopic neoplastic thyroid tissue has been identified in about 4% of hyperthyroid cats.⁵⁵ Changes in the thyroid gland suggestive of malignant transformation including distorted margins, multiple foci of uptake, extension into the thoracic inlet, and

metastasis can also be evaluated, although scintigraphy cannot reliably differentiate benign from malignant disease.^{56,91,92} In one study of thyroid carcinoma determined by histopathology, thyroid scintigraphy could not distinguish between benign and malignant tissue in 7/8 cases.⁹⁴

Several methods have been used to evaluate the thyroid gland quantitatively using scintigraphy. The most commonly used and simplest method to quantify thyroid function is the thyroid-to-salivary intensity ratio (T:S). The comparison is performed by placing ROIs to obtain a count density ratio of the thyroid to the zygomatic salivary glands.^{93,95} The ROI is determined by manually drawing an ROI around each thyroid and salivary image. The T:S intensity ratio is calculated by dividing the mean count density within the thyroid glands ROI by the mean count density of the salivary glands ROI.^{92,96} Normal cats have a T:S intensity ratio of 0.87:1 with a range of 0.6:1 to 1.03:1.^{58,62,93,96} There is the potential for the T:S intensity ratio to be influenced by either thyroid or salivary dysfunction.⁹⁶ Thyroid-background (T:B) ratios have been another method used as they are not affected by salivary dysfunction, and reduce operator error by using only one ROI.^{55,97} The location of the background ROI has been shown to affect values for background activity. Usually the T:B ratio is determined by one ROI in the axillary/shoulder area, however it has been shown that when the one ROI is drawn over the heart it has the best correlation with plasma radioactivity.⁹⁸ T:B ratios are determined by dividing the average thyroid count density by the average background count density.⁵⁵ The normal mean T:B ratio is 2.76:1 with a range of 1.7 to 4.⁹⁹

Measuring percent dose uptake of radionuclide is another quantitative method of evaluating feline thyroid disease. The percent dose uptake is calculated by dividing depth corrected thyroid counts (counts per minute) of all visible thyroid tissue by the amount of radionuclide injected (counts per minute).^{62,100} The main disadvantage is that it is cumbersome

to calculate, but it is a direct test of thyroid uptake, independent of salivary uptake of pertechnetate. The T:S intensity ratio has been shown to have the highest overall test sensitivity for diagnosis of hyperthyroidism, while percent dose uptake may be the best method to determine the functional and metabolic status of the thyroid.¹⁰⁰ The intensity of thyroid uptake correlates well with serum T4 concentrations.⁹⁶

Methimazole administration within 14 days of a thyroid scan can increase thyroid uptake of radionuclide. In cats that are affected unilaterally, this increased uptake may result in a false diagnosis of bilateral disease.^{62,63} This occurs because of a lack of suppression of normal thyroid tissue when TSH levels increase with methimazole therapy. This artifact may result in increased radiation dose to the remaining normal thyroid cells because of increased uptake of ¹³¹I during radioiodine treatment, and increase the risk of post-treatment hypothyroidism.^{62,91}

G. Radioiodine dose determination

The goal of radioiodine treatment of hyperthyroidism is to return a cat to a euthyroid state while avoiding hypothyroidism or persistent hyperthyroidism. Radioiodine can be administered orally, intravenously or subcutaneously. Though the intravenous and subcutaneous routes are equally effective, the subcutaneous route is preferred due to ease of administration, and the lack of a need to place an IV catheter.^{77,101,102} The oral route is not recommended as cats may require relatively higher doses than for people which can lead to vomiting and environmental contamination.¹⁰³ There are three distinct methods in which radioiodine dose can be determined: tracer technique, fixed dose, and modified fixed dose. Variations of each method have been used to achieve optimal dosing.

The tracer technique utilizes a small dose of ¹³¹I to estimate the percentage of iodine

uptake and rate of disappearance from the thyroid gland, and requires an estimate of the weight of the thyroid gland.^{81,104} The tracer technique is designed to predict the individual kinetics of therapeutic ¹³¹I treatment in each cat. The tracer technique is no longer used due to errors in accurately estimating thyroid weight, the need for expensive, sophisticated equipment, and the requirement that a cat be imaged multiple times to determine biologic clearance rates.^{79,86,104}

The fixed dose technique, where cats are treated with a fixed empirical dose of radioiodine, regardless of the severity of their disease, is perhaps the most frequently used technique. The radioiodine dose administered is typically 4-5 mCi per cat.^{79,91} This dosing method is preferred by many facilities since it does not require nuclear imaging or special dose calculations.^{83,84} Currently, a fixed dose method is the preferred method of treating toxic nodular goiter in humans.^{105,106} The development of hypothyroidism after radioiodine treatment in humans is considered acceptable with as many as 90% of patients becoming hypothyroid.⁷⁸ Though some studies have shown fixed dose to be a safe and effective method of treating cats with radioactive iodine, there is a concern that hypothyroidism develops in an excessive number of cats with this method.^{56,83,84} In addition, the excessive administration of ¹³¹I exposes both the cat and veterinary personnel to unnecessary levels of radiation.^{56,79} One study of 62 cats using a fixed dose of 4 mCi found that 8% of cats remained persistently hyperthyroid and 5% were hypothyroid 6 months post treatment.⁸⁴ A study of 65 cats using a fixed dose of 4.0 mCi per cat found 5% to be persistently hyperthyroid and 8% to be hypothyroid after treatment.⁸³ Another study of 9 cats using a fixed dose of 3-4 mCi per cat had 8/9 cats become euthyroid. The one persistently hyperthyroid cat had been on carbimazole up to the point of treatment¹⁰², possibly inhibiting radioiodine efficacy.⁸¹ Another study of 193 cats using a fixed dose of 4 mCi had a 2% rate of persistent hyperthyroidism and 9% rate of clinical hypothyroidism requiring

levothyroxine supplementation post treatment. In this study, they did not find a significant difference in efficacy of treatment, based upon when methimazole was discontinued.⁸⁷

The modified fixed dose technique for determining radioiodine dosing may be performed by assessing a number of factors including T4 concentration, clinical signs, thyroid size and volume, and patient weight.^{85,86,107-109} Depending upon the factors evaluated, a tiered dose of ¹³¹I may be administered on a sliding scale (usually approximately 3, 4, or 5 mCi). In radioiodine treatment of Graves' disease in humans, a tiered dosing regimen has been shown to be more effective than a fixed dose method.¹¹⁰ This method attempts to minimize the amount of radiation each patient receives while still being safe and effective.

A study using a modified fixed dose of radioiodine from 2.0 to 5.0 mCi based strictly upon the volume of hyper-functioning thyroid tissue as determined by thyroid scintigraphy found that 88% returned to euthyroidism and that 12 % remained hyperthyroid. It was found that cats with significantly higher T4 concentrations, and higher volumes of functioning thyroid tissue were more prone to remain hyperthyroid, suggesting that in these cats, calculating a dose based upon thyroid volume as determined by scintigraphy may be inadequate. Also a number of these cats were dosed orally, suggesting oral dosing of radioiodine may be less effective.¹⁰⁸ Another study using a modified fixed dose protocol based upon severity of clinical signs, estimated size of thyroid nodule, and serum T4 concentration used doses of radioiodine ranging from 3.5 mCi to 24.0 mCi. In this study there was a 30% rate of hypothyroidism, with cats having bilateral disease being at increased risk. The follow up period varied with some cats receiving follow up for as little as 3 months or up to 5 years, which may have influenced the results. This study also used much higher doses of radioiodine than other studies.⁸⁹ Another study using a modified fixed dose technique that used the same evaluation parameters as the Nykamp study, dosed cats

with from 2.0 mCi to 6.0 mCi. In this study 1.5% of cats remained persistently hyperthyroid, 2.1% became clinically hypothyroid, and 10.9% had biochemical hypothyroidism. Of the 10.9% of cats with a low T4, 43.5% had detectable non-thyroidal illness which may have lowered their T4. This study had a consistent follow up period of 6 months and noted those cats with significantly higher T4 concentrations and larger thyroid volumes to be more likely to remain persistently hyperthyroid.⁸⁶ Another study using a modified fixed dose ranging from 1.88 mCi to 10.06 mCi based upon severity of clinical signs, total T4, and the T:S intensity ratio on scintigraphy found that 9.5% were persistently hyperthyroid and 24% were hypothyroid. There was an increased rate of persistent hyperthyroidism in the group with the largest thyroid gland size, however the majority of patients that developed hypothyroidism also presented with larger thyroid volumes than the group that became euthyroid. As in the Nykamp study, bilateral disease was considered a risk for the development of hypothyroidism. This is likely due to more thyroid tissue being destroyed with radioiodine, with less functioning tissue left after treatment.¹¹¹ Another study used a modified fixed dose of 2.8 mCi to 8.9 mCi based upon the number of thyroid nodules present and weight of the cat. In this long-term follow up study, 85% of treated cats became euthyroid, with 4% remaining hyperthyroid, and 9% developed hypothyroidism. This study also had a variable period of follow up likely affecting results.⁸⁵

Even though thyroid carcinomas account for fewer than 5% of hyperthyroidism in cats, they pose a special challenge as much higher doses of radioiodine are required for successful treatment. Treatment was successful in 6/8 cats with thyroid carcinoma (7 out of 8 had mitotic activity) using a single high dose of radioiodine of 30 mCi.⁹⁴ Another study reported 7 cats treated with high dose radioiodine (30 mCi) for thyroid carcinoma that had previously undergone surgery. All 7 cats in this study became hypothyroid.¹¹² Another study reported a metastatic

rate of up to 71% in feline thyroid carcinomas, most commonly to the lungs and lymph nodes.¹¹³ Metastatic disease can be difficult to differentiate from ectopic thyroid tissue. It is possible that in some cases carcinomas may be the underlying reason for treatment failures. Despite being the treatment of choice, no one definitive method of radioiodine dose calculation has been found to consistently and reliably result in obtaining euthyroidism.

Since no study has been able to determine the best method of radioiodine dose determination and a variety of results have been obtained from both the fixed and modified fixed dose determination methods, an investigation into defining an ideal method of radioiodine dose determination in the cat was needed, as has been performed in people.¹¹⁴ Previous studies have lacked specific criteria for dose determination and there has never been a direct comparison between the modified fixed dose and fixed dose methods. Thus, a study directly comparing the fixed dose method and modified fixed dose method using the specific criteria of percent dose uptake and thyroid size was designed.

Chapter 2: Investigation of a Novel Modified Fixed Dose Determination Protocol for Radioiodine Treatment of Feline Hyperthyroidism

A. Introduction

Radioactive iodine is the treatment of choice for feline hyperthyroidism. However, the optimal method for calculation of the dose of ^{131}I has not been determined. None of the three methods of radioiodine dose determination: tracer technique, fixed dose method or modified fixed dose method has consistently resulted in euthyroidism. The efficacy of various modified fixed dose methods is difficult to assess because of substantial variation and often, incomplete description of how the dose was determined. The technique uses one or more variables, including severity of clinical signs, thyroid tumor size or volume, and serum thyroxine (T4) concentration to determine a tiered dose of radioiodine. A study using a modified fixed dose technique based upon thyroid volume alone, was ineffective for cats with severe hyperthyroidism.¹⁰⁸ Another study using a modified dose technique based upon the number of thyroid nodules present and body weight resulted in 4% remaining hyperthyroid and 9% hypothyroid.⁸⁵ Studies determining radioiodine dose based upon severity of clinical signs, thyroid size, and serum T4 concentration resulted in 1.5-9.5% being persistently hyperthyroid and 2.1-30% becoming hypothyroid. Cats with bilateral disease and larger thyroid volumes were at an increased risk for developing hypothyroidism.^{86,89,107,111} These studies had variable criteria for classifying hypothyroidism, different means of measuring thyroid volume, varying tiered dosing schemes, and inconsistent follow up periods that make comparisons problematic. The development of hypothyroidism is of particular concern because of its association with the advent or worsening of azotemia, potentially leading reduced survival.⁹⁰ A more objective method of radioiodine dose determination is warranted.

In an attempt to improve outcome of radioiodine treatment, the treatment protocol in our hospital was changed from a fixed dose of 4.5 mCi ^{131}I to a modified fixed dose treatment with cats tiered to receive 3, 3.5, or 4.5 mCi based on evaluation of scintigraphic findings. This presented the unique opportunity to compare the two methods of radioiodine dose determination. The goal of this study was to evaluate an objective, repeatable, and transferable method of radioiodine dose determination for treating hyperthyroidism in cats. This novel method of radioiodine dose determination was based on thyroid size and percent pertechnetate ($^{99\text{m}}\text{TcO}_4^-$) uptake by the thyroid gland using thyroid scintigraphy. The rate of persistent hyperthyroidism and the development of hypothyroidism was determined using this method. Secondly, the treatment outcomes were compared to a standard fixed dose protocol using 4.5 mCi of radioiodine that had been used by our facility previously. We tested the hypothesis that our novel method of radioiodine dose determination, based upon thyroid size and percent pertechnetate uptake, would be superior to a fixed dose technique in achieving euthyroidism in cats treated with radioactive iodine. Specifically, we expected to see a reduced proportion of cats remain hyperthyroid and have a lower occurrence of hypothyroidism than the fixed dose group.

B. Materials and Methods

Animals

Cats referred to the Virginia Maryland College of Veterinary Medicine (VMCVM) for radioiodine treatment of hyperthyroidism were eligible for this study. Cats were determined to be hyperthyroid based upon elevated serum concentrations of T4 with concurrent thyroid stimulating hormone (TSH) below the detection limit of the assay, appropriate clinical signs of hyperthyroidism including one or more of the following (palpable thyroid enlargement, tachycardia, history of weight loss, history of hyperactivity, polyphagia, polyuria/polydipsia,

heart murmur, hypertension), and having characteristics diagnostic of hyperthyroidism (increased thyroid size, increased radionuclide uptake) on nuclear scintigraphy. Exclusion criteria included a documented major illness other than hyperthyroidism or a history of consumption of y/d diet. Cats that had previously been treated with methimazole were included, although treatment had to have been discontinued for a minimum of 2 weeks prior to presentation. The study was approved by the Virginia Tech Institutional Animal Care and Use Committee, VMCVM Veterinary Teaching Hospital Board, and informed consent was obtained from all clients.

Experimental protocol

Cats were initially evaluated with a history, physical examination, CBC, biochemical profile, urinalysis, serum T4 and TSH concentrations, and urine culture. All laboratory testing was performed in the VMCVM clinical pathology laboratory. Serum T4 and TSH concentrations were measured using chemiluminescent enzyme immunoassays^a as previously described.⁹⁸ Cats confirmed to have hyperthyroidism without any of the aforementioned exclusion criteria had thyroid scintigraphy performed. If needed for restraint, cats were sedated with butorphanol (0.3 mg/kg IM), alfaxalone (2-4 mg/kg IM), propofol (6 mg/kg IV) or another drug deemed appropriate by the attending veterinarian. An intravenous catheter was placed in all cats. Following the VMCVM radiation safety protocols, cats were administered 3 mCi $^{99m}\text{TcO}_4^-$ intravenously. Each dose was verified by measuring the activity of the $^{99m}\text{TcO}_4^-$ in the syringe before and after administration using a dose calibrator.^b The syringe was placed on a gamma camera^c before and after dose administration to measure counts per minute for calculation of percent dose uptake. Twenty minutes after $^{99m}\text{TcO}_4^-$ administration, each cat underwent a thyroid scan by being placed on a LEAP collimator^d, first in ventral recumbency, then right and

left lateral recumbency. Thyroid images were obtained using a large-field-of-view scintillation gamma camera.^c

Scintigraphic findings were evaluated for functional status (hyperthyroid, hypothyroid, euthyroid), presence of intrathoracic or extrathoracic ectopic thyroid tissue, and pattern of uptake (unilateral, bilateral, asymmetrical bilateral, or atypical which included cystic or patterns suggestive of malignancy) (Figure 1). For each cat, percent uptake of $^{99m}\text{TcO}_4^-$ in the thyroid gland as well as thyroid gland size were calculated using Nuclear MAC software.^c Regions of interest (ROI) were drawn around each thyroid lobe, both zygomatic salivary glands, and an area adjacent to the thyroid for determining background activity as previously described, by a single individual to avoid any inter-observer variability (Figure 2).^{62,100} A specially designed, inverted-gray-scale lookup-table with a red-color threshold set at 10% of maximum thyroid counts was used to assist in the drawing of the ROIs around the thyroid gland (Figure 3). The imaging software determined the amount of radioactivity within each ROI. The radioactivity within the thyroid was corrected by removing background activity (the radioactivity measured within the thyroid ROI but originating deep and superficial to the gland), radioactive decay, and soft tissue attenuation (absorption of radioactivity by the tissues between the thyroid and the gamma camera). The percent dose uptake was determined by the following formula:

$$\text{Percent Dose Uptake} = \frac{\text{Corrected Counts (cpm) in Thyroid} \times 100}{\text{Radionuclide Dose in cpm}}$$

Thyroid gland size was expressed as a ratio comparing thyroid size to the zygomatic salivary gland using the following formula:

$$\text{TS size ratio: } \frac{\text{Number of Pixels in Thyroid ROI}}{\text{Number of Pixels in Salivary ROI}}$$

Normal percent dose uptake was considered to be 0.2-0.4%.⁶² Cats were grouped as follows for radioiodine dose determination: percent uptake of <5% (mild), 5-10% (moderate), and >10% (severe). Once percent uptake was calculated, thyroid size was stratified by calculating the T:S size ratio with a T:S size ratio of <5 being considered mild, 5-10 moderate, and > 10 severe. The dose of ¹³¹I that was given was first determined by measuring percent uptake. If the thyroid size fell into a higher dosing category than percent uptake, the dose was increased to coincide with the category for size. Dosages were as follows: Low dose (mild) 3.0 mCi, medium dose (moderate) 3.5 mCi, and high dose (severe) 4.5 mCi. Cats in the fixed dose group were all administered 4.5 mCi.

The ¹³¹I was administered subcutaneously. The dose administered was verified using the activity of ¹³¹I in the syringe before and after administration using a dose calibrator.^b Following ¹³¹I dose administration, each cat was placed in radioactive quarantine in the VMCVM. Cats were released from quarantine after the dose of radiation emitted dropped below 2mR/hr at 1 meter and after 4 days in isolation. At home care followed standard radiation safety protocols for 2 weeks.

Animal follow up

Re-evaluation was performed by the referring veterinarian at 1, 3, and 6 months following treatment when blood and urine were collected for serum chemistries, T4, TSH, and urinalysis. All samples were analyzed in the VMCVM clinical pathology laboratory. Treatment outcome was determined by serum T4 concentration 6 months after treatment. Cats were categorized as euthyroid, hypothyroid, or persistently hyperthyroid if serum T4 concentrations were within, below, or above the reference interval, respectively. During the follow-up period, any cat with a low T4 at the 1 or 3-month recheck and development or worsening of azotemia

was administered 0.05 to 0.1 mg of levothyroxine PO daily. These cats were classified as hypothyroid at the 6 month follow-up if they were still receiving supplementation. Cats were excluded if they did not have a 6-month follow-up.

Statistical analysis

Statistical analysis was performed using a commercial statistical software program.^f Normal probability plots showed that age and body weight followed a normal distribution while T4 concentrations, TSH concentrations, percent dose uptake, T:S ratio, and average and maximal intensity were skewed. Chi-square analysis was used to determine the association between dose determination method and outcome. When comparing the novel and fixed dose group characteristics, Chi square was used for categorical data (gender, scintigraphic pattern of uptake, and ectopic tissue). Normally distributed data were analyzed using the t-test (age, weight), while non-parametric data used the Wilcoxon rank sum test (initial T4 concentration, percent dose uptake, T:S size ratio, and average and maximal intensity). Comparison of dosage groups within the novel dose group was performed by use of Chi Square for categorical data (gender, scintigraphic pattern of uptake), one-way analysis of variance (ANOVA) for normally distributed data, and for non-parametric data using the Kruskal-Wallis test (percent dose uptake, T:S size ratio, average and maximal intensity, initial and 6-month T4, and 6-month TSH). Association with outcome for both groups was determined using Chi-square for categorical data (gender, scintigraphic pattern of uptake, ectopic tissue), one-way ANOVA for normally distributed data (age, weight), while non-parametric data was evaluated using the Kruskal-Wallis test (initial T4, percent dose uptake, T:S size ratio, and average and maximal intensity). Logistic regression analysis tested the association of initial T4 concentration, percent dose uptake, T:S size ratio, average and maximal intensity, scintigraphic pattern of uptake, age, weight, and gender, for

hypothyroid outcome vs euthyroid outcome and for hyperthyroid outcome vs euthyroid outcome in both the novel and fixed dose groups. Significance of all tests was set at $p < 0.05$.

C. Results

Study Groups

A total of 94 cats were treated with radioiodine at the VMCVM from November of 2013 through July of 2016, the study period for the novel radioiodine dose group. Seventy-seven cats were enrolled in the study, with 17 being excluded due to prior treatment with radioiodine (3), lymphoma diagnosed at time of treatment (1), consumption of y/d diet (3), and treatment using a different treatment protocol than that defined in this study (10). Of the 77 cats enrolled in the study, 20 were excluded due to incomplete data collection (13), receiving inaccurate radioiodine dose (2), and death during the 6-month follow-up period (5). Cause of death was neoplasia in one cat and unknown in four cats. Of the 57 cats in the novel radioiodine dose group that completed the study, 32 were neutered males and 25 were spayed females. Breeds represented included domestic shorthair (46), domestic longhair (5), Siamese (2), Himalayan (2), Maine Coon (1), and Russian Blue (1). The mean age was 12.0 years (SD +/- 2.) and the mean weight was 4.4 kg (SD +/- 1.3). All cats had an elevated serum T4 concentration above the reference range (16-37.7 nmol/L) and a TSH concentration below the detectable limit of the assay (0.03 ng/mL). The median initial serum T4 concentration was 157 nmol/L with a range of 39.4 to 411 nmol/L.

Twenty-three cats treated with radioiodine at VMCVM between June 2011 and March 2013 meeting the same criteria as the study group, comprised the control group that received a fixed target dose of 4.5 mCi of ^{131}I . These cats have been described in part in another study comparing methods of scintigraphic thyroid quantifications to serum T4 concentrations.

(Bettencourt 2016) Fifty-six cats were initially included in the study, with 33 cats being excluded due to incomplete data collection (30), consumption of y/d diet (2), and euthanasia during the 6-month follow-up period (1). There were 12 neutered males and 11 spayed females. Breeds represented included the domestic shorthair (15) and domestic longhair (8). The mean age was 12.2 years (SD +/- 2.8) and the mean weight was 4.1 kg (SD +/- 0.9). All cats had an elevated serum T4 concentration above the reference range (16-37.7 nmol/L) and a TSH concentration below the detectable limit of the assay (0.03 ng/mL). The median initial serum T4 concentration was 131 nmol/L with a range of 48 to 263 nmol/L. There were no significant differences between the groups in respect to initial T4 concentration, age, weight or gender.

Scintigraphic Characteristics

All cats had increased thyroid uptake of pertechnetate noted on their scintigraphy scans. There were no significant differences in the scintigraphic pattern of disease or the presence of ectopic tissue between the novel dose and fixed dose groups (Table 1). There were three cats with atypical scans in the novel dose group and two in the fixed dose group. In the novel dose group, two of the atypical scans were classified as cystic. One cat in the novel and two in the fixed dose groups were classified as atypical with a concern for malignancy. Eight cats (14%) in the novel dose group had the presence of ectopic tissue compared with one cat (4%) in the fixed dose group.

There were no differences between the groups in percent dose uptake or maximal intensity (Table 2). The T:S size ratio ($p < .0001$) and the average intensity ($p = .028$) in the fixed dose group were greater than in the novel dose group. (Table 2)

Response to treatment

One-month post treatment, data were available for 55/57 cats in the novel dose group

and 22/23 cats in the fixed dose group (Table 3). One cat in the novel treatment group was administered methimazole because of persistent clinical and biochemical hyperthyroidism and was included as hyperthyroid at 6 months, but not included in the 3-month follow-up. Another cat in the novel dose group developed azotemia with concurrent hypothyroidism and was treated with levothyroxine. This cat was included in the hypothyroid group as it continued to receive levothyroxine at the 6-month follow-up, but T4 concentrations were censored at 3 and 6 months.

Three-months post treatment, data were available for 53/57 cats in the novel dose group and 17/23 cats in the fixed dose group (Table 3). Four cats in the novel dose group were determined to be hypothyroid and had worsening of azotemia; they were administered levothyroxine and considered hypothyroid at 6 months. Serum T4 concentrations were not included in data analysis at 6 months as the cats were receiving levothyroxine. In the novel dose group, all 9 cats that were hyperthyroid at 3 months had been hyperthyroid at 1 month. Of the 34 cats euthyroid at 3 months, 2 (6%) were hyperthyroid, 18 (53%) were euthyroid, 12 (35%) were hypothyroid, and data were not available at 1 month in 2 cats. Of the 10 cats hypothyroid at 3 months, (10%) were euthyroid, and (90%) were hypothyroid at 1 month. Three of four cats that did not have data available at 3 months were euthyroid, and one was hyperthyroid at 1 month. In the fixed dose group, the only cat hyperthyroid at 3 months, had been hyperthyroid at 1 month. Of the 11 cats euthyroid at 3 months, 1 (9%) was hyperthyroid, 5 (46%) were euthyroid, 4 (36%) were hypothyroid, and data were not available in 1 (9%). Of the 5 cats hypothyroid at 3 months, all were hypothyroid at 1 month. Six cats that did not have available data at 3 months included 1 cat that was hyperthyroid, 3 that were euthyroid, and 2 that were hypothyroid at 1 month.

At 6 months after treatment, data were available for all cats in both groups (Table 3).

Euthyroidism was present in 33/57 (58%) and 14/23 (61%), hypothyroidism in 15/57 (26%) and 7/23 (30%), and persistent hyperthyroidism in 9/57 (16%) and 2/23 (9%) of cats in the novel and fixed dose groups, respectively. The 5 cats in the novel dose group that were administered levothyroxine because of hypothyroidism and azotemia were classified as hypothyroid since they continued to receive supplementation. The cat previously started on methimazole continued to be hyperthyroid despite medical treatment and was classified as such; these 6 cats were censored for calculation of group median and range data for serum T4 and TSH. In the novel dose group, all 9 cats hyperthyroid at 6 months were hyperthyroid at 1 month, and 8 were hyperthyroid at 3 months; data were not available in 1 cat at 3 months (Figure 4). Of the 33 cats that were euthyroid at 6 months in the novel dose group, 3 (9%) were hyperthyroid, 19 (58%) were euthyroid, and 11 (33%) were hypothyroid at 1 month, and 27 (82%) were euthyroid, 3 (9%) were hypothyroid, and 3 (9%) data were not available at 3 months (Figure 5). Of the 15 cats hypothyroid at 6 months, 3 (20%) were euthyroid, 10 (67%) were hypothyroid, and 2 (13%) did not have data available at one month, and 9 (60%) were euthyroid, and 6 (40%) were hypothyroid at 3 months (Figure 6).

In the fixed dose group, both cats that had persistent hyperthyroidism at 6 months were hyperthyroid at 1 month, and the one cat evaluated at 3 months was hyperthyroid then as well (Figure 7). The 14 cats in the fixed dose group that were euthyroid at 6 months included 1 (7%) that was hyperthyroid, 5 (36%) and 10 (71%) that were euthyroid, and 7 (50%) and 2 (14%) were hypothyroid at 1 and 3 months, respectively (Figure 8). Data were not available at 1 month for 1 cat and at 3 months for 2 cats. The 7 cats in the fixed dose group that were hypothyroid at 6 months included 3 (43%) and 1 (14%) that were euthyroid, and 4 (57%) and 3 (43%) that were hypothyroid at 1 and 3 months, respectively (Figure 9). Data was not available for 3 cats at 3

months.

The three cats with atypical scans in the novel dose group remained persistently hyperthyroid, one each in low, medium, and high dose categories. The two cats in the fixed dose group that had an atypical scan also remained persistently hyperthyroid. Two (25%) of the cats in the novel dose group with ectopic thyroid tissue remained persistently hyperthyroid, and three (38%) became hypothyroid. The one cat in the fixed dose group with ectopic thyroid tissue remained persistently hyperthyroid.

Comparing the novel and fixed dose groups, there was no significant difference in outcome between the two groups at 6 months after treatment (Figure 10).

Novel dosage group characteristics and response to treatment

The low, medium, and high doses of ^{131}I were administered to 29 (51%), 19 (33%), and 9 (16%) of the 57 cats in the novel dose group, respectively (Table 4). The mean dose of radioiodine administered to cats in the low, medium, and high dose groups was 3.07 mCi (SD +/- 0.14), 3.51 mCi (SD +/- 0.07), and 4.46 mCi (SD +/- 0.17), respectively. There was no difference between the dosage groups in regards to age, gender, weight, pattern of disease, 6-month T4, and final outcome. Despite this, the low dose group had the lowest occurrence rate of hyperthyroidism (10%), the medium dose group the lowest occurrence of hypothyroidism (16%), and the medium and high dose groups each had the highest occurrence of hyperthyroidism (21% and (22%) respectively, although these differences were not statistically significant. As a result of study design, as percent dose uptake increased, the dosage level increased ($p < 0.0001$), and as T:S size ratio increased dosage level increased ($p < 0.0001$). As average intensity ($p < 0.0001$) and maximal intensity ($p < 0.0001$) increased, the dosage level increased. The initial serum T4 concentration was directly correlated with the dosage level ($p < 0.0001$) (Figure 11).

There was no association of outcome in the novel dosage groups with respect to age, gender, pattern of disease, if ectopic disease was present, T:S size ratio, average and maximal intensity, or % dose uptake (Table 5). There were outcomes that approached significance with initial T4 serum concentration, as cats with the highest initial T4 concentrations tended to remain persistently hyperthyroid and those with the lowest initial T4 concentrations tended to become hypothyroid ($p = 0.0506$)(Figure 12).

Fixed dose group characteristics and response to treatment

The mean dose of radioiodine in the fixed dose group was 4.45 mCi (SD +/- 0.09). There was no association with outcome in the fixed dose group with respect to gender, age, initial mean serum T4 concentration, mean percent dose uptake, mean average intensity or mean maximal intensity, or presence of ectopic tissue (Table 6). There was an association with T:S size ratio as larger thyroid size had an increased rate of persistent hyperthyroidism ($p = 0.0211$) in the fixed dose group (Figure 13). Also the pattern of disease was significant ($p = 0.0055$), as an 8/9 cats with an asymmetrical bilateral pattern of disease became euthyroid and 2/2 cats with an atypical pattern of disease in the fixed dose group remained persistently hyperthyroid. No associations were found in either group when combining symmetrical bilateral disease with asymmetrical bilateral disease and comparing to unilateral disease for outcomes.

D. Discussion

This study failed to detect differences in outcome between our novel method of dosing based on thyroid scintigraphy and administration of a fixed dose of radioiodine in hyperthyroid cats. Our hypothesis that radioiodine dosing utilizing percent dose uptake and thyroid size as determined by scintigraphy would increase euthyroidism while decreasing the development of hypothyroidism post radioiodine treatment was not supported.

It was noted in the novel dose group that the highest T4 concentrations were associated with persistent hyperthyroidism and the lowest T4 concentrations with hypothyroidism. The high prevalence of hypothyroidism in the novel dose group was likely the result of using insufficient dose tiers, considering that 10 (67%) of the 15 cats that became hypothyroid were in the low dose group. Other studies have used protocols ranging anywhere from 2.0 – 24 mCi.^{86,89} Recently, it was shown that a ¹³¹I dose of 2mCi was highly efficacious in treating cats with mild hyperthyroidism, defined as serum T4 concentration <167 nmol/L (3.24 times the upper limit of reference range).¹¹⁵ In the present study, 16 cats in the novel dose group receiving a low dose of radioiodine had a pre-treatment serum T4 concentration that was <3.24 times the upper limit of the reference range. Nine of these 16 cats became hypothyroid, indicating that a low dose of 3mCi was excessive for the most mildly affected cats. The increased prevalence of higher T4 concentrations being associated with persistent hyperthyroidism is consistent with other studies. However, it is likely not the sole factor as some cats with markedly elevated T4 concentrations in other studies have also become euthyroid or hypothyroid.^{81,84,86} It is likely that markedly increased serum T4 concentrations are often but not always associated with increased thyroid size. Cats with markedly increased T4 concentrations and smaller thyroid size have been shown to become hypothyroid, suggesting thyroid size needs to be assessed along with serum T4.⁸¹

It was noted in the fixed dose group that the largest thyroid sizes were associated with persistent hyperthyroidism suggesting that a 4.5 mCi dose is inadequate for more severe disease, further supporting the need for a >4.5 mCi dose for more severely affected cats. This is consistent with a previous study in which 7 cats with greatly enlarged thyroid glands treated with a dose of 4.0 mCi – 6.0 mCi remained persistently hyperthyroid.⁸⁶

It was noted that 8/9 cats with asymmetrical bilateral disease in the fixed dose group had

a euthyroid outcome, suggesting that this pattern of disease has a protective effect against the development of hypothyroidism, because suppressed thyroid tissue was spared damage and regained normal function. The fixed dose method may be more appropriate for moderately affected cats with this pattern of disease. Unilateral disease should also have a protective effect, by a similar mechanism. Though the present study did not demonstrate a protective benefit with unilateral disease, perhaps the small sample size, 4/23 (17%) unilateral vs 8/23 (35%) in the asymmetrical bilateral group was not large enough to demonstrate a difference. This is consistent with what has been reported; unilateral disease has been shown to be less common than bilateral disease of any form.¹⁹ Most of the cats in the present study that had a euthyroid outcome in both groups had asymmetrical bilateral or unilateral disease.

While numerous studies have evaluated the fixed dose and modified fixed dose methods of radioiodine dose determination, the authors are not aware of studies directly comparing these techniques. In addition, the novel method used in this study is unique in its use of percent dose uptake and thyroid size based on scintigraphy as the sole parameters to determine radioiodine dose. Other studies investigating modified fixed dose method techniques have based dosing on criteria including T4 concentration, clinical signs, and thyroid size, but have not used well-described and repeatable methods of dose calculation.^{85,86,89,111}

The proportion of cats developing hypothyroidism with the fixed dose method in our study was 30%, which is substantially higher than other studies that have shown rates as little as 5-8%.^{83,84,87} The proportion of cats developing hypothyroidism with our novel dose method was also high. Prevalence of hypothyroidism in some studies was as low as 9-11%^{85,86} though other studies had outcomes (hypothyroidism in 24-30%) comparable to the present study.^{89,111} Explanations for these differences include variations in the definition of hypothyroidism

(clinical vs biochemical), the presence of concurrent illness, varying time of follow-up, varying dosage ranges, and differences in the severity and pattern of disease. Cats with bilateral disease have been shown to have a higher rate of hypothyroidism, from presumed destruction of normal thyroid tissue.^{89,111} The novel and fixed dosage groups in the present study had a low percentage of symmetrical bilateral disease at 11% and 13% respectively, with the majority having asymmetrical bilateral disease which might be suspected to result in less destruction of atrophied thyroid tissue, decreasing the development of hypothyroidism. Many previous studies have not differentiated between asymmetrical bilateral and symmetric bilateral disease. When combining asymmetrical bilateral disease and symmetrical bilateral disease together and comparing them to unilateral disease for outcome, no difference was noted in either group. Since asymmetrical bilateral disease alone was associated with a euthyroid outcome in the fixed dose method, this does suggest a protective benefit of this pattern of disease.

Hyperthyroidism persisted after treatment in 16% of cats in the novel dose group, which is higher than the 1.5-12% noted in studies using different modified fixed dose methods.^{85,86,108,111} Cats with more severe disease as indicated by significantly higher T4 concentrations and higher volumes of hyper-functioning thyroid tissue have been shown to have a higher rate of persistent hyperthyroidism.¹⁰⁸ Additionally, carcinomas have been shown to respond poorly to standard doses of radioiodine.⁹⁴ All three cats in our novel group and the two cats in the fixed dose group that had an atypical scintigraphic pattern of disease remained hyperthyroid suggesting an atypical pattern to be associated with persistent hyperthyroidism which may have contributed to the increased frequency of persistent hyperthyroidism seen in our study. The presence of ectopic tissue has also been shown to affect outcome in previous studies.¹¹¹ In the present study, there was no association with the presence of ectopic tissue in

either dosage group with outcome, though ectopic tissue was not quantified as in the Volckaert study.

As the lowest dosage group had the lowest occurrence of persistent hyperthyroidism, and the medium dosage group the lowest occurrence of hypothyroidism, the parameters used to set the division between the low and medium dose appears to be valid. This may not be the case for the parameters determining the division between the medium and higher dosage groups, as 21% of the cats in the medium dosage group remained hyperthyroid after treatment, suggesting a higher dose should have been used in at least some of these cats. These cats had varying T4 concentrations and patterns of disease that lacked definitive parameters suggesting a higher dose. Perhaps, other parameters such as clinical signs and duration of disease should be used to attempt to determine an ideal dose.

Cats were followed 6 months in our study to determine post-treatment serum T4 concentration which has been shown to be an adequate amount of time to determine thyroid status.⁸⁶ Hypothyroidism noted at one month after radioiodine administration was common, but resolved in many cases. In the novel dose group 38% of cats were hypothyroid at one month compared to 26% at 6 months, and in the fixed dose group 50% of cats were hypothyroid at one month compared to 30% at 6 months (Figure 14). This is likely due to atrophied thyroid tissue regaining function over time after treatment. Ten (19%) cats in the novel dose group were hypothyroid at 3 months compared to 15(26%) at 6 months. This stresses the importance of following cats for a minimum of 6 months. Two cats at 3 months had values borderline between euthyroid and hypothyroid, so it is possible normal fluctuation in thyroid hormone or non-thyroidal illness may have affected results in these two cats. All but one cat that was euthyroid at 3 months and hypothyroid at 6 months, had an elevated TSH at 3 months suggesting TSH may

be used to predict the development of hypothyroidism. Two cats were hyperthyroid at one month and went on to become euthyroid at three and 6 months. This suggests a delay in thyroid tissue destruction. One cat had an initial T4 concentration (>250 nmol/L) and one cat had a borderline T4 concentration at one month, suggesting severe disease and normal T4 fluctuation may have been contributing factors to this result. Three of the cats hypothyroid at 3 months became euthyroid, suggesting a delay in regaining thyroid function. This is important because some may be inclined to supplement levothyroxine if biochemically hypothyroid at 3 months. These results indicate supplementation may not be necessary unless the cat is clinically hypothyroid or showing signs of worsening azotemia.

The scintigraphic measurements of the novel compared to the fixed dose group showed a larger thyroid size and higher average scintigraphic intensity of uptake in the fixed dose group. This would suggest more chronic disease in the fixed dose group, requiring a higher dose of radioiodine to achieve euthyroidism. A recent study suggests disease progresses with increased duration.¹⁹ Since disease duration was not recorded in the present study, this is purely speculative. Another recent study suggests that increased thyroid volume (by number of foci) is associated with a hypothyroid outcome.¹¹¹ Since less than 25% of the cats in the fixed dose group had symmetric bilateral disease and only one had ectopic foci, this should have made hypothyroidism less likely. Three of the eight cats (37.5%) in the novel dose group with ectopic tissue were hypothyroid after treatment. This is slightly higher than the overall prevalence of hypothyroidism in this group. If the cats with ectopic tissue were removed from the analysis only 23.5% of cats were hypothyroid which further supports the Volckaert study.

It was noted that as average and maximal intensity increased, dosage increased, suggesting that intensity increased with disease severity. A direct correlation was also seen with

initial serum T4 concentration as the dose correspondingly increased with T4 concentration. T4 concentration was directly associated with percent dose uptake which is consistent with prior studies identifying a strong correlation between percent dose uptake and T4 concentration.^{98,116} Despite these correlations, T4 concentration approached significance with outcome while percent dose uptake and intensity did not. This suggests that serum T4 concentration may be substituted for percent dose uptake and intensity in dose calculations.

One of the limitations of this study was the small size of the study group. Statistical analysis resulted in a number of variables that neared significance, and a larger sample size may have altered results. Many cats were excluded because they were lost to follow-up for unknown reasons. It is possible the population studied does not adequately represent the general population of cats undergoing radioiodine treatment, as many cats were referred for radioiodine treatment due to difficulty in controlling their hyperthyroidism, thereby suggesting more severe disease. Another limitation is that the duration of hyperthyroidism was not recorded. Duration of disease has been suggested to increase disease severity, and possibly response to treatment.¹⁹

Treatment outcome in the present study was classified on the basis of T4 concentration alone. Although not presented, many cats in the present study that had a normal T4 had concurrent elevation of TSH. These cats were classified as euthyroid. It has been shown that a normal T4 with an undetectable TSH is associated with the development of hyperthyroidism,⁵⁴ but it is unclear what an elevated TSH signifies. Additionally, some cats had T4 concentrations that were borderline to being classified into a different outcome category. Normal fluctuations in serum thyroid hormone have been shown to occur in hyperthyroid cats.⁵¹ These fluctuations may have affected how cats were classified. Establishing euthyroidism from subclinical hypothyroidism is difficult and it is unclear if TSH is beneficial. Cats were not evaluated with

radiographs or ultrasound that may have potentially revealed underlying concurrent diseases that may have altered either initial or follow-up T4 concentrations. Cats were not consistently examined to further screen for the development of disease during the follow-up period. This may have skewed results such that some euthyroid sick cats were deemed hypothyroid or some persistently hyperthyroid cats were considered euthyroid. Also, the distinction between biochemical and clinical hypothyroidism was not made, so significance of a subnormal T4 in these cats is unclear.

Extending the follow-up period beyond 6 months, would have also been ideal to monitor for clinical manifestations of hypothyroidism that developed later. As previously mentioned, 26% of cats were biochemically hypothyroid at 6 months compared to 19% at 3 months in the novel dose group. Perhaps extending the period further may have detected more cats to have become hypothyroid. This may have also allowed comparison of survival times in each group.

The ROI was determined consistently by one individual in this study for consistency purposes. As there is some subjectivity in the drawing of the ROI, it is possible, another individual may have drawn a different ROI, leading to some different classification of dosing category.

Chapter 3: Conclusions and Further Research

In summary, our novel method of radioiodine dosing based upon percent dose uptake and thyroid size did not result in improved overall outcomes compared to a standard fixed dose method. In fact rates of post treatment persistent hyperthyroidism and the development of hypothyroidism exceeded those found in other studies of modified fixed dosing methods.^{86,89,107,111} Our study was unique in that it made direct comparisons within a population at one institution, rather than comparing results to other studies. It is likely populations differed among other studies which is likely to have affected results.

Despite these results, associations were found that might help future studies looking to define a better method of radioiodine dose determination. A correlation between thyroid size, percent dose uptake, and uptake intensity was found with dosage level in the novel dose group. Initial serum T4 concentration was approached significance with outcome. This suggests that serum T4 concentration may be used as a predictor for dosing radioiodine.

In our fixed dosage group, asymmetrical bilateral disease was associated with a higher number of cats achieving euthyroidism, suggesting a protective effect. Unilateral disease should offer this same protective effect, but likely due to sample size and the prevalence of unilateral disease, was not demonstrated in this study. Investigation of a fixed dose protocol specifically in asymmetrical bilateral and unilateral disease may be of benefit, as cats with these patterns of disease may be less susceptible to the development of hypothyroidism.

Cats with more severe disease (and likely increased disease duration) with higher T4 concentrations had a higher percentage of persistent hyperthyroidism despite the majority being treated with the high dose in our novel protocol. This suggests a higher dosage is needed in

these cats. At the same time, many cats with mild disease and lower T4 concentrations treated with the lower dose tended towards the development of hypothyroidism suggesting a lower dose is needed in these cats. The investigation of a modified fixed dose method of radioiodine dosing utilizing current dosages along with a higher and lower dose may help increase the rate of euthyroidism after treatment. Perhaps including serum T4 concentration which was associated with outcomes in the novel group, along with thyroid size and pattern of disease which was associated with outcome in the fixed dose group in a larger sample size with an expanded tiered dosing method may be of benefit.

Many additional variables may affect outcomes of radioiodine treatment. Continued investigations into these variables will hopefully uncover a consistent and successful method of radioiodine dose determination.

FOOTNOTES

- a. Immulite Canine Total T4 and Immulite Canine TSH, Immulite 1000; Siemens Healthcare Diagnostics, Tarrytown, NY
- b. Atomlab 500, Biodex Medical Systems, Shirley NY
- c. Omega 500, Technicare Inc., Solon OH
- d. Mirage, Segami Corporation, Columbia MD
- e. Scientific Imaging, PO Box 3691, Crested Butte CO 81224.
- f. SAS Version 9.4, Cary NC

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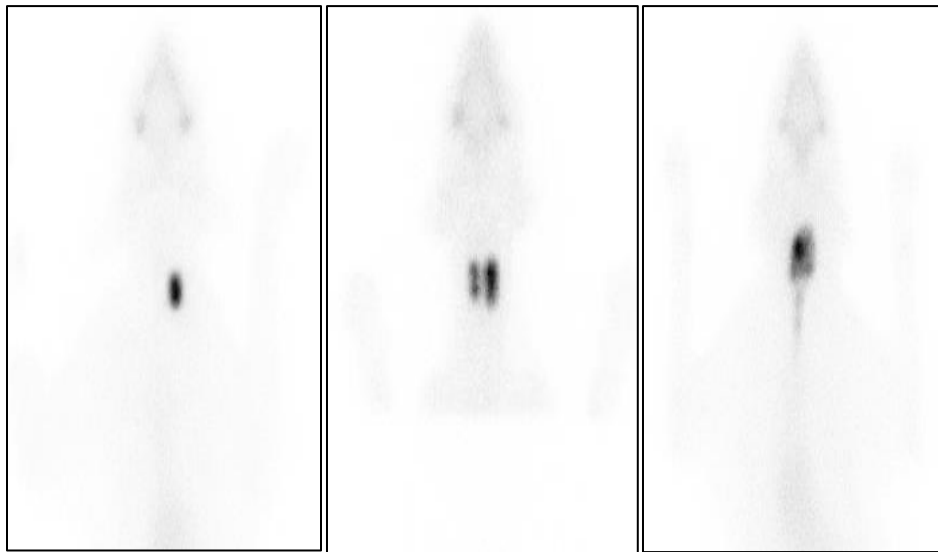
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APPENDIX A: FIGURES

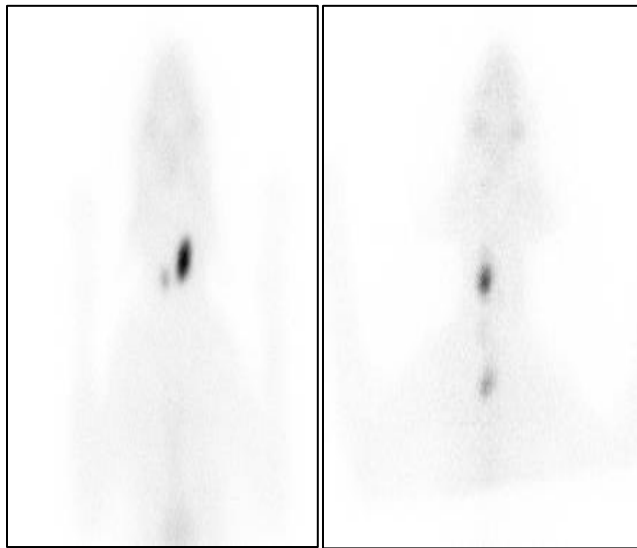
Figure 1. Scintigraphic Patterns of Uptake



Unilateral

Bilateral

Atypical



Asymmetrical bilateral

Ectopic

Figure 2. ROI for percent dose uptake and T:S size ratio calculation

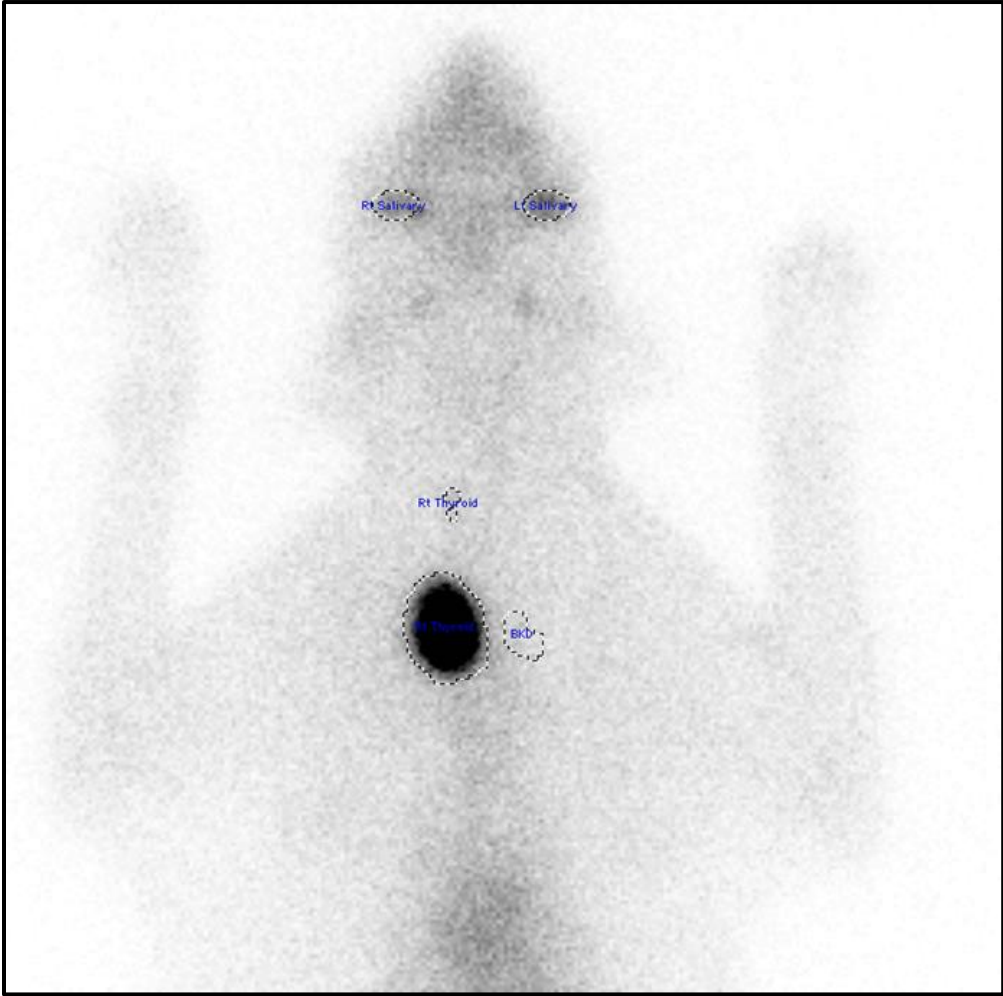


Figure 3. Standard vs. 10% threshold thyroid scintigraphy image

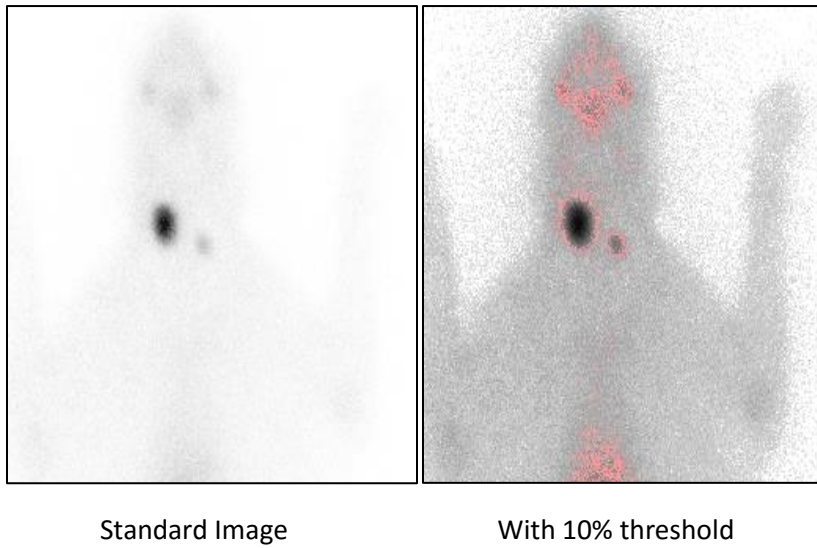
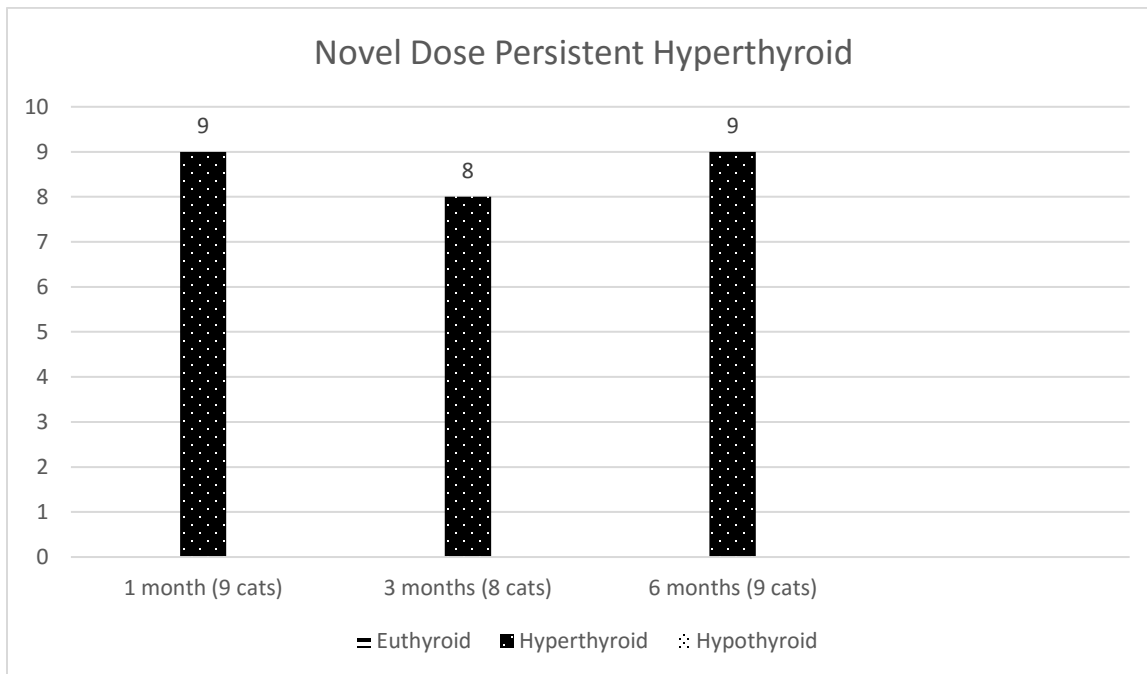
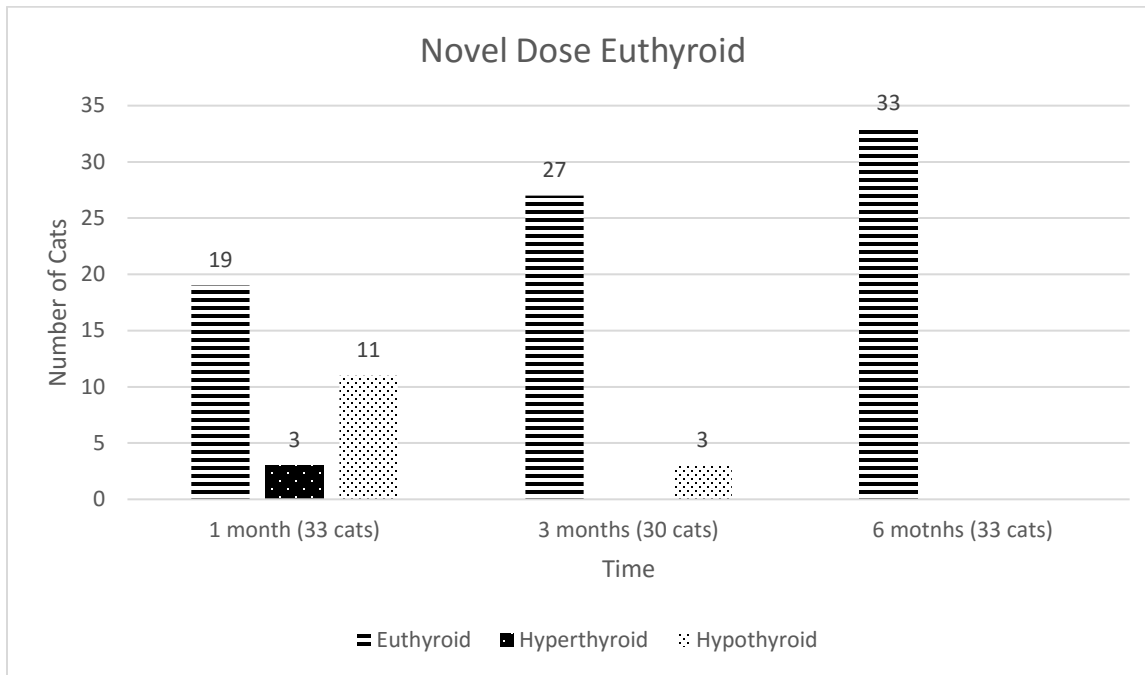


Figure 4. Cats with a hyperthyroid outcome receiving novel radioiodine dose: 1, 3, and 6 month responses to treatment



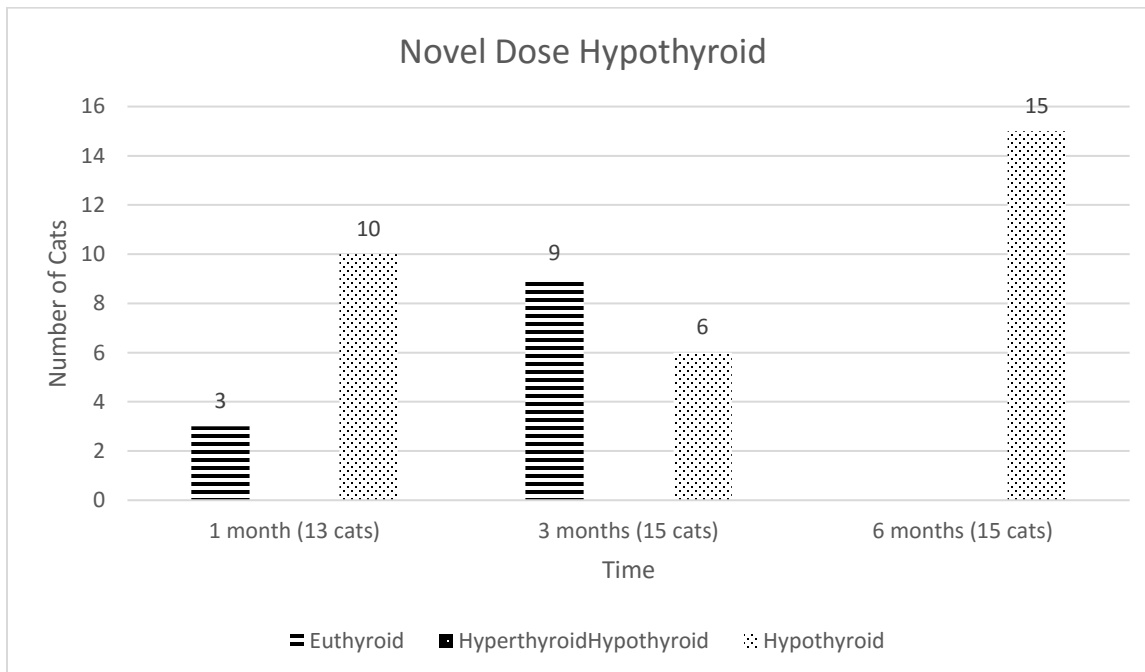
*Data not available for 1 cat at 3 months

Figure 5. Cats with a euthyroid outcome receiving novel radioiodine dose: 1, 3, and 6 month responses to treatment



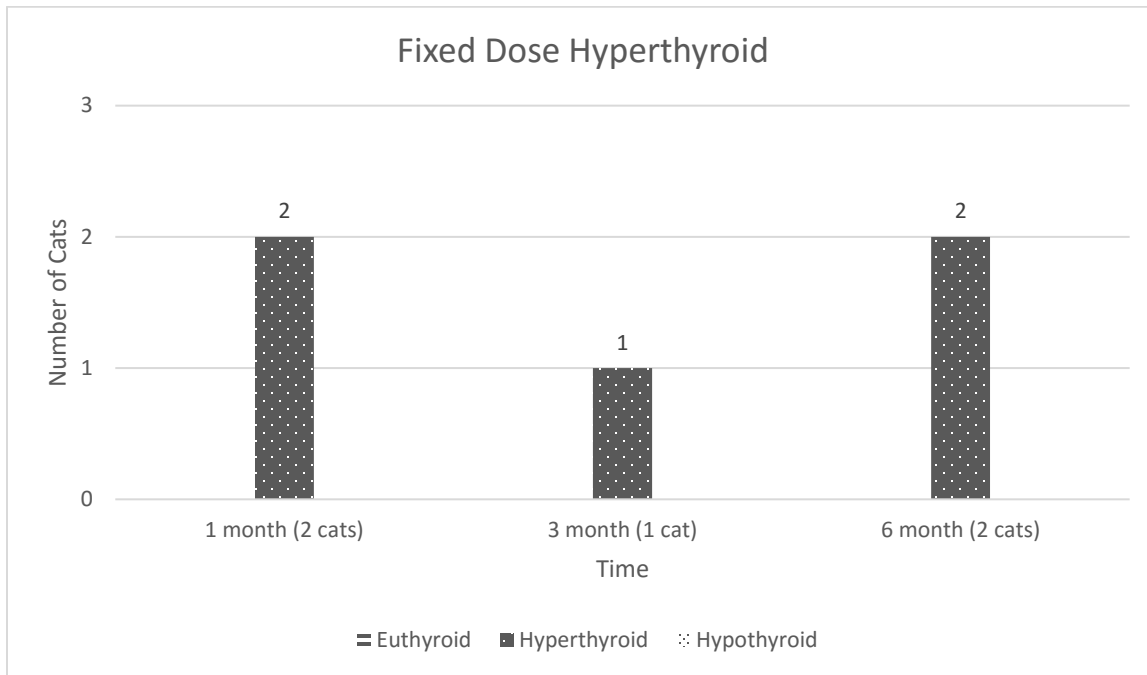
*Data not available for 3 cats at 3 months

Figure 6. Cats with a hypothyroid outcome receiving a novel radioiodine dose: 1, 3, and 6 month responses to treatment



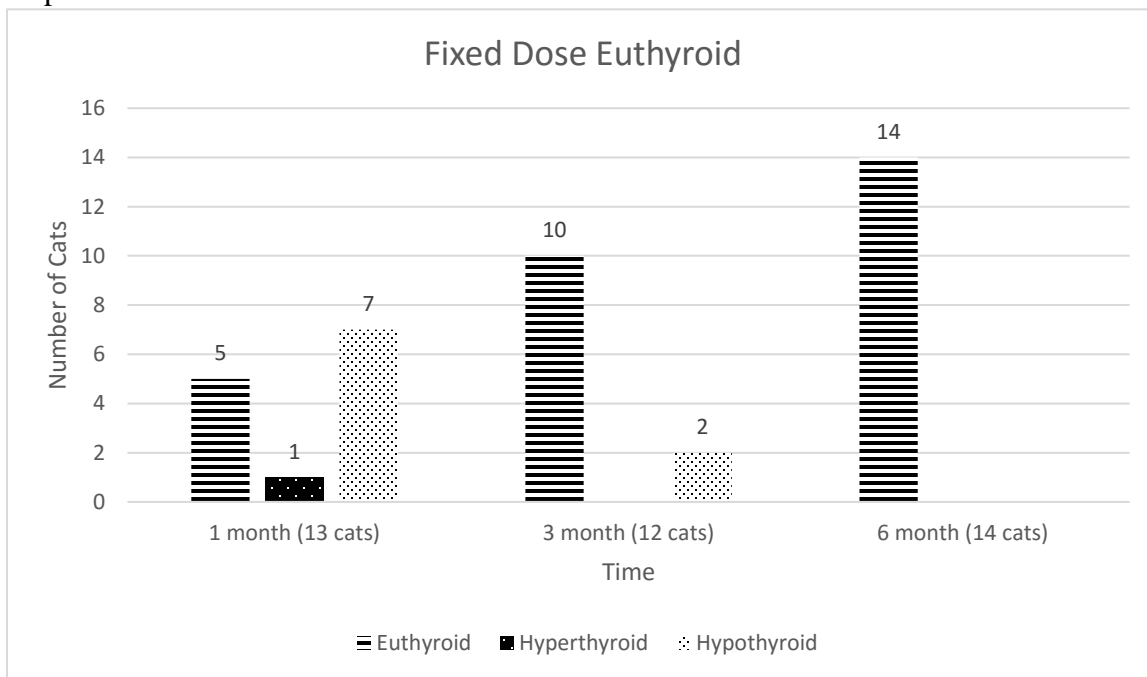
*Data not available for 2 cats at 1 month

Figure 7. Cats with a hyperthyroid outcome receiving a fixed dose of radioiodine: 1, 3, and 6 month responses to treatment



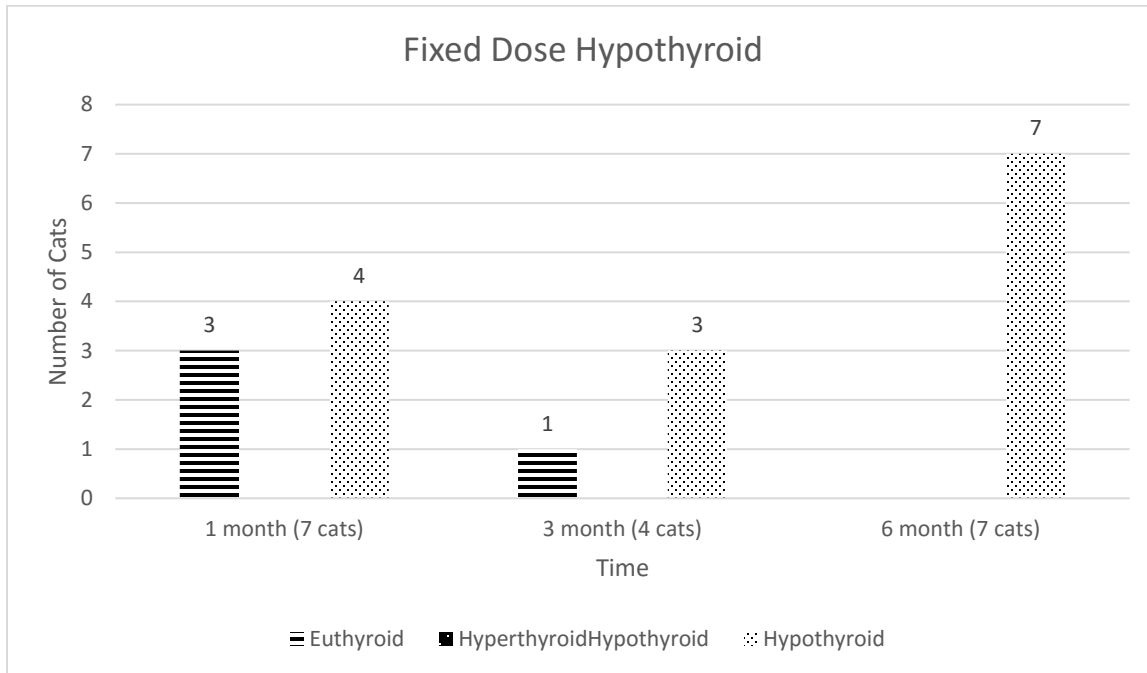
*Data not available for 1 cat at 3 months

Figure 8. Cats with a euthyroid outcome receiving a fixed dose of radioiodine: 1, 3, and 6 month responses to treatment



*Data not available for 1 cat at 1 month and 2 cats at 3 months

Figure 9: Cats with a hypothyroid outcome receiving a fixed dose of radioiodine: 1, 3, and 6 month responses to treatment



*Data not available for 3 cats at 3 months

Figure 10. Comparison of 6-month Outcomes: Novel and Fixed Dose Radioiodine Groups of Cats.

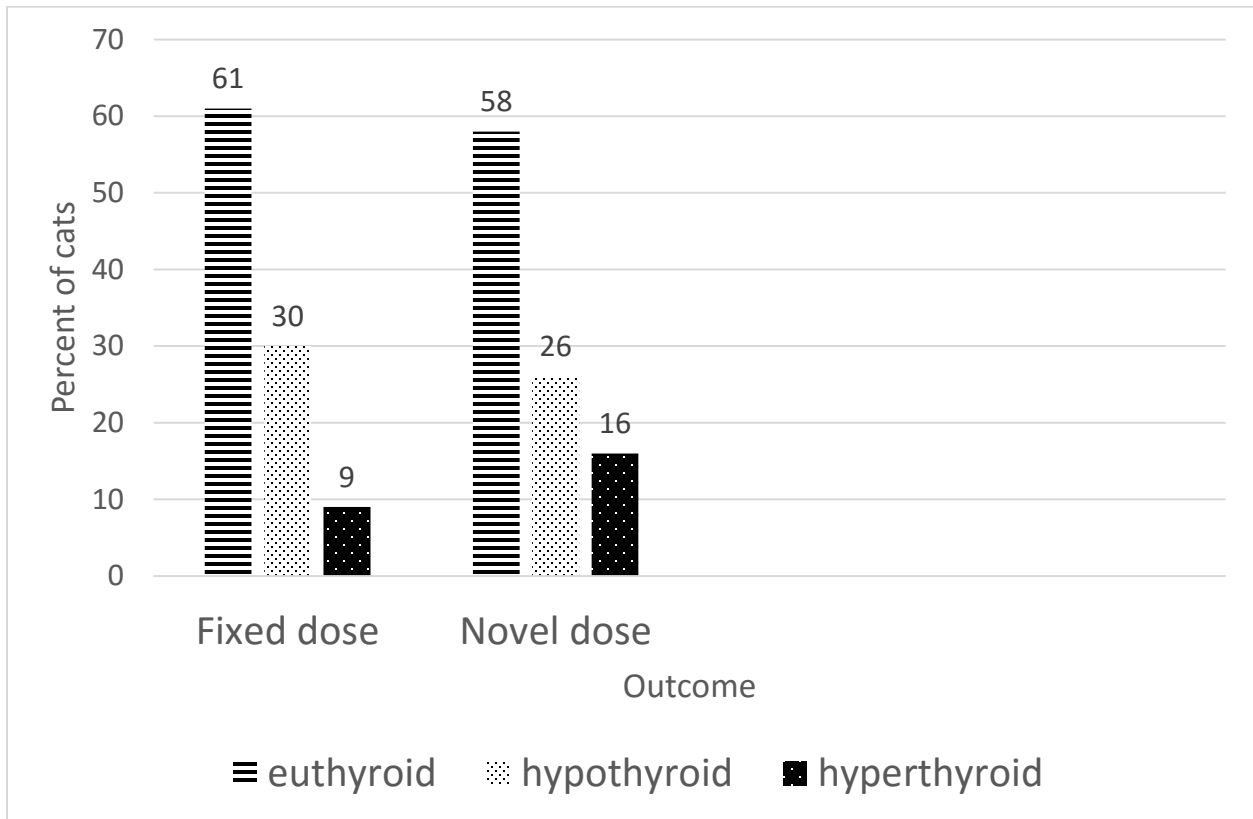


Figure 11. Initial T4 concentrations in relation to 3.0, 3.5 mCi, and 4.5 mCi novel dose group

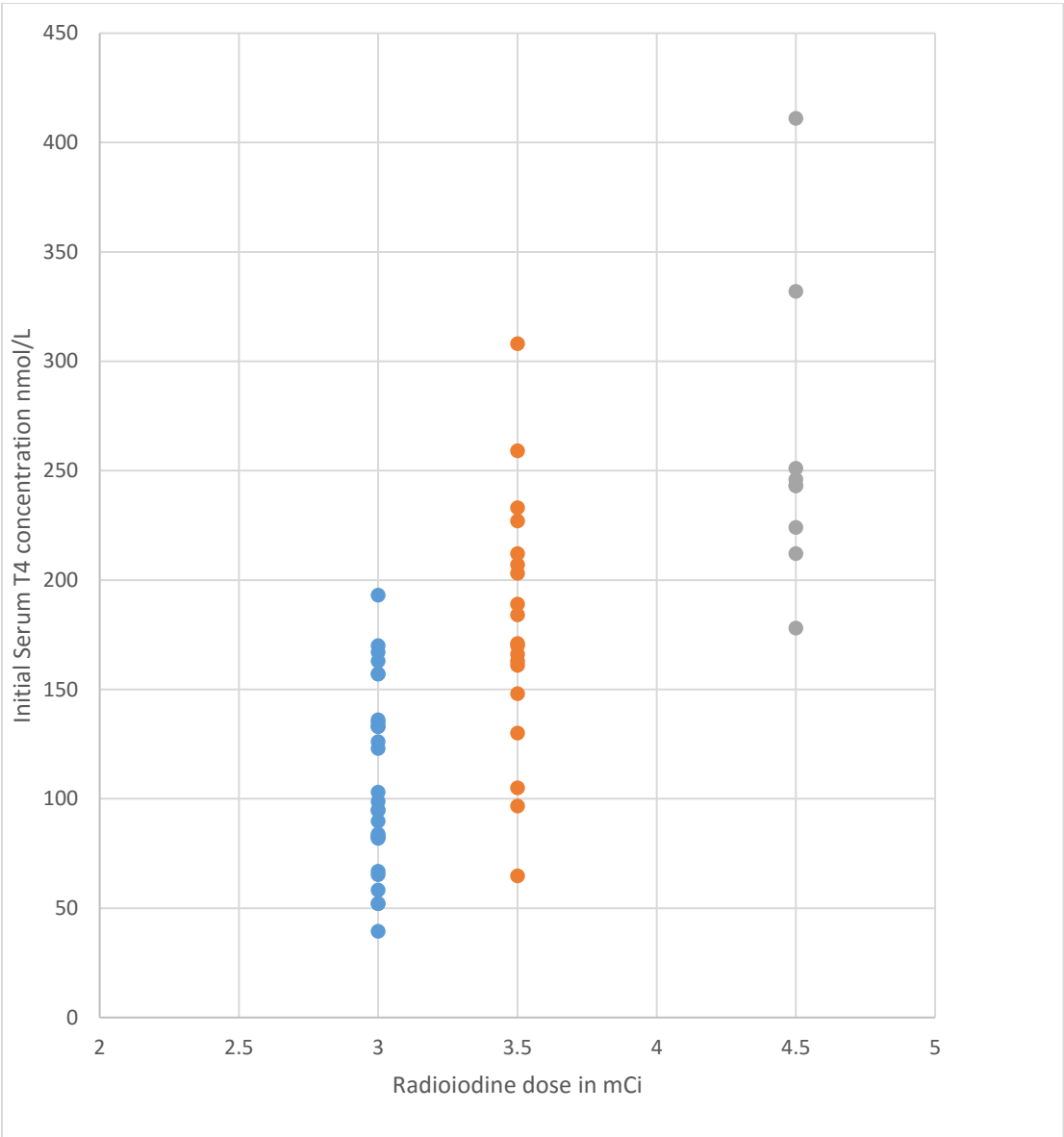
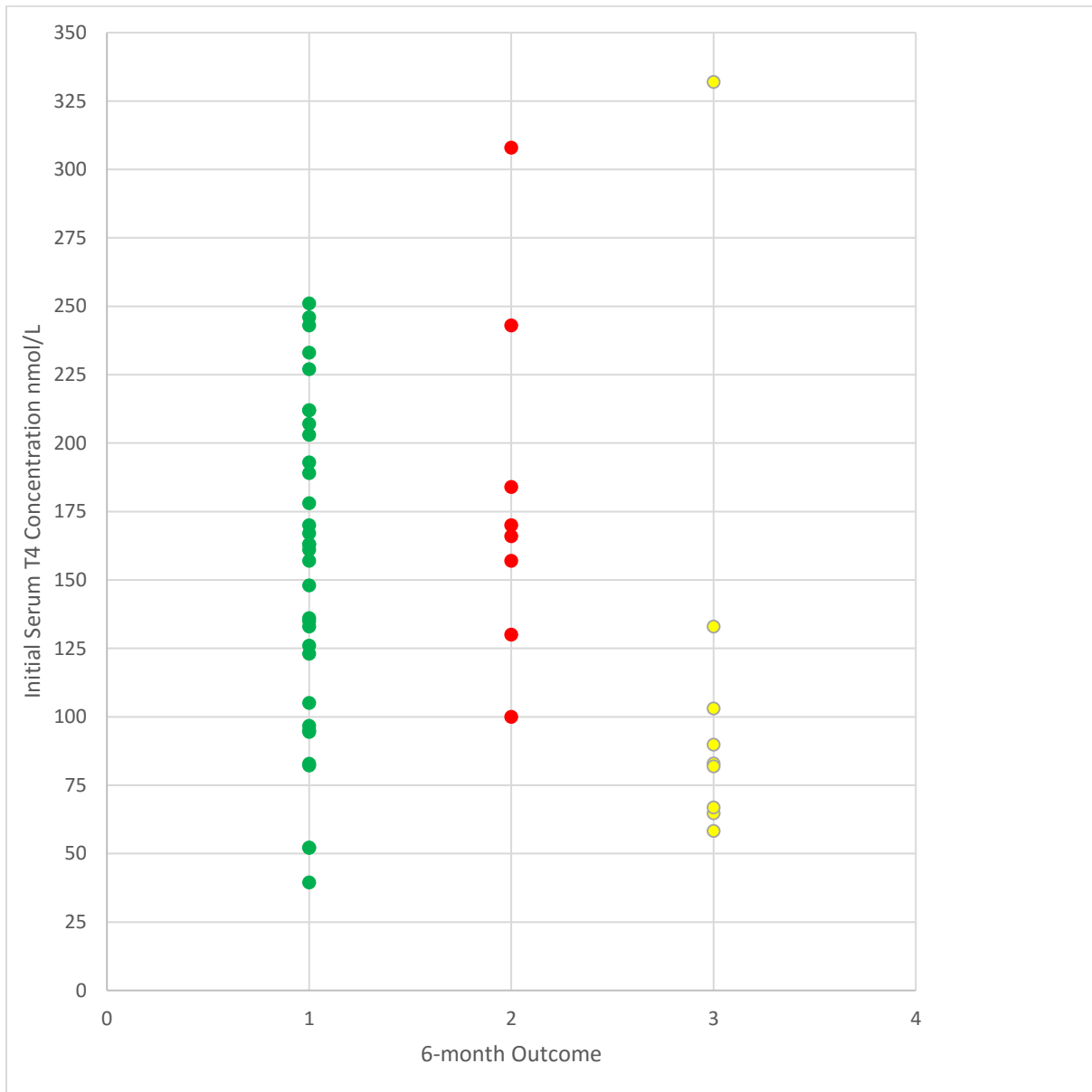
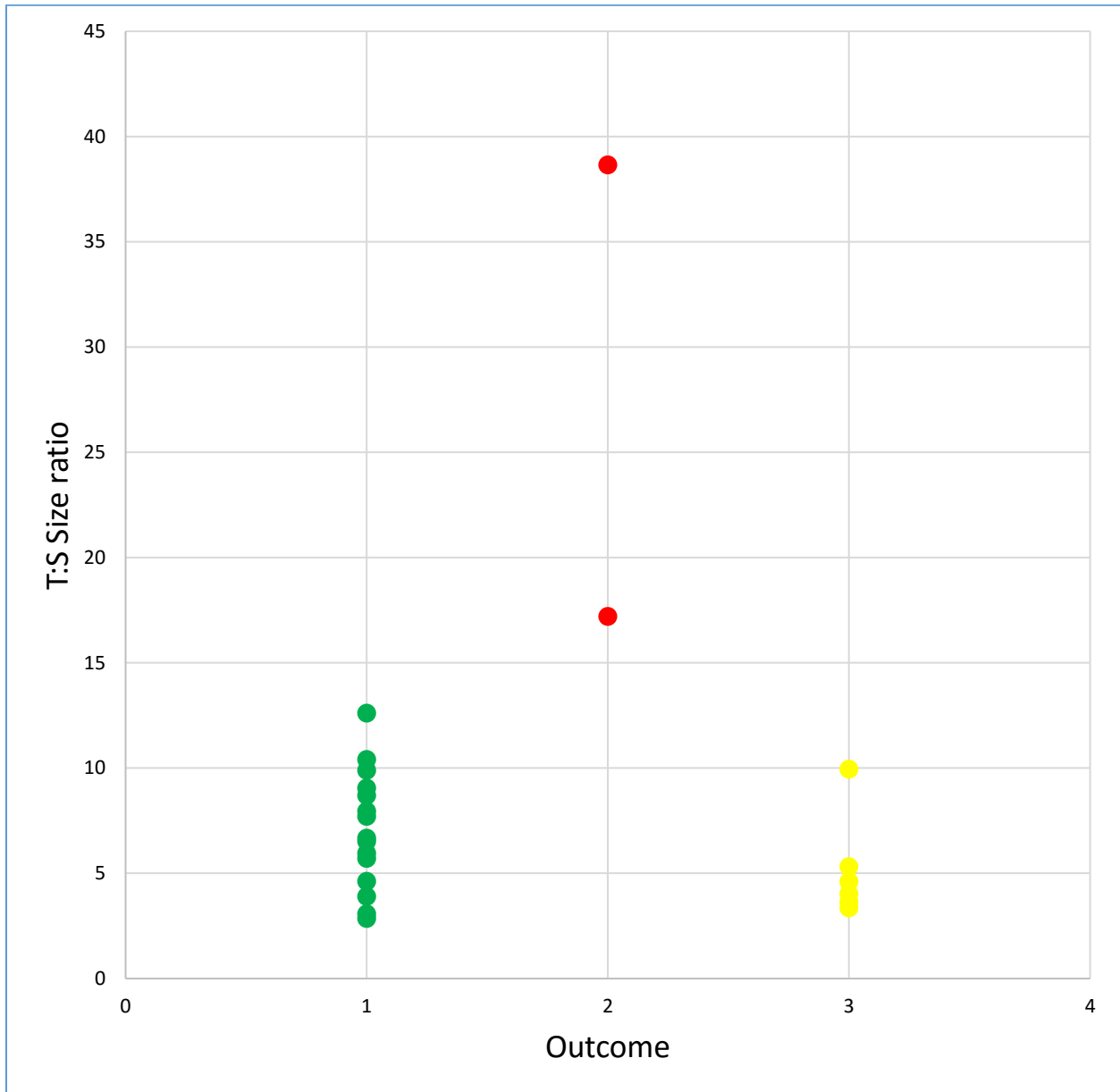


Figure 12. Initial T4 concentration in relation to euthyroid, persistently hyperthyroid, and hypothyroid outcome in novel dose group



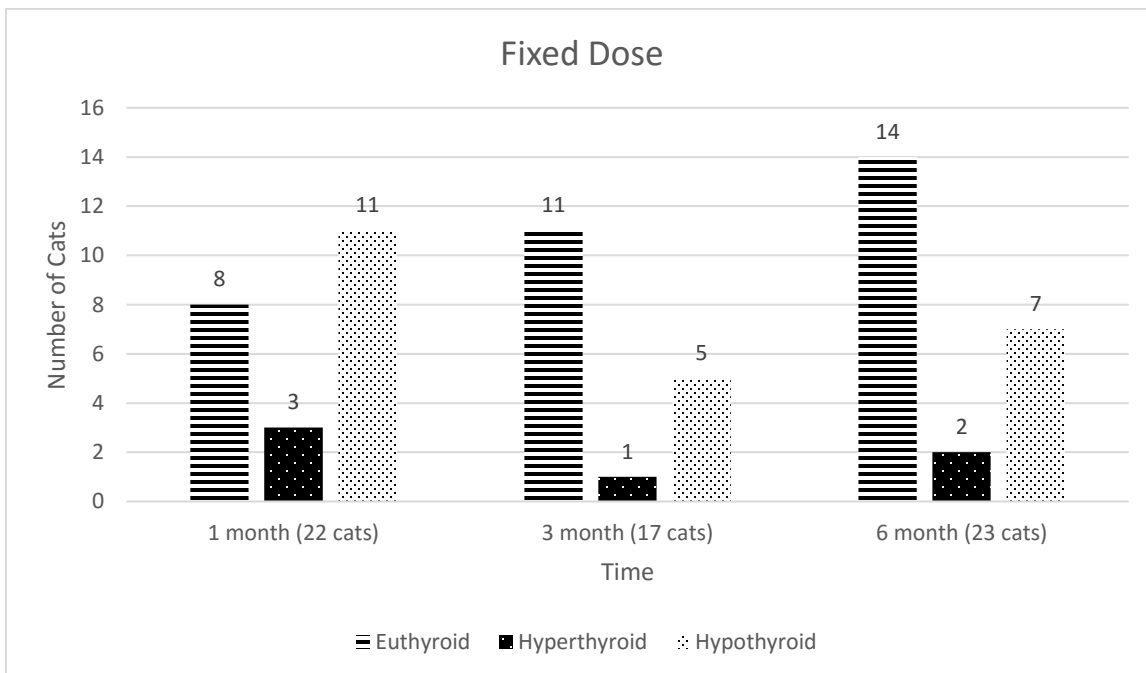
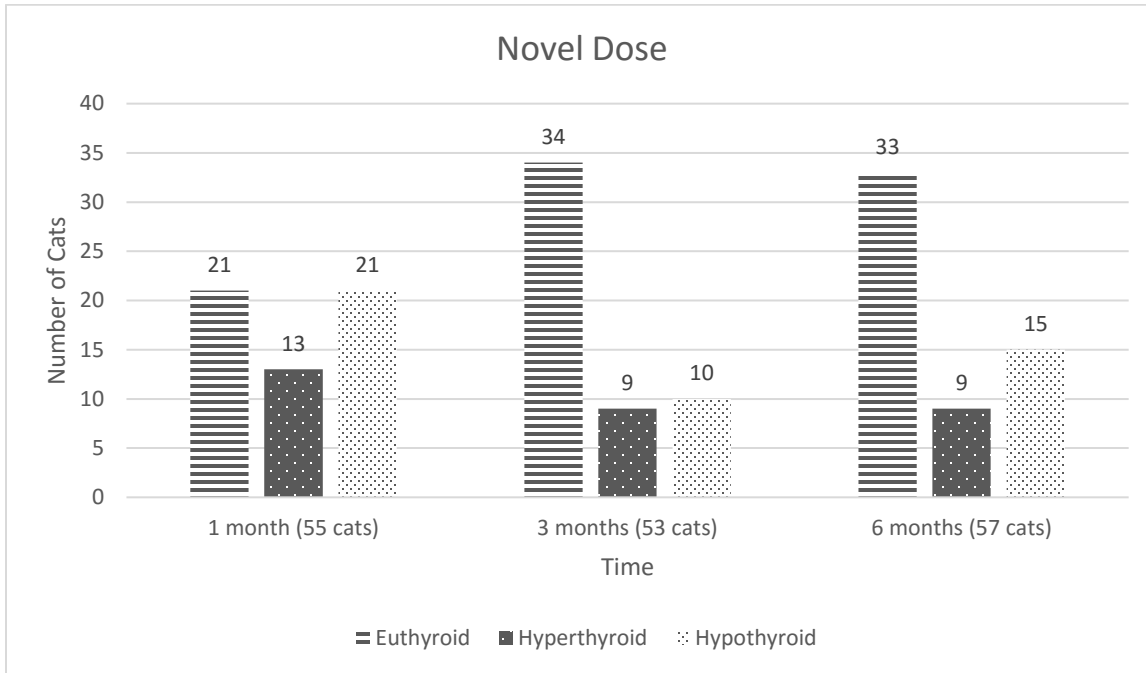
1 =Euthyroid, 2= Hyperthyroid, 3=Hypothyroid

Figure 13. Fixed dose thyroid size in relation to euthyroid, persistently hyperthyroid, and hypothyroid outcome



1=Euthyroid, 2=Hyperthyroid, 3=Hypothyroid

Figure 14. Overall Responses to Treatment for Novel and Fixed Dose Groups at 1, 3, and 6 months



APPENDIX B: TABLES

Table 1. Scintigraphic characteristics: Pattern of Disease and Ectopic Tissue Fixed and Novel Group Comparisons

Patten of Disease	Novel dose (number/%)	Fixed dose (number/%)	<i>p</i> - value*
Unilateral	21 (37%)	9 (39%)	1.000
Bilateral	6 (11%)	3 (13%)	0.7116
Asymmetrical bilateral	27 (47%)	9 (39%)	0.6213
Atypical	3 (5%)	2 (9%)	0.6221
Ectopic thyroid tissue	8 (14%)	1 (4%)	0.4344

*Using Fisher's exact test

Table 2. Scintigraphic characteristics: % Dose uptake, Average and Maximal Intensity Comparison of Fixed and Novel Dose Groups

Characteristic	Novel dose (median/range)	Fixed dose median/range	<i>p</i> value*
% dose uptake	3.8 (0.3-34.5)	4.4 (0.5-14.2)	<i>p</i> = 0.9958
T:S size ratio	3.9:1 (1.4-17.2)	6.5:1 (2.9-38.7)	<i>p</i> < 0.0001
Average intensity	2.9 (0.8-23.3)	4.4 (0.9-16.8)	<i>p</i> = 0.0277
Maximal intensity	4.3 (0.8-23.3)	5.6 (1.2-20.2)	<i>p</i> = 0.1891

*Using Wilcoxon Two-Sample Test

Table 3. Response to treatment for novel and fixed doses of radioiodine at 1, 3, and 6 months

	Follow up	1 Mon			3 Mon			6 mon		
	Outcome	Number (%)	T4 *	TSH**	Number (%)	T4*	TSH**	Number (%)	T4*	TSH**
N O V E L	Euthyroid	21 (38%)	20.2	0.17	34	22.1	0.10	33	22.8	0.13
			16.1-29.7	<0.03-0.64	(64%)	16.1-36.9	<0.03-1.41	(58%)	16.2-35.9	<0.03-0.88
	Hyperthyroid	13 (24%)	78.9	<0.03	9	77.2	<0.03	9	61.2	<0.03
			43.9-265	<0.03	(17%)	39-88	<0.03	(16%)	40.5-102	<0.03
	Hypothyroid	21 (38%)	8.8	0.10	10	10.4	0.88	15	12.8	2.09
			<6.4-15.7	<0.03-1.79	(19%)	<6.4-15.8	0.12->12.0	(26%)	<6.4-14.3	0.09->12.0
F I X E D	Euthyroid	8	19.3	0.13	11	22.5	0.10	14	22.6	0.16
		(36%)	16.9-28.1	<0.03-0.58	(65%)	16.9-33.8	<0.03-1.12	(61%)	17-33.7	0.06-2.97
	Hyperthyroid	3	80.3	<0.03	1	282	<0.03	2	126.6	<0.03
		(14%)	52.8-400	<0.03	(6%)	282	<0.03	(9%)	60.2-193	<0.03
	Hypothyroid	11 (50%)	10.7	0.12	5	8.9	0.80	7	14.5	0.74
			<6.4-14.3	<0.03-2.07	(29%)	<6.4-13.8	0.63-4.33	(30%)	6.4-15.3	0.21-4.48

*= median and range nmol/L, ** = median and range ng/ml

Table 4. Novel Dose Group Characteristics and Response to Treatment

	Low dose (3.0 mCi) 29 cats (51%)	Medium dose (3.5 mCi) 19 cats (33%)	High dose (4.5 mCi) 9 cats (16%)	p value
Gender	15 nm 14 sf	14 nm 5 sf	3nm 6sf	<i>p</i> = 0.1051+
Age**	12.2 (+/- 2.2)	11.6 (+/- 2.3)	12.1(+/-1.8)	<i>p</i> = 0.6563++
% Dose Uptake*	2.3 (0.3-4.6)	5.7 (1.7-9.6)	16.3 (11.5-34.5)	<i>p</i> < 0.0001+++
T:S size ratio*	2.9:1 (1.4-5.5)	4.3:1 (1.9-7.4)	6:1 (4.1-17.2)	<i>p</i> < 0.0001+++
Average Intensity*	2.4 (0.8-5.4)	3.7 (1.6-7.7)	8.3 (4.5-23.3)	<i>p</i> < 0.0001+++
Maximal Intensity*	3.4 (0.8-7.1)	5.3 (2.0-7.7)	13.3 (5.6-23.3)	<i>p</i> < 0.0001+++
Pattern of Disease***	U: 12 (41%) B: 1 (3.5%) AB: 15(52%) A: 1 (3.5%)	U: 7 (37%) B: 2 (11%) AB: 9 (47%) A: 1 (5%)	U: 2 (22%) B: 3 (33.5%) AB: 3 (33.5%) A: 1 (11%)	<i>p</i> = 0.2405++++
Initial T4 * nmol/L	98.7 (39.4-193)	171 (64.7-308)	243 (178-411)	<i>p</i> < 0.0001+++
6-month T4 * Nmol/L	22.7 (10-102)	22.7 (13.2-65.6)	26 (6.4-62.0)	<i>p</i> = 0.7533+++
Outcome	Euth: 16 (55%) Hyper: 3 (10%) Hypo: 10 (35%)	Euth: 12(63%) Hyper: 4(21%) Hypo: 3(16%)	Euth: 5(56%) Hyper: 2(22%) Hypo: 2(22%)	<i>p</i> = 0.5699++++

*= median/range **= mean/SD in years

***U=Unilateral, B=Bilateral, AB=Asymmetric Bilateral, A= Asymmetric Bilateral

+Chi-square, ++One Way ANOVA, +++Kruskal Wallis test,++++Fisher's exact test

Table 5. Novel Dose Group Outcome Associations

	<u>Euthyroid</u> 33 cats (58%)	<u>Hyperthyroid</u> 9 cats (16%)	<u>Hypothyroid</u> 15 cats (26%)	<u>p value</u>
Gender	19 nm 14 sf	6 nm 3 sf	7 nm 8 sf	<i>p</i> = 0.6158+
Age**	11.8 (+/- 2.0)	11.9 (+/-3.1)	12.5 (+/-1.8)	<i>p</i> = 0.5323++
% Dose Uptake*	4.5 (0.3-24.2)	5.5 (0.6-34.5)	2.23 (0.98-21.8)	<i>p</i> = 0.1897+++
T:S size ratio*	3.8:1 (1.4-8.5)	5.3:1 (1.9-17.2)	4:1 (1.9-6.4)	<i>p</i> = 0.4798+++
Average Intensity*	3.4 (1.2-23.3)	4.9 (0.8-12.1)	2.2 (1.5-13.9)	<i>p</i> = 0.0835+++
Maximal Intensity*	5.1 (1.5-23.3)	4.9 (0.8-13.3)	3.2 (1.7-19.6)	<i>p</i> = 0.0975+++
Pattern of Disease***	U: 14 (42%) B: 3 (9%) AB: 16 (49%) A: 0 (0%)	U: 2 (22%) B: 1 (12%) AB: 3 (33%) A: 3 (33%)	U: 5 (33%) B: 2 (13%) AB: 8 (53%) A: 0 (0%)	<i>p</i> = 0.0615++++
Ectopic thyroid tissue present	3 (9%)	2 (22%)	3 (20%)	<i>p</i> = 0.3655+
Initial T4 nmol/L *	161 (39.4-251)	170 (100-411)	89.8 (58.3-332)	<i>p</i> = 0.0506+++

*=median and range, **=mean and SD in years

***U=Unilateral, B=Bilateral, AB=Asymmetric bilateral, A=Atypical

+Fisher's Exact test, ++ One way ANOVA, +++Kruskal-Wallis test

Table 6. Fixed Dose Group Outcome Associations

	<u>Euthyroid</u> 14 cats (61%)	<u>Hyperthyroid</u> 2 cats (9%)	<u>Hypothyroid</u> 7 cats (30%)	<u>p value</u>
Gender	6 nm 8sf	2nm 0sf	4 nm 3sf	$p = 0.4847+$
Age**	12.6 (+/-2.5)	14 (+/- 0)	11.0 (+/-3.7)	$P = 0.3339++$
% Dose Uptake*	4.5 (1.2-14.8)	7.5 (4.1-11)	4.1 (0.5-5.00)	$p = 0.4516+++$
T:S size ratio*	7.2:1 (2.9-12.6)	27.9:1 (17.2-38.7)	4.6.1 (3.4-10.0)	$P = \mathbf{0.0211+++}$
Average Intensity*	4.3 (2.3-16.8)	4.5 (3.0-5.9)	4.4 (0.9-8.4)	$p = 0.7112+++$
Maximal Intensity*	5.9 (2.4-20.2)	4.8 (3.4-6.3)	5.2 (1.2-11.5)	$p = 0.6394+++$
Pattern of Disease***	U: 4 (29%) B: 2 (14%) AB: 8 (57%) A: 0 (0%)	U: 0 (0%) B: 1 (50%) AB: 0 0%) A: 2 (100%)	U: 5 (72%) B: 1 (14%) AB: 1 (14%) A: 0 (0%)	$p = \mathbf{0.0055++++}$
Ectopic thyroid tissue present	0 (0%)	1 (50%)	0 (0%)	$p = 0.0870+$
Initial T4 concentration*	139 (48-263)	177.5 (172-183)	108 (87-184)	$P = 0.3113+++$

*=median and range, **mean and SD in years

***U=Unilateral, B=Bilateral,AB=Asymmetric bilateral, A=Atypical

+Fisher's Exact test, ++ One way ANOVA, +++Kruskal-Wallis test