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MEDICATION WITHDRAWAL GUIDE FOR ALLERGEN-SPECIFIC SEROLOGICAL TESTING

SERUM SAMPLE STORAGE FREE

Nextmune offers a **FREE** of charge **serum storage facility** to enable you to sample before medicating and then test if and when you are ready.

For more information visit the Nextmune Practice Portal at **nextmunelaboratories.co.uk/login**

PRACTICE PORTAL

MEDICATION (AT LICENSED DOSE IF NOT STATED)	SUGGESTED MINIMUM WITHDRAWAL TIME
GLUCOCORTICOIDS	
Oral short-acting <0.5mg/kg, q24hrs for <2 months (e.g. prednisolone, prednisone, methylprednisolone)	0 days
Oral short acting >0.5mg/kg, q12hrs or >2 months (e.g. prednisolone, prednisone, methylprednisolone)	*see guidance below
Injectable short-acting (e.g. single dose of dexamethasone)	7 days
Injectable long-acting (e.g. methylprednisolone acetate)	28 days
Inhaled - non-licensed (e.g. beclometasone or fluticasone)	14 days
Inhaled - licensed pro-drugs (e.g. Aservo (ciclesonide))	0 days
Topical short-acting (e.g. skin, aural and ophthalmic medications)	0 days
Topical long-acting (e.g. aural medications Neptra® and Osurnia® with sustained residual effects)	14 days
OTHER MEDICATIONS	
Ciclosporin, Oclacitinib (Apoquel®), **Lokivetmab** (Cytopoint®), **antihistamines, essential fatty acids, antibiotics, anti-fungals, ecto and endo-parasiticides, NSAIDs, cardiac and thyroid medications**	0 days

*There is an absence of specific studies looking at the effect of higher doses or a longer duration of corticosteroid use but testing may be affected. We would advise, where possible, to withdraw for a minimum of 4-6 weeks in these cases but particularly if immuno-suppression has occurred, a significantly longer withdrawal may be required.

Where it is not possible to safely withdraw corticosteroids, or use alternative therapies to control the clinical signs sufficiently, we advise reducing the dose as much as possible prior to testing and interpreting the results in light of this. Please also refer to our guidance notes on the optimal timing for testing at nextmunelaboratories.co.uk.

Recommendations are based on the publications below in addition to pharmaceutical company data and dermatologist guidance. There is currently limited available evidence (especially for cats and horses) particularly regarding the effect of the long-term use of the majority of drugs listed above, or when used off license.

- Olivry, T. & Saridomichelakis, M. (2013). Evidence-based guidelines for anti-allergic drug withdrawal times before allergen-specific intradermal testing and IgE serological tests in dogs for the International Taskforce on Allergic Diseases of Animals (ICADA). Veterinary Dermatology 24(2): 225-e49
- Clear, V., Petersen, A., Rosser, E.J. & Ruggiero, V. (2015). Investigation of the effects of 30 day administration of oclacitinib (Apoquel®) on intradermal and allergen-specific IgE serology testing in atopic dogs. In: 29th Proceedings of the North American Veterinary Dermatology Forum (NAVDF), Nashville, Tennessee
- Souza, C.P., Rosychuk, R.A., Contreras, E.T., Schissler, J.R., & Simpson, A.C. (2018). A retrospective analysis of the use of lokivetmab in the management of allergic pruritus in a referral population of 135 dogs in the western USA. Veterinary dermatology 29(6): 489-e164

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REFERENCE GUIDE
OPTIMAL TIMING FOR SEROLOGICAL ALLERGY TESTING

The optimal time to sample for allergen-specific IgE serological testing will vary dependent on:

- the level and timing of allergen exposure (e.g. less exposure to pollens is expected in the winter)
- the individual immune response (which itself will vary and is affected both by age and certain medications)
- how long each animal will take to clear IgE and IgG antibodies from the blood after exposure

The guidelines below can be used to maximise the chances of generating meaningful test results.

1. SAMPLE WHEN FULLY SYMPTOMATIC

Unless clinical signs are controlled ONLY using medications that are not thought to affect testing, we would advise:

- always sampling when the animal is fully symptomatic; and
- either before starting on medication or after withdrawing it for a sufficient time period*.

* Please see our **Medication Withdrawal Guidelines** for further information and see overleaf for details of our serum storage facility.

2. AGE

We advise ideally testing from 12-15 months of age in order to allow:

- maternally derived antibody levels to subside;
- exposure to a complete allergy season, regardless of the time of year they were born.

If earlier testing is felt necessary, we would advise re-testing when over 12 months of age to capture any further hypersensitivities that may develop.

3. ACUTE ONSET CASES

For cases with a rapid onset of clinical signs that may only last for a few days, such as acute urticaria, we would advise:

- testing at least 24 hours after symptoms start; but
- ensuring that sampling takes place before signs have started to clinically improve.

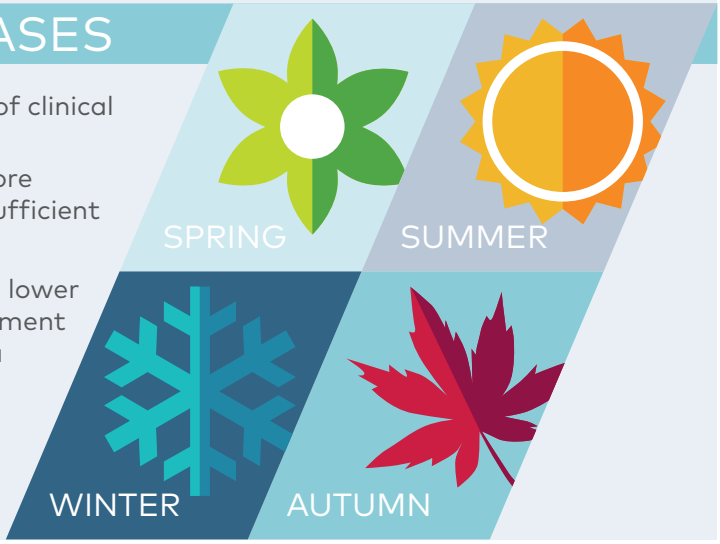
Sampling within this window increases the chances of serological IgE being sufficiently raised after exposure to allergens so that it can be detected, but before the IgE is on the decline due to clearance from the blood.

4. CHRONIC SEASONAL CASES

For chronic seasonal cases with a more gradual onset of clinical signs, we would advise:

- waiting at least 2 weeks after symptoms start before environmental allergen testing, to ensure there is sufficient levels of allergen-specific IgE in the blood.

This is because the initial signs may be in response to a lower allergen burden (fewer pollens or insects in the environment in the early season for example), which could result in a more gradual increase in the level of antibodies in the blood. See also point 6 overleaf.



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5. ALLERGEN AVOIDANCE

If a level of allergen avoidance has already been implemented (e.g. a restricted diet or stabling to avoid pollens / insects), then these restrictions must be removed prior to testing:

- for environmental allergens this should be for at least 2 weeks;
- for food allergens this should be at least 2 months (as the food testing also measures allergen-specific IgG which may take longer to increase).

Minimising recent exposure prior to testing can lead to lower levels of allergen-specific IgE or IgG in the blood and therefore potentially produce false negative results.

6. SEASONAL SYMPTOMS

If animals have a definite seasonal component to their clinical signs, we would advise sampling at the time of year in which their clinical signs are at their worst.

It can be beneficial to sample towards the end of this period but before their signs have started to wane. This way you know:

- the animal has been recently / is still being exposed to the relevant allergens;
- the serological IgE should be sufficiently elevated; and
- if appropriate, allergen specific immunotherapy can be started prior to the next season.

REMEMBER!

- Environmental allergy testing is used after the diagnosis of atopic dermatitis has been made (following the appropriate clinical rule-outs) to identify potential allergens.
- Food allergy testing is used to help identify suitable ingredients for a dietary trial; the dietary trial itself is used for the diagnosis of adverse food reactions.
- All results should be interpreted alongside the history, clinical signs and environmental considerations.

Sampling at the optimum time will increase the chance of getting meaningful results. Nextmune offers a **FREE** of charge **serum storage facility** (initially for 3 months) to enable you to sample before medicating and then test if and when you are ready.

For more information visit the Nextmune Practice Portal at **nextmunelaboratories.co.uk/login**

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REFERENCE GUIDE
REDUCING THE RISK OF NON-VIABLE SAMPLES

Occasionally we receive samples that unfortunately cannot be used for testing. To try to reduce the risk of this situation occurring, listed below are some suggestions for the top 4 sample issues we encounter.

1. HAEMOLYSED - CAN PREVENT TESTING IF EXTREME

- Book ample time for sampling, making use of topical anaesthetic creams for more anxious patients.
- Do not use excessive alcohol to swab the site and ensure it is dry prior to sampling; if aspirated, alcohol can damage cells within the blood.
- Occlude the vein for the minimum time possible.
- Use a 21-gauge needle in dogs and 23-gauge needle in cats if possible.
- Avoid moving in and out of the vein, and excessive suction, when collecting blood; use a different site if a haematoma forms rather than trying to sample through it.
- After sampling, remove the needle from the syringe entirely before slowly decanting the blood into the tubes; avoid shaking the sample.
- Allow the blood to clot for approximately 30 minutes at room temperature before spinning down; balance the centrifuge before use, and separate the serum from the sample as soon as completed.
- Always send serum rather than whole blood, where possible. Whole blood can be sent but double the volume is required and it is much more likely to haemolyse.
- Store the sample in the fridge and post as soon as possible. Avoid posting on a Friday, to minimise extended time in the post where temperature is not controlled.

2. LIPAEMIC - CAN PREVENT TESTING IF EXTREME

- For dogs and cats, fasting for 8 - 12 hours (an overnight fast with free access to water) is often helpful to reduce the likelihood of lipaemia.
- Repeat sampling a couple of hours later may produce a less lipaemic sample.
- Collecting and centrifuging a larger volume of blood may yield a sufficient sample between the lipid layer and red blood cells; refrigeration of the sample can also help the separation.
- Centrifugation at a higher-than-normal speed can assist in clearing the lipid layer.

3. INSUFFICIENT SAMPLE

- Remember the volume stated on the submission form is the minimum required, more is always good!
- Separated serum is always preferable to whole blood to reduce the risk of haemolysis. If you have to send whole blood, it should be at least double the volume required.
- When using gel tubes for serum, remember to spin them down and ensure that the gel isn't included in the volume.
- Send sufficient serum for all requested tests, and ideally any additional tests you may decide to run depending on the results.

4. WRONG BLOOD COLLECTION TUBES

- Samples must be submitted in either plain tubes (white top, or red top if using a vacutainer) or serum gel tubes (brown top).

To check you are using the correct type of tube, visit the FAQs section of the Nextmune Practice Portal at **nextmunelaboratories.co.uk/login**

REMEMBER!

If possible, before discharging the patient, check the separated serum volume is sufficient; although we can run some haemolysed and lipaemic samples, ideally it should also be clear of these issues.

Nextmune offers a **free of charge serum storage facility** so you can sample at your convenience and test when you are ready. Just tick the storage option on your submission form.

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