Murine models of endometriosis corelates cystic load with pain response along with observable signs of anxiety and depression in C57/Bl6 mice and Balb/c mice

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SCAN ME

BACKGROUND

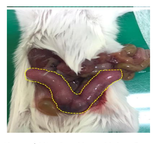
Endometriosis is a womans health issue and a condition that involves the ovaries, fallopian tubes and pelvic tissue. The condition rarely spreads outside of the pelvis and is characterized by pain of varying degrees, infertility if left untreated and can result in debilitation from chronic pain. There are currently treatments available, but these often don't fully address the condition in women suffering from endometriosis. Endometriosis modelling within the preclinical sphere involves the creation of rodent modelling paradigms that seek to recreate the growth of endometrial tissue (endometriomas) and also to evoke some of the responses and behaviors associated with pelvic and abdominal pain. In the course of internal validation work we observed that certain models seem to evoke a stronger response to pain as well as manifesting various behavioral responses to pain like social isolation and disruption of circadian rhythm.

METHODS

Various methods are involved in the paradigms employed to induce endometriosis. Among these there are:

- Surgical implantation of donor tissue into the recipient
- Syngeneic innoculation model whereupon prepared donor tissue is "sprayed" into the recipient endometrium of the donor
- A **refined** model whereby the endometrial tissue from the donor is placed into the recipient and secured with surgical glue

Placement of P4 Silastic Implant



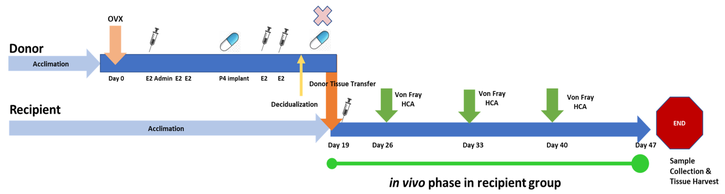
Decidualization of Uterine Horn Using Sesame Oil

Sham groups are created according to the methodology employed to induce endometriosis i.e. for the adhesive (glue) model a surgical incision is created at the same time as the transfer of tissue or innoculation and several spots of surgical glue are applied in the same pattern as donor tissue would be applied to the wall of the endometrium. In the case of the syngeneic "spray" model a suture to the secum is created after the opening of the abdomen in a WT mouse.

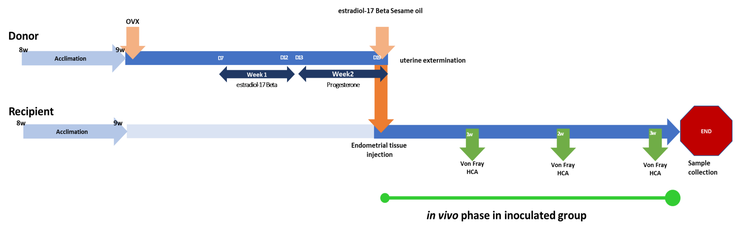
Home cage monitoring in group housed conditions was instituted using the **Actual HCA™** Home Cage Monitor (Actual Analytics Ltd., Edinburgh, UK) animals were tracked in their home cage in a socially housed manner and a particular set of parameters is obtained from this extended locomotor activity monitoring.

- Currently animal models are limited to mainly rodents and often focused on a short-term cystic treatment or reduction in number of cysts.
- The use of a SHAM control group (suture to colonic mesentery) is rarely used in modeling, and this is largely required for addressing nociception within the model.
- There are various methods of modelling endometriosis but for exploring the associated pain with endometriosis as well as having a cohort large enough the Balb/c mouse model of inoculation of donor tissue seems to be about the best strain for pain studies.

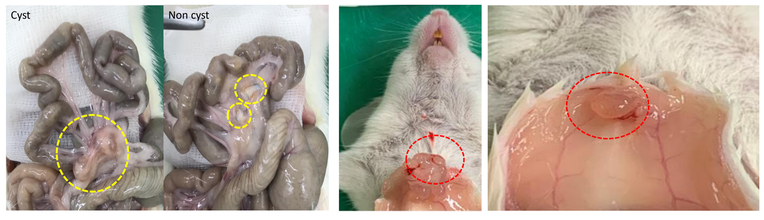
STUDY PARADIGMS for INDUCTION of ENDOMETRIOSIS



Adhesive inoculation model using surgical glue to secure donor tissue in the recipient mice



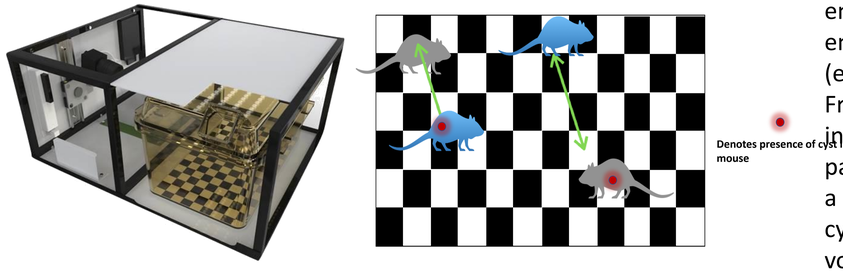
Syngeneic model using harvested and decidualized tissue to prepare an inoculate solution that is introduced to the recipient mice



80% - 90% of inoculated animals will develop cysts – the locations will vary and randomly appear in various locations within the peritoneal space

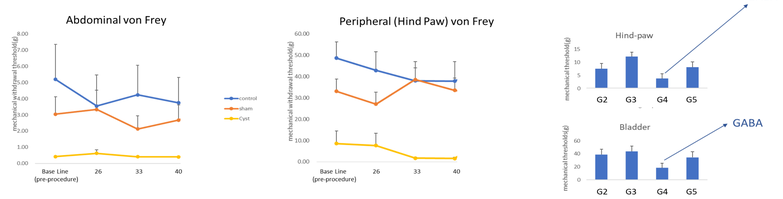
Average	long axis (mm)	short axis (mm)	cyst volume (mm)
Cyst size (Balb/c)	3.7±0.47	2.0±0.13	12.8±2.65

Dimensions of cysts after inoculation and harvest of cystic tissue. It should be noted that the presence of non-cystic tissue that may contribute to endometriosis and the associated pain as well as functional and behavioral changes from chronic pain is not measured in these studies – only the presence of, amount and dimensions of cysts harvested from the rodent.

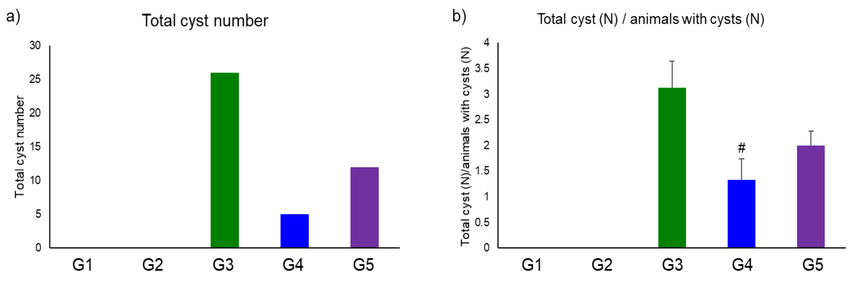


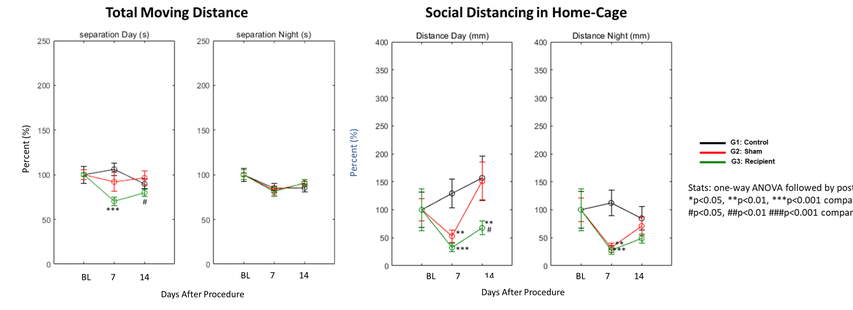
Monitoring of animals movement and behaviour in a longitudinal fashion allows for the observation and quantification of the amount of time animals stay (or attempt to stay) isolated from the other cage-mates.

OBSERVATIONS



von Frey testing in BAlB/c mice. It can be plainly seen that the pain threshold is decreased in both the sham and the cystic (inoculated groups) – the value of using the SHAM animals as a reference can be plainly seen from the





CONCLUSIONS

Over the course of 4 years we have performed many studies for endometriosis in mice. In order to fully validate the disease model, Naason Science employed various ways of inducing disease-like symptoms in mice and rats. In general, the end-points of any study in endometriosis focus on the growth of abberant endometrial tissue or, rather, the actual number of endometriomal growths (endometriomas) found in the peritoneal space. The evoked pain signals, like von Frey measurement, can be seen to track inline with cystic tissue load in the inoculated animals – as cystic load is found to increase there are decreases in the pain threshold in these animals. At first this data was relatively anecdotal but after a review of various studies it seems that an increase in cystic load (number of cysts) will manifest itself through a decrease of pain thresholds as measured by von Frey. There are also salient non-evoked signs of changes in locomotion, home cage social interaction as well as the previously mentioned evoked pain response changes which are measured in both the abdominal region and the periphery (hind paw). It shows that the model can be used to study endometriosis and the formation of endometriomas but is also a suitable model for the study of abdominal/visceral pain.