'Investigating the epidemiology of Borrelia Spirochaetes in Dorset'

- This project is a collaboration between The Thomas Hardye School and the University of Exeter
- This project has been supported by the following organisations

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THE ROYAL SOCIETY

Introduction

- 1-10% ticks *Borrelia* positive in different areas of UK (according to PHL England)
- Many individuals bitten by ticks do not go on to develop Lyme disease

Aims

- Build students' technical and team working skills beyond curriculum requirements
- Carry out authentic science in conjunction with university scientists
  - Focussing on Dorset we aim to try to map the distribution of *Borrelia* positive ticks
  - Correlate positive ticks with tick gender, size and location
- Raise awareness of Lyme disease within Dorset and further afield

What is GENEsis Club?

Methodology: Tick collection

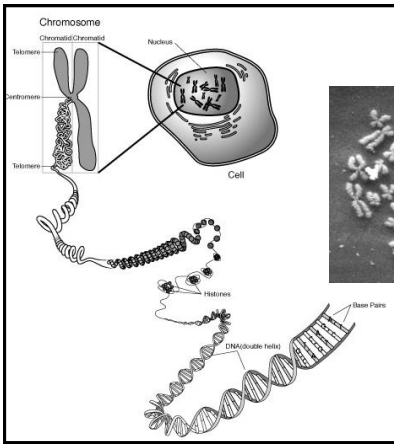
- Distribute "tick boxes" across Dorset to individuals who come into contact with ticks
- Ticks are placed in 70% ethanol and details about their collection recorded

Methodology: Processing ticks

- Ticks returned to Thomas Hardye School
- 6th Form students photograph ticks and record details of ticks
- Students then extract DNA, carry out PCR and gel electrophoresis to identify if tick contains Borrelia burgdorferi

What techniques have we been using?

- Making up buffers
- Accurate pipetting
- Molecular Biology techniques:
  - DNA extraction
  - PCR
  - DNA gel electrophoresis



DNA

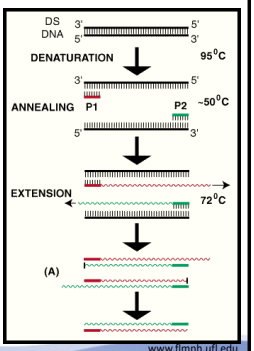
Polymerase chain reaction (PCR)

3 steps in PCR (30-40 cycles):

1.Denaturation
double DNA strand melts open

2.Annealing
primers bind to DNA and polymerase attaches and starts copying DNA

3.Extension
at 72°C: optimum temperature for *taq* polymerase and extension of fragment



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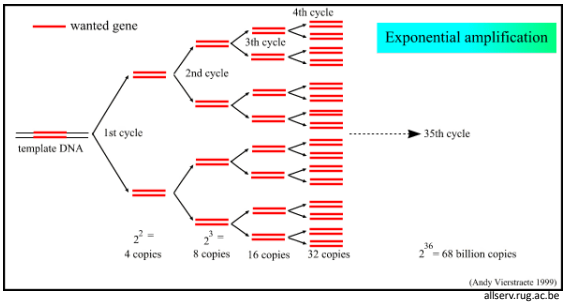
Polymerase chain reaction (PCR)



allserv.rug.ac.be

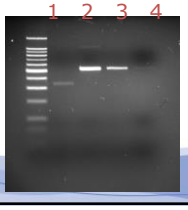
Polymerase chain reaction (PCR)

Gel electrophoresis

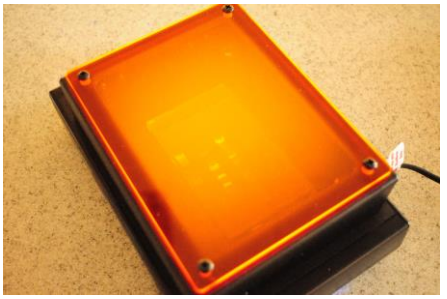
1 2 3 4

DNA ladder

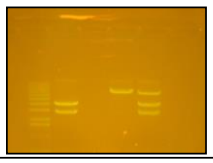
1000bp
500bp
100bp



Using Safe Blue to stain DNA and make it fluoresce under UV (using a safe light box)



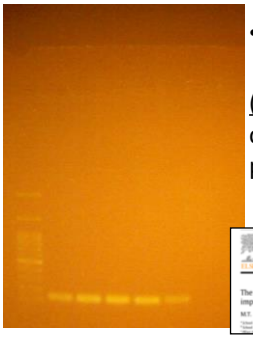
Initial work analysing plasmid then our own DNA



We used "Nature's Dice" DNA kit to create a pedigree on inheritance of Sickle Cell disease.

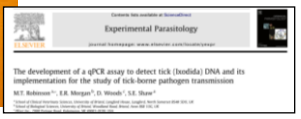
We linked PTC taste sensitivity to our probable genotypes. This involved DNA extraction, PCR, restriction enzyme digestion and DNA gel electrophoresis.

Extracted DNA from ticks and confirmed extraction using a specific primer for tick DNA

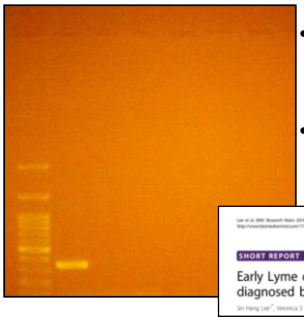


- TKcyt1.3 Tick DNA control PCR results

(I. ricinus: using cytochrome C oxidase primer)



All samples with confirmed tick DNA were then tested using primers for Borrelia burgdorferi DNA



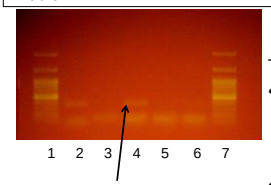
- *B. burgdorferi* 16S rRNA primers:

LD2: (5'-GACTTATCACCGGCAGTCTTA) and TEC1 (5'-CTGGGGAGTATGCTCGCAAGA)

- 129base product

Finally after 10 gels we got our first "hit"

- Lanes 1,7 are DNA ladders.
- Lane 2 is a positive control for *Borrelia* DNA (129bp)
- Lane 3,4,5,6 are samples from different ticks.

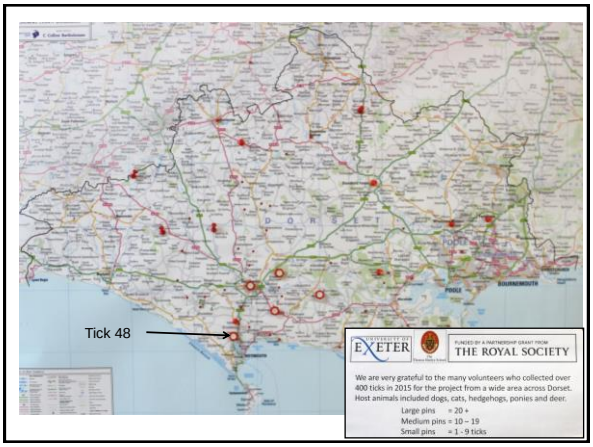


1 2 3 4 5 6 7

An extra band indicates a positive result for Lyme disease DNA.

The GENEsis club first positive result!

- After testing 60 ticks, on tick number 48 (see above) we found our first tick positive for Lyme disease.
- Students will continue testing to try and find the next.



What have our students learnt?

- *'Genesis has granted me the opportunity to work like a professional scientist. Even by cutting off the heads of ticks I have become better accustomed to conducting experiments and working with specialist apparatus.'*
- *'It has reinforced my decision to do a career in science.'*
- *'The project has really boosted my confidence, independence and analytical skills: three things that could not have been so well improved from just studying my A-level courses.'*
- *'I have never been so engaged in a project, I think it was because we were all involved in something that actually had a real-life outcome unlike our class practicals – I really have enjoyed being part of it.'*

Developing technical skills

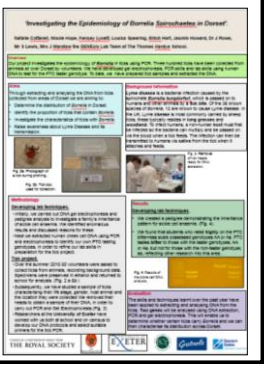
Using pipettes, PCR and gel electrophoresis

These are all techniques normally used at a university, not in a school!

Interacting with the wider scientific community

We met 3rd year Undergraduates at Exeter University.

Presenting at The Royal Society



Raising awareness of Lyme disease

Within-school events for:

- younger students
- visiting teachers
- outreach providers

Out and about at:

'The Family Festival of Science' Oct 2015 and 'The Big Bang Fair South West' March 2016

- Students asked members of the public and visiting pupils to play games including 'identify the "cuddly" tick with Lyme disease by analysing chromatography

Networking at Dorset County Show

- Students spoke with members of the public and local vets about Lyme Disease transmission and symptoms

Extra Publicity

- Radio Solent
- Dorset Echo
- Twitter
- Association of Science Education
- Lyme Disease Action Conference (!)

The school benefits from….

- Engaging and extending student beyond the curriculum
- Preparing students for research careers? (more 6th form students progressing to top universities to study related courses)
- Making new and strengthened community and academic links e.g. Exeter and FRS lecturers
- Updating teachers' knowledge, skills and enthusiasm
- Gaining equipment and reagent for wider curriculum use and disseminating skills to colleagues

Future plans for 2016/17

- Collect ticks from farmers as well as pet owners and environmental sector
- Y12 to analyse another 50+ ticks this year
- Y13 use primers for other tick-borne pathogens such as Anaplasma to use on extracted DNA to plot distribution across Dorset
- Write up investigation as a report/paper to publish in appropriate journal
- Hold a celebration in April for project partners
- Secure funding to continue next year…..

Presented by:

- Suzannah Hall
- Adele Mair
- Joselyn Savill
- Holly Townsend
- Eleanor Wilberforce
- Dr Jeremy Rowe Thomas Hardye School

This project has been supported by the following organisations:

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ERNEST COOK TRUST
Learning from the best

The Thomas Hardye School

UNIVERSITY OF EXETER

LDA
Lyme Disease Action

Gratnells

Severn Sales
Laboratory Equipment

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- Mrs Judith Wardlaw, Thomas Hardye School
- Mr Simon Lewis,Thomas Hardye School.