



Breakthrough Cancer Detection Method

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Our mission is to save lives and improve outcomes for millions of people and animals worldwide.

Looking for
cancer in a
liquid biopsy is
like looking for
a needle in a
haystack

**...and the
needle is
made of hay!**



**But we found the
way to find it**

The Potential Opportunity

It's BIG!

\$Billions\$

The Commercial Strategy

Licensing

We will seek

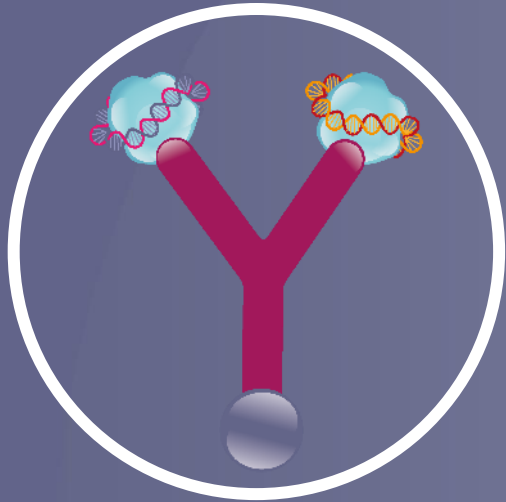
- Upfront payments
- Milestone payments
- Royalties and
- Sales of Key Components

Early cancer detection by plasma CTCF transcription factor analysis

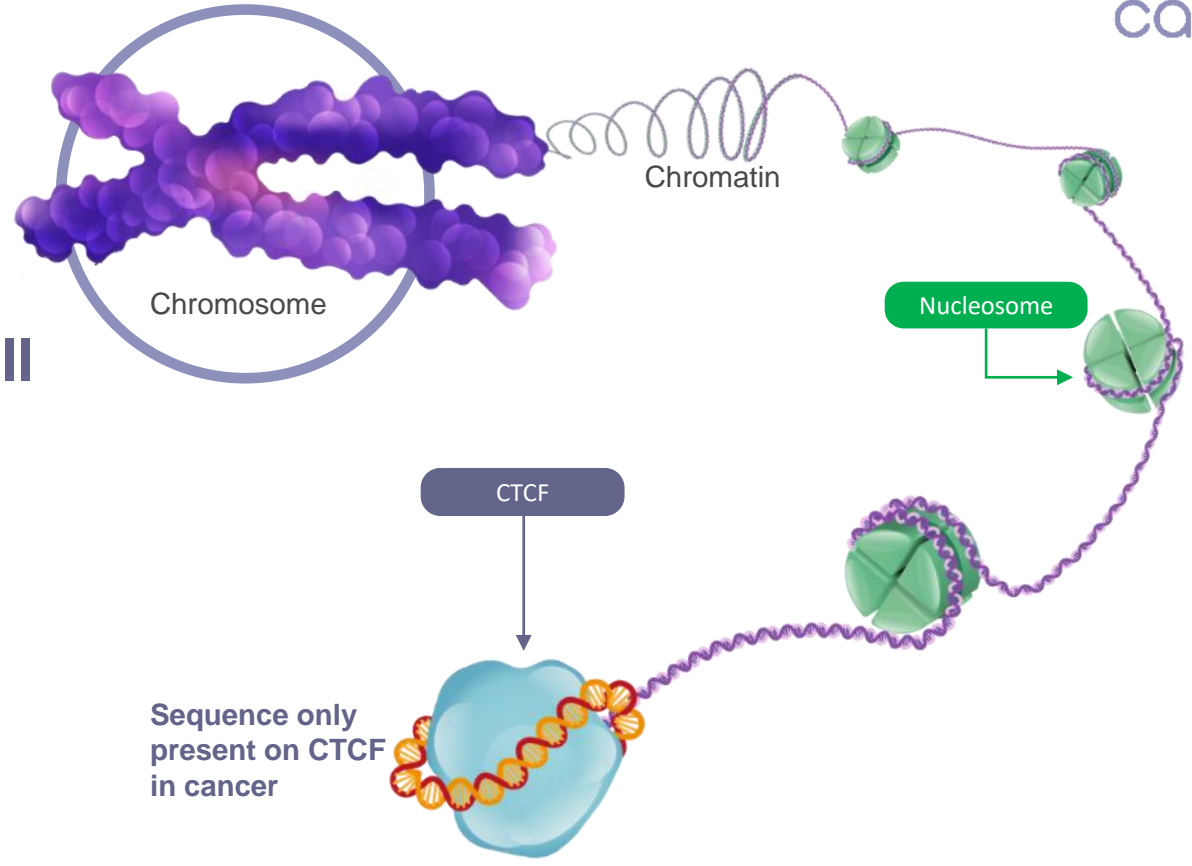
Dr Jake Micallef

Chief Scientific Officer





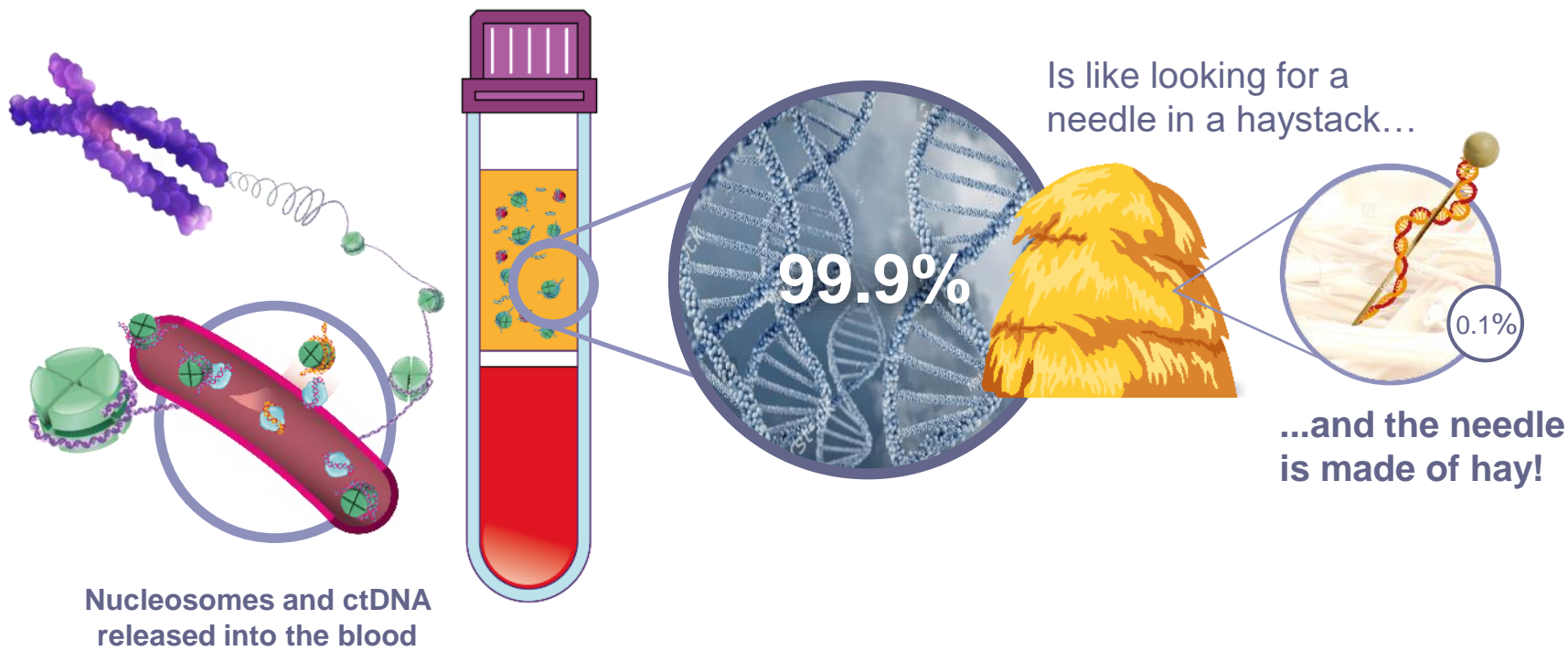
Background



Cancer and cell death cause chromatin fragmentation

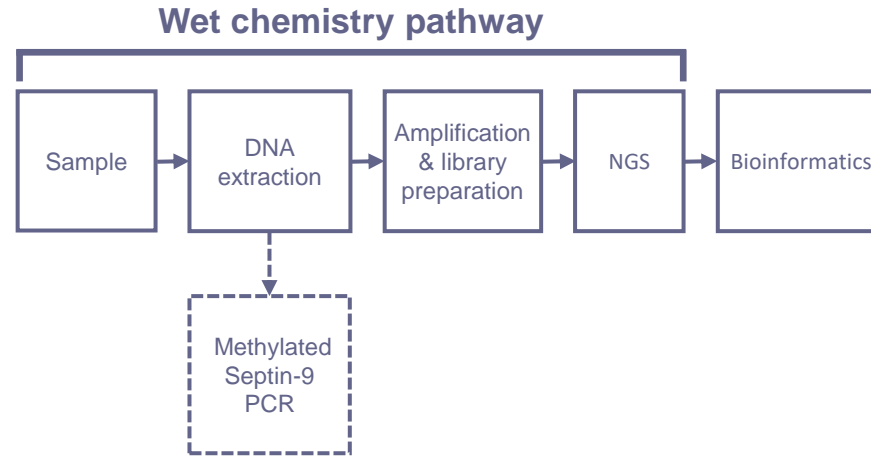
Liquid biopsy for ctDNA

cancer



Liquid biopsy for ctDNA

cancer

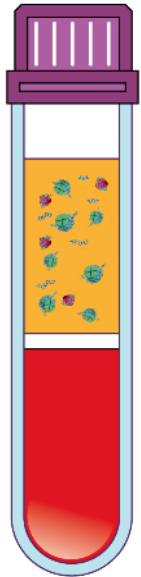


Method unchanged since liquid biopsy first conceived

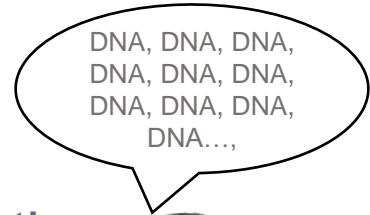
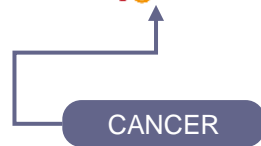
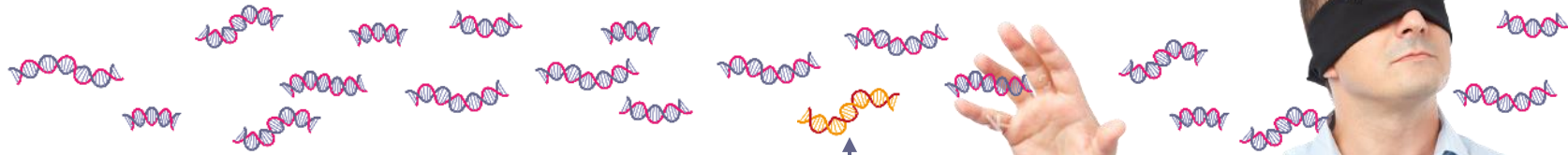
What can you do with tens of millions of DNA sequences – other than sequence them?

Liquid biopsy for ctDNA circulating tumor chromosome fragments

cancer

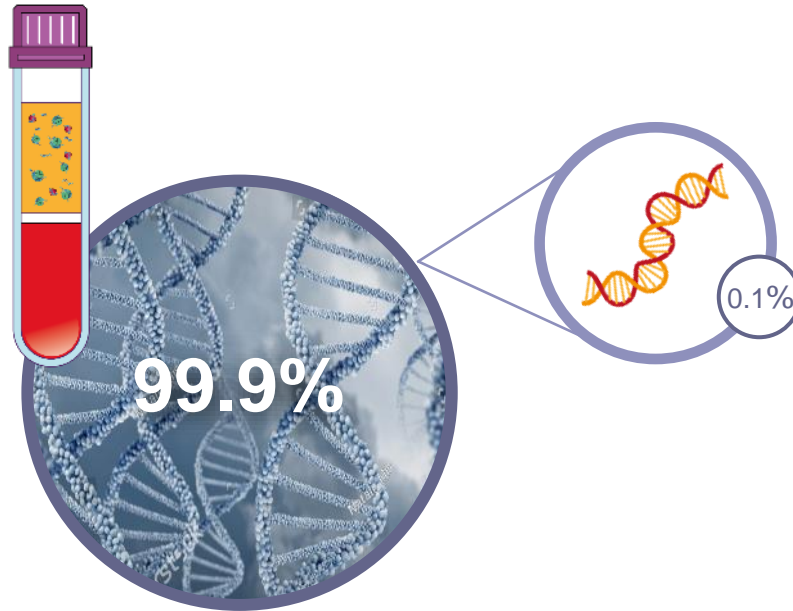


But DNA is DNA – you can't separate out the bit you want!



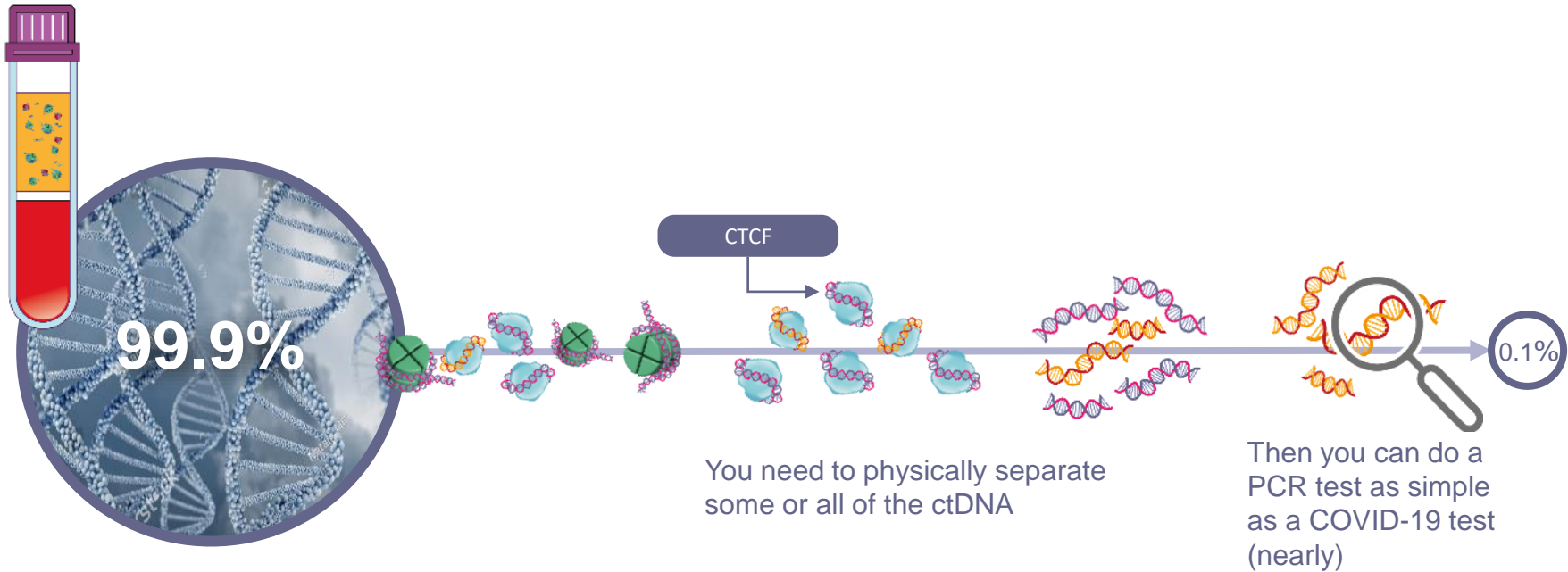
Liquid biopsy for ctDNA... ...the real answer is KISS

cancer



Liquid biopsy for ctDNA

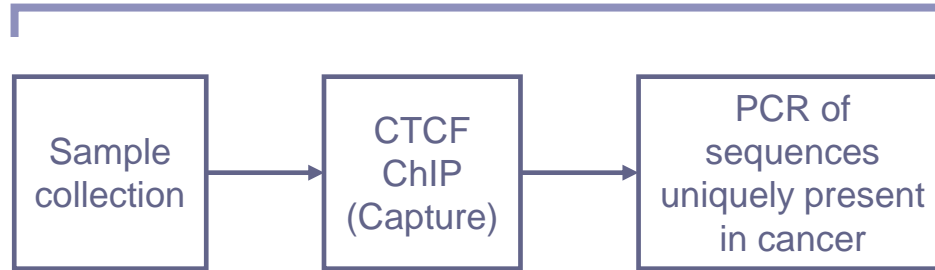
cancer



Novel wet chemistry pathway for ctDNA analysis

cancer

NEW wet chemistry pathway



First novel wet chemistry pathway for liquid biopsy

First report of plasma TF-ChIP (Capture)

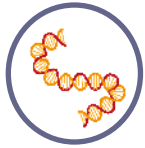


Alterations to the binding site occupation pattern of CTCF is a functional epigenomic signature of cancer including both gain and loss of CTCF occupied sites in cancer cells.¹

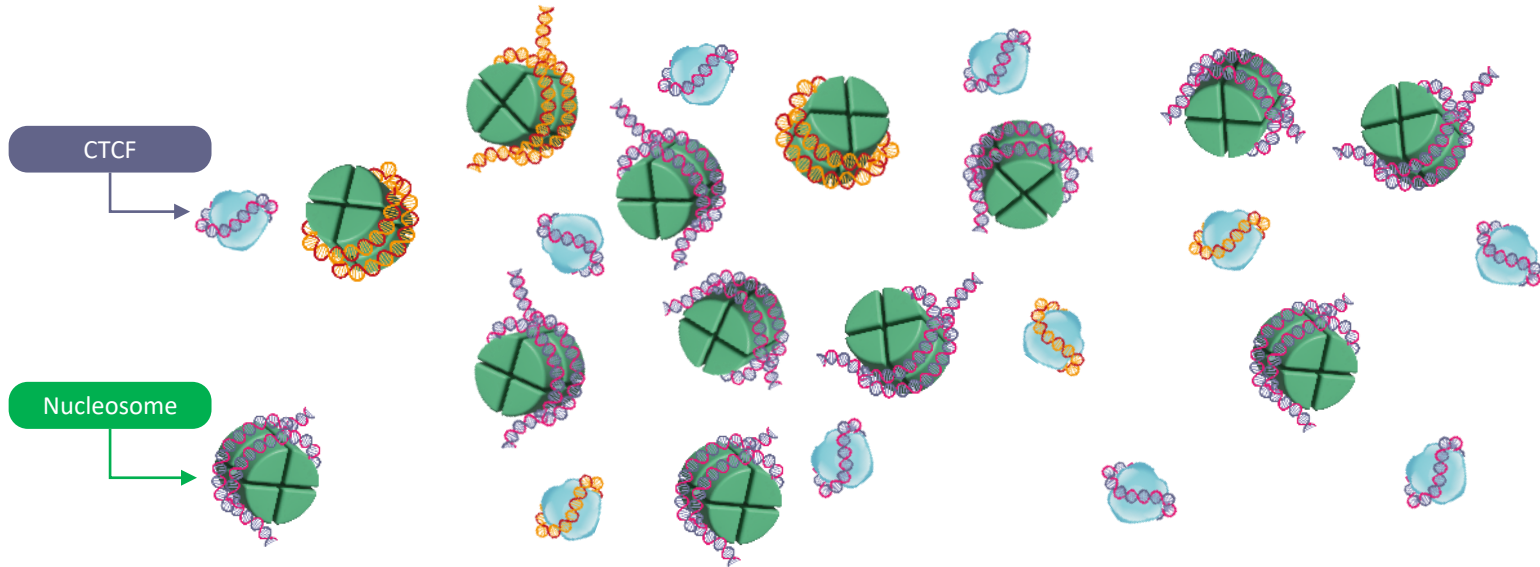
1. Fang et al. Genome Biology (2020) 21:247

Hypothesis

cancer

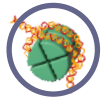


= CTCF binding site with gain of occupancy in **cancer**

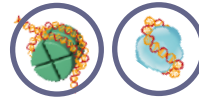


Hypothesis

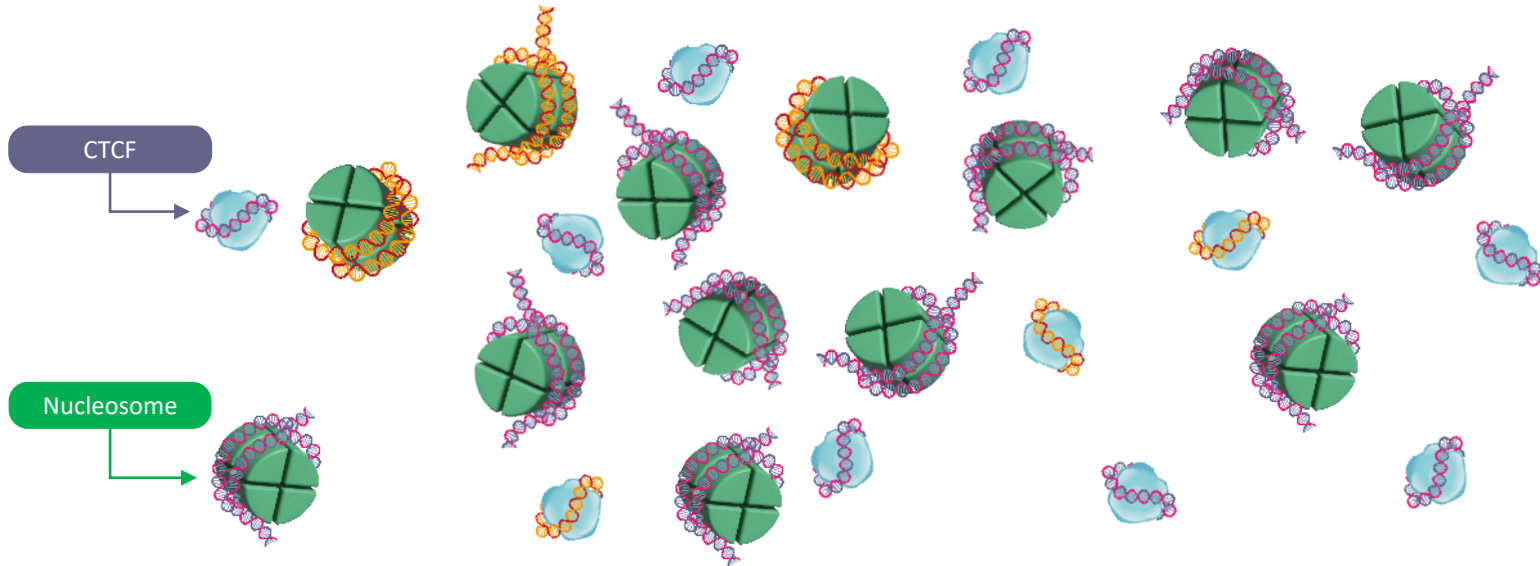
cancer



Sequence present on nucleosomes
in **healthy**



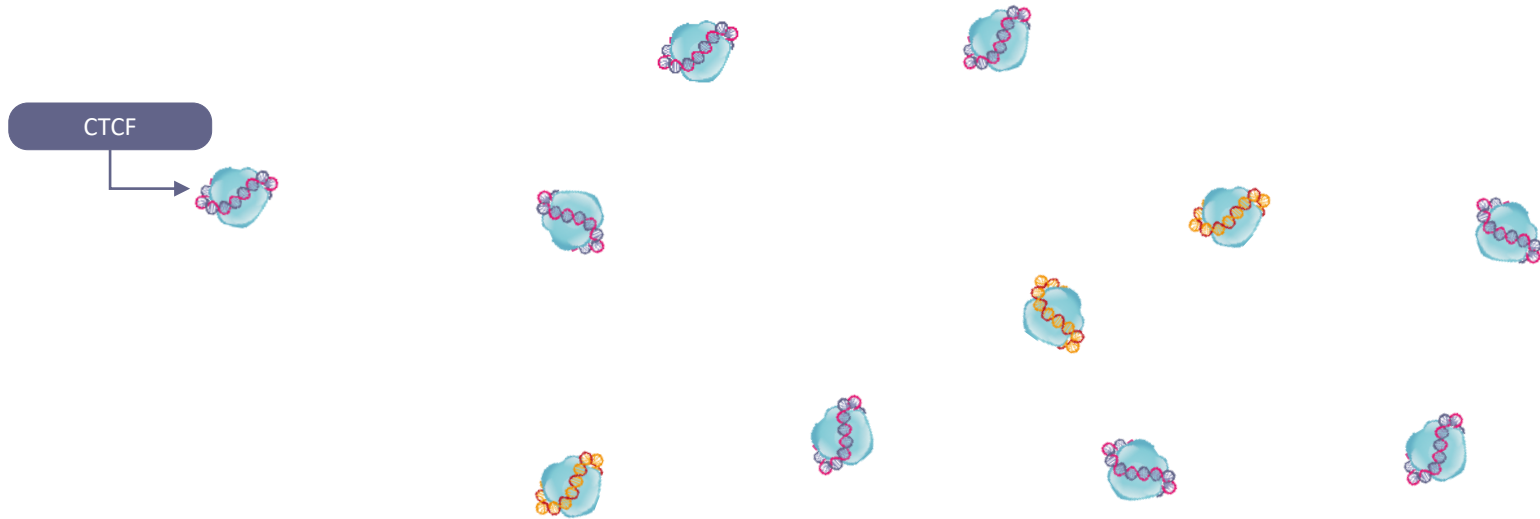
Sequence is present in nucleosomes
and in CTCF **cancer**



Hypothesis

cancer

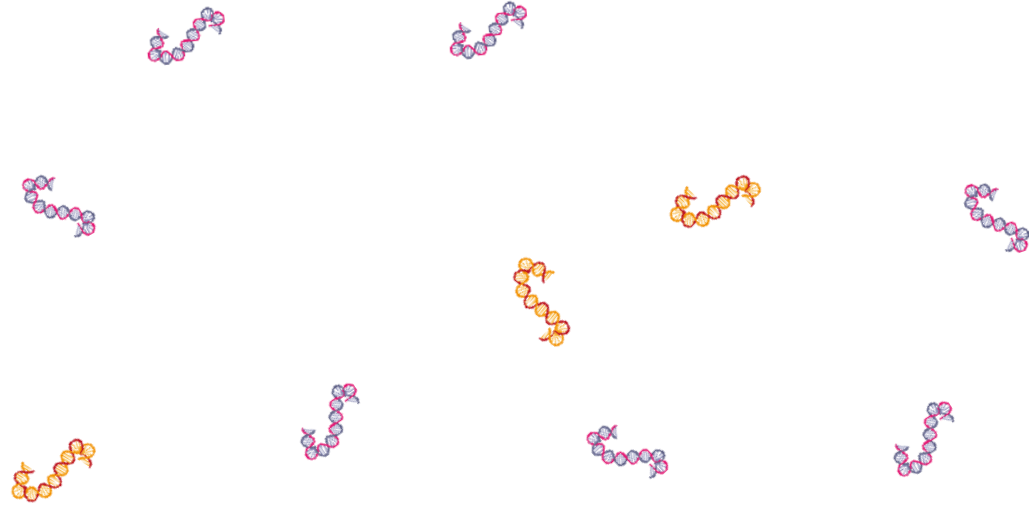
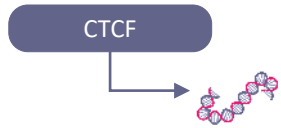
Physically separate the CTCF from all the nucleosomes



Hypothesis

cancer

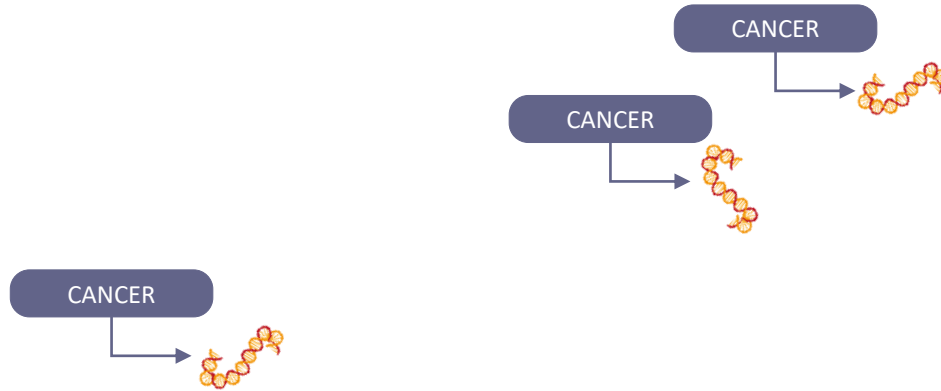
Extract the DNA



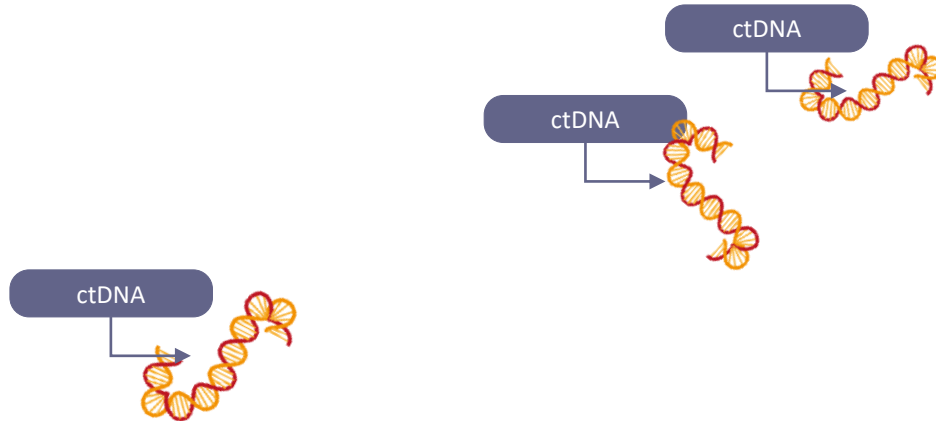
Hypothesis

cancer

Then cfDNA fragment with that sequence tumour derived

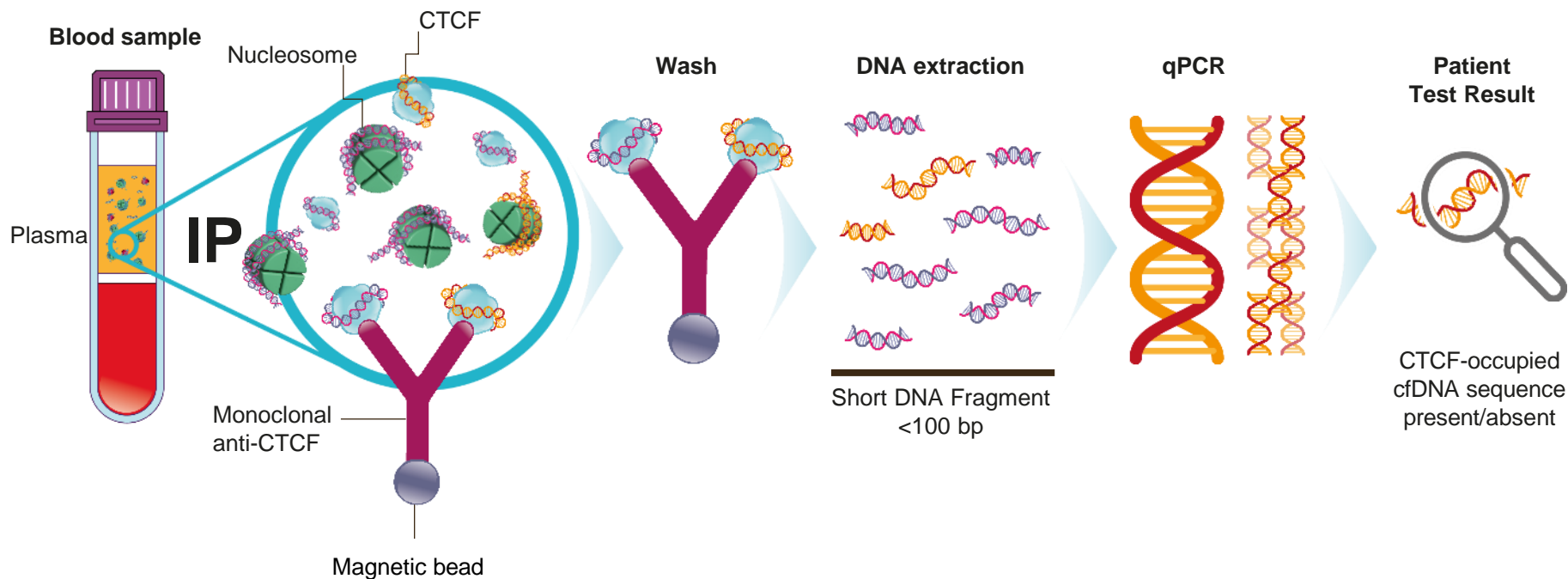


ctDNA

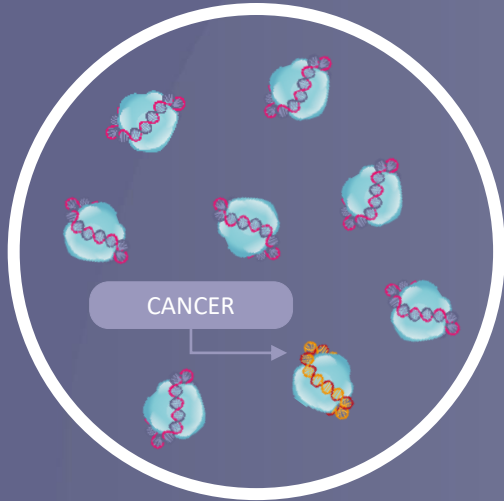


Method: CTCF-IP / qPCR Liquid Biopsy

cancer



This method for the *first time* completely physically isolates ctDNA from non-tumour derived background cfDNA of the same sequence, thereby removing the background cfDNA problem of liquid biopsy.



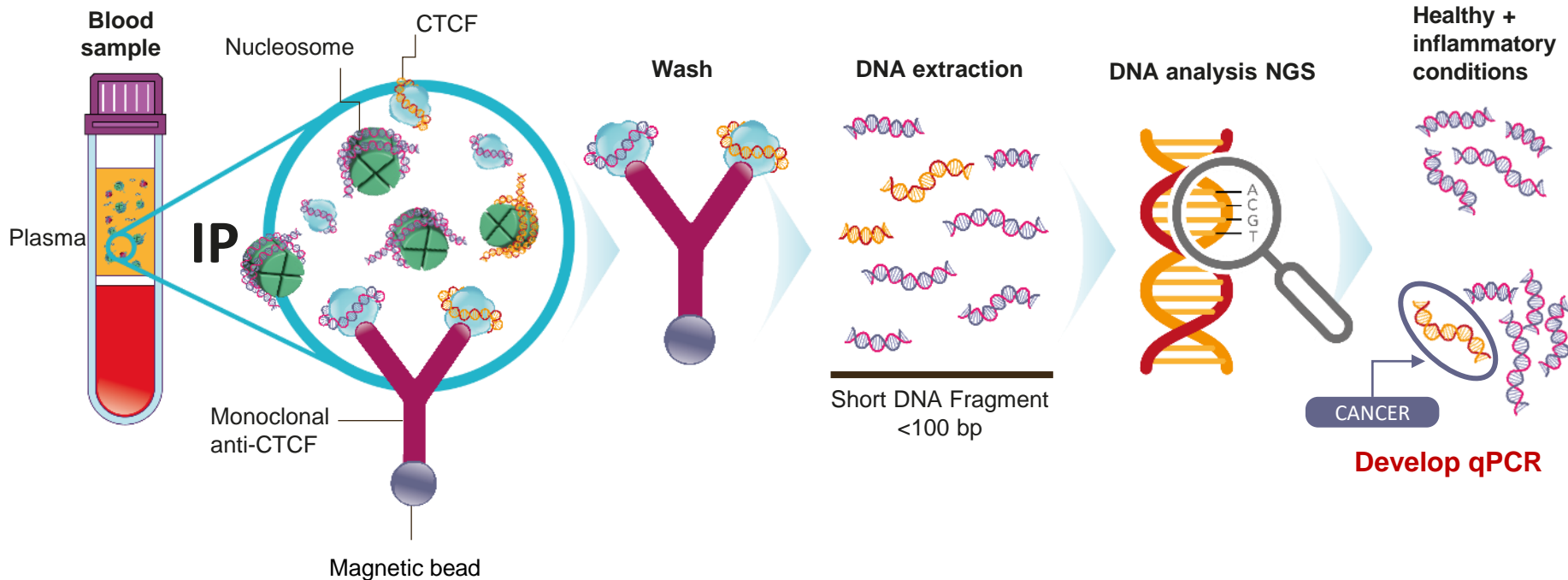
Biomarker Discovery

Finding CTCF Binding sites occupied in cancer ctDNA

– nucleosome covered in healthy and inflammatory plasma cfDNA

Method: Discovery

cancer





Hypothesis in action

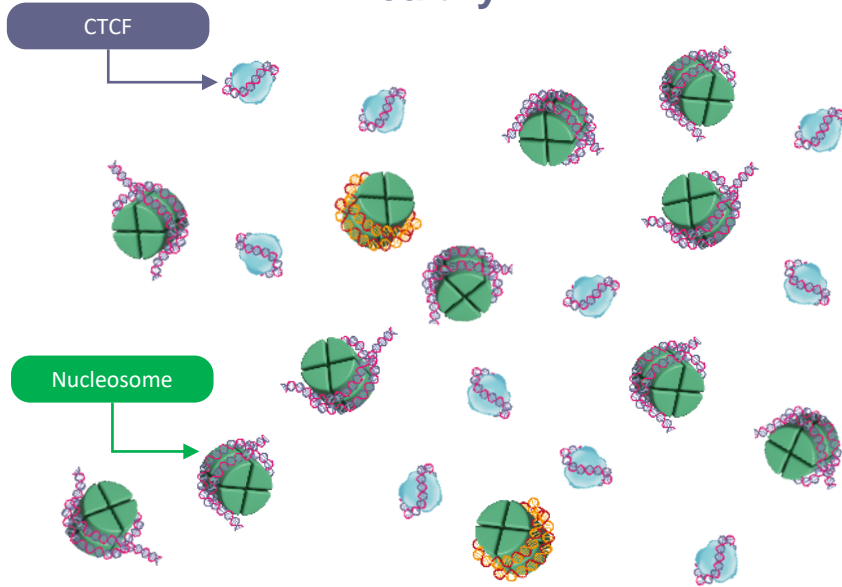
Hypothesis in action

cancer

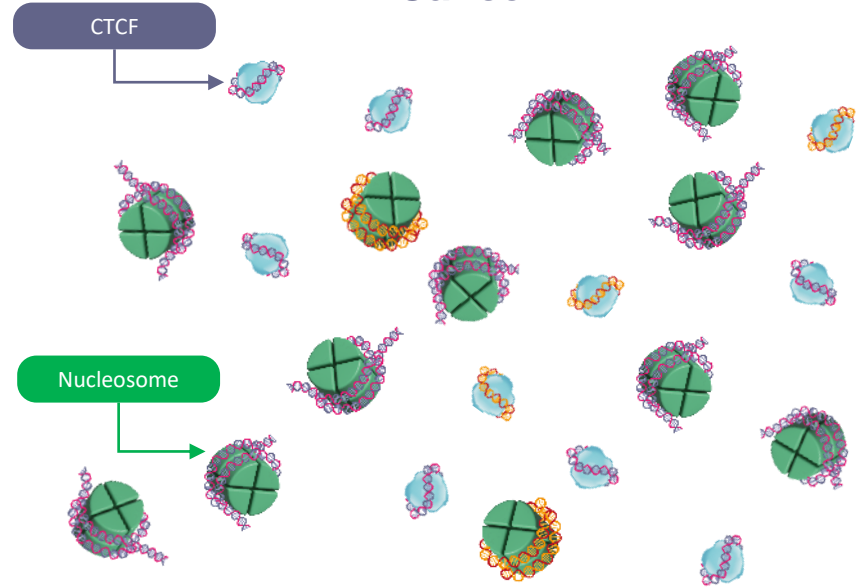


= CTCF binding site with gain of occupancy in cancer

Healthy

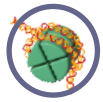


Cancer

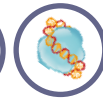
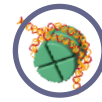


Hypothesis in action

cancer

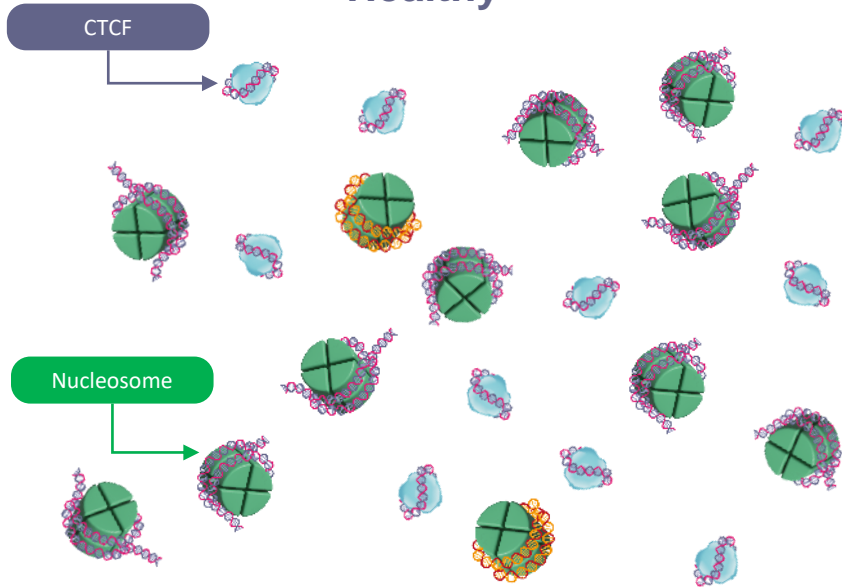


Sequence present on nucleosomes in healthy

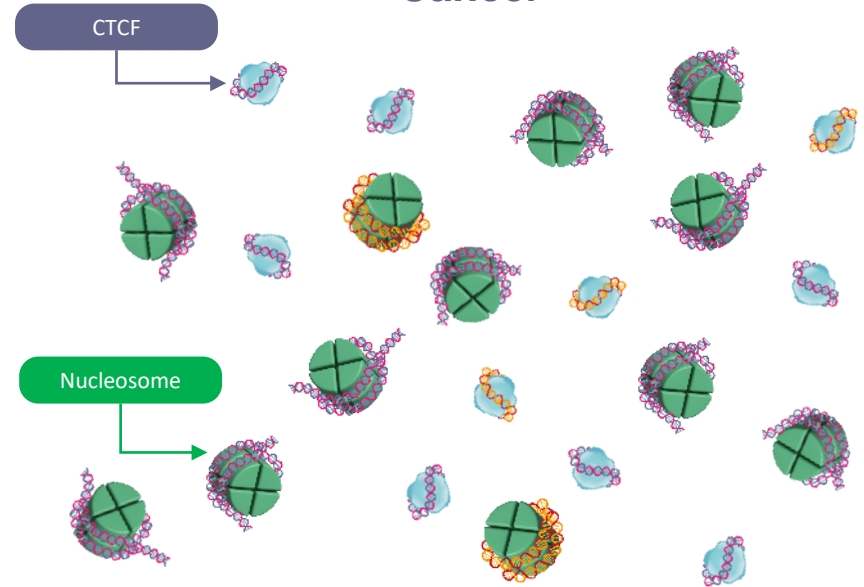


Sequence is present in nucleosomes and in CTCF cancer

Healthy



Cancer

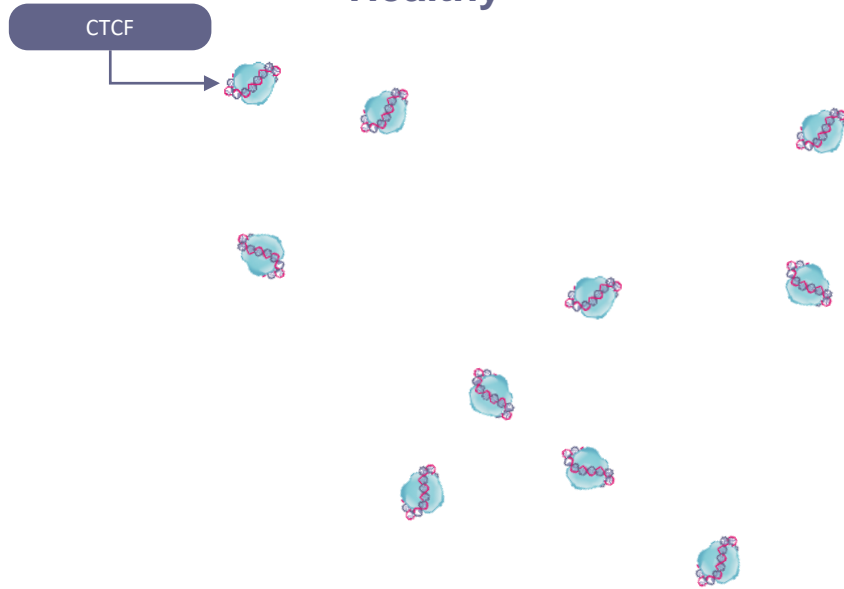


Hypothesis in action

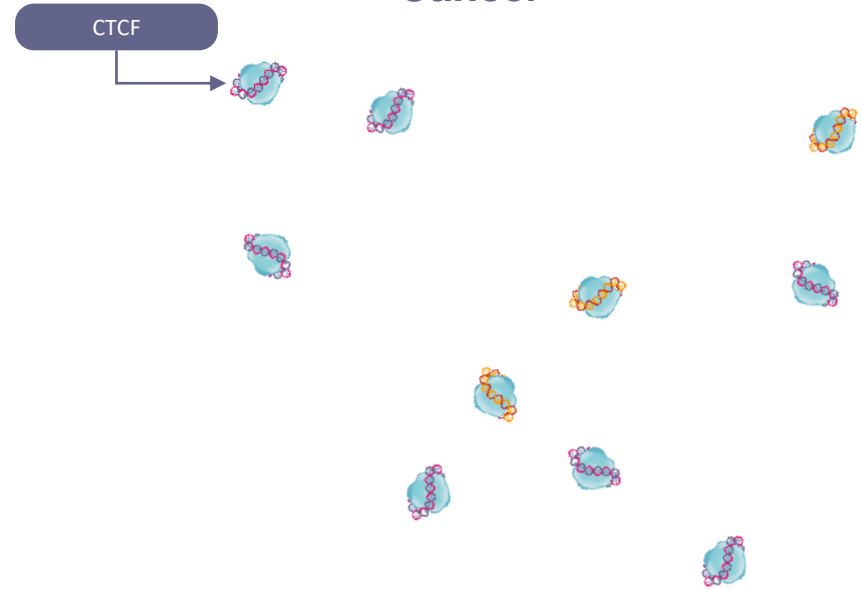
cancer

Physically separate the CTCF from all the nucleosomes

Healthy



Cancer

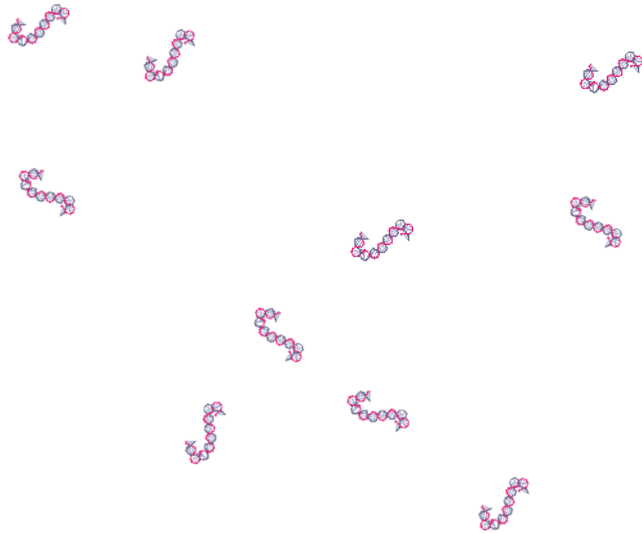


Hypothesis in action

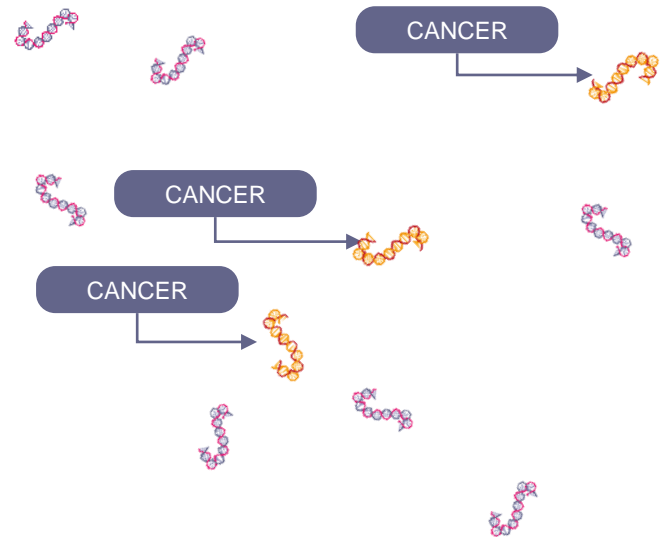
cancer

Then cfDNA fragment with that sequence tumour derived

Healthy



Cancer



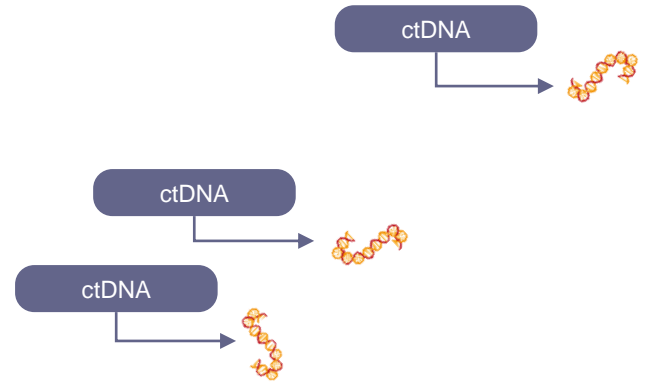
Hypothesis in action

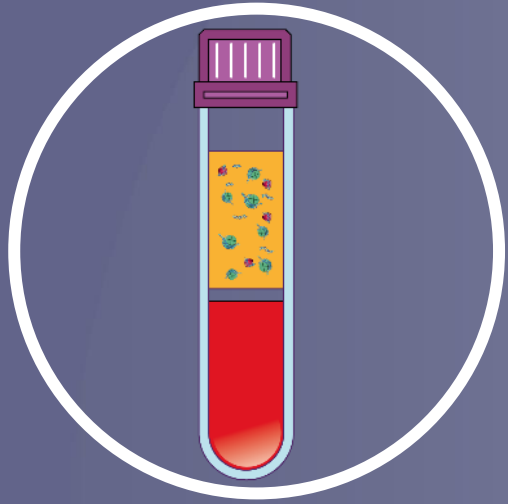
cancer

Healthy

ctDNA

Cancer





Clinical Experiment Results

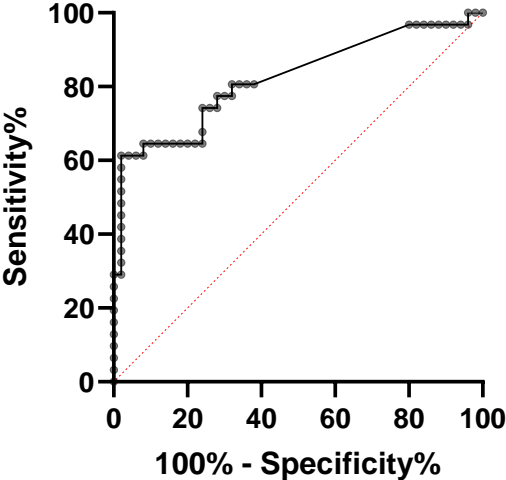
All 10 qPCR assays using CTCF-cfDNA biomarkers discovered for leukemia were effective for detection of leukemia

- Some biomarkers were quite specific for leukemia
- Most biomarkers were also effective for discovery of a variety of common solid cancers.
- Different biomarkers were effective for different solid cancers – including at early stage
- This means it is possible that we will be able to produce tests for particular cancers e.g., a breast cancer test or a prostate cancer test
- We are currently developing biomarkers for breast, prostate, colorectal, lung and liver cancers
- Biomarker discovery is rapid and low cost
- **As CTCF has close to 100,000 genomic binding site loci, the technology opens a whole new field of hitherto unknown potential biomarkers**

A single qPCR assay discovered on leukemia detected 61% of leukemia cases at 98% specificity



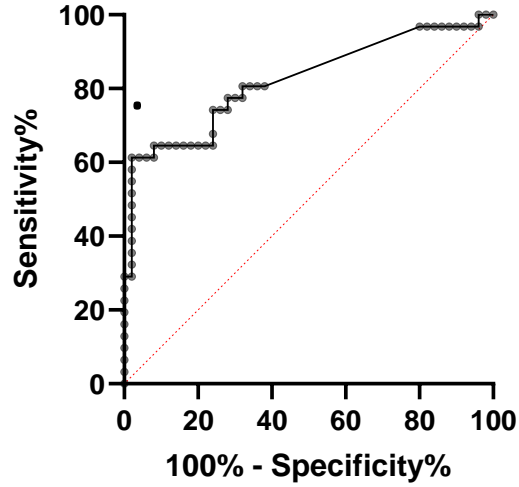
ROC curve: Leukaemia (single qPCR assay)



Healthy	Cancer
1	1
1	0.249
1	1
1	1
0.28	352.139
1	137.663
1	35.383
3.89	4.155
1	1
6.17	41.643
2.85	30.274
6.39	20.252
0.83	4.857
0.66	11.314
5.37	1.58
1.439	1
1	36.504
1.292	10.267
1	985.697
11.043	115.76
0.297	216.77
6.453	36.25
1	142.52
10.703	57.68
10.629	69.55
1	
5.756	
0.745	
61.82	
6.681	
1	
1	
3.149	
6.543	
3.959	
1	
0.299	
6.658	
1	
0.225	
1	
0.25	
1	
0.247	

A single qPCR assay discovered 61% of leukemia detected 98% specificity

ROC curve: Leukaemia (single qPCR assay)

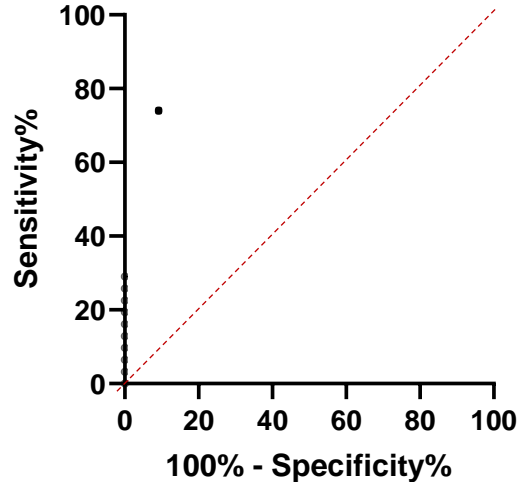


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1	
0.299	
6.658	
1	
0.225	
1	
0.25	
1	
0.247	

Adding a second qPCR detected 74% of leukemia cases at 96% specificity

Another 2-qPCR assay detected 77% of colorectal cancer cases at 92% specificity

ROC curve: Leukaemia (single qPCR assay)



Blood test almost as good as fecal FIT test for colorectal cancer with biomarkers developed on leukemia.

We are now developing individual biomarkers specific for CRC.
Expected Q1 2024.

And more tests for lung, prostate, breast etc

Proof of principle for multi-cancer early detection using a 2-qPCR assay (90% specificity)

cancer

Solid cancers	patients	positive	sensitivity
• CRC	13	9	69%
• Breast	10	5	50%
• Prostate	10	5	50%
• Liver	10	6	60%
TOTAL	43	25	58%

Solid cancers	patients	positive	sensitivity
• Stage I	9	4	44%
• Stage II	9	3	33%
• Stage III	9	6	67%
• Stage IV	16	12	75%
TOTAL	43	25	58%

Comparison CHIP/PCR with traditional NGS/ ctDNA assays

cancer

Traditional NGS/ ctDNA assay

- Cost >\$1000
- Too complex to be fully automated
- High technology labs only
- Turnaround - days or weeks
- Library preparation
- Sequencing
- Bioinformatic computer analysis
- Computer provides the answer

CHIP/PCR

- ~\$100
- Can be automated
- Any hospital lab
- Turnaround 1 day (less if automated)
- No library preparation
- No sequencing
- No computer
- Simple +/- PCR answer (like a COVID-test)



Dr Andrew Retter

Hematologist, Intensive Care, ECMO & Thrombosis
Consultant, Guys and St Thomas' NHS Foundation Trust, UK.

Conflicts of interest to declare

cancer

Consultant to VolitionRX Limited

No conflicts of interests. But just to confirm that these are my own opinions, and I am speaking in an independent capacity from the NHS Trust where I work as a consultant in hematology and intensive care

Cancer is the *second* leading
cause of death worldwide.

Cancer Burden – new cases

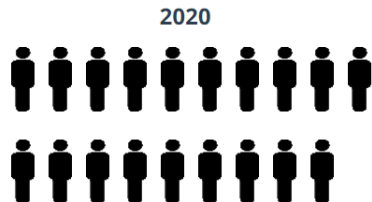
cancer



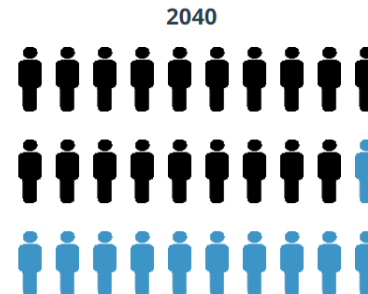
Estimated number of new cases from 2020 to 2040, Both sexes, age [0-85+]

All cancers

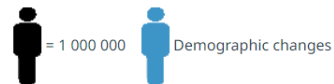
World



19.3M



30.2M



Cancer Burden – death rate

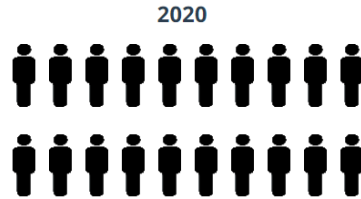
cancer



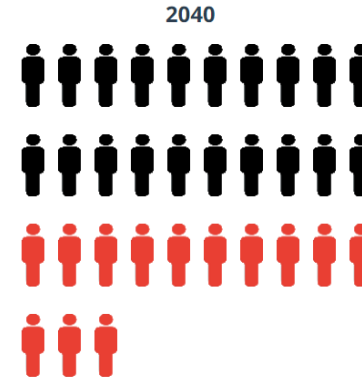
Estimated number of deaths from 2020 to 2040, Both sexes, age [0-85+]

All cancers

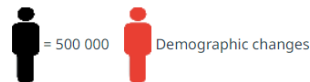
World



9.96M



16.3M



Some further facts and figures...

cancer

- Five-year survival rates are **improving** (from 49% in the mid-1970's to 68% today)
- **Stage at diagnosis** of cancer is an important factor that affects cancer outcomes.
- **Earlier** diagnosis, when cancers are diagnosed at stages I and II as opposed to stages III and IV, is associated with **better** prognosis on average.
- Yet shockingly, ~45% of cancers diagnosed in the UK in 2018 were late-stage i.e. stage III and IV.

How might Volition's technology help?

cancer

- Diagnostic enrichment – early-stage diagnosis
- Monitoring of disease progression and response to treatment

...if we can diagnose earlier and monitor more efficiently outcomes should improve

What's next?

cancer

- Clinical Paper submission, peer review and publication
- Development of specific biomarkers for Lung, CRC, Prostate, Breast and Liver cancers
- Cancer specific abstracts and posters at conferences throughout 2024
- Development of Centers of Excellence and Key Opinion Leaders

In Summary

- Exciting breakthrough method
- A number of “firsts”
- Significant Clinical and Commercial opportunity

The background features a dark blue gradient with several light blue circles of varying sizes. A large, dark blue quote icon is positioned behind the text. The text is centered and reads "Question & Answer Session" in a white, sans-serif font.

**Question
&
Answer Session**