

Code ▾

Galaxy Liver Mets Kataoka et al_08052024 Clinical analysis

```
library(swimplot) library(coxphf) library(grid) library(gtable) library(readr) library(mosaic) library(dplyr) library(survival)
library(survminer) library(gridtext) library(ggplot2) library(scales) library(officer) library(ggthemes) library(tidyverse)
library(gtsummary) library(flextable) library(parameters) library(car) library(grid) library(ComplexHeatmap)
library(readxl) library(janitor) library(rms) library(DT)
```

#Demographics Table

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```

rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]

circ_data_subset <- circ_data %>%
  select(
    Age,
    Gender,
    ECOG,
    PrimSite,
    Mets.Type,
    Hepatectomy,
    NumLiverMetsGroup,
    SizeLiverMetsmmGroup,
    pT,
    pN,
    ACT,
    Oxaliplatin.History,
    Postop.Complication,
    BRAF.V600E,
    RAS,
    MSI,
    RFS.Event,
    OS.months) %>%
  mutate(
    Age = as.numeric(Age),
    Gender = factor(Gender, levels = c("Male", "Female")),
    ECOG = factor(ECOG, levels = c(0, 1)),
    PrimSite = factor(PrimSite, levels = c("Right-sided colon", "Left-sided colon")),
    Mets.Type = factor(Mets.Type, levels = c("Synchronous", "Metachronous")),
    Hepatectomy = factor(Hepatectomy, levels = c("Minor", "Major")),
    NumLiverMetsGroup = factor(NumLiverMetsGroup, levels = c("1", "≥2")),
    SizeLiverMetsmmGroup = factor(SizeLiverMetsmmGroup, levels = c("<50", "≥50")),
    pT = factor(pT, levels = c("T1-T2", "T3-T4")),
    pN = factor(pN, levels = c("N0", "N1-N2")),
    ACT = factor(ACT, levels = c("TRUE", "FALSE"), labels = c("Adjuvant Chemotherapy",
    "Observation")),
    Oxaliplatin.History = factor(Oxaliplatin.History, levels = c("No", "Yes")),
    Postop.Complication = factor(Postop.Complication, levels = c("No", "Yes")),
    BRAF.V600E = factor(BRAF.V600E, levels = c("WT", "MUT"), labels = c("BRAF wt", "BRAF
    V600E")),
    RAS = factor(RAS, levels = c("WT", "MUT"), labels = c("RAS wt", "RAS mut")),
    MSI = factor(MSI, levels = c("MSS", "MSI-High")),
    RFS.Event = factor(RFS.Event, levels = c("TRUE", "FALSE"), labels = c("Recurrence",
    "No Recurrence")),
    OS.months = as.numeric(OS.months))
  table1 <- circ_data_subset %>%
   tbl_summary(
    statistic = list(
      all_continuous() ~ "{median} ({min} - {max})",
      all_categorical() ~ "{n} ({p}%)") %>%

```

```
bold_labels()
table1
```

Characteristic	N = 190 ¹
Age	68 (34 - 85)
Gender	
Male	118 (62%)
Female	72 (38%)
ECOG	
0	186 (98%)
1	4 (2.1%)
PrimSite	
Right-sided colon	50 (26%)
Left-sided colon	140 (74%)
Mets.Type	
Synchronous	64 (34%)
Metachronous	126 (66%)
Hepatectomy	
Minor	159 (84%)
Major	31 (16%)
NumLiverMetsGroup	
1	121 (64%)
≥2	69 (36%)
SizeLiverMetsmmGroup	
<50	180 (95%)
≥50	10 (5.3%)
pT	
T1-T2	23 (12%)

¹ Median (Range); n (%)

Characteristic	N = 190¹
T3-T4	167 (88%)
pN	
N0	77 (41%)
N1-N2	113 (59%)
ACT	
Adjuvant Chemotherapy	48 (25%)
Observation	142 (75%)
Oxaliplatin.History	50 (26%)
Postop.Complication	23 (12%)
BRAF.V600E	
BRAF wt	188 (99%)
BRAF V600E	2 (1.1%)
RAS	
RAS wt	106 (56%)
RAS mut	84 (44%)
MSI	
MSS	188 (99%)
MSI-High	2 (1.1%)
RFS.Event	
Recurrence	95 (50%)
No Recurrence	95 (50%)
OS.months	24 (1 - 48)

¹ Median (Range); n (%)

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```
fit1 <- as_flex_table(
  table1,
  include = everything(),
  return_calls = FALSE,
  strip_md_bold = TRUE)
```

Warning: The `strip_md_bold` argument of `as_flex_table()` is deprecated as of gtsummary 1.6.0.
 This warning is displayed once every 8 hours.
 Call `lifecycle::last_lifecycle_warnings()` to see where this warning was generated.

[Hide](#)

fit1

Characteristic	N = 190 ¹
Age	68 (34 - 85)
Gender	
Male	118 (62%)
Female	72 (38%)
ECOG	
0	186 (98%)
1	4 (2.1%)
PrimSite	
Right-sided colon	50 (26%)
Left-sided colon	140 (74%)
Mets.Type	
Synchronous	64 (34%)
Metachronous	126 (66%)
Hepatectomy	
Minor	159 (84%)
Major	31 (16%)
NumLiverMetsGroup	
1	121 (64%)
≥2	69 (36%)
SizeLiverMetsmmGroup	

¹Median (Range); n (%)

Characteristic	N = 190¹
<50	180 (95%)
≥50	10 (5.3%)
pT	
T1-T2	23 (12%)
T3-T4	167 (88%)
pN	
N0	77 (41%)
N1-N2	113 (59%)
ACT	
Adjuvant Chemotherapy	48 (25%)
Observation	142 (75%)
Oxaliplatin.History	50 (26%)
Postop.Complication	23 (12%)
BRAF.V600E	
BRAF wt	188 (99%)
BRAF V600E	2 (1.1%)
RAS	
RAS wt	106 (56%)
RAS mut	84 (44%)
MSI	
MSS	188 (99%)
MSI-High	2 (1.1%)
RFS.Event	
Recurrence	95 (50%)
No Recurrence	95 (50%)
OS.months	24 (1 - 48)

¹Median (Range); n (%)

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```
save_as_docx(fit1, path= "~/Downloads/table1.docx")
```

#Demographics Table by MRD ctDNA Status

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```

rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]

circ_data_subset1 <- circ_data %>%
  select(
    Age,
    Gender,
    ECOG,
    PrimSite,
    Mets.Type,
    Hepatectomy,
    NumLiverMetsGroup,
    SizeLiverMetsmmGroup,
    pT,
    pN,
    ACT,
    Oxaliplatin.History,
    Postop.Complication,
    BRAF.V600E,
    RAS,
    MSI) %>%
  mutate(
    Age = as.numeric(Age),
    Gender = factor(Gender, levels = c("Male", "Female")),
    ECOG = factor(ECOG, levels = c(0, 1)),
    PrimSite = factor(PrimSite, levels = c("Right-sided colon", "Left-sided colon")),
    Mets.Type = factor(Mets.Type, levels = c("Synchronous", "Metachronous")),
    Hepatectomy = factor(Hepatectomy, levels = c("Minor", "Major")),
    NumLiverMetsGroup = factor(NumLiverMetsGroup, levels = c("1", "≥2")),
    SizeLiverMetsmmGroup = factor(SizeLiverMetsmmGroup, levels = c("<50", "≥50")),
    pT = factor(pT, levels = c("T1-T2", "T3-T4")),
    pN = factor(pN, levels = c("N0", "N1-N2")),
    ACT = factor(ACT, levels = c("TRUE", "FALSE"), labels = c("Adjuvant Chemotherapy",
    "Observation")),
    Oxaliplatin.History = factor(Oxaliplatin.History, levels = c("No", "Yes")),
    Postop.Complication = factor(Postop.Complication, levels = c("No", "Yes")),
    BRAF.V600E = factor(BRAF.V600E, levels = c("WT", "MUT"), labels = c("BRAF wt", "BRAF
    V600E")),
    RAS = factor(RAS, levels = c("WT", "MUT"), labels = c("RAS wt", "RAS mut")),
    MSI = factor(MSI, levels = c("MSS", "MSI-High")))

circ_data1 <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data1 <- circ_data1[circ_data1$LiverMets=="TRUE",]

circ_data_subset2 <- circ_data %>%
  select(
    Age,
    Gender,
    ECOG,
    PrimSite,

```

```

Mets.Type,
Hepatectomy,
NumLiverMetsGroup,
SizeLiverMetsmmGroup,
pT,
pN,
ACT,
Oxaliplatin.History,
Postop.Complication,
BRAF.V600E,
RAS,
MSI,
ctDNA.MRD) %>%
mutate(
  Age = as.numeric(Age),
  Gender = factor(Gender, levels = c("Male", "Female")),
  ECOG = factor(ECOG, levels = c(0, 1)),
  PrimSite = factor(PrimSite, levels = c("Right-sided colon", "Left-sided colon")),
  Mets.Type = factor(Mets.Type, levels = c("Synchronous", "Metachronous")),
  Hepatectomy = factor(Hepatectomy, levels = c("Minor", "Major")),
  NumLiverMetsGroup = factor(NumLiverMetsGroup, levels = c("1", "≥2")),
  SizeLiverMetsmmGroup = factor(SizeLiverMetsmmGroup, levels = c("<50", "≥50")),
  pT = factor(pT, levels = c("T1-T2", "T3-T4")),
  pN = factor(pN, levels = c("N0", "N1-N2")),
  ACT = factor(ACT, levels = c("TRUE", "FALSE"), labels = c("Adjuvant Chemotherapy",
  "Observation")),
  Oxaliplatin.History = factor(Oxaliplatin.History, levels = c("No", "Yes")),
  Postop.Complication = factor(Postop.Complication, levels = c("No", "Yes")),
  BRAF.V600E = factor(BRAF.V600E, levels = c("WT", "MUT"), labels = c("BRAF wt", "BRAF
  V600E")),
  RAS = factor(RAS, levels = c("WT", "MUT"), labels = c("RAS wt", "RAS mut")),
  MSI = factor(MSI, levels = c("MSS", "MSI-High")),
  ctDNA.MRD = factor(ctDNA.MRD, levels = c("NEGATIVE", "POSITIVE")))
Overall <- circ_data_subset1 %>%
 tbl_summary(
  statistic = list(
    all_continuous() ~ "{median} ({min} - {max})",
    all_categorical() ~ "{n} ({p}%)") %>%
  bold_labels()
Overall

```

Characteristic	N = 190 ¹
Age	68 (34 - 85)
Gender	
Male	118 (62%)
Female	72 (38%)

¹ Median (Range); n (%)

Characteristic	N = 190 ¹
ECOG	
0	186 (98%)
1	4 (2.1%)
PrimSite	
Right-sided colon	50 (26%)
Left-sided colon	140 (74%)
Mets.Type	
Synchronous	64 (34%)
Metachronous	126 (66%)
Hepatectomy	
Minor	159 (84%)
Major	31 (16%)
NumLiverMetsGroup	
1	121 (64%)
≥2	69 (36%)
SizeLiverMetsmmGroup	
<50	180 (95%)
≥50	10 (5.3%)
pT	
T1-T2	23 (12%)
T3-T4	167 (88%)
pN	
N0	77 (41%)
N1-N2	113 (59%)
ACT	
Adjuvant Chemotherapy	48 (25%)

¹ Median (Range); n (%)

Characteristic	N = 190 ¹
Observation	142 (75%)
Oxaliplatin.History	50 (26%)
Postop.Complication	23 (12%)
BRAF.V600E	
BRAF wt	188 (99%)
BRAF V600E	2 (1.1%)
RAS	
RAS wt	106 (56%)
RAS mut	84 (44%)
MSI	
MSS	188 (99%)
MSI-High	2 (1.1%)

¹ Median (Range); n (%)

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```
ByctDNA_MRD <- circ_data_subset2 %>%
 tbl_summary(
  by = ctDNA.MRD, # Subgroup by ctDNA.MRD
  statistic = list(
    all_continuous() ~ "{median} ({min} - {max})",
    all_categorical() ~ "{n} ({p}%)") %>%
  add_p(test = all_categorical() ~ "fisher.test", pvalue_fun = ~style_pvalue(p.adjust(.,
  x, method = "bonferroni"))) %>%
  bold_labels()
ByctDNA_MRD
```

Characteristic	NEGATIVE, N = 129 ¹	POSITIVE, N = 61 ¹	p-value ²
Age	69 (37 - 85)	68 (34 - 84)	>0.9
Gender			>0.9
Male	83 (64%)	35 (57%)	

¹ Median (Range); n (%)

² Wilcoxon rank sum test; Fisher's exact test

Characteristic	NEGATIVE, N = 129 ¹	POSITIVE, N = 61 ¹	p-value ²
Female	46 (36%)	26 (43%)	
ECOG			>0.9
0	127 (98%)	59 (97%)	
1	2 (1.6%)	2 (3.3%)	
PrimSite			0.6
Right-sided colon	40 (31%)	10 (16%)	
Left-sided colon	89 (69%)	51 (84%)	
Mets.Type			0.015
Synchronous	33 (26%)	31 (51%)	
Metachronous	96 (74%)	30 (49%)	
Hepatectomy			>0.9
Minor	112 (87%)	47 (77%)	
Major	17 (13%)	14 (23%)	
NumLiverMetsGroup			0.010
1	93 (72%)	28 (46%)	
≥2	36 (28%)	33 (54%)	
SizeLiverMetsmmGroup			>0.9
<50	123 (95%)	57 (93%)	
≥50	6 (4.7%)	4 (6.6%)	
pT			>0.9
T1-T2	16 (12%)	7 (11%)	
T3-T4	113 (88%)	54 (89%)	
pN			0.3
N0	60 (47%)	17 (28%)	
N1-N2	69 (53%)	44 (72%)	

¹ Median (Range); n (%)² Wilcoxon rank sum test; Fisher's exact test

Characteristic	NEGATIVE, N = 129 ¹	POSITIVE, N = 61 ¹	p-value ²
ACT	>0.9		
Adjuvant Chemotherapy	33 (26%)	15 (25%)	
Observation	96 (74%)	46 (75%)	
Oxaliplatin.History	39 (30%)	11 (18%)	>0.9
Postop.ComPLICATION	10 (7.8%)	13 (21%)	0.2
BRAF.V600E	>0.9		
BRAF wt	127 (98%)	61 (100%)	
BRAF V600E	2 (1.6%)	0 (0%)	
RAS	>0.9		
RAS wt	74 (57%)	32 (52%)	
RAS mut	55 (43%)	29 (48%)	
MSI	>0.9		
MSS	128 (99%)	60 (98%)	
MSI-High	1 (0.8%)	1 (1.6%)	

¹ Median (Range); n (%)² Wilcoxon rank sum test; Fisher's exact testHide

```
merged_table <-tbl_merge(tbls=list(Overall, ByctDNA_MR))  
merged_table
```

Characteristic	Table 1		Table 2	
	N = 190 ¹	NEGATIVE, N = 129 ¹	POSITIVE, N = 61 ¹	p-value ²
Age	68 (34 - 85)	69 (37 - 85)	68 (34 - 84)	>0.9
Gender				>0.9
Male	118 (62%)	83 (64%)	35 (57%)	
Female	72 (38%)	46 (36%)	26 (43%)	

¹ Median (Range); n (%)

² Wilcoxon rank sum test; Fisher's exact test

Characteristic	Table 1		Table 2	
	N = 190 ¹	NEGATIVE, N = 129 ¹	POSITIVE, N = 61 ¹	p-value ²
ECOG				>0.9
0	186 (98%)	127 (98%)	59 (97%)	
1	4 (2.1%)	2 (1.6%)	2 (3.3%)	
PrimSite				0.6
Right-sided colon	50 (26%)	40 (31%)	10 (16%)	
Left-sided colon	140 (74%)	89 (69%)	51 (84%)	
Mets.Type				0.015
Synchronous	64 (34%)	33 (26%)	31 (51%)	
Metachronous	126 (66%)	96 (74%)	30 (49%)	
Hepatectomy				>0.9
Minor	159 (84%)	112 (87%)	47 (77%)	
Major	31 (16%)	17 (13%)	14 (23%)	
NumLiverMetsGroup				0.010
1	121 (64%)	93 (72%)	28 (46%)	
≥2	69 (36%)	36 (28%)	33 (54%)	
SizeLiverMetsmmGroup				>0.9
<50	180 (95%)	123 (95%)	57 (93%)	
≥50	10 (5.3%)	6 (4.7%)	4 (6.6%)	
pT				>0.9
T1-T2	23 (12%)	16 (12%)	7 (11%)	
T3-T4	167 (88%)	113 (88%)	54 (89%)	
pN				0.3
N0	77 (41%)	60 (47%)	17 (28%)	
N1-N2	113 (59%)	69 (53%)	44 (72%)	

¹ Median (Range); n (%)² Wilcoxon rank sum test; Fisher's exact test

Characteristic	Table 1		Table 2	
	N = 190 ¹	NEGATIVE, N = 129 ¹	POSITIVE, N = 61 ¹	p-value ²
ACT	>0.9			
Adjuvant Chemotherapy	48 (25%)	33 (26%)	15 (25%)	
Observation	142 (75%)	96 (74%)	46 (75%)	
Oxaliplatin.History	50 (26%)	39 (30%)	11 (18%)	>0.9
Postop.Complication	23 (12%)	10 (7.8%)	13 (21%)	0.2
BRAF.V600E	>0.9			
BRAF wt	188 (99%)	127 (98%)	61 (100%)	
BRAF V600E	2 (1.1%)	2 (1.6%)	0 (0%)	
RAS	>0.9			
RAS wt	106 (56%)	74 (57%)	32 (52%)	
RAS mut	84 (44%)	55 (43%)	29 (48%)	
MSI	>0.9			
MSS	188 (99%)	128 (99%)	60 (98%)	
MSI-High	2 (1.1%)	1 (0.8%)	1 (1.6%)	

¹ Median (Range); n (%)² Wilcoxon rank sum test; Fisher's exact test[Hide](#)

```
fit1 <- as_flex_table(
  merged_table,
  include = everything(),
  return_calls = FALSE,
  strip_md_bold = TRUE)
fit1
```

Characteristic	Table 1		Table 2	
	N = 190 ¹	NEGATIVE, N = 129 ¹	POSITIVE, N = 61 ¹	p-value ²
Age	68 (34 - 85)	69 (37 - 85)	68 (34 - 84)	>0.9
Gender				>0.9

¹Median (Range); n (%)²Wilcoxon rank sum test; Fisher's exact test

Characteristic	Table 1	Table 2		p-value²
	N = 190¹	NEGATIVE, N = 129¹	POSITIVE, N = 61¹	
Male	118 (62%)	83 (64%)	35 (57%)	
Female	72 (38%)	46 (36%)	26 (43%)	
ECOG				>0.9
0	186 (98%)	127 (98%)	59 (97%)	
1	4 (2.1%)	2 (1.6%)	2 (3.3%)	
PrimSite				0.6
Right-sided colon	50 (26%)	40 (31%)	10 (16%)	
Left-sided colon	140 (74%)	89 (69%)	51 (84%)	
Mets.Type				0.015
Synchronous	64 (34%)	33 (26%)	31 (51%)	
Metachronous	126 (66%)	96 (74%)	30 (49%)	
Hepatectomy				>0.9
Minor	159 (84%)	112 (87%)	47 (77%)	
Major	31 (16%)	17 (13%)	14 (23%)	
NumLiverMetsGroup				0.010
1	121 (64%)	93 (72%)	28 (46%)	
≥2	69 (36%)	36 (28%)	33 (54%)	
SizeLiverMetsmmGroup				>0.9
<50	180 (95%)	123 (95%)	57 (93%)	
≥50	10 (5.3%)	6 (4.7%)	4 (6.6%)	
pT				>0.9
T1-T2	23 (12%)	16 (12%)	7 (11%)	
T3-T4	167 (88%)	113 (88%)	54 (89%)	
pN				0.3
N0	77 (41%)	60 (47%)	17 (28%)	
N1-N2	113 (59%)	69 (53%)	44 (72%)	
ACT				>0.9
Adjuvant Chemotherapy	48 (25%)	33 (26%)	15 (25%)	
Observation	142 (75%)	96 (74%)	46 (75%)	

¹Median (Range); n (%)²Wilcoxon rank sum test; Fisher's exact test

Characteristic	Table 1		Table 2	
	N = 190 ¹	NEGATIVE, N = 129 ¹	POSITIVE, N = 61 ¹	p-value ²
Oxaliplatin.History	50 (26%)	39 (30%)	11 (18%)	>0.9
Postop.Complication	23 (12%)	10 (7.8%)	13 (21%)	0.2
BRAF.V600E				>0.9
BRAF wt	188 (99%)	127 (98%)	61 (100%)	
BRAF V600E	2 (1.1%)	2 (1.6%)	0 (0%)	
RAS				>0.9
RAS wt	106 (56%)	74 (57%)	32 (52%)	
RAS mut	84 (44%)	55 (43%)	29 (48%)	
MSI				>0.9
MSS	188 (99%)	128 (99%)	60 (98%)	
MSI-High	2 (1.1%)	1 (0.8%)	1 (1.6%)	

¹Median (Range); n (%)

²Wilcoxon rank sum test; Fisher's exact test

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```
save_as_docx(fit1, path= "~/Downloads/merged_table.docx")
```

#ctDNA Detection Rates by Window and Stages

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```

#ctDNA at Baseline
rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$ctDNA.Baseline!="",]
circ_data$ctDNA.Baseline <- factor(circ_data$ctDNA.Baseline, levels=c("NEGATIVE","POSITIVE"))
circ_data$Stage <- factor(circ_data$Stage, levels=c("IV"))
positive_counts_by_stage <- aggregate(circ_data$ctDNA.Baseline == "POSITIVE", by=list(circ_data$Stage), FUN=sum)
total_counts_by_stage <- aggregate(circ_data$ctDNA.Baseline, by=list(circ_data$Stage), FUN=length)
combined_data <- data.frame(
  Stage = total_counts_by_stage$Group.1,
  Total_Count = total_counts_by_stage$x,
  Positive_Count = positive_counts_by_stage$x,
  Rate = (positive_counts_by_stage$x / total_counts_by_stage$x) * 100 # Convert to percentage
)
combined_data$Rate <- sprintf("%.2f%%", combined_data$Rate)
overall_total_count <- nrow(circ_data)
overall_positive_count <- nrow(circ_data[circ_data$ctDNA.Baseline == "POSITIVE",])
overall_positivity_rate <- (overall_positive_count / overall_total_count) * 100 # Convert to percentage
overall_row <- data.frame(
  Stage = "Overall",
  Total_Count = overall_total_count,
  Positive_Count = overall_positive_count,
  Rate = sprintf("%.2f%%", overall_positivity_rate)
)
combined_data <- rbind(combined_data, overall_row)
print(combined_data)

```

Stage	Total_Count	Positive_Count	Rate
<fctr>	<int>	<int>	<chr>
IV	189	186	98.41%
Overall	189	186	98.41%
2 rows			

Hide

```
#ctDNA at MRD Window
rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$ctDNA.MRD!="",]
circ_data$ctDNA.MRD <- factor(circ_data$ctDNA.MRD, levels=c("NEGATIVE","POSITIVE"))
circ_data$Stage <- factor(circ_data$Stage, levels=c("IV"))
positive_counts_by_stage <- aggregate(circ_data$ctDNA.MRD == "POSITIVE", by=list(circ_data$Stage), FUN=sum)
total_counts_by_stage <- aggregate(circ_data$ctDNA.MRD, by=list(circ_data$Stage), FUN=length)
combined_data <- data.frame(
  Stage = total_counts_by_stage$Group.1,
  Total_Count = total_counts_by_stage$x,
  Positive_Count = positive_counts_by_stage$x,
  Rate = (positive_counts_by_stage$x / total_counts_by_stage$x) * 100 # Convert to percentage
)
combined_data$Rate <- sprintf("%.2f%%", combined_data$Rate)
overall_total_count <- nrow(circ_data)
overall_positive_count <- nrow(circ_data[circ_data$ctDNA.MRD == "POSITIVE",])
overall_positivity_rate <- (overall_positive_count / overall_total_count) * 100 # Convert to percentage
overall_row <- data.frame(
  Stage = "Overall",
  Total_Count = overall_total_count,
  Positive_Count = overall_positive_count,
  Rate = sprintf("%.2f%%", overall_positivity_rate)
)
combined_data <- rbind(combined_data, overall_row)
print(combined_data)
```

Stage	Total_Count	Positive_Count	Rate
		<int>	<chr>
IV	190	61	32.11%
Overall	190	61	32.11%
2 rows			

Hide

```
#ctDNA at Surveillance Window
rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data$ctDNA.Surveillance <- factor(circ_data$ctDNA.Surveillance, levels=c("NEGATIVE", "POSITIVE"))
circ_data <- subset(circ_data, ctDNA.Surveillance %in% c("NEGATIVE", "POSITIVE"))
circ_data$Stage <- factor(circ_data$Stage, levels=c("IV"))
positive_counts_by_stage <- aggregate(circ_data$ctDNA.Surveillance == "POSITIVE", by=list(circ_data$Stage), FUN=sum)
total_counts_by_stage <- aggregate(circ_data$ctDNA.Surveillance, by=list(circ_data$Stage), FUN=length)
combined_data <- data.frame(
  Stage = total_counts_by_stage$Group.1,
  Total_Count = total_counts_by_stage$x,
  Positive_Count = positive_counts_by_stage$x,
  Rate = (positive_counts_by_stage$x / total_counts_by_stage$x) * 100 # Convert to percentage
)
combined_data$Rate <- sprintf("%.2f%%", combined_data$Rate)
overall_total_count <- nrow(circ_data)
overall_positive_count <- nrow(circ_data[circ_data$ctDNA.Surveillance == "POSITIVE",])
overall_positivity_rate <- (overall_positive_count / overall_total_count) * 100 # Convert to percentage
overall_row <- data.frame(
  Stage = "Overall",
  Total_Count = overall_total_count,
  Positive_Count = overall_positive_count,
  Rate = sprintf("%.2f%%", overall_positivity_rate)
)
combined_data <- rbind(combined_data, overall_row)
print(combined_data)
```

Stage	Total_Count	Positive_Count	Rate
		<int>	<chr>
IV	154	55	35.71%
Overall	154	55	35.71%
2 rows			

#ctDNA Detection Rates by Window and Synchronicity

[Hide](#)

```

#ctDNA at Baseline
rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$ctDNA.Baseline!="",]
circ_data$ctDNA.Baseline <- factor(circ_data$ctDNA.Baseline, levels=c("NEGATIVE", "POSITIVE"))
circ_data$Mets.Type <- factor(circ_data$Mets.Type, levels = c("Metachronous", "Synchronous"))
positive_counts_by_stage <- aggregate(circ_data$ctDNA.Baseline == "POSITIVE", by=list(circ_data$Mets.Type), FUN=sum)
total_counts_by_stage <- aggregate(circ_data$ctDNA.Baseline, by=list(circ_data$Mets.Type), FUN=length)
combined_data <- data.frame(
  Stage = total_counts_by_stage$Group.1,
  Total_Count = total_counts_by_stage$x,
  Positive_Count = positive_counts_by_stage$x,
  Rate = (positive_counts_by_stage$x / total_counts_by_stage$x) * 100 # Convert to percentage
)
combined_data$Rate <- sprintf("%.2f%%", combined_data$Rate)
overall_total_count <- nrow(circ_data)
overall_positive_count <- nrow(circ_data[circ_data$ctDNA.Baseline == "POSITIVE",])
overall_positivity_rate <- (overall_positive_count / overall_total_count) * 100 # Convert to percentage
overall_row <- data.frame(
  Stage = "Overall",
  Total_Count = overall_total_count,
  Positive_Count = overall_positive_count,
  Rate = sprintf("%.2f%%", overall_positivity_rate)
)
combined_data <- rbind(combined_data, overall_row)
print(combined_data)

```

Stage	Total_Count	Positive_Count	Rate
<fctr>	<int>	<int>	<chr>
Metachronous	125	122	97.60%
Synchronous	64	64	100.00%
Overall	189	186	98.41%
3 rows			

Hide

```

#ctDNA at MRD Window
rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$ctDNA.MRD!="",]
circ_data$ctDNA.MRD <- factor(circ_data$ctDNA.MRD, levels=c("NEGATIVE","POSITIVE"))
circ_data$Mets.Type <- factor(circ_data$Mets.Type, levels = c("Metachronous", "Synchronous"))
positive_counts_by_stage <- aggregate(circ_data$ctDNA.MRD == "POSITIVE", by=list(circ_data$Mets.Type), FUN=sum)
total_counts_by_stage <- aggregate(circ_data$ctDNA.MRD, by=list(circ_data$Mets.Type), FUN=length)
combined_data <- data.frame(
  Mets.Type = total_counts_by_stage$Group.1,
  Total_Count = total_counts_by_stage$x,
  Positive_Count = positive_counts_by_stage$x,
  Rate = (positive_counts_by_stage$x / total_counts_by_stage$x) * 100 # Convert to percentage
)
combined_data$Rate <- sprintf("%.2f%%", combined_data$Rate)
overall_total_count <- nrow(circ_data)
overall_positive_count <- nrow(circ_data[circ_data$ctDNA.MRD == "POSITIVE",])
overall_positivity_rate <- (overall_positive_count / overall_total_count) * 100 # Convert to percentage
overall_row <- data.frame(
  Mets.Type = "Overall",
  Total_Count = overall_total_count,
  Positive_Count = overall_positive_count,
  Rate = sprintf("%.2f%%", overall_positivity_rate)
)
combined_data <- rbind(combined_data, overall_row)
print(combined_data)

```

Mets.Type	Total_Count	Positive_Count	Rate
		<int>	<chr>
Metachronous	126	30	23.81%
Synchronous	64	31	48.44%
Overall	190	61	32.11%
3 rows			

Hide

```

#ctDNA at Surveillance Window
rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data$ctDNA.Surveillance <- factor(circ_data$ctDNA.Surveillance, levels=c("NEGATIVE", "POSITIVE"))
circ_data <- subset(circ_data, ctDNA.Surveillance %in% c("NEGATIVE", "POSITIVE"))
circ_data$Mets.Type <- factor(circ_data$Mets.Type, levels = c("Metachronous", "Synchronous"))
positive_counts_by_stage <- aggregate(circ_data$ctDNA.Surveillance == "POSITIVE", by=list(circ_data$Mets.Type), FUN=sum)
total_counts_by_stage <- aggregate(circ_data$ctDNA.Surveillance, by=list(circ_data$Mets.Type), FUN=length)
combined_data <- data.frame(
  Mets.Type = total_counts_by_stage$Group.1,
  Total_Count = total_counts_by_stage$x,
  Positive_Count = positive_counts_by_stage$x,
  Rate = (positive_counts_by_stage$x / total_counts_by_stage$x) * 100 # Convert to percentage
)
combined_data$Rate <- sprintf("%.2f%%", combined_data$Rate)
overall_total_count <- nrow(circ_data)
overall_positive_count <- nrow(circ_data[circ_data$ctDNA.Surveillance == "POSITIVE",])
overall_positivity_rate <- (overall_positive_count / overall_total_count) * 100 # Convert to percentage
overall_row <- data.frame(
  Mets.Type = "Overall",
  Total_Count = overall_total_count,
  Positive_Count = overall_positive_count,
  Rate = sprintf("%.2f%%", overall_positivity_rate)
)
combined_data <- rbind(combined_data, overall_row)
print(combined_data)

```

Mets.Type	Total_Count	Positive_Count	Rate
	<int>	<int>	<chr>
Metachronous	102	30	29.41%
Synchronous	52	25	48.08%
Overall	154	55	35.71%
3 rows			

#Barplot with Detection Rates at the MRD and Surveillance Windows by Synchronicity

Hide

```
#Detection rate at the Surveillance Window
rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]

circ_data$ctDNA.MRD <- factor(circ_data$ctDNA.MRD, levels = c("NEGATIVE", "POSITIVE"), labels = c("Negative", "Positive"))
circ_data$Mets.Type <- factor(circ_data$Mets.Type, levels = c("Metachronous", "Synchronous"))
contingency_table <- table(circ_data$Mets.Type, circ_data$ctDNA.MRD)
chi_square_test <- chisq.test(contingency_table)
print(chi_square_test)
```

Pearson's Chi-squared test with Yates' continuity correction

```
data: contingency_table
X-squared = 10.707, df = 1, p-value = 0.001067
```

[Hide](#)

```
fisher_exact_test <- fisher.test(contingency_table)
print(fisher_exact_test)
```

Fisher's Exact Test for Count Data

```
data: contingency_table
p-value = 0.0009273
alternative hypothesis: true odds ratio is not equal to 1
95 percent confidence interval:
1.507768 5.977774
sample estimates:
odds ratio
2.987004
```

[Hide](#)

```
print(contingency_table)
```

	Negative	Positive
Metachronous	96	30
Synchronous	33	31

[Hide](#)

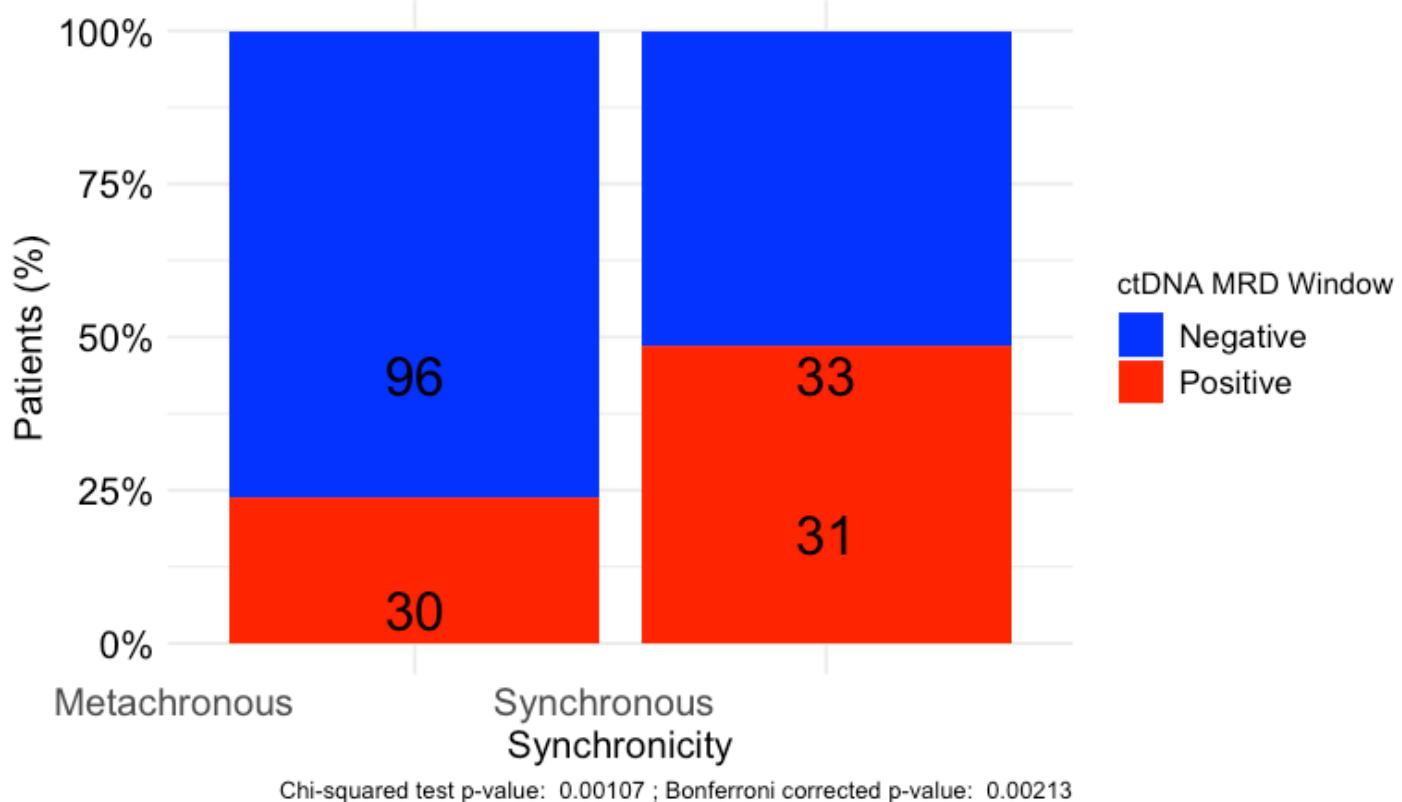
```
p_values <- c(chi_square_test$p.value, fisher_exact_test$p.value)
p_adjusted <- p.adjust(p_values, method = "bonferroni")
names(p_adjusted) <- c("Chi-Square Test", "Fisher's Exact Test")
print(p_adjusted)
```

```
Chi-Square Test Fisher's Exact Test
0.002134653 0.001854561
```

[Hide](#)

```
table_df <- as.data.frame(contingency_table)
table_df$Total <- ave(table_df$Freq, table_df$Var1, FUN = sum)
table_df$Percentage <- table_df$Freq / table_df$Total
table_df$MiddlePercentage <- table_df$Percentage / 2
ggplot(table_df, aes(x = Var1, y = Percentage, fill = Var2)) +
  geom_bar(stat = "identity") +
  geom_text(aes(y = MiddlePercentage, label = Freq), position = "stack", color = "black", vjust = 1.5, size = 7) +
  theme_minimal() +
  labs(title = "Patients with CLM - ctDNA MRD Window",
       x = "Synchronicity",
       y = "Patients (%)",
       fill = "ctDNA MRD Window",
       caption = paste("Chi-squared test p-value: ", format.pval(chi_square_test$p.value), "; Bonferroni corrected p-value: ", format.pval(p_adjusted["Chi-Square Test"]))) +
  scale_y_continuous(labels = scales::percent_format()) +
  scale_fill_manual(values = c("Negative" = "blue", "Positive" = "red")) + # define custom colors
  theme(axis.text.x = element_text(angle = 0, hjust = 1.5, size = 14), # increase x-axis text size
        axis.text.y = element_text(size = 14, color = "black"), # increase y-axis text size
        axis.title.x = element_text(size = 14, color = "black"), # increase x-axis label size
        axis.title.y = element_text(size = 14, color = "black"), # increase y-axis label size
        legend.text = element_text(size = 12, color = "black")) # increase Recurrence label size
```

Patients with CLM - ctDNA MRD Window



Hide

```
#Detection rate at the Surveillance Window
rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]

circ_data$ctDNA.Surveillance <- factor(circ_data$ctDNA.Surveillance, levels = c("NEGATIVE", "POSITIVE"), labels = c("Negative", "Positive"))
circ_data$Mets.Type <- factor(circ_data$Mets.Type, levels = c("Metachronous", "Synchronous"))
contingency_table <- table(circ_data$Mets.Type, circ_data$ctDNA.Surveillance)
chi_square_test <- chisq.test(contingency_table)
print(chi_square_test)
```

Pearson's Chi-squared test with Yates' continuity correction

```
data: contingency_table
X-squared = 4.4449, df = 1, p-value = 0.03501
```

Hide

```
fisher_exact_test <- fisher.test(contingency_table)
print(fisher_exact_test)
```

Fisher's Exact Test for Count Data

```
data: contingency_table
p-value = 0.03232
alternative hypothesis: true odds ratio is not equal to 1
95 percent confidence interval:
1.048856 4.688132
sample estimates:
odds ratio
2.210114
```

[Hide](#)

```
print(contingency_table)
```

	Negative	Positive
Metachronous	72	30
Synchronous	27	25

[Hide](#)

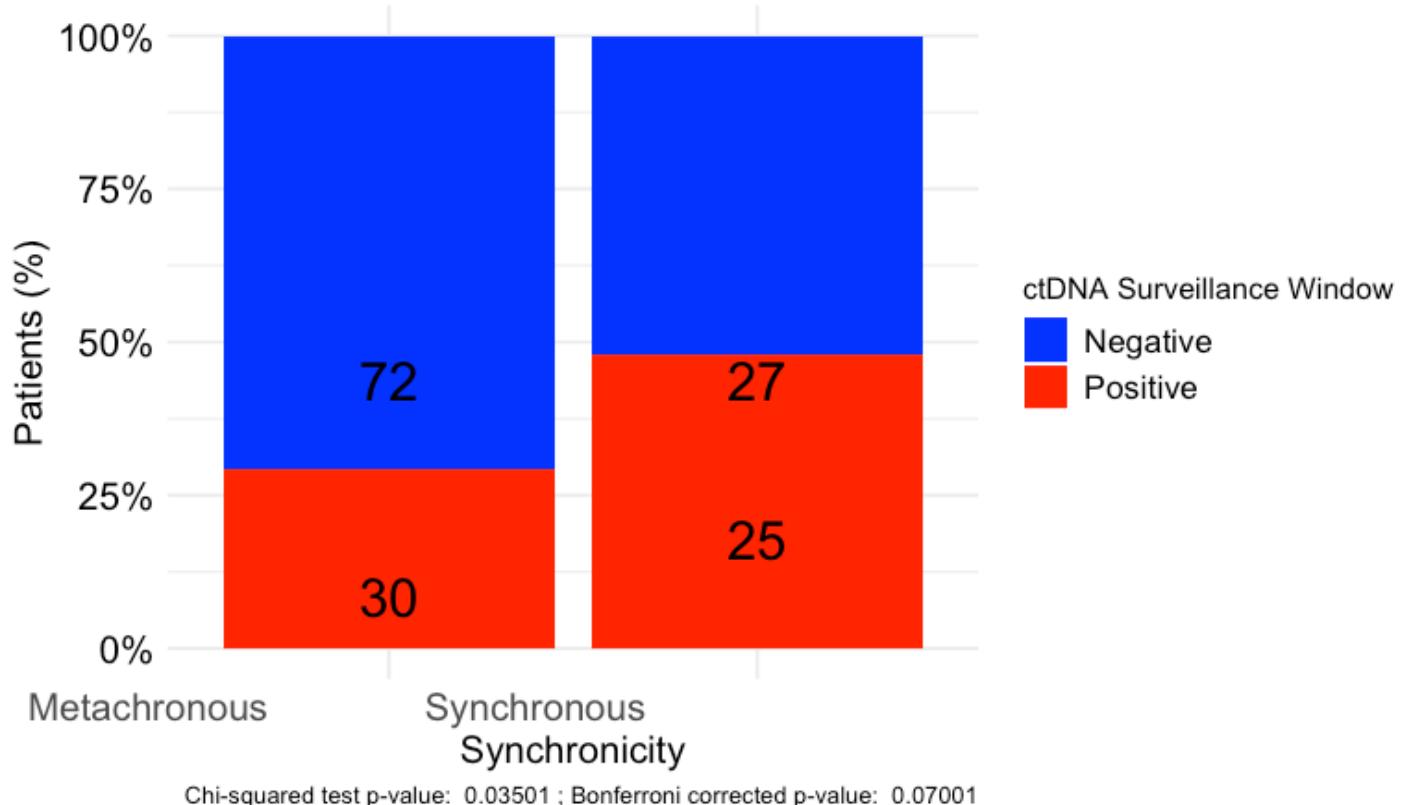
```
p_values <- c(chi_square_test$p.value, fisher_exact_test$p.value)
p_adjusted <- p.adjust(p_values, method = "bonferroni")
names(p_adjusted) <- c("Chi-Square Test", "Fisher's Exact Test")
print(p_adjusted)
```

Chi-Square Test	Fisher's Exact Test
0.07001175	0.06464745

[Hide](#)

```
table_df <- as.data.frame(contingency_table)
table_df$Total <- ave(table_df$Freq, table_df$Var1, FUN = sum)
table_df$Percentage <- table_df$Freq / table_df$Total
table_df$MiddlePercentage <- table_df$Percentage / 2
ggplot(table_df, aes(x = Var1, y = Percentage, fill = Var2)) +
  geom_bar(stat = "identity") +
  geom_text(aes(y = MiddlePercentage, label = Freq), position = "stack", color = "black", vjust = 1.5, size = 7) +
  theme_minimal() +
  labs(title = "Patients with CLM - ctDNA Surveillance Window",
       x = "Synchronicity",
       y = "Patients (%)",
       fill = "ctDNA Surveillance Window",
       caption = paste("Chi-squared test p-value: ", format.pval(chi_square_test$p.value), "; Bonferroni corrected p-value: ", format.pval(p.adjusted["Chi-Square Test"]))) +
  scale_y_continuous(labels = scales::percent_format()) +
  scale_fill_manual(values = c("Negative" = "blue", "Positive" = "red")) + # define custom colors
  theme(axis.text.x = element_text(angle = 0, hjust = 1.5, size = 14), # increase x-axis text size
        axis.text.y = element_text(size = 14, color = "black"), # increase y-axis text size
        axis.title.x = element_text(size = 14, color = "black"), # increase x-axis label size
        axis.title.y = element_text(size = 14, color = "black"), # increase y-axis label size
        legend.text = element_text(size = 12, color = "black")) # increase Recurrence label size
```

Patients with CLM - ctDNA Surveillance Window



#Barplot with Detection Rates at pre-op and ctDNA MRD by pre-OP CEA

[Hide](#)

```
rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]

circ_data$ctDNA.Baseline <- factor(circ_data$ctDNA.Baseline, levels = c("NEGATIVE", "POSITIVE"), labels = c("Negative", "Positive"))
circ_data$CEA.Baseline <- factor(circ_data$CEA.Baseline, levels=c("NEGATIVE","POSITIVE"), labels = c("Normal", "Elevated"))
contingency_table <- table(circ_data$CEA.Baseline, circ_data$ctDNA.Baseline)
chi_square_test <- chisq.test(contingency_table)
```

Warning in stats::chisq.test(x, y, ...) :
Chi-squared approximation may be incorrect

[Hide](#)

```
print(chi_square_test)
```

```
Pearson's Chi-squared test with Yates' continuity correction
```

```
data: contingency_table
X-squared = 0.038105, df = 1, p-value = 0.8452
```

[Hide](#)

```
fisher_exact_test <- fisher.test(contingency_table)
print(fisher_exact_test)
```

```
Fisher's Exact Test for Count Data
```

```
data: contingency_table
p-value = 0.5858
alternative hypothesis: true odds ratio is not equal to 1
95 percent confidence interval:
 0.1293152 150.9263898
sample estimates:
odds ratio
 2.524321
```

[Hide](#)

```
print(contingency_table)
```

	Negative	Positive
Normal	2	82
Elevated	1	104

[Hide](#)

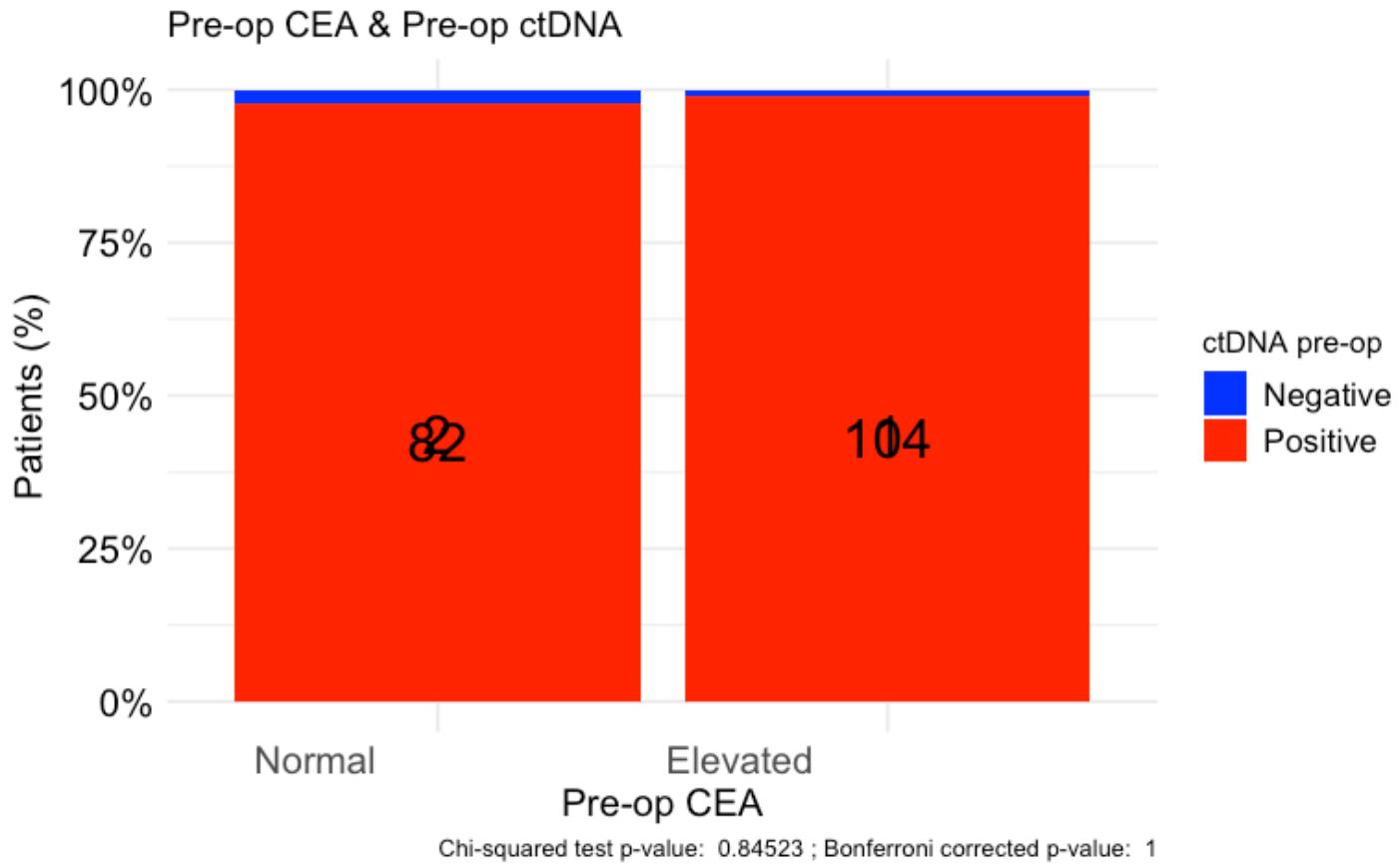
```
p_values <- c(chi_square_test$p.value, fisher_exact_test$p.value)
p_adjusted <- p.adjust(p_values, method = "bonferroni")
names(p_adjusted) <- c("Chi-Square Test", "Fisher's Exact Test")
print(p_adjusted)
```

```
Chi-Square Test Fisher's Exact Test
```

```
1 1
```

[Hide](#)

```
table_df <- as.data.frame(contingency_table)
table_df$Total <- ave(table_df$Freq, table_df$Var1, FUN = sum)
table_df$Percentage <- table_df$Freq / table_df$Total
table_df$MiddlePercentage <- table_df$Percentage / 2
ggplot(table_df, aes(x = Var1, y = Percentage, fill = Var2)) +
  geom_bar(stat = "identity") +
  geom_text(aes(y = MiddlePercentage, label = Freq), position = "stack", color = "black", vjust = 1.5, size = 7) +
  theme_minimal() +
  labs(title = "Pre-op CEA & Pre-op ctDNA",
       x = "Pre-op CEA",
       y = "Patients (%)",
       fill = "ctDNA pre-op",
       caption = paste("Chi-squared test p-value: ", format.pval(chi_square_test$p.value), "; Bonferroni corrected p-value: ", format.pval(p.adjusted["Chi-Square Test"]))) +
  scale_y_continuous(labels = scales::percent_format()) +
  scale_fill_manual(values = c("Negative" = "blue", "Positive" = "red")) + # define custom colors
  theme(axis.text.x = element_text(angle = 0, hjust = 1.5, size = 14), # increase x-axis text size
        axis.text.y = element_text(size = 14, color = "black"), # increase y-axis text size
        axis.title.x = element_text(size = 14, color = "black"), # increase x-axis label size
        axis.title.y = element_text(size = 14, color = "black"), # increase y-axis label size
        legend.text = element_text(size = 12, color = "black")) # increase Recurrence label size
```



Hide

```
rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]

circ_data$ctDNA.MRD <- factor(circ_data$ctDNA.MRD, levels = c("NEGATIVE", "POSITIVE"), labels = c("Negative", "Positive"))
circ_data$CEA.Baseline <- factor(circ_data$CEA.Baseline, levels=c("NEGATIVE","POSITIVE"), labels = c("Normal", "Elevated"))
contingency_table <- table(circ_data$CEA.Baseline, circ_data$ctDNA.MRD)
chi_square_test <- chisq.test(contingency_table)
print(chi_square_test)
```

Pearson's Chi-squared test with Yates' continuity correction

```
data: contingency_table
X-squared = 2.8368e-30, df = 1, p-value = 1
```

Hide

```
fisher_exact_test <- fisher.test(contingency_table)
print(fisher_exact_test)
```

Fisher's Exact Test for Count Data

```
data: contingency_table
p-value = 1
alternative hypothesis: true odds ratio is not equal to 1
95 percent confidence interval:
0.5168345 1.9328504
sample estimates:
odds ratio
0.9969296
```

[Hide](#)

```
print(contingency_table)
```

	Negative	Positive
Normal	57	27
Elevated	72	34

[Hide](#)

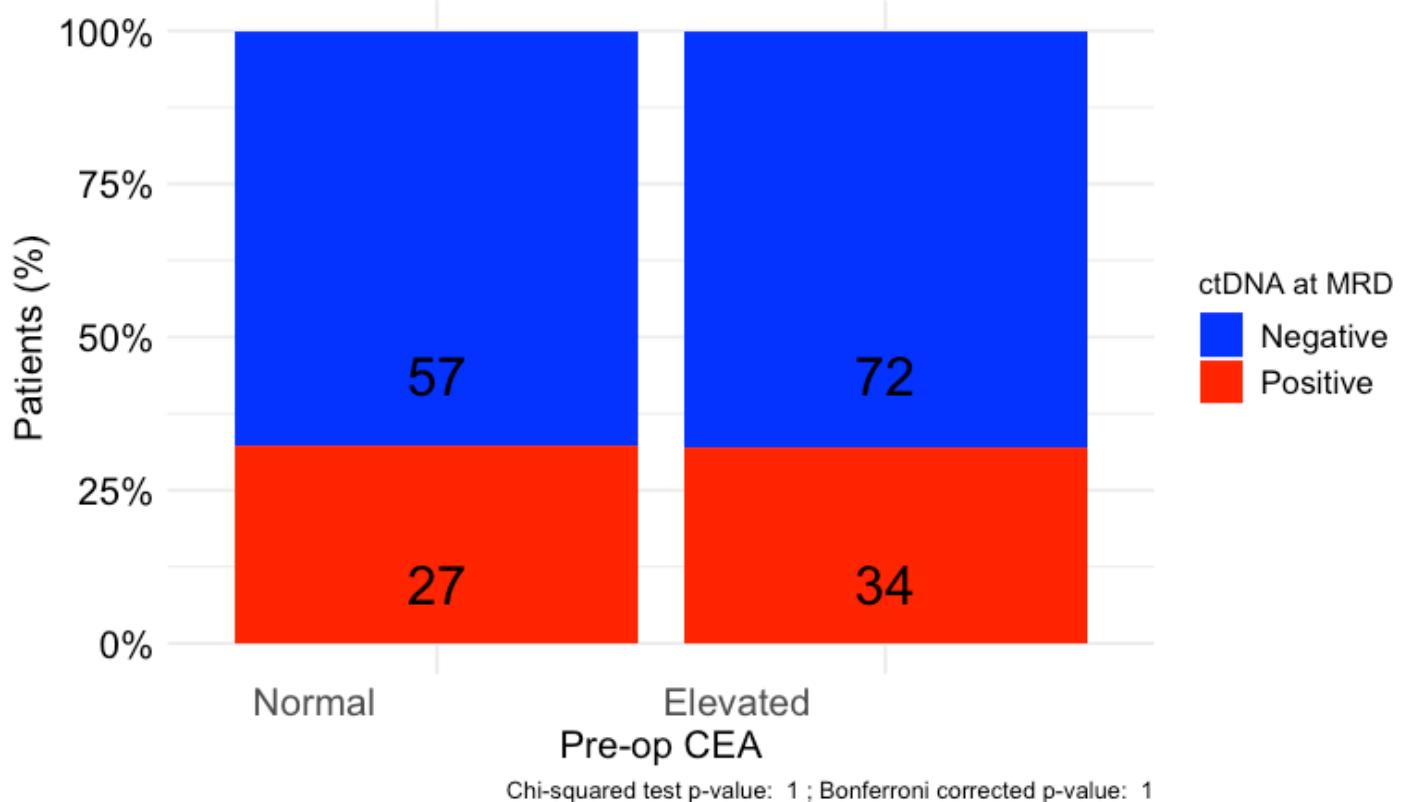
```
p_values <- c(chi_square_test$p.value, fisher_exact_test$p.value)
p_adjusted <- p.adjust(p_values, method = "bonferroni")
names(p_adjusted) <- c("Chi-Square Test", "Fisher's Exact Test")
print(p_adjusted)
```

```
Chi-Square Test Fisher's Exact Test
1 1
```

[Hide](#)

```
table_df <- as.data.frame(contingency_table)
table_df$Total <- ave(table_df$Freq, table_df$Var1, FUN = sum)
table_df$Percentage <- table_df$Freq / table_df$Total
table_df$MiddlePercentage <- table_df$Percentage / 2
ggplot(table_df, aes(x = Var1, y = Percentage, fill = Var2)) +
  geom_bar(stat = "identity") +
  geom_text(aes(y = MiddlePercentage, label = Freq), position = "stack", color = "black", vjust = 1.5, size = 7) +
  theme_minimal() +
  labs(title = "Pre-op CEA & ctDNA MRD",
       x = "Pre-op CEA",
       y = "Patients (%)",
       fill = "ctDNA at MRD",
       caption = paste("Chi-squared test p-value: ", format.pval(chi_square_test$p.value), "; Bonferroni corrected p-value: ", format.pval(p.adjusted["Chi-Square Test"]))) +
  scale_y_continuous(labels = scales::percent_format()) +
  scale_fill_manual(values = c("Negative" = "blue", "Positive" = "red")) + # define custom colors
  theme(axis.text.x = element_text(angle = 0, hjust = 1.5, size = 14), # increase x-axis text size
        axis.text.y = element_text(size = 14, color = "black"), # increase y-axis text size
        axis.title.x = element_text(size = 14, color = "black"), # increase x-axis label size
        axis.title.y = element_text(size = 14, color = "black"), # increase y-axis label size
        legend.text = element_text(size = 12, color = "black")) # increase Recurrence label size
```

Pre-op CEA & ctDNA MRD



#DFS by ctDNA at the MRD Window - Landmark MRD timepoint

Hide

```
rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$ctDNA.MRD!="",]
circ_data <- circ_data[circ_data$DFS.MRD.months>=0,]
circ_datadf <- as.data.frame(circ_data)

survfit(Surv(time = circ_data$DFS.MRD.months, event = circ_data$DFS.Event)~ctDNA.MRD, data = circ_data)
```

Call: `survfit(formula = Surv(time = circ_data$DFS.MRD.months, event = circ_data$DFS.Event) ~ ctDNA.MRD, data = circ_data)`

	n	events	median	0.95LCL	0.95UCL
ctDNA.MRD=NEGATIVE	129	45	NA	32	NA
ctDNA.MRD=POSITIVE	60	53	2.56	2	4.6

Hide

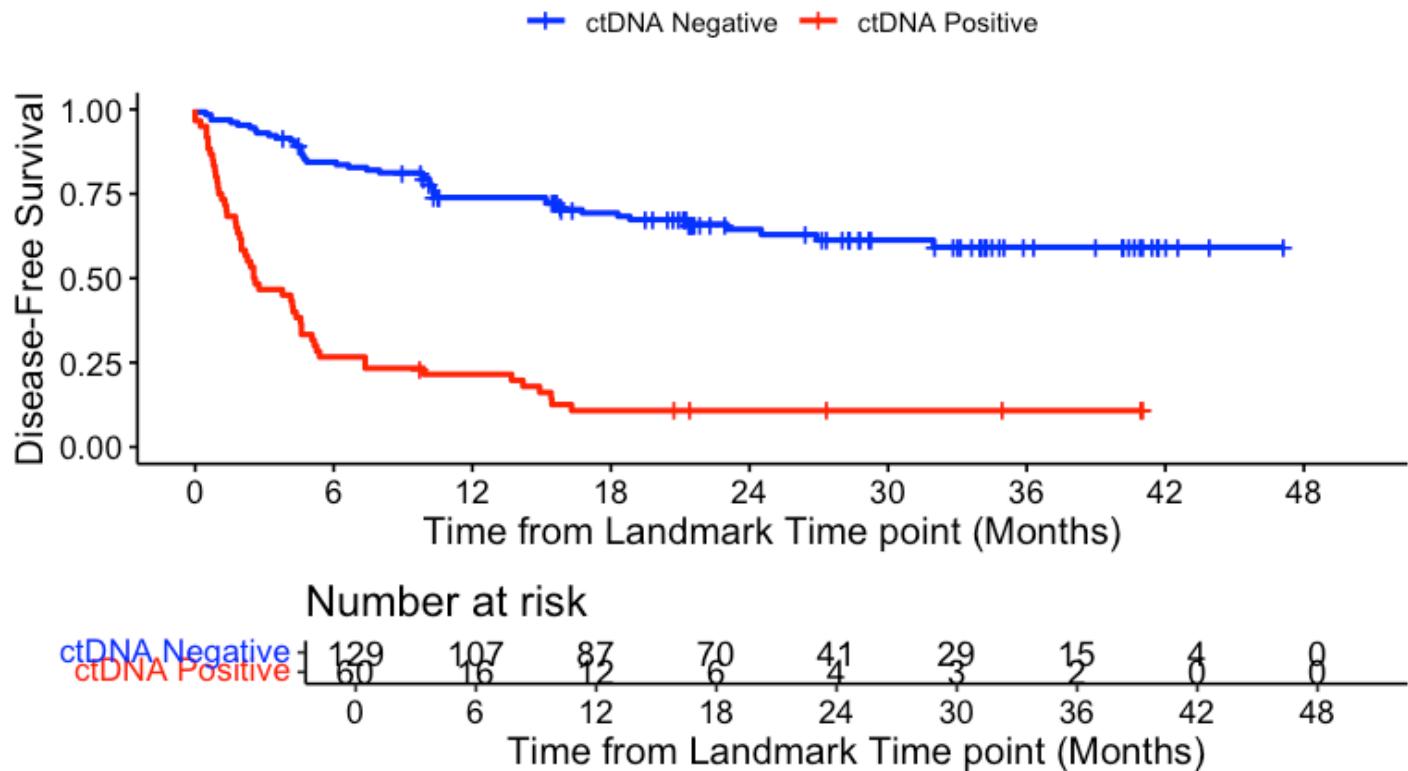
```
event_summary <- circ_data %>%
  group_by(ctDNA.MRD) %>%
  summarise(
    Total = n(),
    Events = sum(DFS.Event),
    Fraction = Events / n(),
    Percentage = (Events / n()) * 100
  )
print(event_summary)
```

ctDNA.MRD	Total	Events	Fraction	Percentage
<chr>	<int>	<int>	<dbl>	<dbl>
NEGATIVE	129	45	0.3488372	34.88372
POSITIVE	60	53	0.8833333	88.33333
2 rows				

Hide

```
surv_object <- Surv(time = circ_data$DFS.MRD.months, event = circ_data$DFS.Event)
KM_curve <- survfit(surv_object ~ ctDNA.MRD, data = circ_data, conf.int=0.95, conf.type="log-log")
ggsurvplot(KM_curve, data = circ_data, pval = FALSE, conf.int = FALSE, risk.table = TRUE,
break.time.by=6, palette=c("blue","red"), title="DFS - ctDNA MRD window", ylab= "Disease-Free Survival", xlab="Time from Landmark Time point (Months)", legend.labs=c("ctDNA Negative", "ctDNA Positive"), legend.title="")
```

DFS - ctDNA MRD window



```
summary(KM_curve, times= c(24, 30, 36))
```

```
Call: survfit(formula = surv_object ~ ctDNA.MRD, data = circ_data,
  conf.int = 0.95, conf.type = "log-log")
```

ctDNA.MRD=NEGATIVE

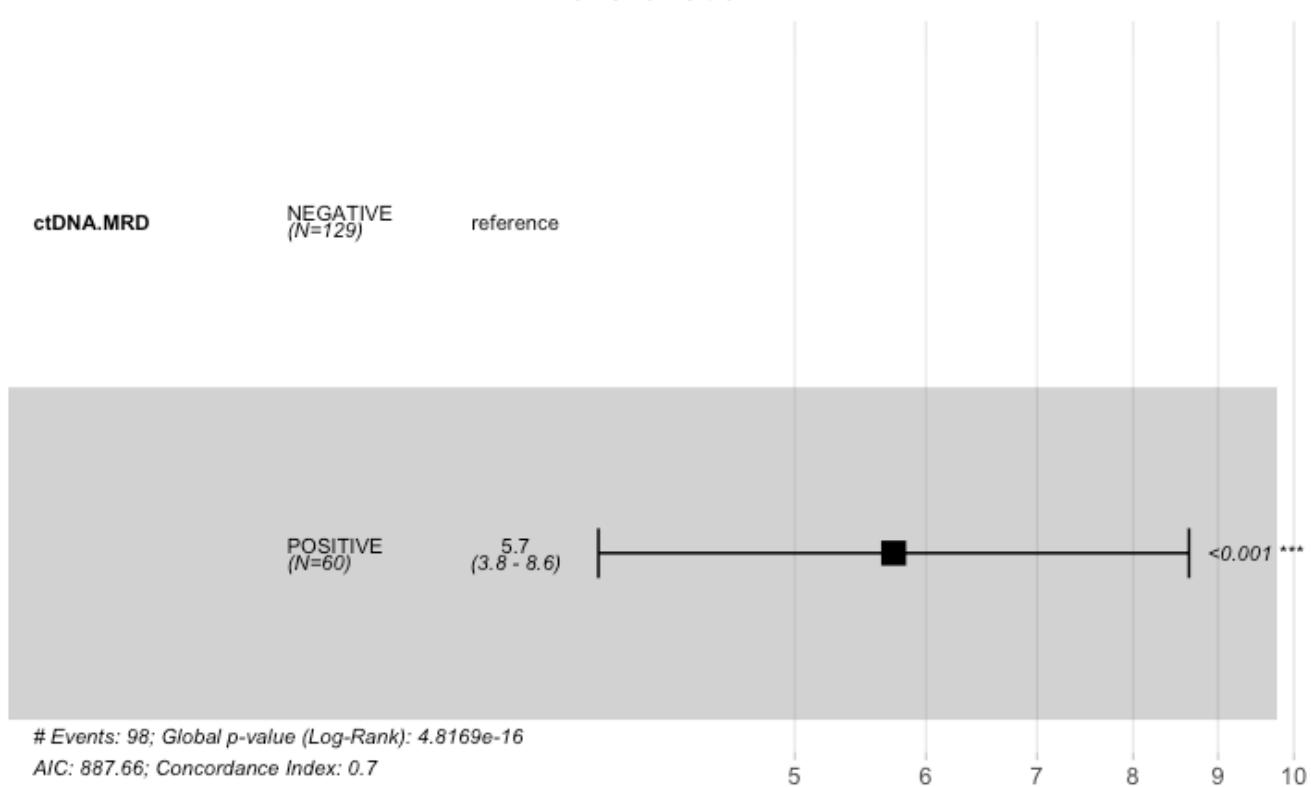
time	n.risk	n.event	survival	std.err	lower	95% CI	upper	95% CI
24	41	42	0.645	0.0456	0.548		0.726	
30	29	2	0.613	0.0486	0.511		0.700	
36	15	1	0.592	0.0513	0.484		0.684	

ctDNA.MRD=POSITIVE

time	n.risk	n.event	survival	std.err	lower	95% CI	upper	95% CI
24	4	53	0.108	0.0409	0.0445		0.203	
30	3	0	0.108	0.0409	0.0445		0.203	
36	2	0	0.108	0.0409	0.0445		0.203	

```
circ_data$ctDNA.MRD <- factor(circ_data$ctDNA.MRD, levels=c("NEGATIVE","POSITIVE"))
cox_fit <- coxph(surv_object ~ ctDNA.MRD, data=circ_data)
ggforest(cox_fit, data = circ_data)
```

Hazard ratio



Hide

```
summary(cox_fit)
```

Call:

```
coxph(formula = surv_object ~ ctDNA.MRD, data = circ_data)
```

```
n= 189, number of events= 98
```

	coef	exp(coef)	se(coef)	z	Pr(> z)
ctDNA.MRDPOSITIVE	1.7470	5.7371	0.2092	8.352	<2e-16 ***

```
Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

	exp(coef)	exp(-coef)	lower .95	upper .95
ctDNA.MRDPOSITIVE	5.737	0.1743	3.808	8.645

```
Concordance= 0.701 (se = 0.021 )
```

```
Likelihood ratio test= 65.87 on 1 df, p=5e-16
```

```
Wald test = 69.75 on 1 df, p=<2e-16
```

```
Score (logrank) test = 86.49 on 1 df, p=<2e-16
```

Hide

```
cox_fit_summary <- summary(cox_fit)

# Extract values for HR, 95% CI, and p-value
HR <- cox_fit_summary$coefficients[2]
lower_CI <- cox_fit_summary$conf.int[3]
upper_CI <- cox_fit_summary$conf.int[4]
p_value <- cox_fit_summary$coefficients[5]
label_text <- paste0("HR = ", round(HR, 2), " (", round(lower_CI, 2), "-", round(upper_CI, 2), "); p = ", round(p_value, 3))
print(label_text)
```

```
[1] "HR = 5.74 (3.81-8.64); p = 0"
```

[Hide](#)

```
#Fisher test for DFS percentages at 24, 30, and 36 months
dfs_times <- c(24, 30, 36)
p_values <- sapply(dfs_times, function(time) {
  neg_count <- sum(circ_data$ctDNA.MRD == "NEGATIVE" & circ_data$DFS.MRD.months >= time & circ_data$DFS.Event == 0)
  pos_count <- sum(circ_data$ctDNA.MRD == "POSITIVE" & circ_data$DFS.MRD.months >= time & circ_data$DFS.Event == 0)
  neg_total <- sum(circ_data$ctDNA.MRD == "NEGATIVE")
  pos_total <- sum(circ_data$ctDNA.MRD == "POSITIVE")

  neg_surv <- neg_total - sum(circ_data$ctDNA.MRD == "NEGATIVE" & circ_data$DFS.Event == 1 & circ_data$DFS.MRD.months < time)
  pos_surv <- pos_total - sum(circ_data$ctDNA.MRD == "POSITIVE" & circ_data$DFS.Event == 1 & circ_data$DFS.MRD.months < time)

  surv_matrix <- matrix(c(neg_surv, pos_surv, neg_total - neg_surv, pos_total - pos_surv), nrow = 2)
  test_result <- fisher.test(surv_matrix)
  return(test_result$p.value)
})
names(p_values) <- paste0("p-value at ", dfs_times, " months")
print(p_values)
```

```
p-value at 24 months p-value at 30 months p-value at 36 months
2.248794e-13 8.823505e-13 1.412511e-12
```

[Hide](#)

```
p_values_adjusted <- p.adjust(p_values, method = "bonferroni")
names(p_values_adjusted) <- paste0("Corrected p-value at ", dfs_times, " months")
print(p_values_adjusted)
```

Corrected p-value at 24 months	Corrected p-value at 30 months	Corrected p-value at 36 months
6.746383e-13 e-12	2.647051e-12	4.237533

#OS by ctDNA at the MRD Window - Landmark MRD timepoint

Hide

```
rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$ctDNA.MRD!="",]
circ_data <- circ_data[circ_data$OS.MRD.months>=0,]
circ_datadf <- as.data.frame(circ_data)

survfit(Surv(time = circ_data$OS.MRD.months, event = circ_data$OS.Event)~ctDNA.MRD, data = circ_data)
```

Call: survfit(formula = Surv(time = circ_data\$OS.MRD.months, event = circ_data\$OS.Event)
~
ctDNA.MRD, data = circ_data)

	n	events	median	0.95LCL	0.95UCL
ctDNA.MRD=NEGATIVE	129	5	NA	NA	NA
ctDNA.MRD=POSITIVE	61	11	43.4	NA	NA

Hide

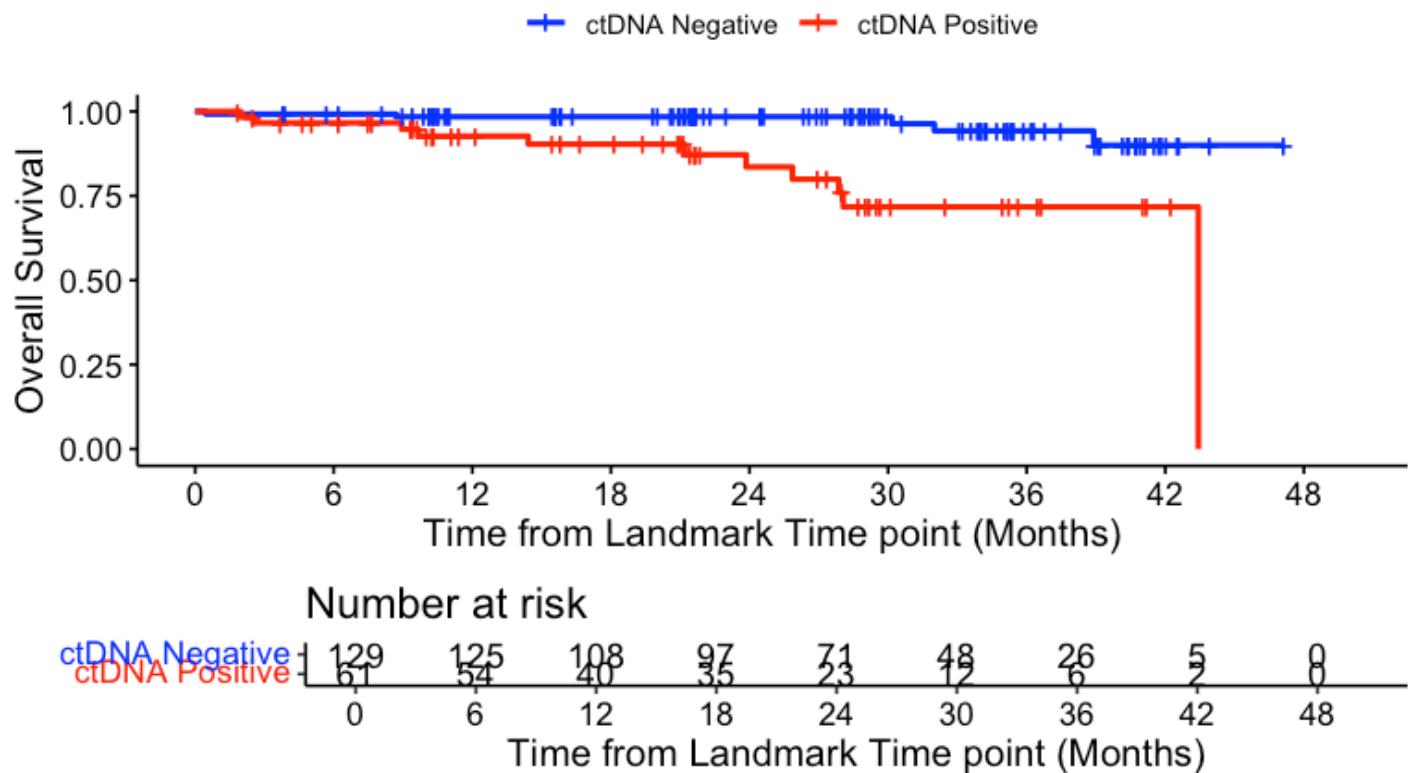
```
event_summary <- circ_data %>%
  group_by(ctDNA.MRD) %>%
  summarise(
    Total = n(),
    Events = sum(OS.Event),
    Fraction = Events / n(),
    Percentage = (Events / n()) * 100
  )
print(event_summary)
```

ctDNA.MRD	Total	Events	Fraction	Percentage
<chr>	<int>	<int>	<dbl>	<dbl>
NEGATIVE	129	5	0.03875969	3.875969
POSITIVE	61	11	0.18032787	18.032787
2 rows				

Hide

```
surv_object <- Surv(time = circ_data$OS.MRD.months, event = circ_data$OS.Event)
KM_curve <- survfit(surv_object ~ ctDNA.MRD, data = circ_data, conf.int=0.95,conf.type="log-log")
ggsurvplot(KM_curve, data = circ_data, pval = FALSE, conf.int = FALSE, risk.table = TRUE,
break.time.by=6, palette=c("blue","red"), title="OS - ctDNA MRD window | All stages",
ylab= "Overall Survival", xlab="Time from Landmark Time point (Months)", legend.labs=c
("ctDNA Negative", "ctDNA Positive"), legend.title="")
```

OS - ctDNA MRD window | All stages



Hide

```
summary(KM_curve, times= c(24, 30, 36))
```

```
Call: survfit(formula = surv_object ~ ctDNA.MRD, data = circ_data,
  conf.int = 0.95, conf.type = "log-log")
```

ctDNA.MRD=NEGATIVE

time	n.risk	n.event	survival	std.err	lower	95% CI	upper	95% CI
24	71	2	0.984	0.0111	0.938		0.996	
30	48	0	0.984	0.0111	0.938		0.996	
36	26	2	0.943	0.0306	0.841		0.980	

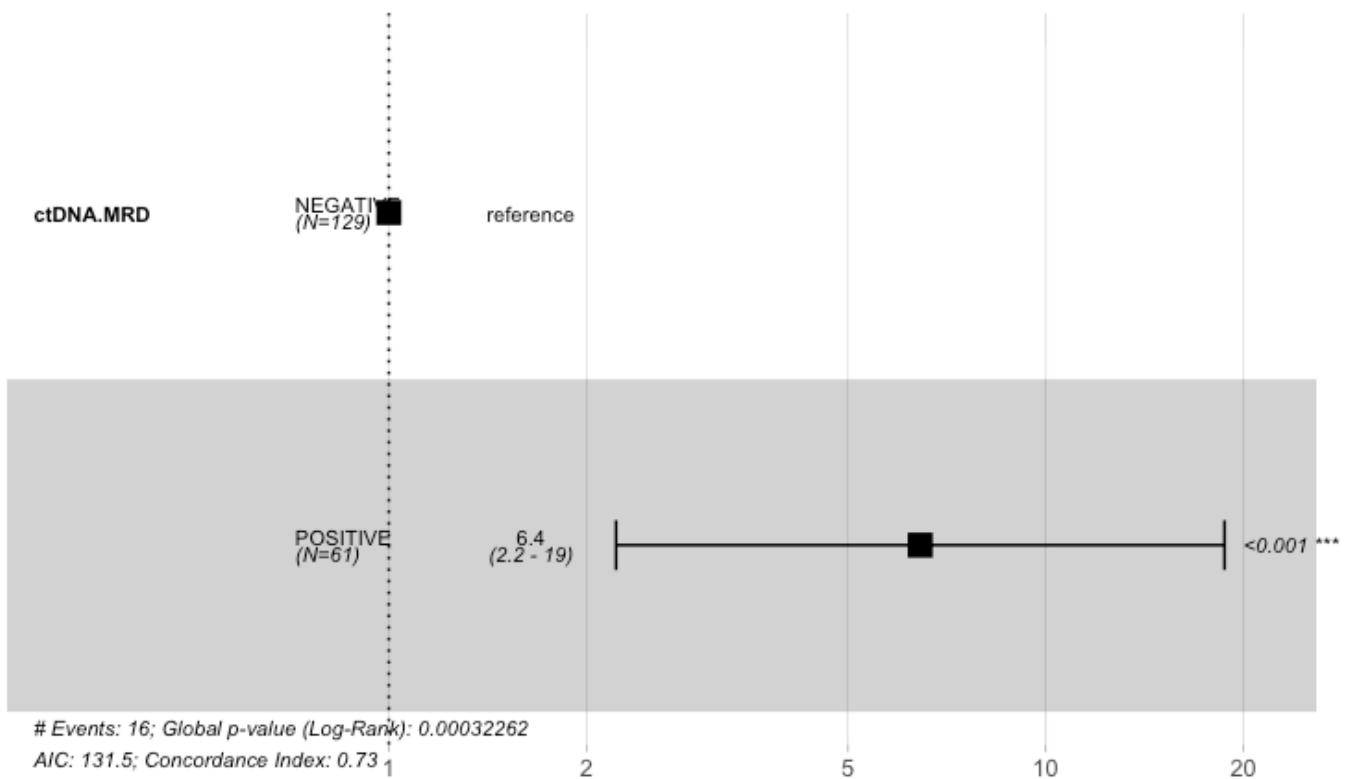
ctDNA.MRD=POSITIVE

time	n.risk	n.event	survival	std.err	lower	95% CI	upper	95% CI
24	23	7	0.836	0.0601	0.674		0.921	
30	12	3	0.717	0.0819	0.521		0.844	
36	6	0	0.717	0.0819	0.521		0.844	

Hide

```
circ_data$ctDNA.MRD <- factor(circ_data$ctDNA.MRD, levels=c("NEGATIVE","POSITIVE"))
cox_fit <- coxph(surv_object ~ ctDNA.MRD, data=circ_data)
ggforest(cox_fit, data = circ_data)
```

Hazard ratio



Hide

```
summary(cox_fit)
```

Call:
`coxph(formula = surv_object ~ ctDNA.MRD, data = circ_data)`

n= 190, number of events= 16

	coef	exp(coef)	se(coef)	z	Pr(> z)
ctDNA.MRDPOSITIVE	1.8630	6.4429	0.5441	3.424	0.000618 ***

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

	exp(coef)	exp(-coef)	lower .95	upper .95
ctDNA.MRDPOSITIVE	6.443	0.1552	2.218	18.72

Concordance= 0.726 (se = 0.064)
 Likelihood ratio test= 12.93 on 1 df, p=3e-04
 Wald test = 11.72 on 1 df, p=6e-04
 Score (logrank) test = 15.32 on 1 df, p=9e-05

Hide

```
cox_fit_summary <- summary(cox_fit)

# Extract values for HR, 95% CI, and p-value
HR <- cox_fit_summary$coefficients[2]
lower_CI <- cox_fit_summary$conf.int[3]
upper_CI <- cox_fit_summary$conf.int[4]
p_value <- cox_fit_summary$coefficients[5]
label_text <- paste0("HR = ", round(HR, 2), " (", round(lower_CI, 2), "-", round(upper_CI, 2), "); p = ", round(p_value, 4))
print(label_text)
```

```
[1] "HR = 6.44 (2.22-18.72); p = 6e-04"
```

[Hide](#)

```
#Fisher test for OS percentages at 24, 30, and 36 months
os_times <- c(24, 30, 36)
p_values <- sapply(os_times, function(time) {
  neg_count <- sum(circ_data$ctDNA.MRD == "NEGATIVE" & circ_data$OS.MRD.months >= time & circ_data$OS.Event == 0)
  pos_count <- sum(circ_data$ctDNA.MRD == "POSITIVE" & circ_data$OS.MRD.months >= time & circ_data$OS.Event == 0)
  neg_total <- sum(circ_data$ctDNA.MRD == "NEGATIVE")
  pos_total <- sum(circ_data$ctDNA.MRD == "POSITIVE")

  neg_surv <- neg_total - sum(circ_data$ctDNA.MRD == "NEGATIVE" & circ_data$OS.Event == 1 & circ_data$OS.MRD.months < time)
  pos_surv <- pos_total - sum(circ_data$ctDNA.MRD == "POSITIVE" & circ_data$OS.Event == 1 & circ_data$OS.MRD.months < time)

  surv_matrix <- matrix(c(neg_surv, pos_surv, neg_total - neg_surv, pos_total - pos_surv), nrow = 2)
  test_result <- fisher.test(surv_matrix)
  return(test_result$p.value)
})
names(p_values) <- paste0("p-value at ", os_times, " months")
print(p_values)
```

```
p-value at 24 months p-value at 30 months p-value at 36 months
0.0054510839 0.0002468767 0.0020570349
```

[Hide](#)

```
p_values_adjusted <- p.adjust(p_values, method = "bonferroni")
names(p_values_adjusted) <- paste0("Corrected p-value at ", os_times, " months")
print(p_values_adjusted)
```

Corrected p-value at 24 months	Corrected p-value at 30 months	Corrected p-value at 36 months
0.0163532517	0.0007406301	0.006171
1046		

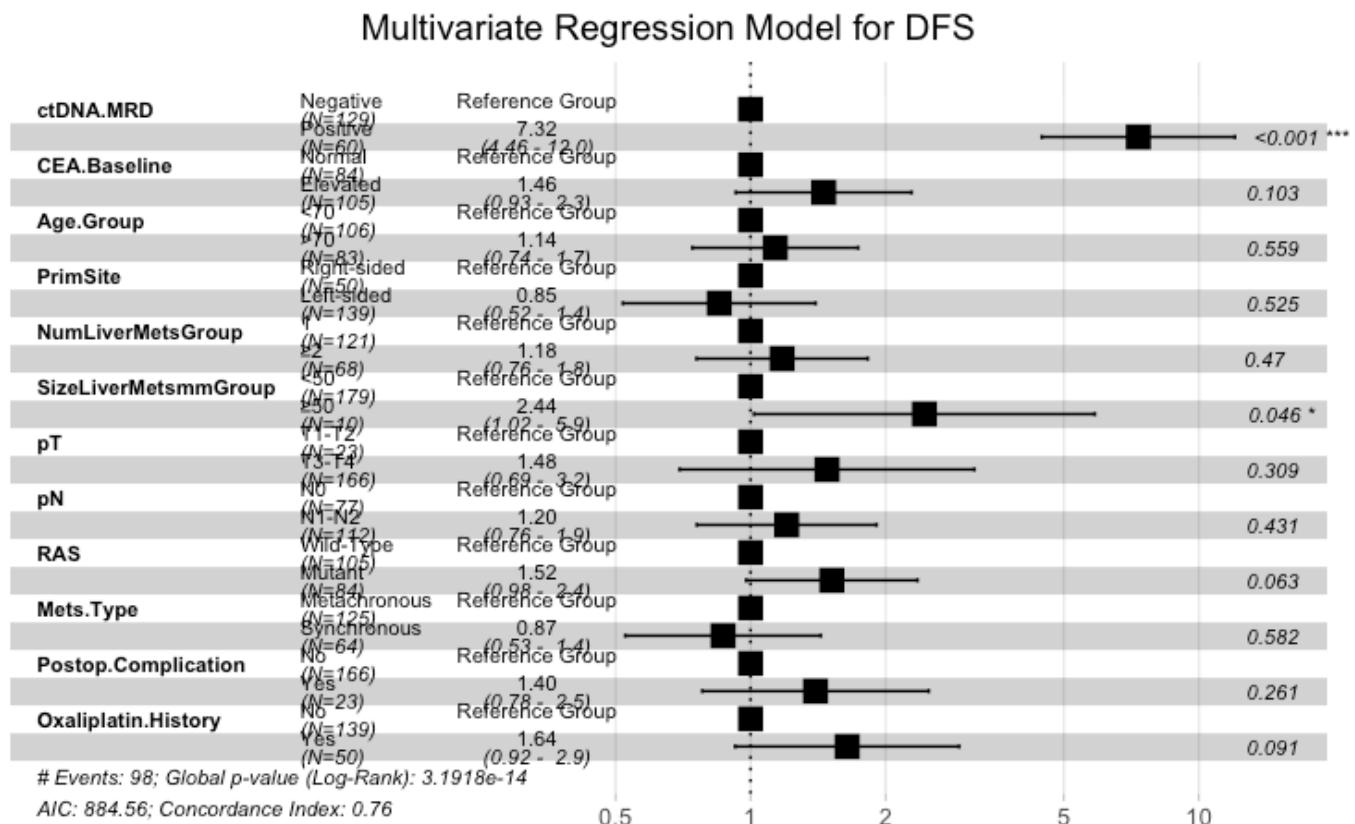
#Multivariate cox regression at MRD Window for DFS - Landmark MRD timepoint

```

rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$ctDNA.MRD!="",]
circ_data <- circ_data[circ_data$DFS.MRD.months>=0,]
circ_dataadf <- as.data.frame(circ_data)

circ_data$ctDNA.MRD <- factor(circ_data$ctDNA.MRD, levels=c("NEGATIVE", "POSITIVE"), labels = c("Negative", "Positive"))
circ_data$CEA.Baseline <- factor(circ_data$CEA.Baseline, levels=c("NEGATIVE", "POSITIVE"), labels = c("Normal", "Elevated"))
circ_data$Gender <- factor(circ_data$Gender, levels = c("Female", "Male"))
circ_data$Age.Group <- factor(circ_data$Age.Group, levels = c("1", "2"), labels = c("<70", ">70"))
circ_data$PrimSite <- factor(circ_data$PrimSite, levels = c("Right-sided colon", "Left-sided colon"), labels = c("Right-sided", "Left-sided"))
circ_data$NumLiverMetsGroup <- factor(circ_data$NumLiverMetsGroup, levels = c("1", "≥2"))
circ_data$SizeLiverMetsmmGroup <- factor(circ_data$SizeLiverMetsmmGroup, levels = c("<50", "≥50"))
circ_data$ECOG <- factor(circ_data$ECOG, levels = c("0", "1"))
circ_data$pT <- factor(circ_data$pT, levels = c("T1-T2", "T3-T4"))
circ_data$pN <- factor(circ_data$pN, levels = c("N0", "N1-N2"))
circ_data$ACT <- factor(circ_data$ACT, levels = c("FALSE", "TRUE"), labels = c("Observation", "Chemotherapy"))
circ_data$Postop.Complication <- factor(circ_data$Postop.Complication, levels = c("No", "Yes"))
circ_data$Oxaliplatin.History <- factor(circ_data$Oxaliplatin.History, levels = c("No", "Yes"))
circ_data$Mets.Type <- factor(circ_data$Mets.Type, levels = c("Metachronous", "Synchronous"))
circ_data$MSI <- factor(circ_data$MSI, levels = c("MSS", "MSI-High"), labels = c("MSS", "MSI-High"))
circ_data$BRAF.V600E <- factor(circ_data$BRAF.V600E, levels = c("WT", "MUT"), labels = c("Wild-Type", "V600E"))
circ_data$RAS <- factor(circ_data$RAS, levels = c("WT", "MUT"), labels = c("Wild-Type", "Mutant"))
surv_object <- Surv(time = circ_data$DFS.MRD.months, event = circ_data$DFS.Event)
cox_fit <- coxph(surv_object ~ ctDNA.MRD + CEA.Baseline + Age.Group + PrimSite + NumLiverMetsGroup + SizeLiverMetsmmGroup + pT + pN + RAS + Mets.Type + Postop.Complication + Oxaliplatin.History, data=circ_data)
ggforest(cox_fit, data = circ_data, main = "Multivariate Regression Model for DFS", refLabel = "Reference Group")

```



Hide

```
test.ph <- cox.zph(cox_fit)
```

#Multivariate cox regression at MRD Window for DFS - ctDNA MRD Positive Landmark MRD timepoint

Hide

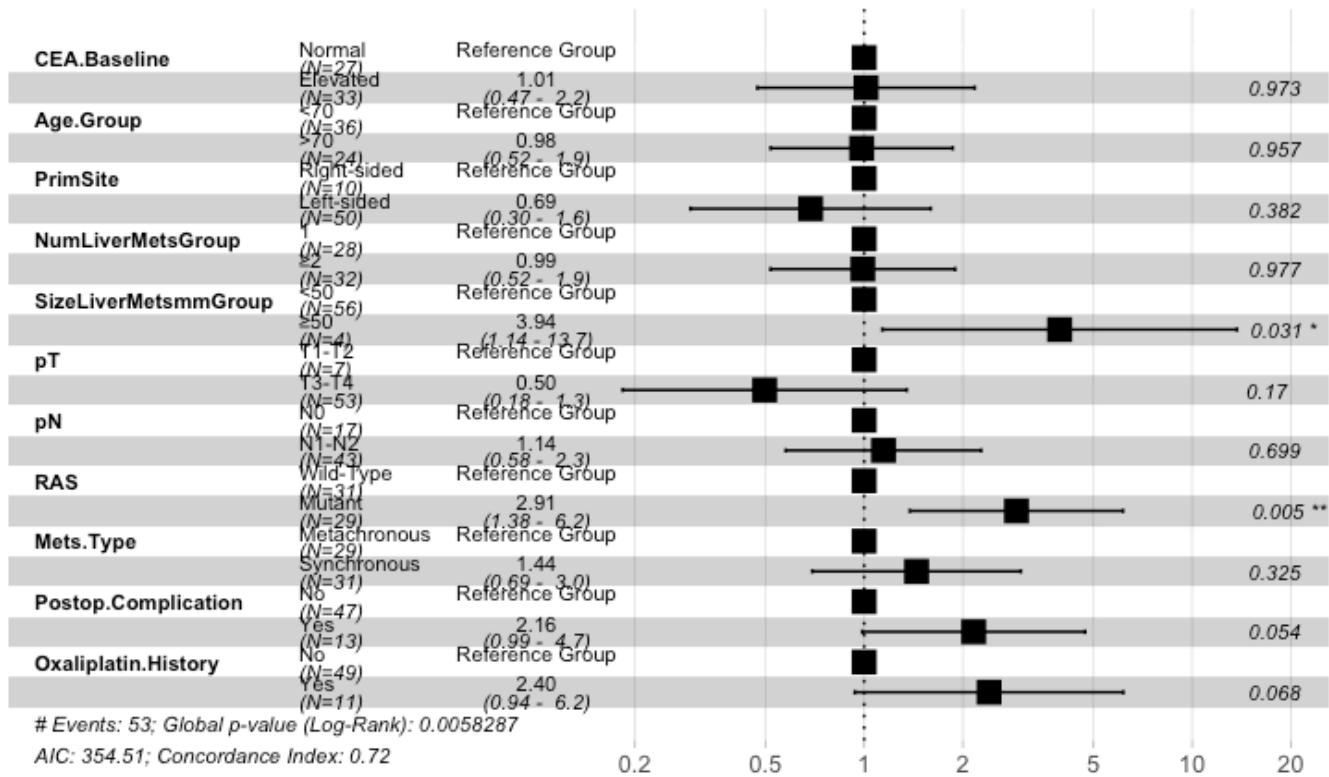
```

rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$ctDNA.MRD!="",]
circ_data <- circ_data[circ_data$DFS.MRD.months>=0,]
circ_data <- circ_data[circ_data$ctDNA.MRD=="POSITIVE",]
circ_datadf <- as.data.frame(circ_data)

circ_data$ctDNA.MRD <- factor(circ_data$ctDNA.MRD, levels=c("NEGATIVE", "POSITIVE"), labels = c("Negative", "Positive"))
circ_data$CEA.Baseline <- factor(circ_data$CEA.Baseline, levels=c("NEGATIVE", "POSITIVE"), labels = c("Normal", "Elevated"))
circ_data$Gender <- factor(circ_data$Gender, levels = c("Female", "Male"))
circ_data$Age.Group <- factor(circ_data$Age.Group, levels = c("1", "2"), labels = c("<70", ">70"))
circ_data$PrimSite <- factor(circ_data$PrimSite, levels = c("Right-sided colon", "Left-sided colon"), labels = c("Right-sided", "Left-sided"))
circ_data$NumLiverMetsGroup <- factor(circ_data$NumLiverMetsGroup, levels = c("1", "≥ 2"))
circ_data$SizeLiverMetsmmGroup <- factor(circ_data$SizeLiverMetsmmGroup, levels = c("<50", "≥50"))
circ_data$ECOG <- factor(circ_data$ECOG, levels = c("0", "1"))
circ_data$pT <- factor(circ_data$pT, levels = c("T1-T2", "T3-T4"))
circ_data$pN <- factor(circ_data$pN, levels = c("N0", "N1-N2"))
circ_data$ACT <- factor(circ_data$ACT, levels = c("FALSE", "TRUE"), labels = c("Observation", "Chemotherapy"))
circ_data$Postop.ComPLICATION <- factor(circ_data$Postop.ComPLICATION, levels = c("No", "Yes"))
circ_data$Oxaliplatin.History <- factor(circ_data$Oxaliplatin.History, levels = c("No", "Yes"))
circ_data$Mets.Type <- factor(circ_data$Mets.Type, levels = c("Metachronous", "Synchronous"))
circ_data$MSI <- factor(circ_data$MSI, levels = c("MSS", "MSI-High"), labels = c("MSS", "MSI-High"))
circ_data$BRAF.V600E <- factor(circ_data$BRAF.V600E, levels = c("WT", "MUT"), labels = c("Wild-Type", "V600E"))
circ_data$RAS <- factor(circ_data$RAS, levels = c("WT", "MUT"), labels = c("Wild-Type", "Mutant"))
surv_object <- Surv(time = circ_data$DFS.MRD.months, event = circ_data$DFS.Event)
cox_fit <- coxph(surv_object ~ CEA.Baseline + Age.Group + PrimSite + NumLiverMetsGroup + SizeLiverMetsmmGroup + pT + pN + RAS + Mets.Type + Postop.ComPLICATION + Oxaliplatin.History, data=circ_data)
ggforest(cox_fit, data = circ_data, main = "Multivariate Regression Model for DFS - ctDNA MRD Positive", refLabel = "Reference Group")

```

Multivariate Regression Model for DFS - ctDNA MRD Positive



Hide

```
test.ph <- cox.zph(cox_fit)
```

#Multivariate cox regression at MRD Window for DFS - ctDNA MRD Negative Landmark MRD timepoint

Hide

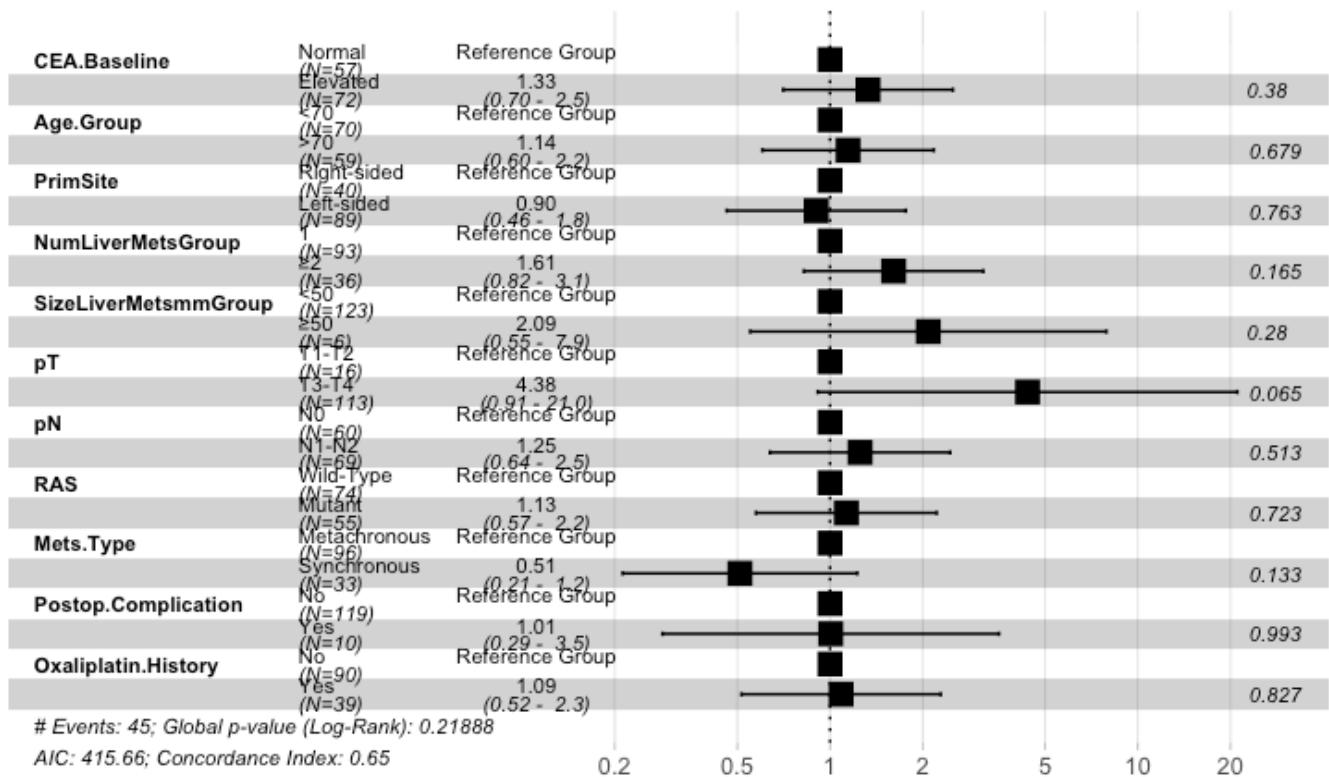
```

rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$ctDNA.MRD!="",]
circ_data <- circ_data[circ_data$DFS.MRD.months>=0,]
circ_data <- circ_data[circ_data$ctDNA.MRD=="NEGATIVE",]
circ_datadf <- as.data.frame(circ_data)

circ_data$CEA.Baseline <- factor(circ_data$CEA.Baseline, levels=c("NEGATIVE", "POSITIVE"), labels = c("Normal", "Elevated"))
circ_data$Gender <- factor(circ_data$Gender, levels = c("Female", "Male"))
circ_data$Age.Group <- factor(circ_data$Age.Group, levels = c("1", "2"), labels = c("<70", ">70"))
circ_data$PrimSite <- factor(circ_data$PrimSite, levels = c("Right-sided colon", "Left-sided colon"), labels = c("Right-sided", "Left-sided"))
circ_data$NumLiverMetsGroup <- factor(circ_data$NumLiverMetsGroup, levels = c("1", "≥2"))
circ_data$SizeLiverMetsmmGroup <- factor(circ_data$SizeLiverMetsmmGroup, levels = c("<50", "≥50"))
circ_data$ECOG <- factor(circ_data$ECOG, levels = c("0", "1"))
circ_data$pT <- factor(circ_data$pT, levels = c("T1-T2", "T3-T4"))
circ_data$pN <- factor(circ_data$pN, levels = c("N0", "N1-N2"))
circ_data$ACT <- factor(circ_data$ACT, levels = c("FALSE", "TRUE"), labels = c("Observation", "Chemotherapy"))
circ_data$Postop.Complication <- factor(circ_data$Postop.Complication, levels = c("No", "Yes"))
circ_data$Oxaliplatin.History <- factor(circ_data$Oxaliplatin.History, levels = c("No", "Yes"))
circ_data$Mets.Type <- factor(circ_data$Mets.Type, levels = c("Metachronous", "Synchronous"))
circ_data$MSI <- factor(circ_data$MSI, levels = c("MSS", "MSI-High"), labels = c("MSS", "MSI-High"))
circ_data$BRAF.V600E <- factor(circ_data$BRAF.V600E, levels = c("WT", "MUT"), labels = c("Wild-Type", "V600E"))
circ_data$RAS <- factor(circ_data$RAS, levels = c("WT", "MUT"), labels = c("Wild-Type", "Mutant"))
surv_object <- Surv(time = circ_data$DFS.MRD.months, event = circ_data$DFS.Event)
cox_fit <- coxph(surv_object ~ CEA.Baseline + Age.Group + PrimSite + NumLiverMetsGroup + SizeLiverMetsmmGroup + pT + pN + RAS + Mets.Type + Postop.Complication + Oxaliplatin.History, data=circ_data)
ggforest(cox_fit, data = circ_data, main = "Multivariate Regression Model for DFS - ctDNA MRD Negative", refLabel = "Reference Group")

```

Multivariate Regression Model for DFS - ctDNA MRD Negative



Hide

```
test.ph <- cox.zph(cox_fit)
```

#DFS by ACT treatment in MRD negative

[Hide](#)

```

rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$ctDNA.MRD!="",]
circ_data <- circ_data[circ_data$ctDNA.MRD=="NEGATIVE",]
circ_data$DFS.months=circ_data$DFS.months-2
circ_data <- circ_data[circ_data$DFS.months>=0,]
circ_datadf <- as.data.frame(circ_data)

survfit(Surv(time = circ_data$DFS.months, event = circ_data$DFS.Event)~ACT, data = circ_data)

```

Call: survfit(formula = Surv(time = circ_data\$DFS.months, event = circ_data\$DFS.Event) ~ ACT, data = circ_data)

	n	events	median	0.95LCL	0.95UCL
ACT=FALSE	92	33	NA	25.6	NA
ACT=TRUE	33	8	NA	NA	NA

[Hide](#)

```

event_summary <- circ_data %>%
  group_by(ACT) %>%
  summarise(
    Total = n(),
    Events = sum(DFS.Event),
    Fraction = Events / n(),
    Percentage = (Events / n()) * 100
  )
print(event_summary)

```

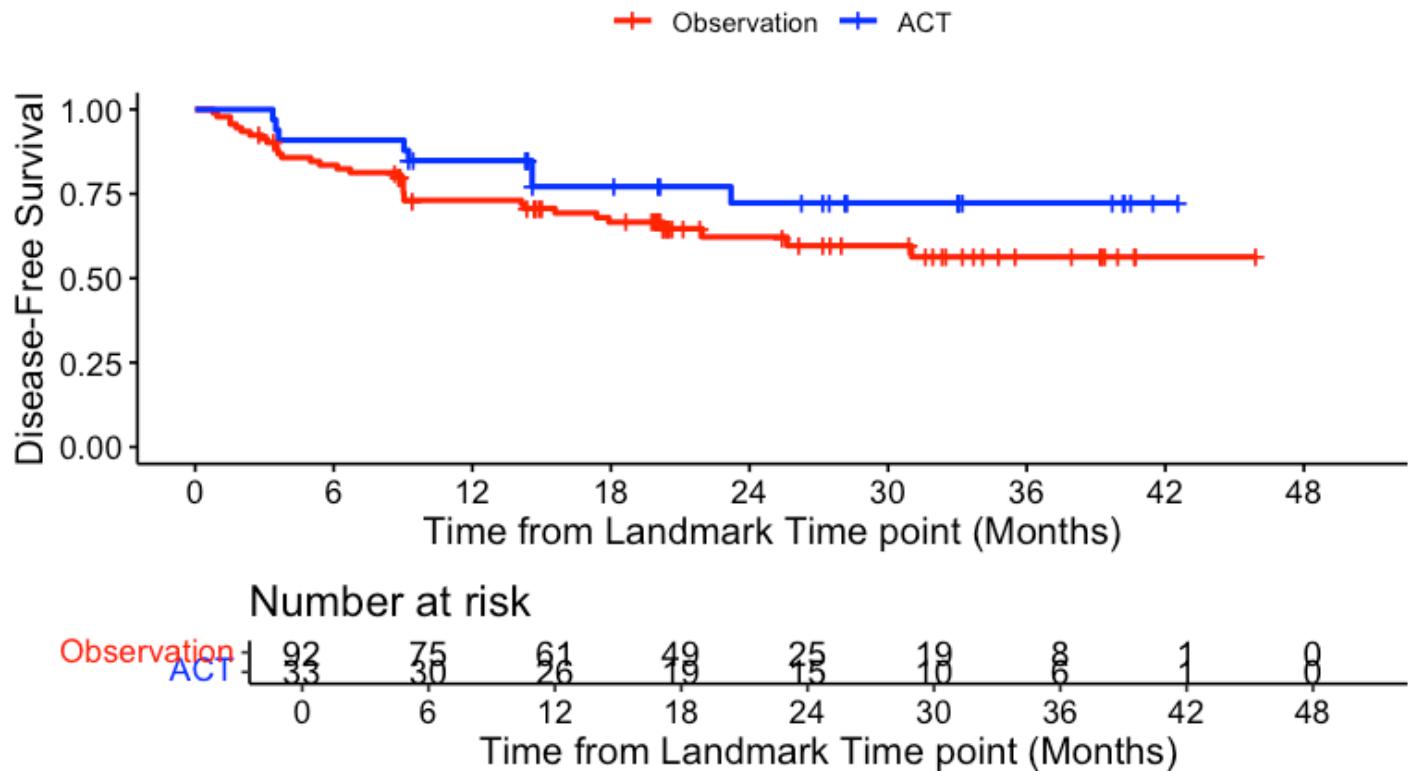
ACT	Total	Events	Fraction	Percentage
	<int>	<int>	<dbl>	<dbl>
FALSE	92	33	0.3586957	35.86957
TRUE	33	8	0.2424242	24.24242

2 rows

[Hide](#)

```
surv_object <- Surv(time = circ_data$DFS.months, event = circ_data$DFS.Event)
KM_curve <- survfit(surv_object ~ ACT, data = circ_data, conf.int=0.95,conf.type="log-log")
ggsurvplot(KM_curve, data = circ_data, pval = FALSE, conf.int = FALSE, risk.table = TRUE,
break.time.by=6, palette=c("red","blue"), title="DFS - ctDNA MRD Negative ACT vs Observation",
ylab= "Disease-Free Survival", xlab="Time from Landmark Time point (Months)",
legend.labs=c("Observation", "ACT"), legend.title="")
```

DFS - ctDNA MRD Negative ACT vs Observation



Hide

```
summary(KM_curve, times= c(24))
```

```
Call: survfit(formula = surv_object ~ ACT, data = circ_data, conf.int = 0.95,
  conf.type = "log-log")
```

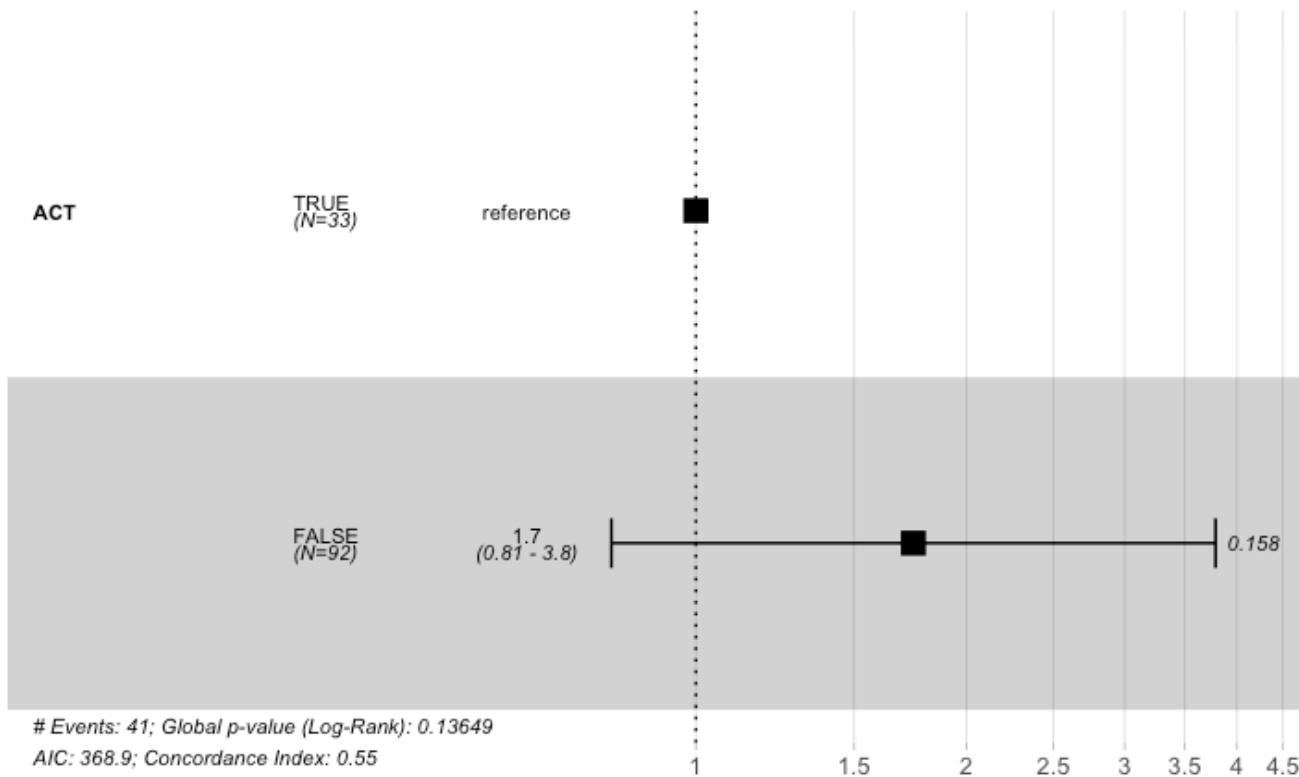
CI	ACT=FALSE						
	time	n.risk	n.event	survival	std.err	lower	95% CI upper
18	24.0000	25.0000	31.0000	0.6222	0.0566	0.5011	0.72

CI	ACT=TRUE						
	time	n.risk	n.event	survival	std.err	lower	95% CI upper
38	24.0000	15.0000	8.0000	0.7231	0.0859	0.5143	0.85

Hide

```
circ_data$ACT <- factor(circ_data$ACT, levels=c("TRUE","FALSE"))
cox_fit <- coxph(surv_object ~ ACT, data=circ_data)
ggforest(cox_fit,data = circ_data)
```

Hazard ratio



```
summary(cox_fit)
```

Call:
 coxph(formula = surv_object ~ ACT, data = circ_data)

n= 125, number of events= 41

	coef	exp(coef)	se(coef)	z	Pr(> z)
ACTFALSE	0.5575	1.7463	0.3948	1.412	0.158

	exp(coef)	exp(-coef)	lower .95	upper .95
ACTFALSE	1.746	0.5727	0.8054	3.786

Concordance= 0.553 (se = 0.032)
 Likelihood ratio test= 2.22 on 1 df, p=0.1
 Wald test = 1.99 on 1 df, p=0.2
 Score (logrank) test = 2.05 on 1 df, p=0.2

```
cox_fit_summary <- summary(cox_fit)

# Extract values for HR, 95% CI, and p-value
HR <- cox_fit_summary$coefficients[2]
lower_CI <- cox_fit_summary$conf.int[3]
upper_CI <- cox_fit_summary$conf.int[4]
p_value <- cox_fit_summary$coefficients[5]
label_text <- paste0("HR = ", round(HR, 2), " (", round(lower_CI, 2), "-", round(upper_CI, 2), "); p = ", round(p_value, 3))
print(label_text)
```

[1] "HR = 1.75 (0.81-3.79); p = 0.158"

[Hide](#)

```
#Fisher test for DFS percentages at 24
dfs_times <- c(24)
p_values <- sapply(dfs_times, function(time) {
  neg_count <- sum(circ_data$ACT == "TRUE" & circ_data$DFS.months >= time & circ_data$DFS.Event == 0)
  pos_count <- sum(circ_data$ACT == "FALSE" & circ_data$DFS.months >= time & circ_data$DFS.Event == 0)
  neg_total <- sum(circ_data$ACT == "TRUE")
  pos_total <- sum(circ_data$ACT == "FALSE")

  neg_surv <- neg_total - sum(circ_data$ACT == "TRUE" & circ_data$DFS.Event == 1 & circ_data$DFS.months < time)
  pos_surv <- pos_total - sum(circ_data$ACT == "FALSE" & circ_data$DFS.Event == 1 & circ_data$DFS.months < time)

  surv_matrix <- matrix(c(neg_surv, pos_surv, neg_total - neg_surv, pos_total - pos_surv), nrow = 2)
  test_result <- fisher.test(surv_matrix)
  return(test_result$p.value)
})
names(p_values) <- paste0("p-value at ", dfs_times, " months")
print(p_values)
```

p-value at 24 months
0.3844277

[Hide](#)

```
p_values_adjusted <- p.adjust(p_values, method = "bonferroni")
names(p_values_adjusted) <- paste0("Corrected p-value at ", dfs_times, " months")
print(p_values_adjusted)
```

Corrected p-value at 24 months
0.3844277

Hide

```

#Adjusted HR "ACT vs no ACT" - age, gender, ECOG and pathological stage - ACT as reference
rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$ctDNA.MRD!="",]
circ_data <- circ_data[circ_data$ctDNA.MRD=="NEGATIVE",]
circ_data$DFS.months=circ_data$DFS.months-2
circ_data <- circ_data[circ_data$DFS.months>=0,]
circ_dataadf <- as.data.frame(circ_data)

circ_data$Gender <- factor(circ_data$Gender, levels = c("Female", "Male"))
circ_data$Age.Group <- factor(circ_data$Age.Group, levels = c("1", "2"), labels = c("<70", ">70"))
circ_data$PrimSite <- factor(circ_data$PrimSite, levels = c("Right-sided colon", "Left-sided colon"))
circ_data$NumLiverMetsGroup <- factor(circ_data$NumLiverMetsGroup, levels = c("1", "≥2"))
circ_data$SizeLiverMetsmmGroup <- factor(circ_data$SizeLiverMetsmmGroup, levels = c("<50", "≥50"))
circ_data$ECOG <- factor(circ_data$ECOG, levels = c("0", "1"))
circ_data$pT <- factor(circ_data$pT, levels = c("T1-T2", "T3-T4"))
circ_data$pN <- factor(circ_data$pN, levels = c("N0", "N1-N2"))
circ_data$ACT <- factor(circ_data$ACT, levels = c("FALSE", "TRUE"))
circ_data$Postop.Complication <- factor(circ_data$Postop.Complication, levels = c("No", "Yes"))
circ_data$Oxaliplatin.History <- factor(circ_data$Oxaliplatin.History, levels = c("No", "Yes"))
circ_data$Mets.Type <- factor(circ_data$Mets.Type, levels = c("Metachronous", "Synchronous"))
circ_data$MSI <- factor(circ_data$MSI, levels = c("MSS", "MSI-High"), labels = c("MSS", "MSI-High"))
circ_data$BRAF.V600E <- factor(circ_data$BRAF.V600E, levels = c("WT", "MUT"), labels = c("Wild-Type", "V600E"))
circ_data$RAS <- factor(circ_data$RAS, levels = c("WT", "MUT"), labels = c("Wild-Type", "Mutant"))
surv_object <- Surv(time = circ_data$DFS.MRD.months, event = circ_data$DFS.Event)
cox_fit <- coxph(surv_object ~ ACT + Gender + Age.Group + NumLiverMetsGroup + SizeLiverMetsmmGroup + RAS + Mets.Type + Oxaliplatin.History, data=circ_data)
summary(cox_fit)

```

Call:

```
coxph(formula = surv_object ~ ACT + Gender + Age.Group + NumLiverMetsGroup +
  SizeLiverMetsmmGroup + RAS + Mets.Type + Oxaliplatin.History,
  data = circ_data)
```

n= 125, number of events= 41

	coef	exp(coef)	se(coef)	z	Pr(> z)
ACTTRUE	-0.3844	0.6809	0.4300	-0.894	0.371
GenderMale	0.3311	1.3925	0.3938	0.841	0.400
Age.Group>70	0.2674	1.3066	0.3279	0.815	0.415
NumLiverMetsGroup≥2	0.2951	1.3432	0.3658	0.807	0.420
SizeLiverMetsmmGroup≥50	0.4474	1.5642	0.6244	0.716	0.474
RASMutant	0.3107	1.3644	0.3582	0.867	0.386
Mets.TypeSynchronous	-0.1946	0.8232	0.4471	-0.435	0.663
Oxaliplatin.HistoryYes	0.2572	1.2934	0.3778	0.681	0.496

	exp(coef)	exp(-coef)	lower .95	upper .95
ACTTRUE	0.6809	1.4687	0.2931	1.581
GenderMale	1.3925	0.7181	0.6436	3.013
Age.Group>70	1.3066	0.7654	0.6871	2.485
NumLiverMetsGroup≥2	1.3432	0.7445	0.6559	2.751
SizeLiverMetsmmGroup≥50	1.5642	0.6393	0.4600	5.319
RASMutant	1.3644	0.7329	0.6761	2.753
Mets.TypeSynchronous	0.8232	1.2148	0.3427	1.977
Oxaliplatin.HistoryYes	1.2934	0.7732	0.6168	2.712

Concordance= 0.638 (se = 0.042)
 Likelihood ratio test= 6.52 on 8 df, p=0.6
 Wald test = 6.23 on 8 df, p=0.6
 Score (logrank) test = 6.37 on 8 df, p=0.6

#DFS by ACT treatment in MRD positive

Hide

```
rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$ctDNA.MRD!="",]
circ_data <- circ_data[circ_data$ctDNA.MRD=="POSITIVE",]
circ_data$DFS.months=circ_data$DFS.months-2
circ_data <- circ_data[circ_data$DFS.months>=0,]
circ_datadf <- as.data.frame(circ_data)

survfit(Surv(time = circ_data$DFS.months, event = circ_data$DFS.Event)~ACT, data = circ_data)
```

```
Call: survfit(formula = Surv(time = circ_data$DFS.months, event = circ_data$DFS.Event) ~ ACT, data = circ_data)
```

	n	events	median	0.95LCL	0.95UCL
ACT=FALSE	31	29	1.45	0.76	3.32
ACT=TRUE	15	10	12.92	4.11	NA

[Hide](#)

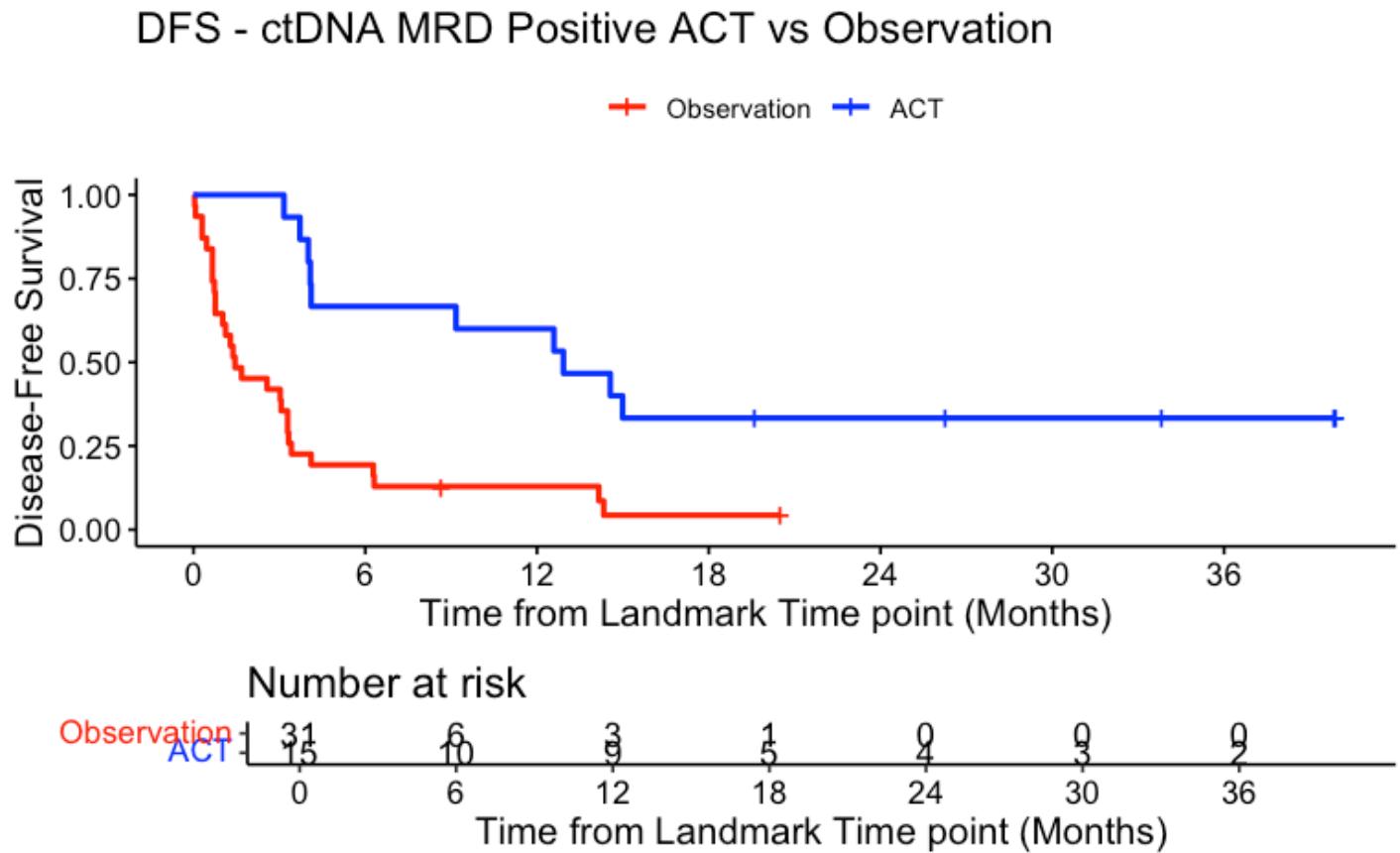
```
event_summary <- circ_data %>%
  group_by(ACT) %>%
  summarise(
    Total = n(),
    Events = sum(DFS.Event),
    Fraction = Events / n(),
    Percentage = (Events / n()) * 100
  )
print(event_summary)
```

ACT	Total	Events	Fraction	Percentage
	<int>	<int>	<dbl>	<dbl>
FALSE	31	29	0.9354839	93.54839
TRUE	15	10	0.6666667	66.66667

2 rows

[Hide](#)

```
surv_object <- Surv(time = circ_data$DFS.months, event = circ_data$DFS.Event)
KM_curve <- survfit(surv_object ~ ACT, data = circ_data, conf.int=0.95, conf.type="log-log")
ggsurvplot(KM_curve, data = circ_data, pval = FALSE, conf.int = FALSE, risk.table = TRUE,
break.time.by=6, palette=c("red","blue"), title="DFS - ctDNA MRD Positive ACT vs Observation",
ylab= "Disease-Free Survival", xlab="Time from Landmark Time point (Months)",
legend.labs=c("Observation", "ACT"), legend.title="")
```



```
summary(KM_curve, times= c(12, 24))
```

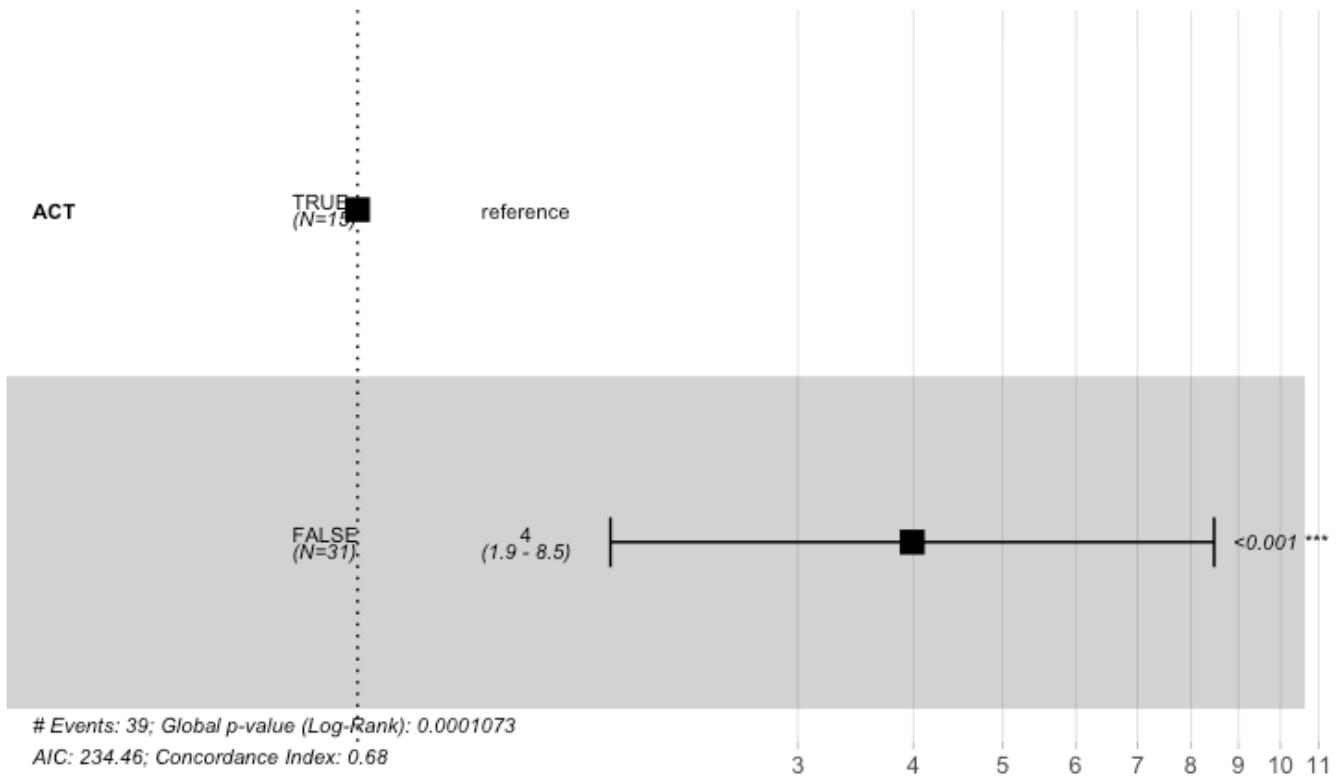
```
Call: survfit(formula = surv_object ~ ACT, data = circ_data, conf.int = 0.95,
  conf.type = "log-log")
```

CI	ACT=FALSE								
	time	n.risk	n.event	survival	std.err	lower	95% CI	upper	95%
12	12.0000	3.0000	27.0000	0.1290	0.0602	0.0407	0.26		
24									
ACT=TRUE									
	time	n.risk	n.event	survival	std.err	lower	95% CI	upper	95% CI
	12	9	6	0.600	0.126	0.318	0.797		
	24	4	4	0.333	0.122	0.122	0.564		

[Hide](#)

```
circ_data$ACT <- factor(circ_data$ACT, levels=c("TRUE","FALSE"))
cox_fit <- coxph(surv_object ~ ACT, data=circ_data)
ggforest(cox_fit,data = circ_data)
```

Hazard ratio



[Hide](#)

```
summary(cox_fit)
```

Call:

coxph(formula = surv_object ~ ACT, data = circ_data)

n= 46, number of events= 39

	coef	exp(coef)	se(coef)	z	Pr(> z)
ACTFALSE	1.3838	3.9899	0.3842	3.602	0.000316 ***

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

	exp(coef)	exp(-coef)	lower .95	upper .95
ACTFALSE	3.99	0.2506	1.879	8.472

Concordance= 0.675 (se = 0.03)

Likelihood ratio test= 15 on 1 df, p=1e-04

Wald test = 12.97 on 1 df, p=3e-04

Score (logrank) test = 14.61 on 1 df, p=1e-04

Hide

cox_fit_summary <- summary(cox_fit)

Extract values for HR, 95% CI, and p-value

HR <- cox_fit_summary\$coefficients[2]

lower_CI <- cox_fit_summary\$conf.int[3]

upper_CI <- cox_fit_summary\$conf.int[4]

p_value <- cox_fit_summary\$coefficients[5]

label_text <- paste0("HR = ", round(HR, 2), " (", round(lower_CI, 2), "-", round(upper_CI, 2), "); p = ", round(p_value, 3))

print(label_text)

[1] "HR = 3.99 (1.88-8.47); p = 0"

Hide

```
#Adjusted HR "ACT vs no ACT" - age, gender, ECOG and pathological stage - ACT as reference
rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$ctDNA.MRD!="",]
circ_data <- circ_data[circ_data$ctDNA.MRD=="POSITIVE",]
circ_data$DFS.months=circ_data$DFS.months-2
circ_data <- circ_data[circ_data$DFS.months>=0,]
circ_datadf <- as.data.frame(circ_data)

circ_data$Gender <- factor(circ_data$Gender, levels = c("Female", "Male"))
circ_data$Age.Group <- factor(circ_data$Age.Group, levels = c("1", "2"), labels = c("<70", ">70"))
circ_data$PrimSite <- factor(circ_data$PrimSite, levels = c("Right-sided colon", "Left-sided colon"))
circ_data$NumLiverMetsGroup <- factor(circ_data$NumLiverMetsGroup, levels = c("1", "≥2"))
circ_data$SizeLiverMetsmmGroup <- factor(circ_data$SizeLiverMetsmmGroup, levels = c("<50", "≥50"))
circ_data$ECOG <- factor(circ_data$ECOG, levels = c("0", "1"))
circ_data$pT <- factor(circ_data$pT, levels = c("T1-T2", "T3-T4"))
circ_data$pN <- factor(circ_data$pN, levels = c("N0", "N1-N2"))
circ_data$ACT <- factor(circ_data$ACT, levels = c("FALSE", "TRUE"))
circ_data$Postop.Complication <- factor(circ_data$Postop.Complication, levels = c("No", "Yes"))
circ_data$Oxaliplatin.History <- factor(circ_data$Oxaliplatin.History, levels = c("No", "Yes"))
circ_data$Mets.Type <- factor(circ_data$Mets.Type, levels = c("Metachronous", "Synchronous"))
circ_data$MSI <- factor(circ_data$MSI, levels = c("MSS", "MSI-High"), labels = c("MSS", "MSI-High"))
circ_data$BRAF.V600E <- factor(circ_data$BRAF.V600E, levels = c("WT", "MUT"), labels = c("Wild-Type", "V600E"))
circ_data$RAS <- factor(circ_data$RAS, levels = c("WT", "MUT"), labels = c("Wild-Type", "Mutant"))
surv_object <- Surv(time = circ_data$DFS.MRD.months, event = circ_data$DFS.Event)
cox_fit <- coxph(surv_object ~ ACT + Gender + Age.Group + NumLiverMetsGroup + SizeLiverMetsmmGroup + RAS + Mets.Type + Oxaliplatin.History, data=circ_data)
summary(cox_fit)
```

Call:

```
coxph(formula = surv_object ~ ACT + Gender + Age.Group + NumLiverMetsGroup +
  SizeLiverMetsmmGroup + RAS + Mets.Type + Oxaliplatin.History,
  data = circ_data)
```

n= 46, number of events= 39

	coef	exp(coef)	se(coef)	z	Pr(> z)						
ACTTRUE	-2.68676	0.06810	0.67360	-3.989	6.65e-05 ***						
GenderMale	-1.04107	0.35308	0.51890	-2.006	0.04483 *						
Age.Group>70	-0.01813	0.98203	0.39282	-0.046	0.96318						
NumLiverMetsGroup≥2	-0.94250	0.38965	0.43212	-2.181	0.02918 *						
SizeLiverMetsmmGroup≥50	1.96295	7.12027	0.91333	2.149	0.03162 *						
RASMutant	1.01046	2.74687	0.36487	2.769	0.00562 **						
Mets.TypeSynchronous	0.62120	1.86116	0.44165	1.407	0.15956						
Oxaliplatin.HistoryYes	0.81261	2.25379	0.57133	1.422	0.15493						

Signif. codes:	0	'***'	0.001	'**'	0.01	'*'	0.05	'.'	0.1	' '	1

	exp(coef)	exp(-coef)	lower .95	upper .95
ACTTRUE	0.0681	14.6840	0.01819	0.2550
GenderMale	0.3531	2.8322	0.12770	0.9762
Age.Group>70	0.9820	1.0183	0.45473	2.1208
NumLiverMetsGroup≥2	0.3897	2.5664	0.16705	0.9089
SizeLiverMetsmmGroup≥50	7.1203	0.1404	1.18869	42.6504
RASMutant	2.7469	0.3641	1.34357	5.6159
Mets.TypeSynchronous	1.8612	0.5373	0.78316	4.4230
Oxaliplatin.HistoryYes	2.2538	0.4437	0.73552	6.9061

Concordance= 0.785 (se = 0.033)
 Likelihood ratio test= 35.06 on 8 df, p=3e-05
 Wald test = 28.56 on 8 df, p=4e-04
 Score (logrank) test = 35.96 on 8 df, p=2e-05

#DFS by ACT treatment in MRD negative - Synchronous metastatic site

Hide

```
rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$ctDNA.MRD!="",]
circ_data <- circ_data[circ_data$ctDNA.MRD=="NEGATIVE",]
circ_data <- circ_data[circ_data$Mets.Type=="Synchronous",]
circ_data$DFS.months=circ_data$DFS.months-2
circ_data <- circ_data[circ_data$DFS.months>=0,]
circ_datadf <- as.data.frame(circ_data)

survfit(Surv(time = circ_data$DFS.months, event = circ_data$DFS.Event)~ACT, data = circ_data)
```

```
Call: survfit(formula = Surv(time = circ_data$DFS.months, event = circ_data$DFS.Event) ~ ACT, data = circ_data)
```

```
  n events median 0.95LCL 0.95UCL
ACT=FALSE 16      5      NA     9.01      NA
ACT=TRUE   17      3      NA      NA      NA
```

[Hide](#)

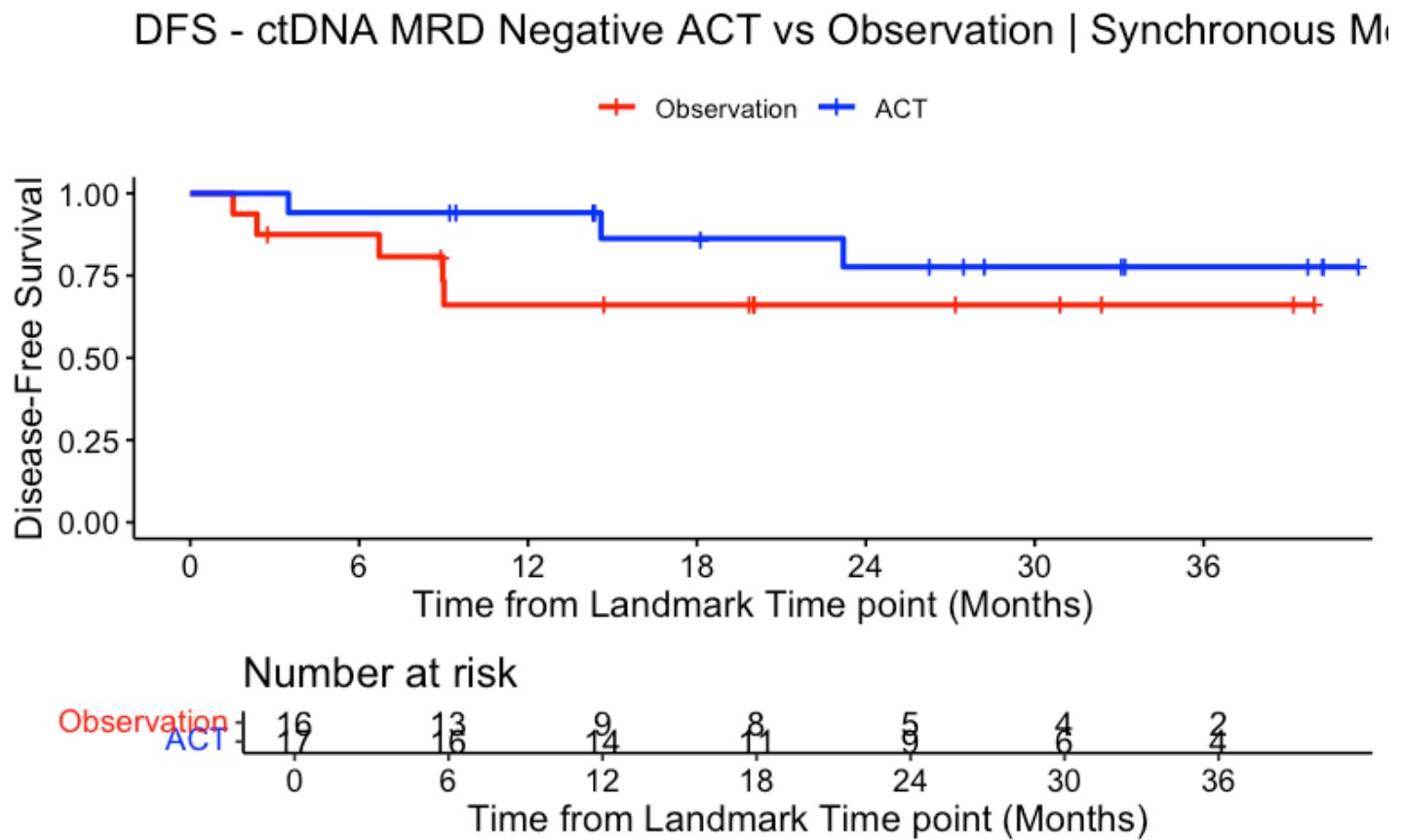
```
event_summary <- circ_data %>%
  group_by(ACT) %>%
  summarise(
    Total = n(),
    Events = sum(DFS.Event),
    Fraction = Events / n(),
    Percentage = (Events / n()) * 100
  )
print(event_summary)
```

ACT	Total	Events	Fraction	Percentage
	<int>	<int>	<dbl>	<dbl>
FALSE	16	5	0.3125000	31.25000
TRUE	17	3	0.1764706	17.64706

2 rows

[Hide](#)

```
surv_object <- Surv(time = circ_data$DFS.months, event = circ_data$DFS.Event)
KM_curve <- survfit(surv_object ~ ACT, data = circ_data, conf.int=0.95, conf.type="log-log")
ggsurvplot(KM_curve, data = circ_data, pval = FALSE, conf.int = FALSE, risk.table = TRUE,
break.time.by=6, palette=c("red","blue"), title="DFS - ctDNA MRD Negative ACT vs Observation | Synchronous Met Site", ylab= "Disease-Free Survival", xlab="Time from Landmark Time point (Months)", legend.labs=c("Observation", "ACT"), legend.title="")
```



```
summary(KM_curve, times= c(24))
```

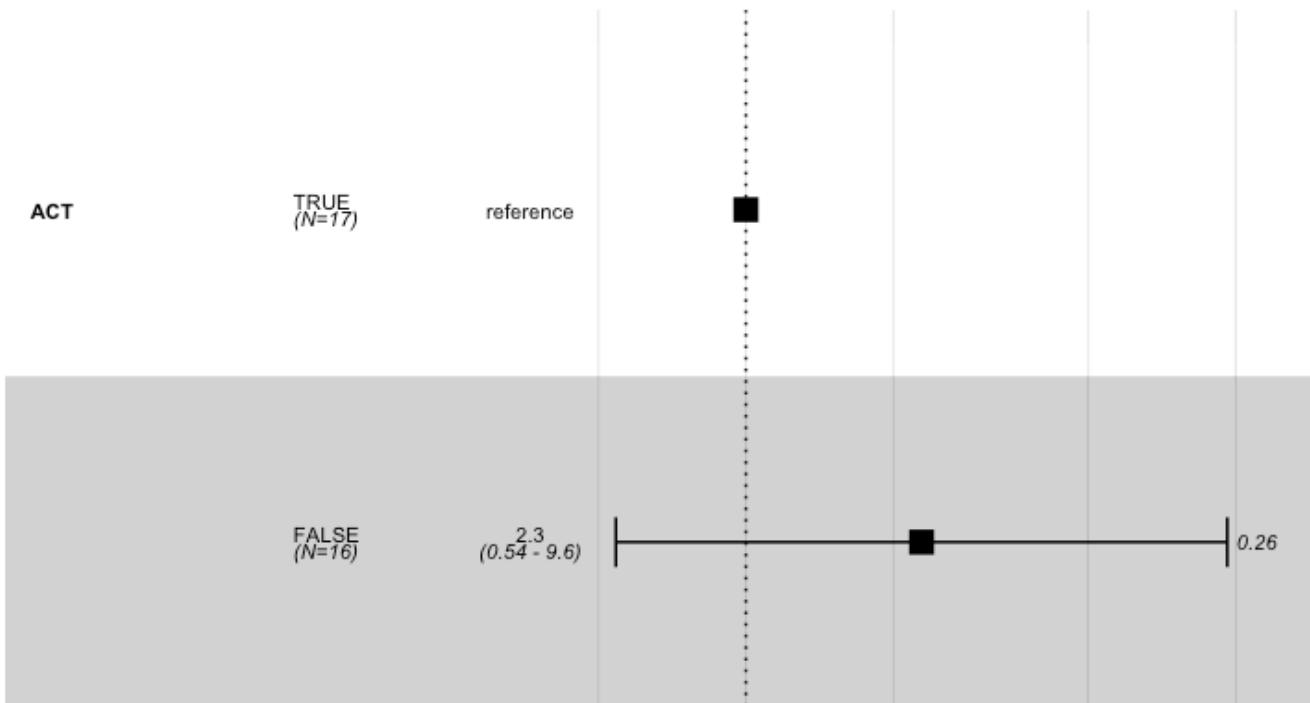
```
Call: survfit(formula = surv_object ~ ACT, data = circ_data, conf.int = 0.95,
  conf.type = "log-log")
```

CI	ACT=FALSE								
	time	n.risk	n.event	survival	std.err	lower	95% CI	upper	95%
44	24.000	5.000	5.000	0.661	0.125	0.364		0.8	
CI	ACT=TRUE								
	time	n.risk	n.event	survival	std.err	lower	95% CI	upper	95%
24	24.000	9.000	3.000	0.776	0.116	0.447		0.9	

[Hide](#)

```
circ_data$ACT <- factor(circ_data$ACT, levels=c("TRUE","FALSE"))
cox_fit <- coxph(surv_object ~ ACT, data=circ_data)
ggforest(cox_fit,data = circ_data)
```

Hazard ratio



[Hide](#)

```
summary(cox_fit)
```

Call:

```
coxph(formula = surv_object ~ ACT, data = circ_data)
```

n= 33, number of events= 8

	coef	exp(coef)	se(coef)	z	Pr(> z)
ACTFALSE	0.8259	2.2838	0.7334	1.126	0.26

	exp(coef)	exp(-coef)	lower .95	upper .95
ACTFALSE	2.284	0.4379	0.5425	9.615

Concordance= 0.634 (se = 0.082)

Likelihood ratio test= 1.32 on 1 df, p=0.3

Wald test = 1.27 on 1 df, p=0.3

Score (logrank) test = 1.34 on 1 df, p=0.2

[Hide](#)

```
cox_fit_summary <- summary(cox_fit)
```

```
# Extract values for HR, 95% CI, and p-value
HR <- cox_fit_summary$coefficients[2]
lower_CI <- cox_fit_summary$conf.int[3]
upper_CI <- cox_fit_summary$conf.int[4]
p_value <- cox_fit_summary$coefficients[5]
label_text <- paste0("HR = ", round(HR, 2), " (", round(lower_CI, 2), "-", round(upper_CI, 2), "); p = ", round(p_value, 3))
print(label_text)
```

```
[1] "HR = 2.28 (0.54-9.62); p = 0.26"
```

[Hide](#)

```
#Fisher test for DFS percentages at 24
dfs_times <- c(24)
p_values <- sapply(dfs_times, function(time) {
  neg_count <- sum(circ_data$ACT == "TRUE" & circ_data$DFS.months >= time & circ_data$DFS.Event == 0)
  pos_count <- sum(circ_data$ACT == "FALSE" & circ_data$DFS.months >= time & circ_data$DFS.Event == 0)
  neg_total <- sum(circ_data$ACT == "TRUE")
  pos_total <- sum(circ_data$ACT == "FALSE")

  neg_surv <- neg_total - sum(circ_data$ACT == "TRUE" & circ_data$DFS.Event == 1 & circ_data$DFS.months < time)
  pos_surv <- pos_total - sum(circ_data$ACT == "FALSE" & circ_data$DFS.Event == 1 & circ_data$DFS.months < time)

  surv_matrix <- matrix(c(neg_surv, pos_surv, neg_total - neg_surv, pos_total - pos_surv), nrow = 2)
  test_result <- fisher.test(surv_matrix)
  return(test_result$p.value)
})
names(p_values) <- paste0("p-value at ", dfs_times, " months")
print(p_values)
```

p-value at 24 months
0.4384333

[Hide](#)

```
p_values_adjusted <- p.adjust(p_values, method = "bonferroni")
names(p_values_adjusted) <- paste0("Corrected p-value at ", dfs_times, " months")
print(p_values_adjusted)
```

Corrected p-value at 24 months
0.4384333

[Hide](#)

```

#Adjusted HR "ACT vs no ACT" - age, gender, ECOG and pathological stage - ACT as reference
rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$ctDNA.MRD!="",]
circ_data <- circ_data[circ_data$ctDNA.MRD=="NEGATIVE",]
circ_data <- circ_data[circ_data$Mets.Type=="Synchronous",]
circ_data$DFS.months=circ_data$DFS.months-2
circ_data <- circ_data[circ_data$DFS.months>=0,]
circ_datadf <- as.data.frame(circ_data)

circ_data$Gender <- factor(circ_data$Gender, levels = c("Female", "Male"))
circ_data$Age.Group <- factor(circ_data$Age.Group, levels = c("1", "2"), labels = c("<70", ">70"))
circ_data$PrimSite <- factor(circ_data$PrimSite, levels = c("Right-sided colon", "Left-sided colon"))
circ_data$NumLiverMetsGroup <- factor(circ_data$NumLiverMetsGroup, levels = c("1", "≥2"))
circ_data$SizeLiverMetsmmGroup <- factor(circ_data$SizeLiverMetsmmGroup, levels = c("<50", "≥50"))
circ_data$ECOG <- factor(circ_data$ECOG, levels = c("0", "1"))
circ_data$pT <- factor(circ_data$pT, levels = c("T1-T2", "T3-T4"))
circ_data$pN <- factor(circ_data$pN, levels = c("N0", "N1-N2"))
circ_data$ACT <- factor(circ_data$ACT, levels = c("FALSE", "TRUE"))
circ_data$Postop.ComPLICATION <- factor(circ_data$Postop.ComPLICATION, levels = c("No", "Yes"))
circ_data$Oxaliplatin.History <- factor(circ_data$Oxaliplatin.History, levels = c("No", "Yes"))
circ_data$Mets.Type <- factor(circ_data$Mets.Type, levels = c("Metachronous", "Synchronous"))
circ_data$MSI <- factor(circ_data$MSI, levels = c("MSS", "MSI-High"), labels = c("MSS", "MSI-High"))
circ_data$BRAF.V600E <- factor(circ_data$BRAF.V600E, levels = c("WT", "MUT"), labels = c("Wild-Type", "V600E"))
circ_data$RAS <- factor(circ_data$RAS, levels = c("WT", "MUT"), labels = c("Wild-Type", "Mutant"))
surv_object <- Surv(time = circ_data$DFS.MRD.months, event = circ_data$DFS.Event)
cox_fit <- coxph(surv_object ~ ACT + Gender + Age.Group + NumLiverMetsGroup + SizeLiverMetsmmGroup + RAS + Oxaliplatin.History, data=circ_data)
summary(cox_fit)

```

Call:

```
coxph(formula = surv_object ~ ACT + Gender + Age.Group + NumLiverMetsGroup +
  SizeLiverMetsmmGroup + RAS + Oxaliplatin.History, data = circ_data)
```

n= 33, number of events= 8

	coef	exp(coef)	se(coef)	z	Pr(> z)
ACTTRUE	-1.5633	0.2094	0.8370	-1.868	0.0618 .
GenderMale	-1.1347	0.3215	0.9708	-1.169	0.2425
Age.Group>70	-0.6898	0.5017	0.8681	-0.795	0.4268
NumLiverMetsGroup≥2	1.0846	2.9583	0.9082	1.194	0.2324
SizeLiverMetsmmGroup≥50	1.7630	5.8302	1.3825	1.275	0.2022
RASMutant	1.5951	4.9288	1.0234	1.559	0.1191
Oxaliplatin.HistoryYes	NA	NA	0.0000	NA	NA

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

	exp(coef)	exp(-coef)	lower .95	upper .95
ACTTRUE	0.2094	4.7744	0.04061	1.080
GenderMale	0.3215	3.1102	0.04796	2.155
Age.Group>70	0.5017	1.9933	0.09151	2.750
NumLiverMetsGroup≥2	2.9583	0.3380	0.49889	17.543
SizeLiverMetsmmGroup≥50	5.8302	0.1715	0.38808	87.588
RASMutant	4.9288	0.2029	0.66321	36.630
Oxaliplatin.HistoryYes	NA	NA	NA	NA

Concordance= 0.824 (se = 0.07)

Likelihood ratio test= 10.15 on 6 df, p=0.1

Wald test = 7.56 on 6 df, p=0.3

Score (logrank) test = 9.31 on 6 df, p=0.2

#DFS by ACT treatment in MRD negative - Metachronous metastatic site

```
rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$ctDNA.MRD!="",]
circ_data <- circ_data[circ_data$ctDNA.MRD=="NEGATIVE",]
circ_data <- circ_data[circ_data$Mets.Type=="Metachronous",]
circ_data$DFS.months=circ_data$DFS.months-2
circ_data <- circ_data[circ_data$DFS.months>=0,]
circ_datadf <- as.data.frame(circ_data)

survfit(Surv(time = circ_data$DFS.months, event = circ_data$DFS.Event)~ACT, data = circ_data)
```

```
Call: survfit(formula = Surv(time = circ_data$DFS.months, event = circ_data$DFS.Event) ~
  ACT, data = circ_data)
```

	n	events	median	0.95LCL	0.95UCL
ACT=FALSE	76	28	NA	25.6	NA
ACT=TRUE	16	5	NA	14.6	NA

[Hide](#)

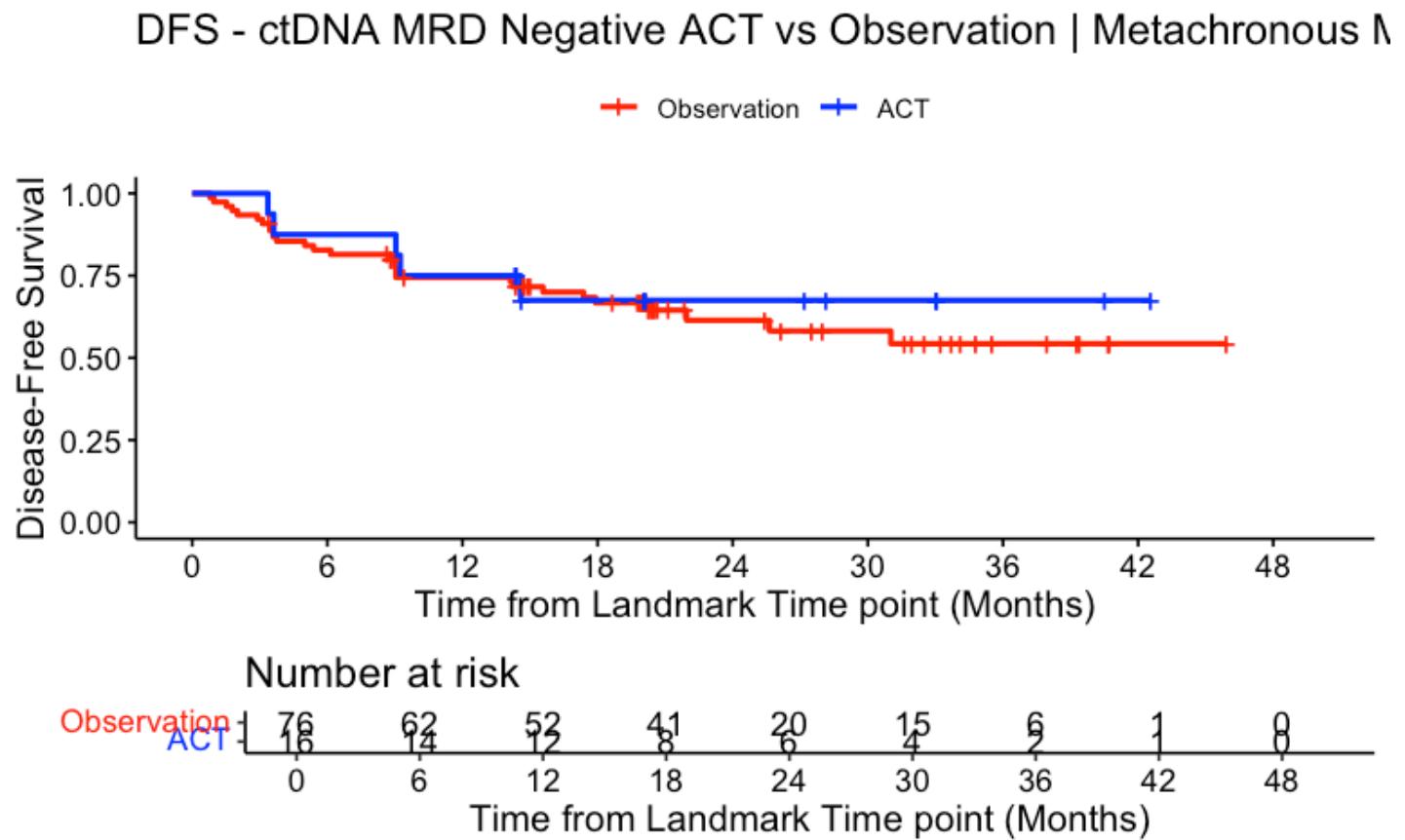
```
event_summary <- circ_data %>%
  group_by(ACT) %>%
  summarise(
    Total = n(),
    Events = sum(DFS.Event),
    Fraction = Events / n(),
    Percentage = (Events / n()) * 100
  )
print(event_summary)
```

ACT	Total	Events	Fraction	Percentage
FALSE	76	28	0.3684211	36.84211
TRUE	16	5	0.3125000	31.25000

2 rows

[Hide](#)

```
surv_object <- Surv(time = circ_data$DFS.months, event = circ_data$DFS.Event)
KM_curve <- survfit(surv_object ~ ACT, data = circ_data, conf.int=0.95, conf.type="log-log")
ggsurvplot(KM_curve, data = circ_data, pval = FALSE, conf.int = FALSE, risk.table = TRUE,
break.time.by=6, palette=c("red","blue"), title="DFS - ctDNA MRD Negative ACT vs Observation | Metachronous Met Site", ylab= "Disease-Free Survival", xlab="Time from Landmark Time point (Months)", legend.labs=c("Observation", "ACT"), legend.title="")
```



```
summary(KM_curve, times= c(24))
```

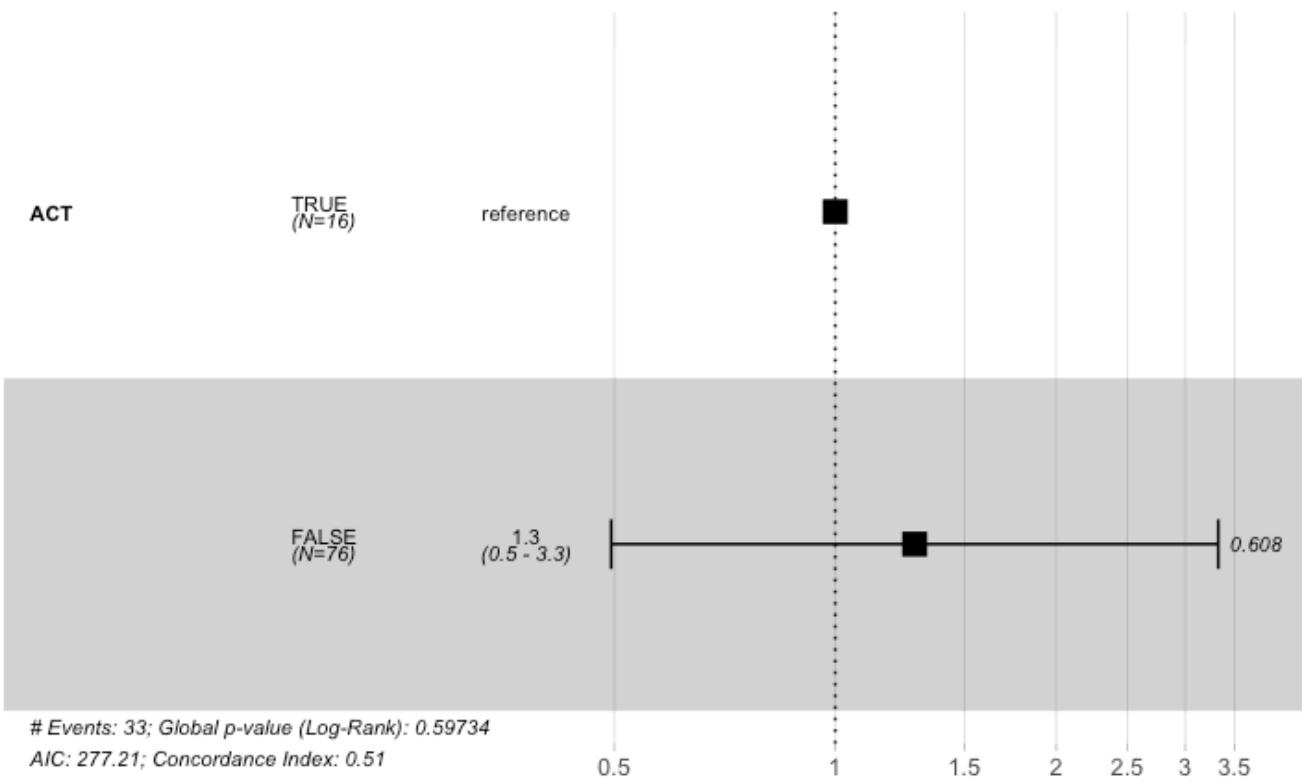
```
Call: survfit(formula = surv_object ~ ACT, data = circ_data, conf.int = 0.95,
  conf.type = "log-log")
```

CI	ACT=FALSE								
	time	n.risk	n.event	survival	std.err	lower	95% CI	upper	95%
46	24.0000	20.0000	26.0000	0.6142	0.0632	0.4784		0.72	
CI	ACT=TRUE								
	time	n.risk	n.event	survival	std.err	lower	95% CI	upper	95%
51	24.00	6.000	5.000	0.675	0.121	0.384		0.8	

Hide

```
circ_data$ACT <- factor(circ_data$ACT, levels=c("TRUE","FALSE"))
cox_fit <- coxph(surv_object ~ ACT, data=circ_data)
ggforest(cox_fit,data = circ_data)
```

Hazard ratio



Hide

```
summary(cox_fit)
```

Call:

coxph(formula = surv_object ~ ACT, data = circ_data)

n= 92, number of events= 33

	coef	exp(coef)	se(coef)	z	Pr(> z)
ACTFALSE	0.2494	1.2833	0.4859	0.513	0.608

	exp(coef)	exp(-coef)	lower .95	upper .95
ACTFALSE	1.283	0.7792	0.4951	3.326

Concordance= 0.514 (se = 0.033)

Likelihood ratio test= 0.28 on 1 df, p=0.6

Wald test = 0.26 on 1 df, p=0.6

Score (logrank) test = 0.26 on 1 df, p=0.6

Hide

cox_fit_summary <- summary(cox_fit)

```
# Extract values for HR, 95% CI, and p-value
HR <- cox_fit_summary$coefficients[2]
lower_CI <- cox_fit_summary$conf.int[3]
upper_CI <- cox_fit_summary$conf.int[4]
p_value <- cox_fit_summary$coefficients[5]
label_text <- paste0("HR = ", round(HR, 2), " (", round(lower_CI, 2), "-", round(upper_CI, 2), "); p = ", round(p_value, 3))
print(label_text)
```

[1] "HR = 1.28 (0.5-3.33); p = 0.608"

Hide

```
#Fisher test for DFS percentages at 24
dfs_times <- c(24)
p_values <- sapply(dfs_times, function(time) {
  neg_count <- sum(circ_data$ACT == "TRUE" & circ_data$DFS.months >= time & circ_data$DFS.Event == 0)
  pos_count <- sum(circ_data$ACT == "FALSE" & circ_data$DFS.months >= time & circ_data$DFS.Event == 0)
  neg_total <- sum(circ_data$ACT == "TRUE")
  pos_total <- sum(circ_data$ACT == "FALSE")

  neg_surv <- neg_total - sum(circ_data$ACT == "TRUE" & circ_data$DFS.Event == 1 & circ_data$DFS.months < time)
  pos_surv <- pos_total - sum(circ_data$ACT == "FALSE" & circ_data$DFS.Event == 1 & circ_data$DFS.months < time)

  surv_matrix <- matrix(c(neg_surv, pos_surv, neg_total - neg_surv, pos_total - pos_surv), nrow = 2)
  test_result <- fisher.test(surv_matrix)
  return(test_result$p.value)
})
names(p_values) <- paste0("p-value at ", dfs_times, " months")
print(p_values)
```

p-value at 24 months

1

[Hide](#)

```
p_values_adjusted <- p.adjust(p_values, method = "bonferroni")
names(p_values_adjusted) <- paste0("Corrected p-value at ", dfs_times, " months")
print(p_values_adjusted)
```

Corrected p-value at 24 months

1

[Hide](#)

```

#Adjusted HR "ACT vs no ACT" - age, gender, ECOG and pathological stage - ACT as reference
rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$ctDNA.MRD!="",]
circ_data <- circ_data[circ_data$ctDNA.MRD=="NEGATIVE",]
circ_data <- circ_data[circ_data$Mets.Type=="Metachronous",]
circ_data$DFS.months=circ_data$DFS.months-2
circ_data <- circ_data[circ_data$DFS.months>=0,]
circ_datadf <- as.data.frame(circ_data)

circ_data$Gender <- factor(circ_data$Gender, levels = c("Female", "Male"))
circ_data$Age.Group <- factor(circ_data$Age.Group, levels = c("1", "2"), labels = c("<70", ">70"))
circ_data$PrimSite <- factor(circ_data$PrimSite, levels = c("Right-sided colon", "Left-sided colon"))
circ_data$NumLiverMetsGroup <- factor(circ_data$NumLiverMetsGroup, levels = c("1", "≥2"))
circ_data$SizeLiverMetsmmGroup <- factor(circ_data$SizeLiverMetsmmGroup, levels = c("<50", "≥50"))
circ_data$ECOG <- factor(circ_data$ECOG, levels = c("0", "1"))
circ_data$pT <- factor(circ_data$pT, levels = c("T1-T2", "T3-T4"))
circ_data$pN <- factor(circ_data$pN, levels = c("N0", "N1-N2"))
circ_data$ACT <- factor(circ_data$ACT, levels = c("FALSE", "TRUE"))
circ_data$Postop.ComPLICATION <- factor(circ_data$Postop.ComPLICATION, levels = c("No", "Yes"))
circ_data$Oxaliplatin.History <- factor(circ_data$Oxaliplatin.History, levels = c("No", "Yes"))
circ_data$Mets.Type <- factor(circ_data$Mets.Type, levels = c("Metachronous", "SynchroNus"))
circ_data$MSI <- factor(circ_data$MSI, levels = c("MSS", "MSI-High"), labels = c("MSS", "MSI-High"))
circ_data$BRAF.V600E <- factor(circ_data$BRAF.V600E, levels = c("WT", "MUT"), labels = c("Wild-Type", "V600E"))
circ_data$RAS <- factor(circ_data$RAS, levels = c("WT", "MUT"), labels = c("Wild-Type", "Mutant"))
surv_object <- Surv(time = circ_data$DFS.MRD.months, event = circ_data$DFS.Event)
cox_fit <- coxph(surv_object ~ ACT + Gender + Age.Group + NumLiverMetsGroup + SizeLiverMetsmmGroup + RAS + Oxaliplatin.History, data=circ_data)
summary(cox_fit)

```

Call:

```
coxph(formula = surv_object ~ ACT + Gender + Age.Group + NumLiverMetsGroup +
  SizeLiverMetsmmGroup + RAS + Oxaliplatin.History, data = circ_data)
```

n= 92, number of events= 33

	coef	exp(coef)	se(coef)	z	Pr(> z)
ACTTRUE	-0.21989	0.80261	0.50394	-0.436	0.6626
GenderMale	0.83345	2.30124	0.47385	1.759	0.0786
Age.Group>70	0.49048	1.63310	0.37182	1.319	0.1871
NumLiverMetsGroup≥2	0.05637	1.05799	0.42226	0.133	0.8938
SizeLiverMetsmmGroup≥50	0.39198	1.47990	0.75395	0.520	0.6031
RASMutant	0.23766	1.26827	0.39924	0.595	0.5517
Oxaliplatin.HistoryYes	0.41547	1.51508	0.39143	1.061	0.2885

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

	exp(coef)	exp(-coef)	lower .95	upper .95
ACTTRUE	0.8026	1.2459	0.2989	2.155
GenderMale	2.3012	0.4345	0.9091	5.825
Age.Group>70	1.6331	0.6123	0.7880	3.385
NumLiverMetsGroup≥2	1.0580	0.9452	0.4624	2.421
SizeLiverMetsmmGroup≥50	1.4799	0.6757	0.3377	6.486
RASMutant	1.2683	0.7885	0.5799	2.774
Oxaliplatin.HistoryYes	1.5151	0.6600	0.7035	3.263

Concordance= 0.623 (se = 0.046)

Likelihood ratio test= 6.09 on 7 df, p=0.5

Wald test = 5.71 on 7 df, p=0.6

Score (logrank) test = 5.8 on 7 df, p=0.6

#DFS by ACT treatment in MRD negative - 1 Liver Met

```
rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$ctDNA.MRD!="",]
circ_data <- circ_data[circ_data$ctDNA.MRD=="NEGATIVE",]
circ_data <- circ_data[circ_data$NumLiverMetsGroup=="1",]
circ_data$DFS.months=circ_data$DFS.months-2
circ_data <- circ_data[circ_data$DFS.months>=0,]
circ_datadf <- as.data.frame(circ_data)

survfit(Surv(time = circ_data$DFS.months, event = circ_data$DFS.Event)~ACT, data = circ_data)
```

```
Call: survfit(formula = Surv(time = circ_data$DFS.months, event = circ_data$DFS.Event) ~
  ACT, data = circ_data)
```

n events median 0.95LCL 0.95UCL					
ACT=FALSE	69	22	NA	31	NA
ACT=TRUE	22	6	NA	NA	NA

[Hide](#)

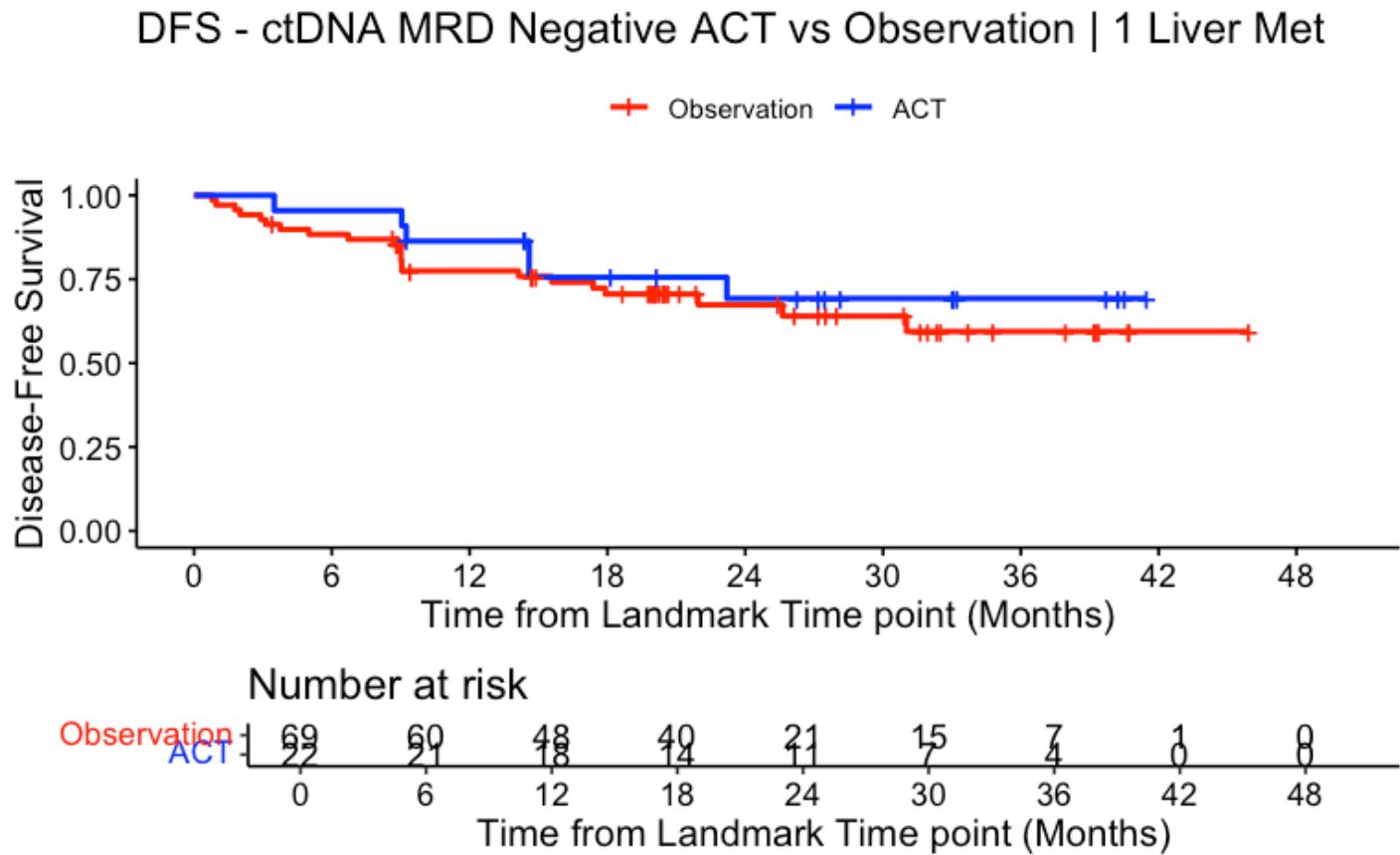
```
event_summary <- circ_data %>%
  group_by(ACT) %>%
  summarise(
    Total = n(),
    Events = sum(DFS.Event),
    Fraction = Events / n(),
    Percentage = (Events / n()) * 100
  )
print(event_summary)
```

ACT	Total	Events	Fraction	Percentage
FALSE	69	22	0.3188406	31.88406
TRUE	22	6	0.2727273	27.27273

2 rows

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```
surv_object <- Surv(time = circ_data$DFS.months, event = circ_data$DFS.Event)
KM_curve <- survfit(surv_object ~ ACT, data = circ_data, conf.int=0.95, conf.type="log-log")
ggsurvplot(KM_curve, data = circ_data, pval = FALSE, conf.int = FALSE, risk.table = TRUE,
break.time.by=6, palette=c("red","blue"), title="DFS - ctDNA MRD Negative ACT vs Observation | 1 Liver Met", ylab= "Disease-Free Survival", xlab="Time from Landmark Time point (Months)", legend.labs=c("Observation", "ACT"), legend.title="")
```



```
summary(KM_curve, times= c(24))
```

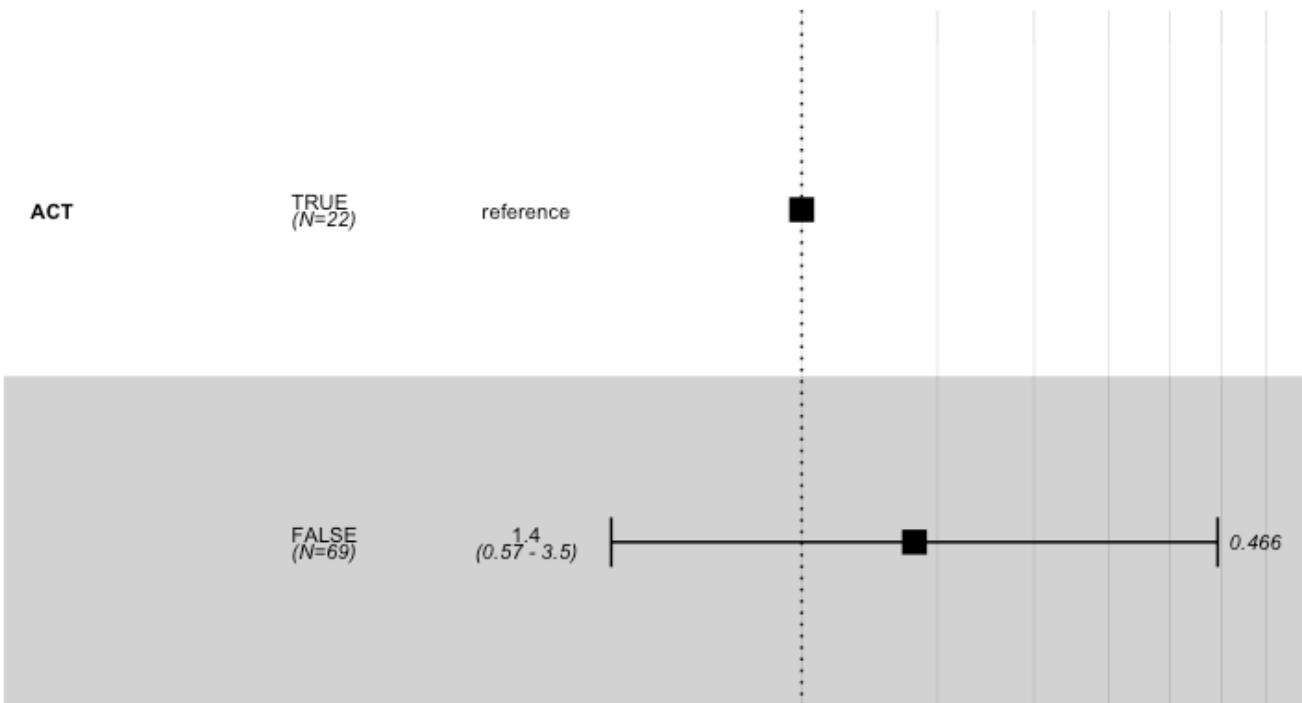
```
Call: survfit(formula = surv_object ~ ACT, data = circ_data, conf.int = 0.95,
  conf.type = "log-log")
```

CI	ACT=FALSE								
	time	n.risk	n.event	survival	std.err	lower	95% CI	upper	95%
80	24.000	21.000	20.000	0.674	0.063	0.534		0.7	
<hr/>									
CI	ACT=TRUE								
	time	n.risk	n.event	survival	std.err	lower	95% CI	upper	95%
51	24.000	11.000	6.000	0.693	0.107	0.434		0.8	

[Hide](#)

```
circ_data$ACT <- factor(circ_data$ACT, levels=c("TRUE","FALSE"))
cox_fit <- coxph(surv_object ~ ACT, data=circ_data)
ggforest(cox_fit,data = circ_data)
```

Hazard ratio

[Hide](#)

```
summary(cox_fit)
```

Call:

```
coxph(formula = surv_object ~ ACT, data = circ_data)
```

n= 91, number of events= 28

	coef	exp(coef)	se(coef)	z	Pr(> z)
ACTFALSE	0.3365	1.4000	0.4618	0.729	0.466

	exp(coef)	exp(-coef)	lower .95	upper .95
ACTFALSE	1.4	0.7143	0.5663	3.461

Concordance= 0.536 (se = 0.038)

Likelihood ratio test= 0.56 on 1 df, p=0.5

Wald test = 0.53 on 1 df, p=0.5

Score (logrank) test = 0.54 on 1 df, p=0.5

[Hide](#)

```
cox_fit_summary <- summary(cox_fit)
```

```
# Extract values for HR, 95% CI, and p-value
HR <- cox_fit_summary$coefficients[2]
lower_CI <- cox_fit_summary$conf.int[3]
upper_CI <- cox_fit_summary$conf.int[4]
p_value <- cox_fit_summary$coefficients[5]
label_text <- paste0("HR = ", round(HR, 2), " (", round(lower_CI, 2), "-", round(upper_CI, 2), "); p = ", round(p_value, 3))
print(label_text)
```

```
[1] "HR = 1.4 (0.57-3.46); p = 0.466"
```

[Hide](#)

```
#Fisher test for DFS percentages at 24
dfs_times <- c(24)
p_values <- sapply(dfs_times, function(time) {
  neg_count <- sum(circ_data$ACT == "TRUE" & circ_data$DFS.months >= time & circ_data$DFS.Event == 0)
  pos_count <- sum(circ_data$ACT == "FALSE" & circ_data$DFS.months >= time & circ_data$DFS.Event == 0)
  neg_total <- sum(circ_data$ACT == "TRUE")
  pos_total <- sum(circ_data$ACT == "FALSE")

  neg_surv <- neg_total - sum(circ_data$ACT == "TRUE" & circ_data$DFS.Event == 1 & circ_data$DFS.months < time)
  pos_surv <- pos_total - sum(circ_data$ACT == "FALSE" & circ_data$DFS.Event == 1 & circ_data$DFS.months < time)

  surv_matrix <- matrix(c(neg_surv, pos_surv, neg_total - neg_surv, pos_total - pos_surv), nrow = 2)
  test_result <- fisher.test(surv_matrix)
  return(test_result$p.value)
})
names(p_values) <- paste0("p-value at ", dfs_times, " months")
print(p_values)
```

p-value at 24 months

1

[Hide](#)

```
p_values_adjusted <- p.adjust(p_values, method = "bonferroni")
names(p_values_adjusted) <- paste0("Corrected p-value at ", dfs_times, " months")
print(p_values_adjusted)
```

Corrected p-value at 24 months

1

[Hide](#)

```

#Adjusted HR "ACT vs no ACT" - age, gender, ECOG and pathological stage - ACT as reference
rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$ctDNA.MRD!="",]
circ_data <- circ_data[circ_data$ctDNA.MRD=="NEGATIVE",]
circ_data <- circ_data[circ_data$NumLiverMetsGroup=="1",]
circ_data$DFS.months=circ_data$DFS.months-2
circ_data <- circ_data[circ_data$DFS.months>=0,]
circ_datadf <- as.data.frame(circ_data)

circ_data$Gender <- factor(circ_data$Gender, levels = c("Female", "Male"))
circ_data$Age.Group <- factor(circ_data$Age.Group, levels = c("1", "2"), labels = c("<70", ">70"))
circ_data$PrimSite <- factor(circ_data$PrimSite, levels = c("Right-sided colon", "Left-sided colon"))
circ_data$NumLiverMetsGroup <- factor(circ_data$NumLiverMetsGroup, levels = c("1", "≥2"))
circ_data$SizeLiverMetsmmGroup <- factor(circ_data$SizeLiverMetsmmGroup, levels = c("<50", "≥50"))
circ_data$ECOG <- factor(circ_data$ECOG, levels = c("0", "1"))
circ_data$pT <- factor(circ_data$pT, levels = c("T1-T2", "T3-T4"))
circ_data$pN <- factor(circ_data$pN, levels = c("N0", "N1-N2"))
circ_data$ACT <- factor(circ_data$ACT, levels = c("FALSE", "TRUE"))
circ_data$Postop.ComPLICATION <- factor(circ_data$Postop.ComPLICATION, levels = c("No", "Yes"))
circ_data$Oxaliplatin.History <- factor(circ_data$Oxaliplatin.History, levels = c("No", "Yes"))
circ_data$Mets.Type <- factor(circ_data$Mets.Type, levels = c("Metachronous", "Synchronous"))
circ_data$MSI <- factor(circ_data$MSI, levels = c("MSS", "MSI-High"), labels = c("MSS", "MSI-High"))
circ_data$BRAF.V600E <- factor(circ_data$BRAF.V600E, levels = c("WT", "MUT"), labels = c("Wild-Type", "V600E"))
circ_data$RAS <- factor(circ_data$RAS, levels = c("WT", "MUT"), labels = c("Wild-Type", "Mutant"))
surv_object <- Surv(time = circ_data$DFS.MRD.months, event = circ_data$DFS.Event)
cox_fit <- coxph(surv_object ~ ACT + Gender + Age.Group + Mets.Type + SizeLiverMetsmmGroup + RAS + Oxaliplatin.History, data=circ_data)
summary(cox_fit)

```

Call:

```
coxph(formula = surv_object ~ ACT + Gender + Age.Group + Mets.Type +
  SizeLiverMetsmmGroup + RAS + Oxaliplatin.History, data = circ_data)
```

n= 91, number of events= 28

	coef	exp(coef)	se(coef)	z	Pr(> z)
ACTTRUE	-0.02522	0.97509	0.52282	-0.048	0.9615
GenderMale	0.82189	2.27480	0.49890	1.647	0.0995 .
Age.Group>70	0.05878	1.06054	0.42162	0.139	0.8891
Mets.TypeSynchronous	-0.38867	0.67796	0.55840	-0.696	0.4864
SizeLiverMetsmmGroup≥50	1.26344	3.53757	0.63719	1.983	0.0474 *
RASMutant	0.82093	2.27261	0.45519	1.803	0.0713 .
Oxaliplatin.HistoryYes	0.25359	1.28864	0.50661	0.501	0.6167

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

	exp(coef)	exp(-coef)	lower .95	upper .95
ACTTRUE	0.9751	1.0255	0.3500	2.717
GenderMale	2.2748	0.4396	0.8556	6.048
Age.Group>70	1.0605	0.9429	0.4641	2.423
Mets.TypeSynchronous	0.6780	1.4750	0.2269	2.025
SizeLiverMetsmmGroup≥50	3.5376	0.2827	1.0147	12.333
RASMutant	2.2726	0.4400	0.9312	5.546
Oxaliplatin.HistoryYes	1.2886	0.7760	0.4774	3.478

Concordance= 0.66 (se = 0.053)

Likelihood ratio test= 8.34 on 7 df, p=0.3

Wald test = 9.02 on 7 df, p=0.3

Score (logrank) test = 9.93 on 7 df, p=0.2

#DFS by ACT treatment in MRD negative - 2≥ Liver Mets

Hide

```
rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$ctDNA.MRD!="",]
circ_data <- circ_data[circ_data$ctDNA.MRD=="NEGATIVE",]
circ_data <- circ_data[circ_data$NumLiverMetsGroup=="≥2",]
circ_data$DFS.months=circ_data$DFS.months-2
circ_data <- circ_data[circ_data$DFS.months>=0,]
circ_datadf <- as.data.frame(circ_data)

survfit(Surv(time = circ_data$DFS.months, event = circ_data$DFS.Event)~ACT, data = circ_data)
```

```
Call: survfit(formula = Surv(time = circ_data$DFS.months, event = circ_data$DFS.Event) ~ ACT, data = circ_data)
```

	n	events	median	0.95LCL	0.95UCL
ACT=FALSE	23	11	20.3	6.15	NA
ACT=TRUE	11	2	NA	NA	NA

[Hide](#)

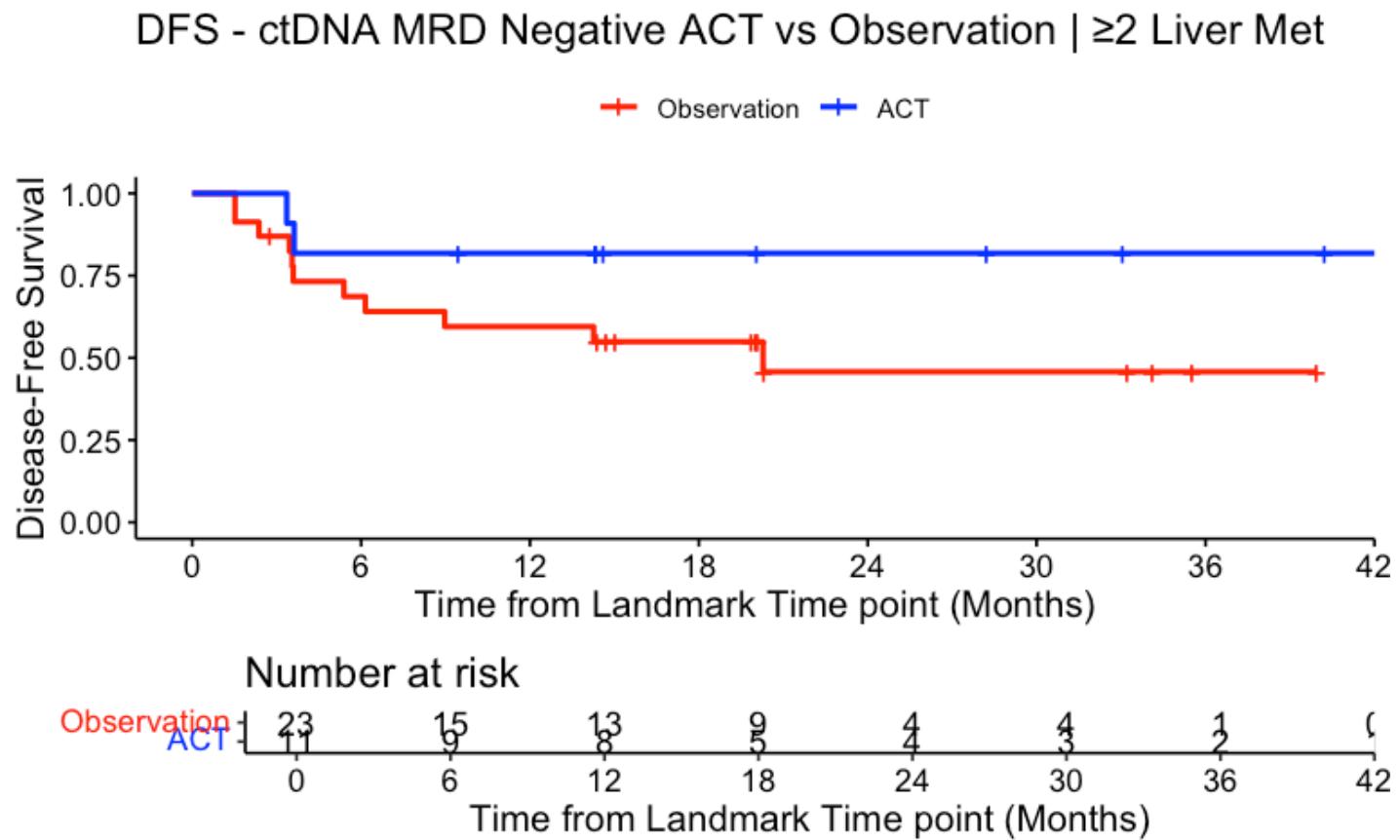
```
event_summary <- circ_data %>%
  group_by(ACT) %>%
  summarise(
    Total = n(),
    Events = sum(DFS.Event),
    Fraction = Events / n(),
    Percentage = (Events / n()) * 100
  )
print(event_summary)
```

ACT	Total	Events	Fraction	Percentage
FALSE	23	11	0.4782609	47.82609
TRUE	11	2	0.1818182	18.18182

2 rows

[Hide](#)

```
surv_object <- Surv(time = circ_data$DFS.months, event = circ_data$DFS.Event)
KM_curve <- survfit(surv_object ~ ACT, data = circ_data, conf.int=0.95, conf.type="log-log")
ggsurvplot(KM_curve, data = circ_data, pval = FALSE, conf.int = FALSE, risk.table = TRUE,
break.time.by=6, palette=c("red","blue"), title="DFS - ctDNA MRD Negative ACT vs Observation | ≥2 Liver Met", ylab= "Disease-Free Survival", xlab="Time from Landmark Time point (Months)", legend.labs=c("Observation", "ACT"), legend.title="")
```



```
summary(KM_curve, times= c(24))
```

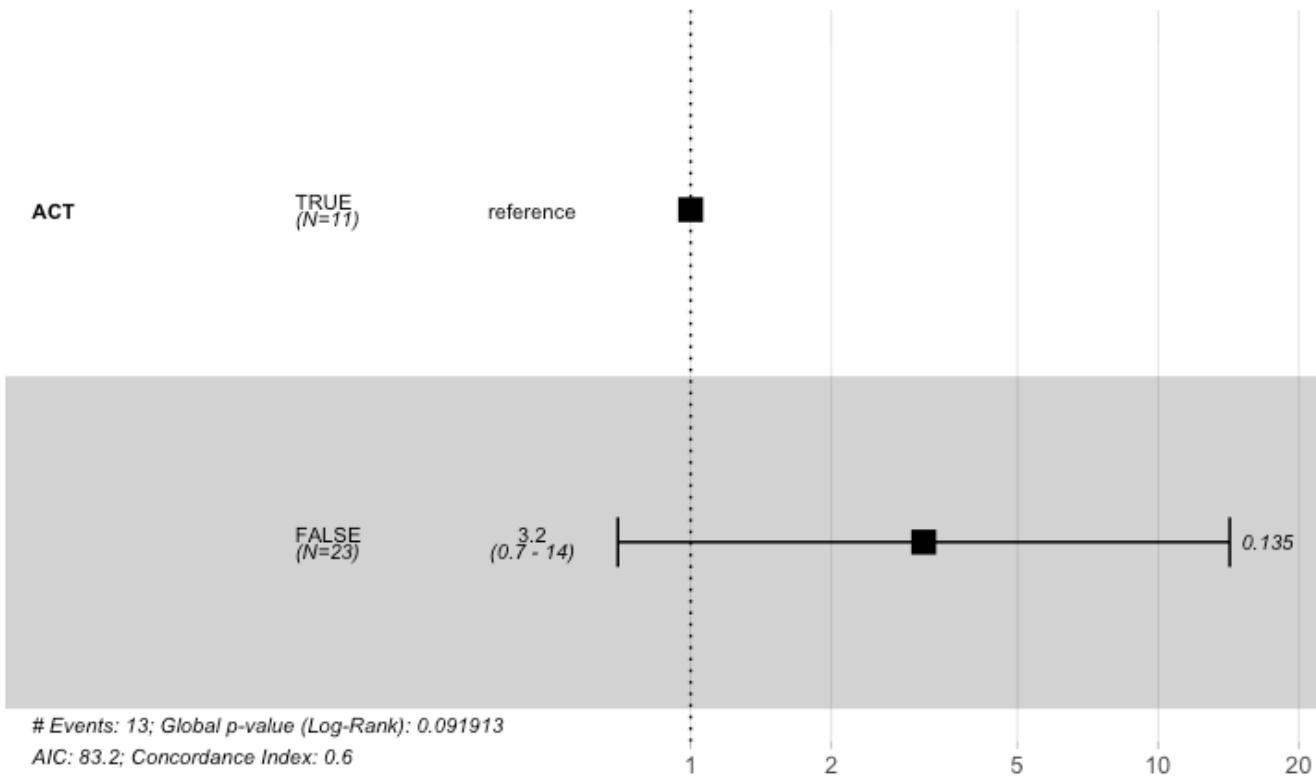
```
Call: survfit(formula = surv_object ~ ACT, data = circ_data, conf.int = 0.95,
  conf.type = "log-log")
```

CI	ACT=FALSE								
	time	n.risk	n.event	survival	std.err	lower	95% CI	upper	95%
69	24.000	4.000	11.000	0.458	0.122	0.218		0.6	
CI	ACT=TRUE								
	time	n.risk	n.event	survival	std.err	lower	95% CI	upper	95%
51	24.000	4.000	2.000	0.818	0.116	0.447		0.9	

[Hide](#)

```
circ_data$ACT <- factor(circ_data$ACT, levels=c("TRUE","FALSE"))
cox_fit <- coxph(surv_object ~ ACT, data=circ_data)
ggforest(cox_fit,data = circ_data)
```

Hazard ratio

[Hide](#)

```
summary(cox_fit)
```

Call:

```
coxph(formula = surv_object ~ ACT, data = circ_data)
```

n= 34, number of events= 13

	coef	exp(coef)	se(coef)	z	Pr(> z)
ACTFALSE	1.1484	3.1533	0.7692	1.493	0.135

	exp(coef)	exp(-coef)	lower .95	upper .95
ACTFALSE	3.153	0.3171	0.6983	14.24

Concordance= 0.602 (se = 0.062)

Likelihood ratio test= 2.84 on 1 df, p=0.09

Wald test = 2.23 on 1 df, p=0.1

Score (logrank) test = 2.48 on 1 df, p=0.1

[Hide](#)

```
cox_fit_summary <- summary(cox_fit)
```

```
# Extract values for HR, 95% CI, and p-value
HR <- cox_fit_summary$coefficients[2]
lower_CI <- cox_fit_summary$conf.int[3]
upper_CI <- cox_fit_summary$conf.int[4]
p_value <- cox_fit_summary$coefficients[5]
label_text <- paste0("HR = ", round(HR, 2), " (", round(lower_CI, 2), "-", round(upper_CI, 2), "); p = ", round(p_value, 3))
print(label_text)
```

```
[1] "HR = 3.15 (0.7-14.24); p = 0.135"
```

[Hide](#)

```
#Fisher test for DFS percentages at 24
dfs_times <- c(24)
p_values <- sapply(dfs_times, function(time) {
  neg_count <- sum(circ_data$ACT == "TRUE" & circ_data$DFS.months >= time & circ_data$DFS.Event == 0)
  pos_count <- sum(circ_data$ACT == "FALSE" & circ_data$DFS.months >= time & circ_data$DFS.Event == 0)
  neg_total <- sum(circ_data$ACT == "TRUE")
  pos_total <- sum(circ_data$ACT == "FALSE")

  neg_surv <- neg_total - sum(circ_data$ACT == "TRUE" & circ_data$DFS.Event == 1 & circ_data$DFS.months < time)
  pos_surv <- pos_total - sum(circ_data$ACT == "FALSE" & circ_data$DFS.Event == 1 & circ_data$DFS.months < time)

  surv_matrix <- matrix(c(neg_surv, pos_surv, neg_total - neg_surv, pos_total - pos_surv), nrow = 2)
  test_result <- fisher.test(surv_matrix)
  return(test_result$p.value)
})
names(p_values) <- paste0("p-value at ", dfs_times, " months")
print(p_values)
```

p-value at 24 months
0.139822

[Hide](#)

```
p_values_adjusted <- p.adjust(p_values, method = "bonferroni")
names(p_values_adjusted) <- paste0("Corrected p-value at ", dfs_times, " months")
print(p_values_adjusted)
```

Corrected p-value at 24 months
0.139822

[Hide](#)

```
#Adjusted HR "ACT vs no ACT" - age, gender, ECOG and pathological stage - ACT as reference
rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$ctDNA.MRD!="",]
circ_data <- circ_data[circ_data$ctDNA.MRD=="NEGATIVE",]
circ_data <- circ_data[circ_data$NumLiverMetsGroup=="≥2",]
circ_data$DFS.months=circ_data$DFS.months-2
circ_data <- circ_data[circ_data$DFS.months≥0,]
circ_datadf <- as.data.frame(circ_data)

circ_data$Gender <- factor(circ_data$Gender, levels = c("Female", "Male"))
circ_data$Age.Group <- factor(circ_data$Age.Group, levels = c("1", "2"), labels = c("<70", ">70"))
circ_data$PrimSite <- factor(circ_data$PrimSite, levels = c("Right-sided colon", "Left-sided colon"))
circ_data$NumLiverMetsGroup <- factor(circ_data$NumLiverMetsGroup, levels = c("1", "≥2"))
circ_data$SizeLiverMetsmmGroup <- factor(circ_data$SizeLiverMetsmmGroup, levels = c("<50", "≥50"))
circ_data$ECOG <- factor(circ_data$ECOG, levels = c("0", "1"))
circ_data$pT <- factor(circ_data$pT, levels = c("T1-T2", "T3-T4"))
circ_data$pN <- factor(circ_data$pN, levels = c("N0", "N1-N2"))
circ_data$ACT <- factor(circ_data$ACT, levels = c("FALSE", "TRUE"))
circ_data$Postop.ComPLICATION <- factor(circ_data$Postop.ComPLICATION, levels = c("No", "Yes"))
circ_data$Oxaliplatin.History <- factor(circ_data$Oxaliplatin.History, levels = c("No", "Yes"))
circ_data$Mets.Type <- factor(circ_data$Mets.Type, levels = c("Metachronous", "Synchronous"))
circ_data$MSI <- factor(circ_data$MSI, levels = c("MSS", "MSI-High"), labels = c("MSS", "MSI-High"))
circ_data$BRAF.V600E <- factor(circ_data$BRAF.V600E, levels = c("WT", "MUT"), labels = c("Wild-Type", "V600E"))
circ_data$RAS <- factor(circ_data$RAS, levels = c("WT", "MUT"), labels = c("Wild-Type", "Mutant"))
surv_object <- Surv(time = circ_data$DFS.MRD.months, event = circ_data$DFS.Event)
cox_fit <- coxph(surv_object ~ ACT + Gender + Age.Group + Mets.Type + RAS + Oxaliplatin.History, data=circ_data)
summary(cox_fit)
```

Call:

```
coxph(formula = surv_object ~ ACT + Gender + Age.Group + Mets.Type +
  RAS + Oxaliplatin.History, data = circ_data)
```

n= 34, number of events= 13

	coef	exp(coef)	se(coef)	z	Pr(> z)
ACTTRUE	-1.350297	0.259163	0.833079	-1.621	0.105
GenderMale	-0.588374	0.555229	0.792352	-0.743	0.458
Age.Group>70	0.399882	1.491649	0.597240	0.670	0.503
Mets.TypeSynchronous	-0.006411	0.993610	0.794551	-0.008	0.994
RASMutant	-0.396021	0.672993	0.629514	-0.629	0.529
Oxaliplatin.HistoryYes	0.149209	1.160916	0.690752	0.216	0.829

	exp(coef)	exp(-coef)	lower .95	upper .95
ACTTRUE	0.2592	3.8586	0.05064	1.326
GenderMale	0.5552	1.8011	0.11750	2.624
Age.Group>70	1.4916	0.6704	0.46269	4.809
Mets.TypeSynchronous	0.9936	1.0064	0.20936	4.716
RASMutant	0.6730	1.4859	0.19596	2.311
Oxaliplatin.HistoryYes	1.1609	0.8614	0.29980	4.495

Concordance= 0.661 (se = 0.086)

Likelihood ratio test= 4.56 on 6 df, p=0.6

Wald test = 3.86 on 6 df, p=0.7

Score (logrank) test = 4.16 on 6 df, p=0.7

#DFS by ctDNA Clearance ACT-treated at 3 months

Hide

```
rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$ctDNA.MRD!="",]
circ_data <- circ_data[circ_data$ACT==TRUE,]
circ_datadf <- as.data.frame(circ_data)

circ_data$ctDNA.Dynamics <- NA #first we create the variable for the ctDNA & NAC combination, and we assign values
circ_data <- circ_data %>%
  mutate(ctDNA.Dynamics = case_when(
    ctDNA.MRD == "POSITIVE" & ctDNA.3months == "NEGATIVE" ~ 1,
    ctDNA.MRD == "POSITIVE" & ctDNA.3months == "POSITIVE" ~ 2
  ))

circ_data <- circ_data[circ_data$DFS.3mo.months>=0,]
survfit(Surv(time = circ_data$DFS.3mo.months, event = circ_data$DFS.Event)~ctDNA.Dynamics, data = circ_data)
```

```
Call: survfit(formula = Surv(time = circ_data$DFS.3mo.months, event = circ_data$DFS.Event) ~
  ctDNA.Dynamics, data = circ_data)

35 observations deleted due to missingness
  n events median 0.95LCL 0.95UCL
ctDNA.Dynamics=1 8      4  14.00  13.80      NA
ctDNA.Dynamics=2 5      5  3.42   3.02      NA
```

[Hide](#)

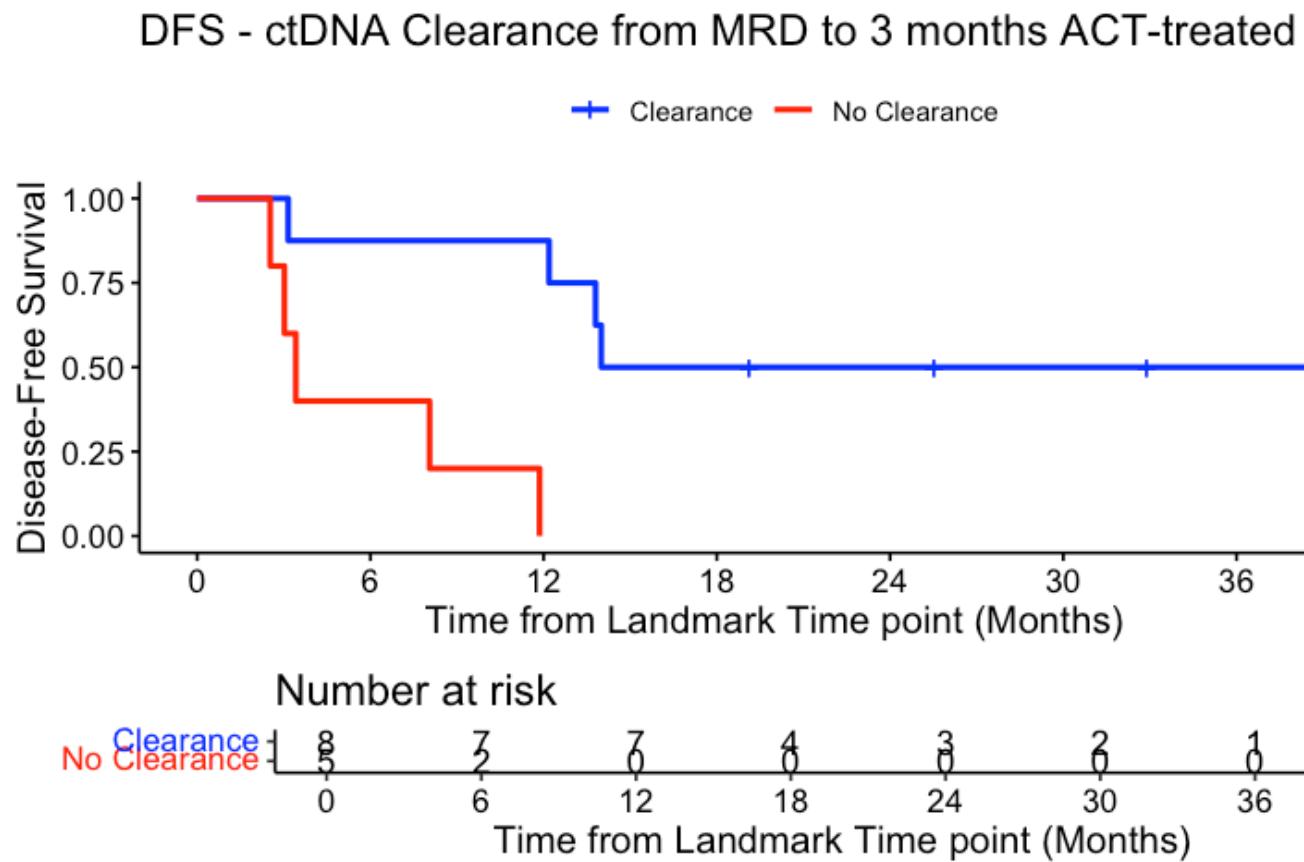
```
event_summary <- circ_data %>%
  group_by(ctDNA.Dynamics) %>%
  summarise(
    Total = n(),
    Events = sum(DFS.Event),
    Fraction = Events / n(),
    Percentage = (Events / n()) * 100
  )
print(event_summary)
```

ctDNA.Dynamics	Total	Events	Fraction	Percentage
	<dbl>	<int>	<dbl>	<dbl>
1	8	4	0.5	50
2	5	5	1.0	100
NA	35	NA	NA	NA

3 rows

[Hide](#)

```
surv_object <- Surv(time = circ_data$DFS.3mo.months, event = circ_data$DFS.Event)
KM_curve <- survfit(surv_object ~ ctDNA.Dynamics, data = circ_data, conf.int=0.95, conf.type="log-log")
ggsurvplot(KM_curve, data = circ_data, pval = FALSE, conf.int = FALSE, risk.table = TRUE, break.time.by=6, palette=c("blue","red"), title="DFS - ctDNA Clearance from MRD to 3 months ACT-treated", ylab= "Disease-Free Survival", xlab="Time from Landmark Time point (Months)", legend.labs=c("Clearance", "No Clearance"), legend.title="")
```



```
summary(KM_curve, times= c(6, 12, 24))
```

```
Call: survfit(formula = surv_object ~ ctDNA.Dynamics, data = circ_data,
  conf.int = 0.95, conf.type = "log-log")
```

35 observations deleted due to missingness

ctDNA.Dynamics=1

time	n.risk	n.event	survival	std.err	lower 95% CI	upper 95% CI
6	7	1	0.875	0.117	0.387	0.981
12	7	0	0.875	0.117	0.387	0.981
24	3	3	0.500	0.177	0.152	0.775

ctDNA.Dynamics=2

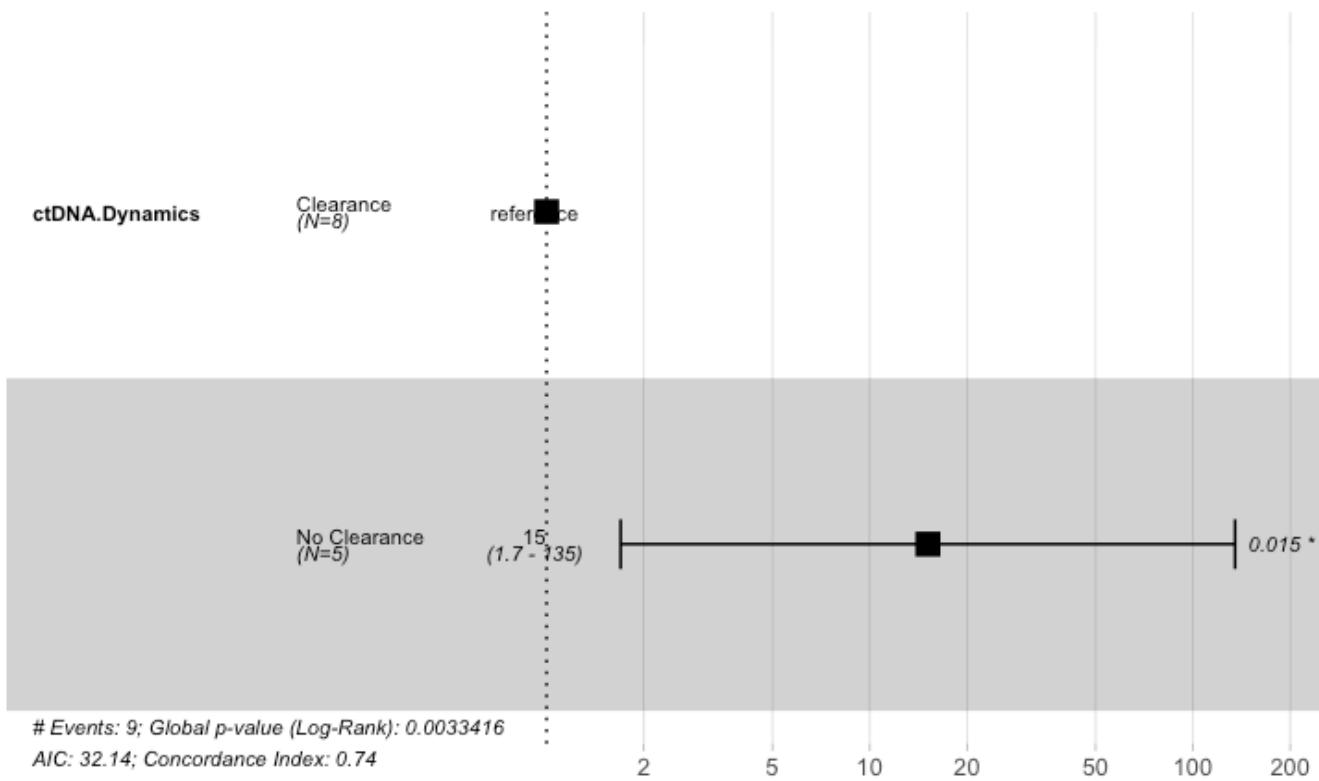
time	n.risk	n.event	survival	std.err	lower 95% CI	upper 95%
6.000	2.000	3.000	0.400	0.219	0.052	0.7

53

[Hide](#)

```
circ_data$ctDNA.Dynamics <- factor(circ_data$ctDNA.Dynamics, levels=c("1","2"), labels =
c("Clearance", "No Clearance"))
cox_fit <- coxph(surv_object ~ ctDNA.Dynamics, data=circ_data)
ggforest(cox_fit,data = circ_data)
```

Hazard ratio



[Hide](#)

```
summary(cox_fit)
```

Call:

```
coxph(formula = surv_object ~ ctDNA.Dynamics, data = circ_data)

n= 13, number of events= 9
(35 observations deleted due to missingness)

            coef exp(coef)  se(coef)      z Pr(>|z|)
ctDNA.DynamicsNo Clearance  2.717     15.131     1.117  2.432    0.015 *
---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

            exp(coef) exp(-coef) lower .95 upper .95
ctDNA.DynamicsNo Clearance     15.13     0.06609     1.695    135.1

Concordance= 0.736 (se = 0.053 )
Likelihood ratio test= 8.61 on 1 df,  p=0.003
Wald test            = 5.92 on 1 df,  p=0.02
Score (logrank) test = 9.83 on 1 df,  p=0.002
```

Hide

```
cox_fit_summary <- summary(cox_fit)

# Extract values for HR, 95% CI, and p-value
HR <- cox_fit_summary$coefficients[2]
lower_CI <- cox_fit_summary$conf.int[3]
upper_CI <- cox_fit_summary$conf.int[4]
p_value <- cox_fit_summary$coefficients[5]
label_text <- paste0("HR = ", round(HR, 2), " (", round(lower_CI, 2), "-", round(upper_CI, 2), "); p = ", round(p_value, 3))
print(label_text)
```

```
[1] "HR = 15.13 (1.69-135.08); p = 0.015"
```

#Levels of MRD MTM/mL in Clearance at 3 months log10 transformation

Hide

```

rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$ctDNA.MRD!="",]
circ_data <- circ_data[circ_data$ACT==TRUE,]
circ_datadf <- as.data.frame(circ_data)

circ_data$ctDNA.Clearance <- NA #first we create the variable for the ctDNA & NAC combination, and we assign values
circ_data <- circ_data %>%
  mutate(ctDNA.Clearance = case_when(
    ctDNA.MRD == "POSITIVE" & ctDNA.3months == "NEGATIVE" ~ 1,
    ctDNA.MRD == "POSITIVE" & ctDNA.3months == "POSITIVE" ~ 2
  ))

circ_data <- circ_data[circ_data$DFS.3mo.months>=0,]
circ_data <- subset(circ_data, !is.na(ctDNA.Clearance))

# Transform p_MR_MTM with log10
circ_data$p_MR_MTM <- as.numeric(as.character(circ_data$p_MR_MTM))
circ_data$ctDNA.Clearance <- factor(circ_data$ctDNA.Clearance, levels=c("1","2"), labels = c("Clearance", "No Clearance"))
median_p_MR_MTM <- aggregate(p_MR_MTM ~ ctDNA.Clearance, data = circ_data, FUN = median)
print(median_p_MR_MTM)

```

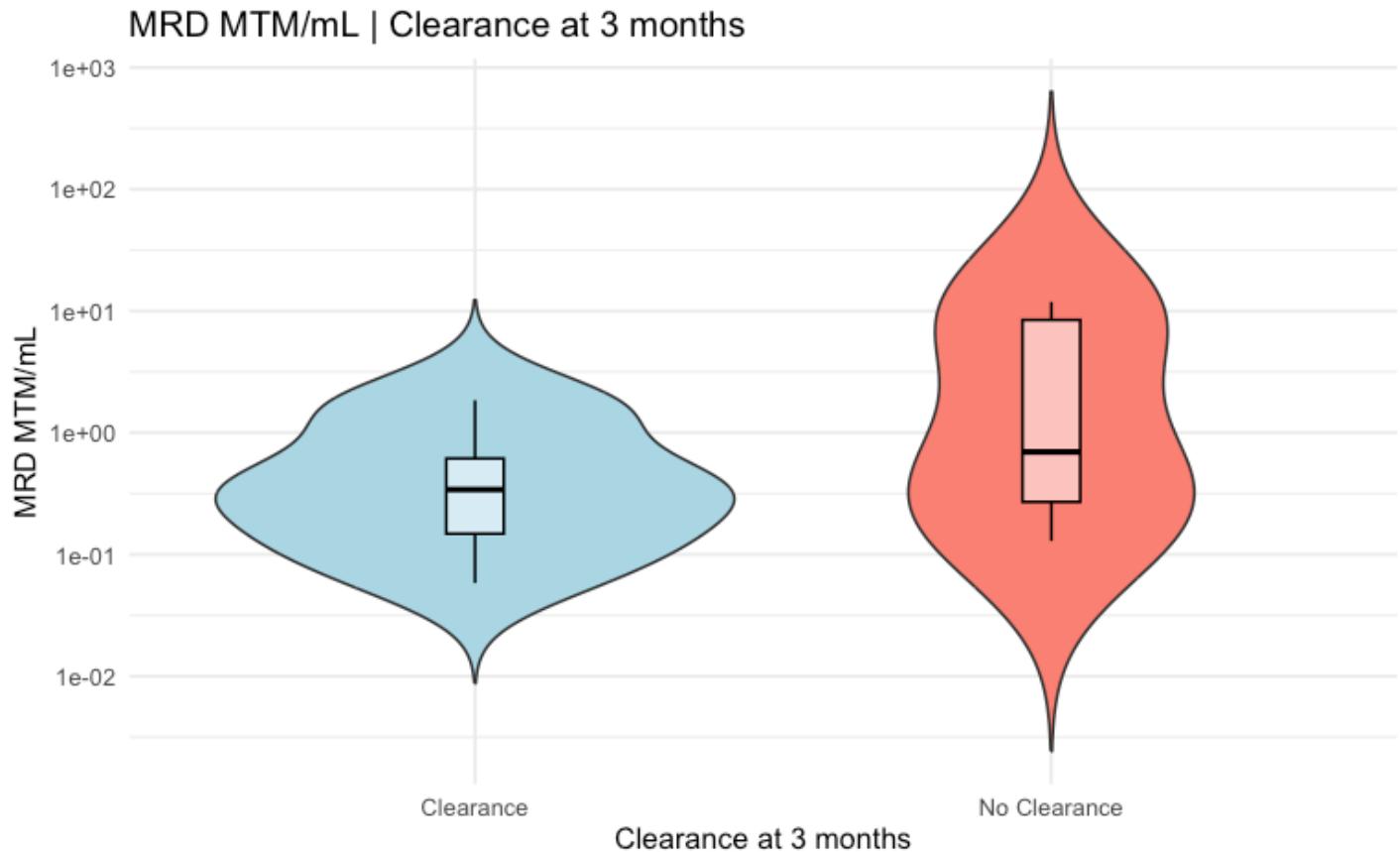
ctDNA.Clearance	p_MR_MTM
<fctr>	<dbl>
Clearance	0.3443529
No Clearance	0.6945222
2 rows	

Hide

```

# Create violin plot with log10 scale on y-axis
ggplot(circ_data, aes(x=ctDNA.Clearance, y=p_MR_MTM, fill=ctDNA.Clearance)) +
  geom_violin(trim=FALSE) +
  scale_fill_manual(values=c("Clearance"="lightblue", "No Clearance"="salmon")) +
  geom_boxplot(width=0.1, fill="white", colour="black", alpha=0.5) +
  scale_y_log10(breaks=c(0.001, 0.01, 0.1, 1, 10, 100, 1000, 10000)) +
  labs(title="MRD MTM/mL | Clearance at 3 months", x="Clearance at 3 months", y="MRD MTM/mL") +
  theme_minimal() +
  theme(legend.position="none")

```



```
m3_1v2 <- wilcox.test(p_MRД_MTМ ~ ctDNA.Clearance,
                       data = circ_data[circ_data$ctDNA.Clearance %in% c("Clearance", "No
Clearance"), ],
                       na.rm = TRUE)
print(m3_1v2)
```

Wilcoxon rank sum exact test

```
data: p_MRД_MTМ by ctDNA.Clearance
W = 13, p-value = 0.3543
alternative hypothesis: true location shift is not equal to 0
```

#DFS by ctDNA Clearance ACT-treated at 6 months

```

rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$ctDNA.MRD!="",]
circ_data <- circ_data[circ_data$ACT==TRUE,]
circ_datadf <- as.data.frame(circ_data)

circ_data$ctDNA.Dynamics <- NA #first we create the variable for the ctDNA & NAC combination, and we assign values
circ_data <- circ_data %>%
  mutate(ctDNA.Dynamics = case_when(
    ctDNA.MRD == "POSITIVE" & ctDNA.6months == "NEGATIVE" ~ 1,
    ctDNA.MRD == "POSITIVE" & ctDNA.6months == "POSITIVE" ~ 2
  ))

circ_data <- circ_data[circ_data$DFS.6mo.months>=0,]
survfit(Surv(time = circ_data$DFS.6mo.months, event = circ_data$DFS.Event)~ctDNA.Dynamics, data = circ_data)

```

Call: `survfit(formula = Surv(time = circ_data$DFS.6mo.months, event = circ_data$DFS.Event) ~ ctDNA.Dynamics, data = circ_data)`

36 observations deleted due to missingness

	n	events	median	0.95LCL	0.95UCL
ctDNA.Dynamics=1	9	5	11.696	9.429	NA
ctDNA.Dynamics=2	3	3	0.789	0.493	NA

[Hide](#)

```

event_summary <- circ_data %>%
  group_by(ctDNA.Dynamics) %>%
  summarise(
    Total = n(),
    Events = sum(DFS.Event),
    Fraction = Events / n(),
    Percentage = (Events / n()) * 100
  )
print(event_summary)

```

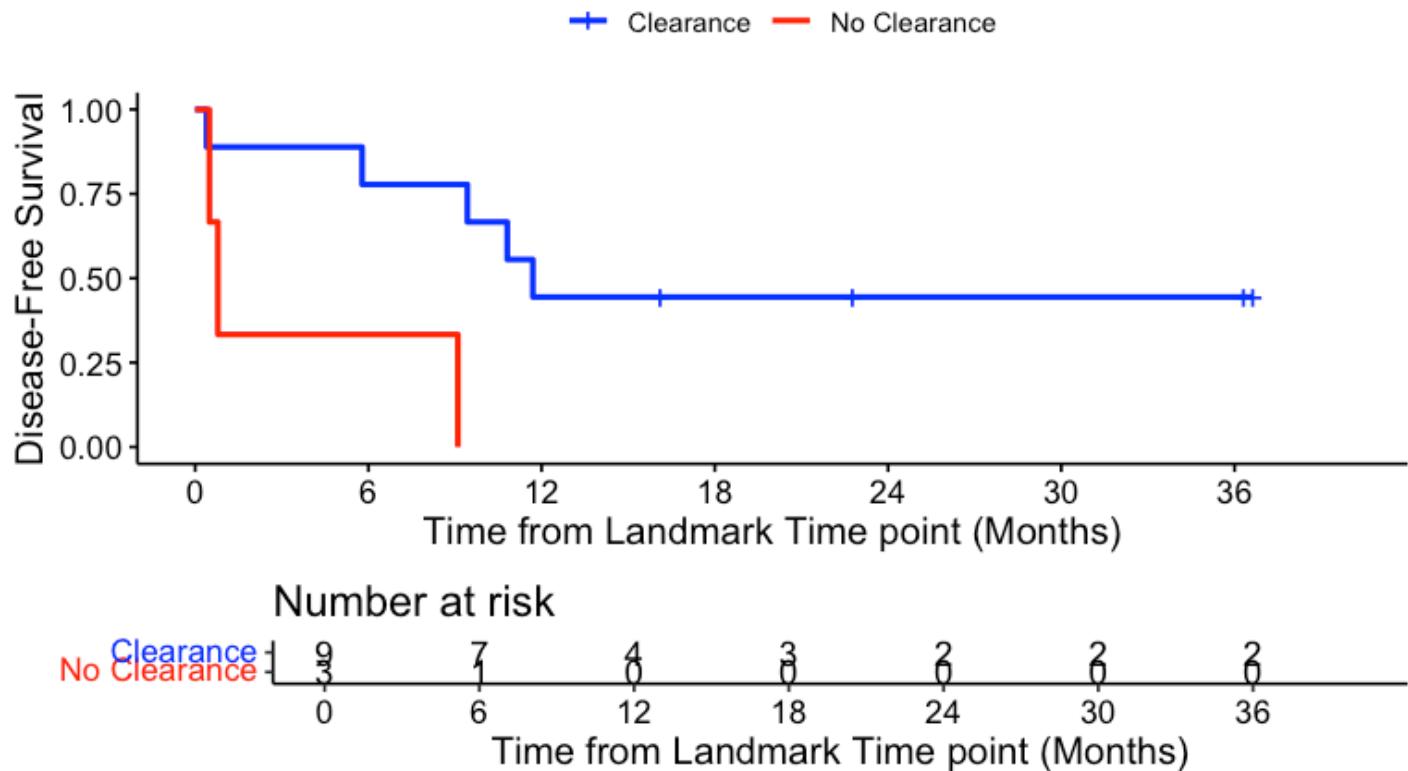
ctDNA.Dynamics	Total	Events	Fraction	Percentage
<dbl>	<int>	<int>	<dbl>	<dbl>
1	9	5	0.5555556	55.555556
2	3	3	1.0000000	100.000000
NA	36	NA	NA	NA

3 rows

Hide

```
surv_object <- Surv(time = circ_data$DFS.6mo.months, event = circ_data$DFS.Event)
KM_curve <- survfit(surv_object ~ ctDNA.Dynamics, data = circ_data, conf.int=0.95,conf.type="log-log")
ggsurvplot(KM_curve, data = circ_data, pval = FALSE, conf.int = FALSE, risk.table = TRUE,
break.time.by=6, palette=c("blue","red"), title="DFS - ctDNA Clearance from MRD to 6 months ACT-treated", ylab= "Disease-Free Survival", xlab="Time from Landmark Time point (Months)", legend.labs=c("Clearance", "No Clearance"), legend.title="")
```

DFS - ctDNA Clearance from MRD to 6 months ACT-treated



```
summary(KM_curve, times= c(6, 24))
```

```
Call: survfit(formula = surv_object ~ ctDNA.Dynamics, data = circ_data,
  conf.int = 0.95, conf.type = "log-log")
```

36 observations deleted due to missingness

ctDNA.Dynamics=1

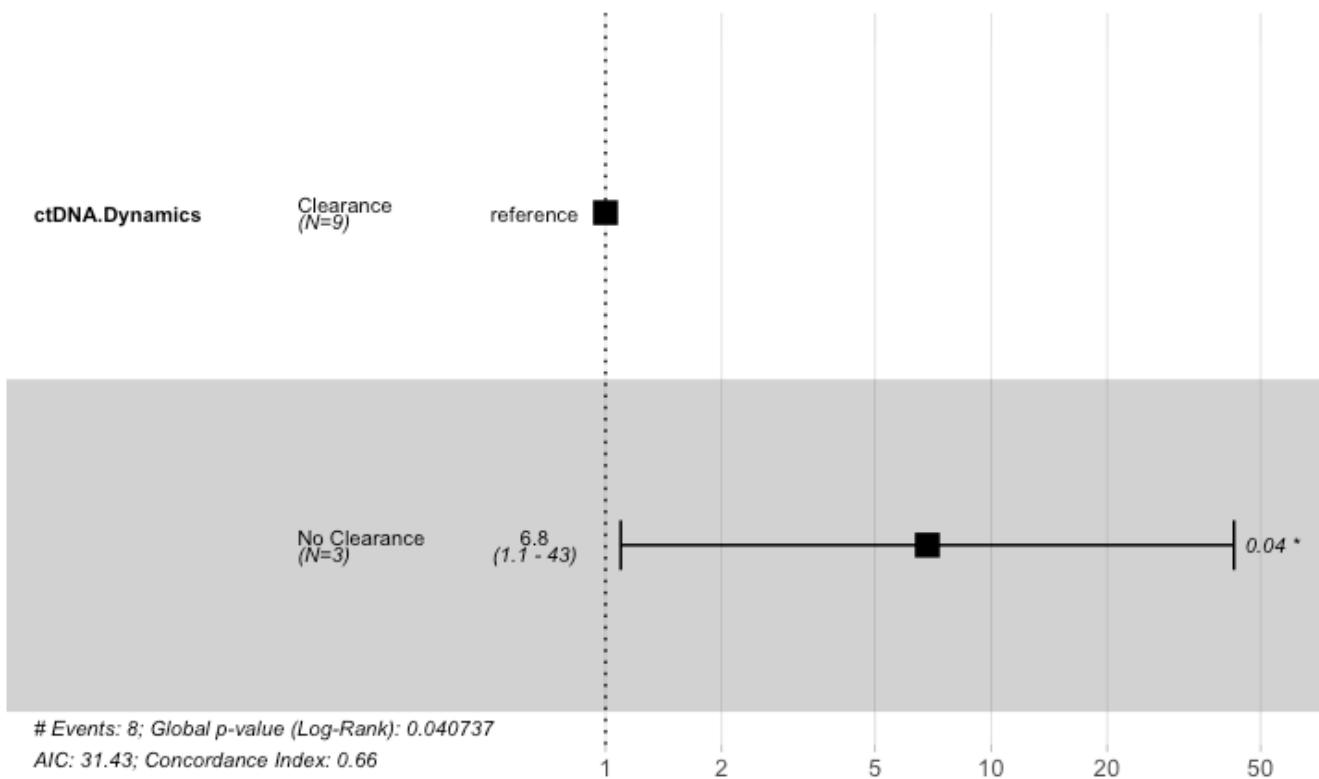
time	n.risk	n.event	survival	std.err	lower	95% CI	upper
6	7	2	0.778	0.139	0.365	0.939	
24	2	3	0.444	0.166	0.136	0.719	

ctDNA.Dynamics=2

time	n.risk	n.event	survival	std.err	lower	95% CI	upper
6.00000	1.00000	2.00000	0.33333	0.27217	0.00896	0.774	
15							

```
circ_data$ctDNA.Dynamics <- factor(circ_data$ctDNA.Dynamics, levels=c("1","2"), labels =
c("Clearance", "No Clearance"))
cox_fit <- coxph(surv_object ~ ctDNA.Dynamics, data=circ_data)
ggforest(cox_fit, data = circ_data)
```

Hazard ratio



Hide

```
summary(cox_fit)
```

Call:

```
coxph(formula = surv_object ~ ctDNA.Dynamics, data = circ_data)
```

```
n= 12, number of events= 8
```

```
(36 observations deleted due to missingness)
```

	coef	exp(coef)	se(coef)	z	Pr(> z)
--	------	-----------	----------	---	----------

ctDNA.Dynamics	No Clearance	1.9221	6.8355	0.9345	2.057	0.0397 *
----------------	--------------	--------	--------	--------	-------	----------

```
---
```

```
Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

	exp(coef)	exp(-coef)	lower .95	upper .95
--	-----------	------------	-----------	-----------

ctDNA.Dynamics	No Clearance	6.836	0.1463	1.095	42.68
----------------	--------------	-------	--------	-------	-------

```
Concordance= 0.658 (se = 0.073 )
```

```
Likelihood ratio test= 4.19 on 1 df, p=0.04
```

```
Wald test = 4.23 on 1 df, p=0.04
```

```
Score (logrank) test = 5.53 on 1 df, p=0.02
```

Hide

```
cox_fit_summary <- summary(cox_fit)

# Extract values for HR, 95% CI, and p-value
HR <- cox_fit_summary$coefficients[2]
lower_CI <- cox_fit_summary$conf.int[3]
upper_CI <- cox_fit_summary$conf.int[4]
p_value <- cox_fit_summary$coefficients[5]
label_text <- paste0("HR = ", round(HR, 2), " (", round(lower_CI, 2), "-", round(upper_CI, 2), "); p = ", round(p_value, 3))
print(label_text)
```

```
[1] "HR = 6.84 (1.09-42.68); p = 0.04"
```

```
#Levels of MRD MTM/mL in Clearance at 6 months log10 transformation
```

[Hide](#)

```
rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$ctDNA.MRD!="",]
circ_data <- circ_data[circ_data$ACT==TRUE,]
circ_datadf <- as.data.frame(circ_data)

circ_data$ctDNA.Clearance <- NA #first we create the variable for the ctDNA & NAC combination, and we assign values
circ_data <- circ_data %>%
  mutate(ctDNA.Clearance = case_when(
    ctDNA.MRD == "POSITIVE" & ctDNA.6months == "NEGATIVE" ~ 1,
    ctDNA.MRD == "POSITIVE" & ctDNA.6months == "POSITIVE" ~ 2
  ))

circ_data <- circ_data[circ_data$DFS.6mo.months>=0,]
circ_data <- subset(circ_data, !is.na(ctDNA.Clearance))

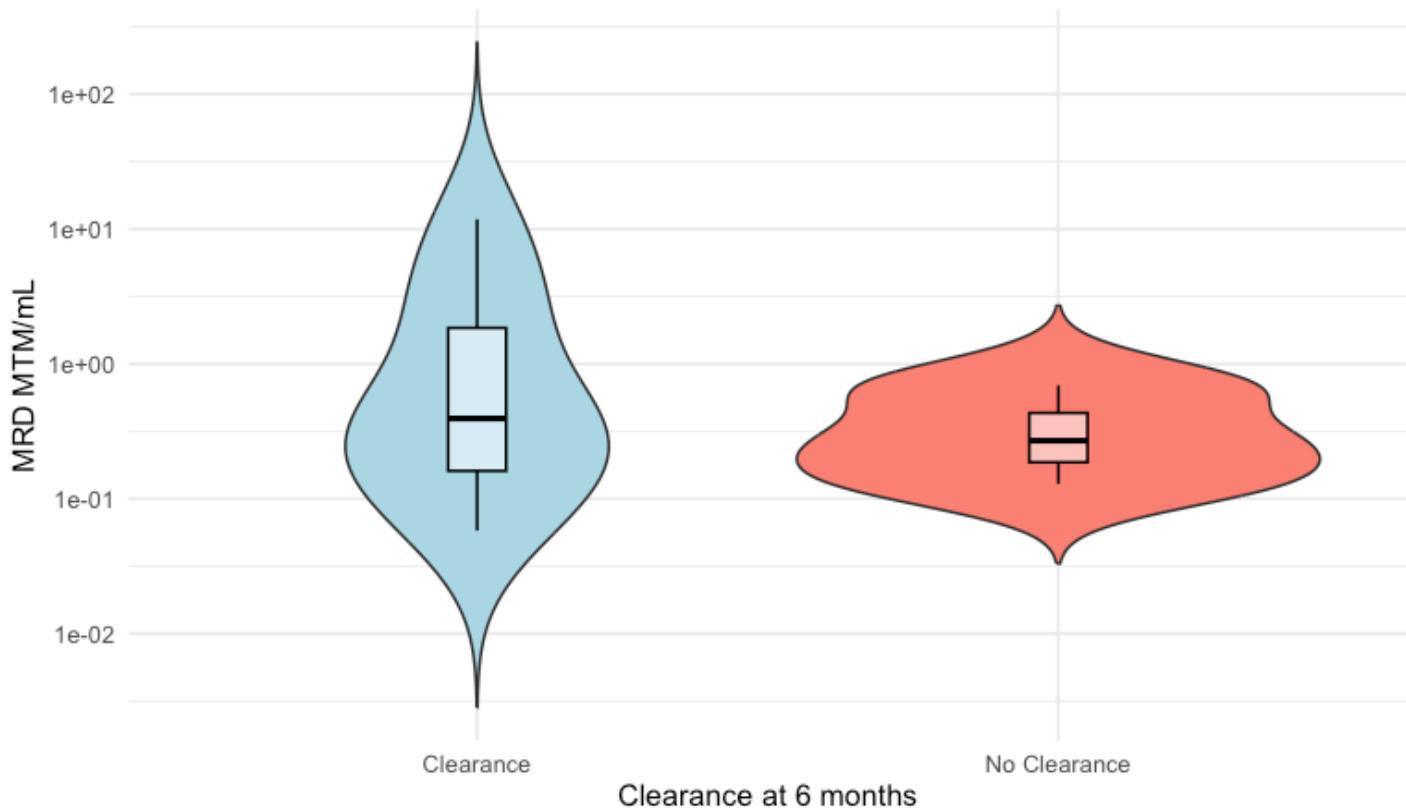
# Transform p_MRД_MTM with log10
circ_data$p_MRД_MTM <- as.numeric(as.character(circ_data$p_MRД_MTM))
circ_data$ctDNA.Clearance <- factor(circ_data$ctDNA.Clearance, levels=c("1","2"), labels = c("Clearance", "No Clearance"))
median_p_MRД_MTM <- aggregate(p_MRД_MTM ~ ctDNA.Clearance, data = circ_data, FUN = median)
print(median_p_MRД_MTM)
```

ctDNA.Clearance	p_MRД_MTM
<fctr>	<dbl>
Clearance	0.3939489
No Clearance	0.2697545
2 rows	

Hide

```
# Create violin plot with log10 scale on y-axis
ggplot(circ_data, aes(x=ctDNA.Clearance, y=p_MRD_MTM, fill=ctDNA.Clearance)) +
  geom_violin(trim=FALSE) +
  scale_fill_manual(values=c("Clearance"="lightblue", "No Clearance"="salmon")) +
  geom_boxplot(width=0.1, fill="white", colour="black", alpha=0.5) +
  scale_y_log10(breaks=c(0.001, 0.01, 0.1, 1, 10, 100, 1000, 10000)) +
  labs(title="MRD MTM/mL | Clearance at 6 months", x="Clearance at 6 months", y="MRD MTM/mL") +
  theme_minimal() +
  theme(legend.position="none")
```

MRD MTM/mL | Clearance at 6 months



Hide

```
m3_1v2 <- wilcox.test(p_MRD_MTM ~ ctDNA.Clearance,
                        data = circ_data[circ_data$ctDNA.Clearance %in% c("Clearance", "No Clearance"), ],
                        na.rm = TRUE)
print(m3_1v2)
```

```
Wilcoxon rank sum exact test
```

```
data: p_MR_D_MTM by ctDNA.Clearance
W = 16, p-value = 0.7273
alternative hypothesis: true location shift is not equal to 0
```

#DFS by ctDNA at the Surveillance Window - Landmark 10 weeks

[Hide](#)

```
rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$ctDNA.Surveillance!="",]
circ_data$DFS.months=circ_data$DFS.months-2.5
circ_data <- circ_data[circ_data$DFS.months>=0,]
circ_datadf <- as.data.frame(circ_data)

survfit(Surv(time = circ_data$DFS.months, event = circ_data$DFS.Event)~ctDNA.Surveillance, data = circ_data)
```

Call: survfit(formula = Surv(time = circ_data\$DFS.months, event = circ_data\$DFS.Event) ~ ctDNA.Surveillance, data = circ_data)

	n	events	median	0.95LCL	0.95UCL
ctDNA.Surveillance=NEGATIVE	99	18	NA	NA	NA
ctDNA.Surveillance=POSITIVE	53	44	5.81	3.05	12.1

[Hide](#)

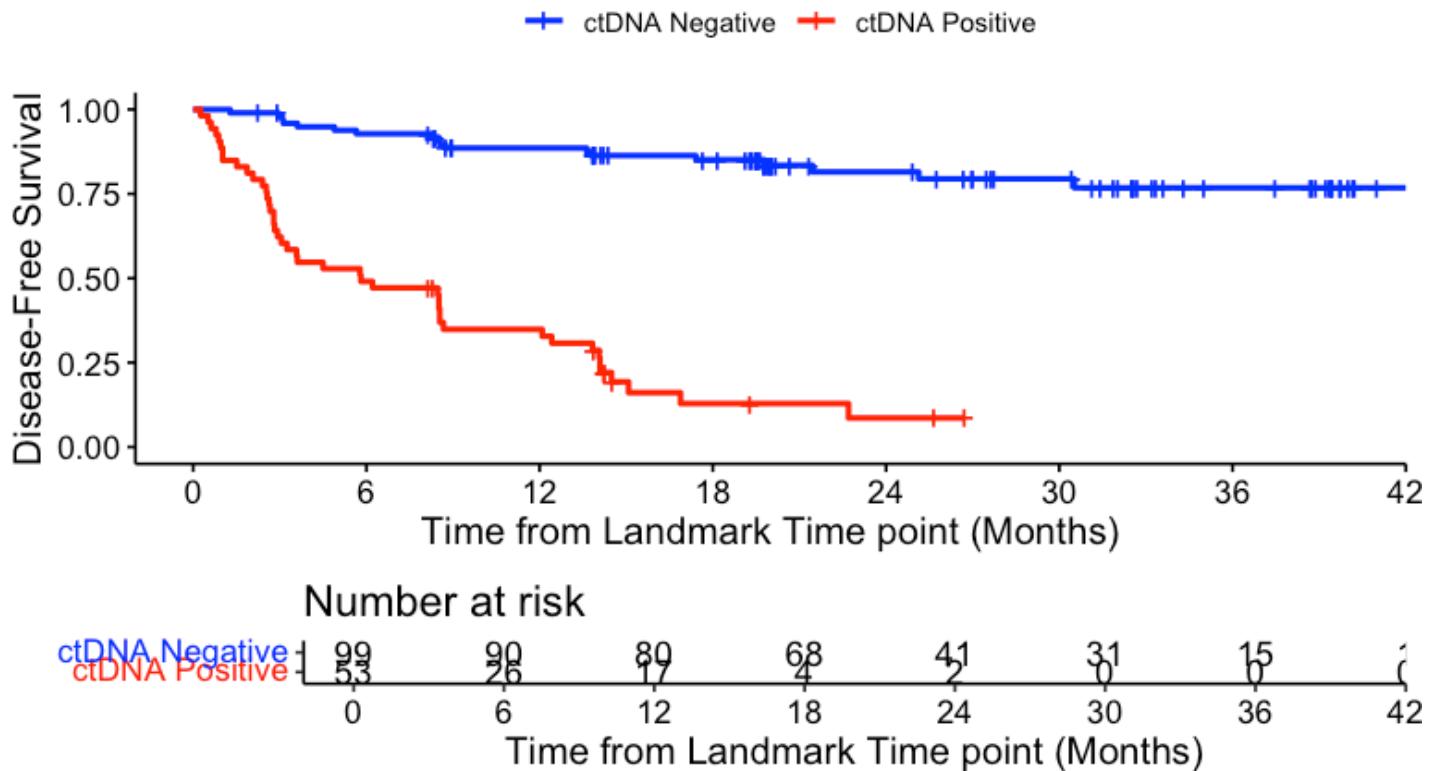
```
event_summary <- circ_data %>%
  group_by(ctDNA.Surveillance) %>%
  summarise(
    Total = n(),
    Events = sum(DFS.Event),
    Fraction = Events / n(),
    Percentage = (Events / n()) * 100
  )
print(event_summary)
```

ctDNA.Surveillance	Total	Events	Fraction	Percentage
<chr>	<int>	<int>	<dbl>	<dbl>
NEGATIVE	99	18	0.1818182	18.18182
POSITIVE	53	44	0.8301887	83.01887
2 rows				

[Hide](#)

```
surv_object <- Surv(time = circ_data$DFS.months, event = circ_data$DFS.Event)
KM_curve <- survfit(surv_object ~ ctDNA.Surveillance, data = circ_data, conf.int=0.95,conf.type="log-log")
ggsurvplot(KM_curve, data = circ_data, pval = FALSE, conf.int = FALSE, risk.table = TRUE, break.time.by=6, palette=c("blue","red"), title="DFS - ctDNA Surveillance window", ylab= "Disease-Free Survival", xlab="Time from Landmark Time point (Months)", legend.labs=c("ctDNA Negative", "ctDNA Positive"), legend.title="")
```

DFS - ctDNA Surveillance window



```
summary(KM_curve, times= c(24, 30, 36))
```

```
Call: survfit(formula = surv_object ~ ctDNA.Surveillance, data = circ_data,
  conf.int = 0.95, conf.type = "log-log")
```

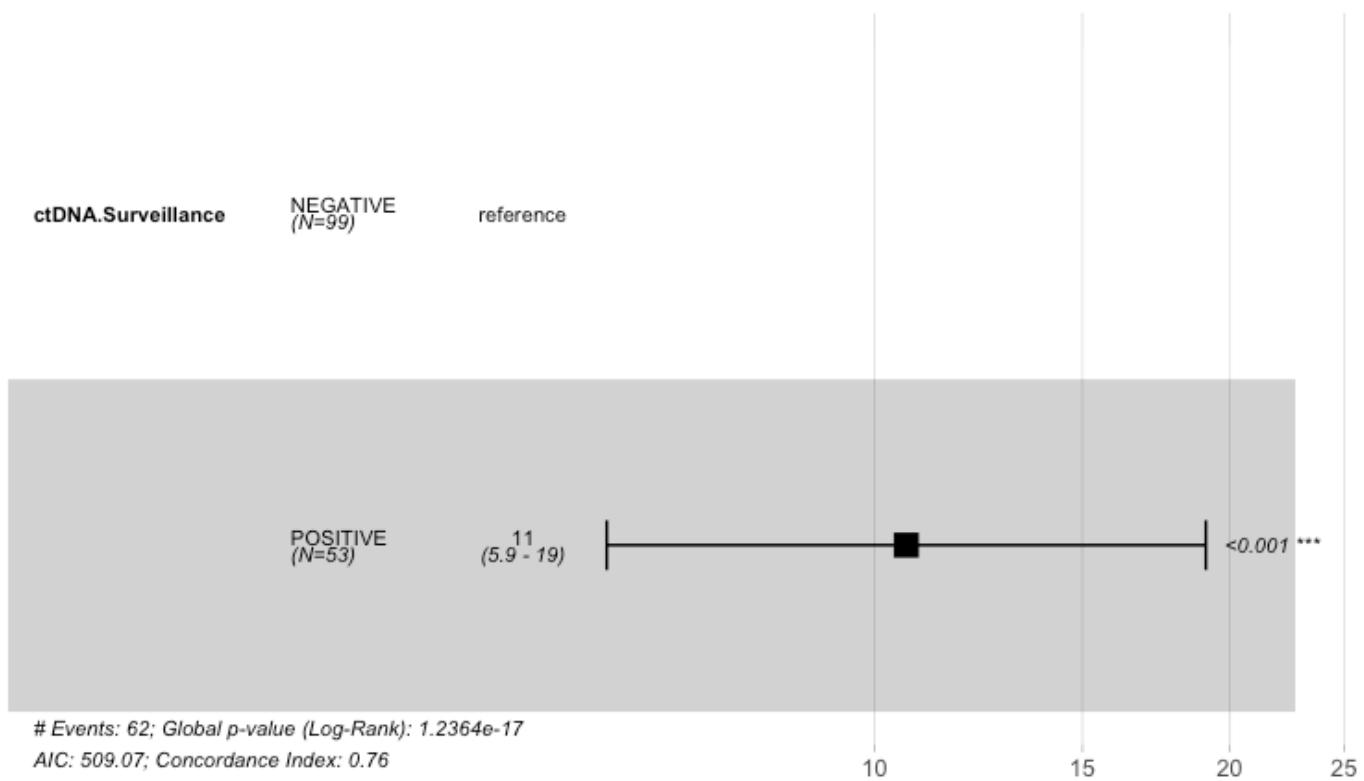
ctDNA.Surveillance=NEGATIVE						
time	n.risk	n.event	survival	std.err	lower	95% CI
24	41	16	0.815	0.0434	0.711	0.884
30	31	1	0.795	0.0468	0.684	0.870
36	15	1	0.768	0.0522	0.646	0.853

ctDNA.Surveillance=POSITIVE						
time	n.risk	n.event	survival	std.err	lower	95% CI

24.0000	2.0000	44.0000	0.0859	0.0501	0.0200	0.21
42						

```
circ_data$ctDNA.Surveillance <- factor(circ_data$ctDNA.Surveillance, levels=c("NEGATIVE", "POSITIVE"))
cox_fit <- coxph(surv_object ~ ctDNA.Surveillance, data=circ_data)
ggforest(cox_fit, data = circ_data)
```

Hazard ratio



```
summary(cox_fit)
```

Call:
`coxph(formula = surv_object ~ ctDNA.Surveillance, data = circ_data)`

n= 152, number of events= 62

	coef	exp(coef)	se(coef)	z	Pr(> z)
ctDNA.SurveillancePOSITIVE	2.3644	10.6382	0.2982	7.929	2.21e-15 ***

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

	exp(coef)	exp(-coef)	lower .95	upper .95
ctDNA.SurveillancePOSITIVE	10.64	0.094	5.93	19.09

Concordance= 0.757 (se = 0.025)
 Likelihood ratio test= 73.09 on 1 df, p=<2e-16
 Wald test = 62.87 on 1 df, p=2e-15
 Score (logrank) test = 91.54 on 1 df, p=<2e-16

```
cox_fit_summary <- summary(cox_fit)

# Extract values for HR, 95% CI, and p-value
HR <- cox_fit_summary$coefficients[2]
lower_CI <- cox_fit_summary$conf.int[3]
upper_CI <- cox_fit_summary$conf.int[4]
p_value <- cox_fit_summary$coefficients[5]
label_text <- paste0("HR = ", round(HR, 2), " (", round(lower_CI, 2), "-", round(upper_CI, 2), "); p = ", round(p_value, 3))
print(label_text)
```

[1] "HR = 10.64 (5.93-19.09); p = 0"

[Hide](#)

```
#Fisher test for DFS percentages at 24, 30, and 36 months
dfs_times <- c(24, 30, 36)
p_values <- sapply(dfs_times, function(time) {
  neg_count <- sum(circ_data$ctDNA.Surveillance == "NEGATIVE" & circ_data$DFS.months >= time & circ_data$DFS.Event == 0)
  pos_count <- sum(circ_data$ctDNA.Surveillance == "POSITIVE" & circ_data$DFS.months >= time & circ_data$DFS.Event == 0)
  neg_total <- sum(circ_data$ctDNA.Surveillance == "NEGATIVE")
  pos_total <- sum(circ_data$ctDNA.Surveillance == "POSITIVE")

  neg_surv <- neg_total - sum(circ_data$ctDNA.Surveillance == "NEGATIVE" & circ_data$DFS.Event == 1 & circ_data$DFS.months < time)
  pos_surv <- pos_total - sum(circ_data$ctDNA.Surveillance == "POSITIVE" & circ_data$DFS.Event == 1 & circ_data$DFS.months < time)

  surv_matrix <- matrix(c(neg_surv, pos_surv, neg_total - neg_surv, pos_total - pos_surv), nrow = 2)
  test_result <- fisher.test(surv_matrix)
  return(test_result$p.value)
})
names(p_values) <- paste0("p-value at ", dfs_times, " months")
print(p_values)
```

p-value at 24 months p-value at 30 months p-value at 36 months
 4.109654e-16 1.518274e-15 4.230750e-15

[Hide](#)

```
p_values_adjusted <- p.adjust(p_values, method = "bonferroni")
names(p_values_adjusted) <- paste0("Corrected p-value at ", dfs_times, " months")
print(p_values_adjusted)
```

Corrected p-value at 24 months	Corrected p-value at 30 months	Corrected p-value at 36 months
1.232896e-15 e-14	4.554823e-15	1.269225

#OS by ctDNA at the Surveillance Window - Landmark 10 weeks

Hide

```
rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$ctDNA.Surveillance!="",]
circ_data$OS.months=circ_data$OS.months-2.5
circ_data <- circ_data[circ_data$OS.months>=0,]
circ_dataadf <- as.data.frame(circ_data)

survfit(Surv(time = circ_data$OS.months, event = circ_data$OS.Event)~ctDNA.Surveillance,
data = circ_data)
```

Call: `survfit(formula = Surv(time = circ_data$OS.months, event = circ_data$OS.Event) ~ ctDNA.Surveillance, data = circ_data)`

	n	events	median	0.95LCL	0.95UCL
ctDNA.Surveillance=NEGATIVE	99	3	NA	NA	NA
ctDNA.Surveillance=POSITIVE	55	6	41.8	37.3	NA

Hide

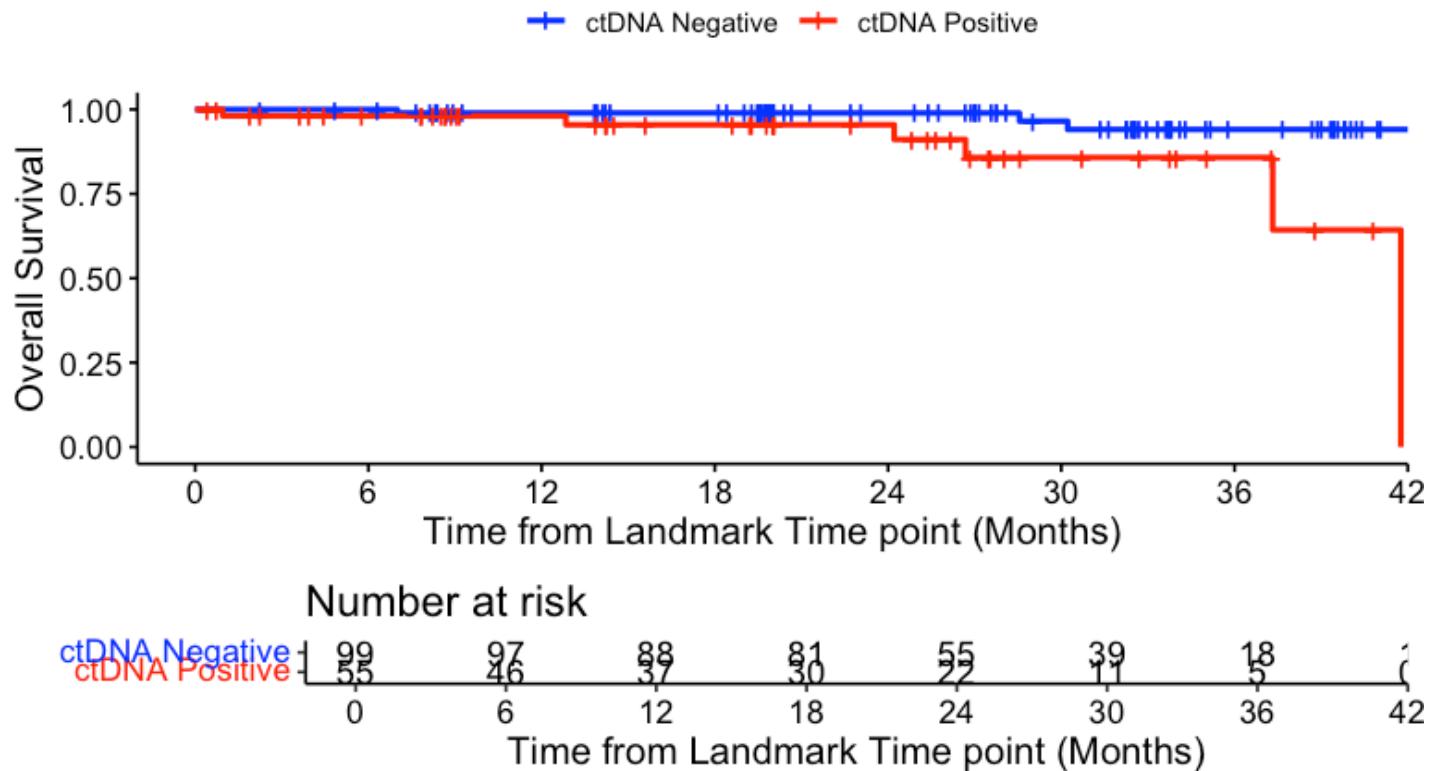
```
event_summary <- circ_data %>%
  group_by(ctDNA.Surveillance) %>%
  summarise(
    Total = n(),
    Events = sum(OS.Event),
    Fraction = Events / n(),
    Percentage = (Events / n()) * 100
  )
print(event_summary)
```

ctDNA.Surveillance	Total	Events	Fraction	Percentage
<chr>	<int>	<int>	<dbl>	<dbl>
NEGATIVE	99	3	0.03030303	3.030303
POSITIVE	55	6	0.10909091	10.909091
2 rows				

Hide

```
surv_object <- Surv(time = circ_data$OS.months, event = circ_data$OS.Event)
KM_curve <- survfit(surv_object ~ ctDNA.Surveillance, data = circ_data, conf.int=0.95,conf.type="log-log")
ggsurvplot(KM_curve, data = circ_data, pval = FALSE, conf.int = FALSE, risk.table = TRUE, break.time.by=6, palette=c("blue","red"), title="OS - ctDNA Surveillance window", ylab= "Overall Survival", xlab="Time from Landmark Time point (Months)", legend.labs=c("ctDNA Negative", "ctDNA Positive"), legend.title="")
```

OS - ctDNA Surveillance window



```
summary(KM_curve, times= c(24, 30, 36))
```

```
Call: survfit(formula = surv_object ~ ctDNA.Surveillance, data = circ_data,
  conf.int = 0.95, conf.type = "log-log")
```

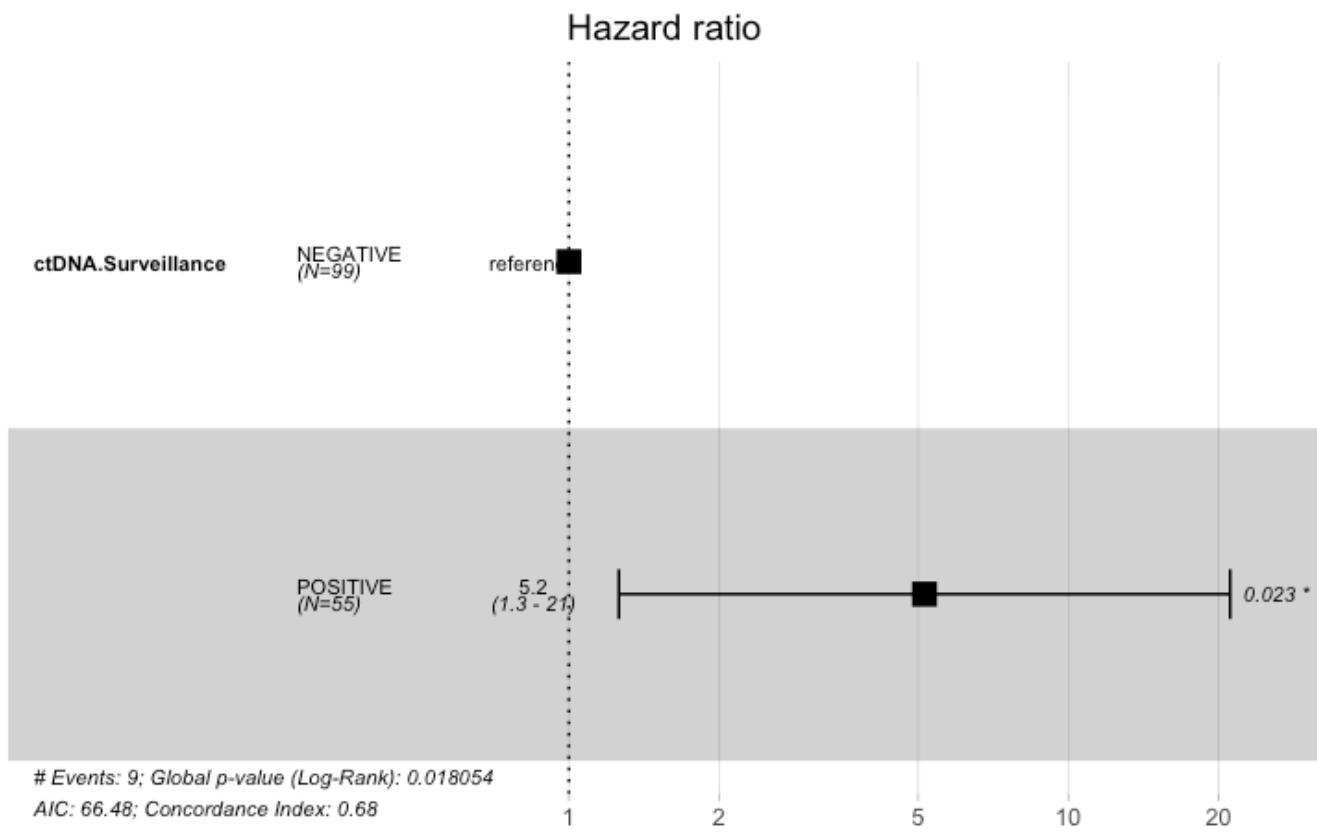
ctDNA.Surveillance=NEGATIVE

time	n.risk	n.event	survival	std.err	lower	95% CI	upper	95% CI
24	55	1	0.990	0.0104	0.928		0.999	
30	39	1	0.965	0.0259	0.855		0.992	
36	18	1	0.941	0.0351	0.817		0.982	

ctDNA.Surveillance=POSITIVE

time	n.risk	n.event	survival	std.err	lower	95% CI	upper	95% CI
24	22	2	0.955	0.0319	0.827		0.989	
30	11	2	0.858	0.0715	0.641		0.948	
36	5	0	0.858	0.0715	0.641		0.948	

```
circ_data$ctDNA.Surveillance <- factor(circ_data$ctDNA.Surveillance, levels=c("NEGATIVE", "POSITIVE"))
cox_fit <- coxph(surv_object ~ ctDNA.Surveillance, data=circ_data)
ggforest(cox_fit, data = circ_data)
```



```
summary(cox_fit)
```

Call:
`coxph(formula = surv_object ~ ctDNA.Surveillance, data = circ_data)`

n= 154, number of events= 9

	coef	exp(coef)	se(coef)	z	Pr(> z)	
ctDNA.Surveillance	POSITIVE	1.6394	5.1523	0.7188	2.281	0.0226 *

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

	exp(coef)	exp(-coef)	lower .95	upper .95	
ctDNA.Surveillance	POSITIVE	5.152	0.1941	1.259	21.08

Concordance= 0.678 (se = 0.094)
 Likelihood ratio test= 5.59 on 1 df, p=0.02
 Wald test = 5.2 on 1 df, p=0.02
 Score (logrank) test = 6.33 on 1 df, p=0.01

```
cox_fit_summary <- summary(cox_fit)

# Extract values for HR, 95% CI, and p-value
HR <- cox_fit_summary$coefficients[2]
lower_CI <- cox_fit_summary$conf.int[3]
upper_CI <- cox_fit_summary$conf.int[4]
p_value <- cox_fit_summary$coefficients[5]
label_text <- paste0("HR = ", round(HR, 2), " (", round(lower_CI, 2), "-", round(upper_CI, 2), "); p = ", round(p_value, 3))
print(label_text)
```

[1] "HR = 5.15 (1.26-21.08); p = 0.023"

[Hide](#)

```
#Fisher test for OS percentages at 24, 30, and 36 months
os_times <- c(24, 30, 36)
p_values <- sapply(os_times, function(time) {
  neg_count <- sum(circ_data$ctDNA.MRD == "NEGATIVE" & circ_data$OS.months >= time & circ_data$OS.Event == 0)
  pos_count <- sum(circ_data$ctDNA.MRD == "POSITIVE" & circ_data$OS.months >= time & circ_data$OS.Event == 0)
  neg_total <- sum(circ_data$ctDNA.MRD == "NEGATIVE")
  pos_total <- sum(circ_data$ctDNA.MRD == "POSITIVE")

  neg_surv <- neg_total - sum(circ_data$ctDNA.MRD == "NEGATIVE" & circ_data$OS.Event == 1 & circ_data$OS.months < time)
  pos_surv <- pos_total - sum(circ_data$ctDNA.MRD == "POSITIVE" & circ_data$OS.Event == 1 & circ_data$OS.months < time)

  surv_matrix <- matrix(c(neg_surv, pos_surv, neg_total - neg_surv, pos_total - pos_surv), nrow = 2)
  test_result <- fisher.test(surv_matrix)
  return(test_result$p.value)
})
names(p_values) <- paste0("p-value at ", os_times, " months")
print(p_values)
```

p-value at 24 months p-value at 30 months p-value at 36 months
 0.12280702 0.02179403 0.04276279

[Hide](#)

```
p_values_adjusted <- p.adjust(p_values, method = "bonferroni")
names(p_values_adjusted) <- paste0("Corrected p-value at ", os_times, " months")
print(p_values_adjusted)
```

Corrected p-value at 24 months	Corrected p-value at 30 months	Corrected p-value at 36 months
8838	0.36842105	0.06538209

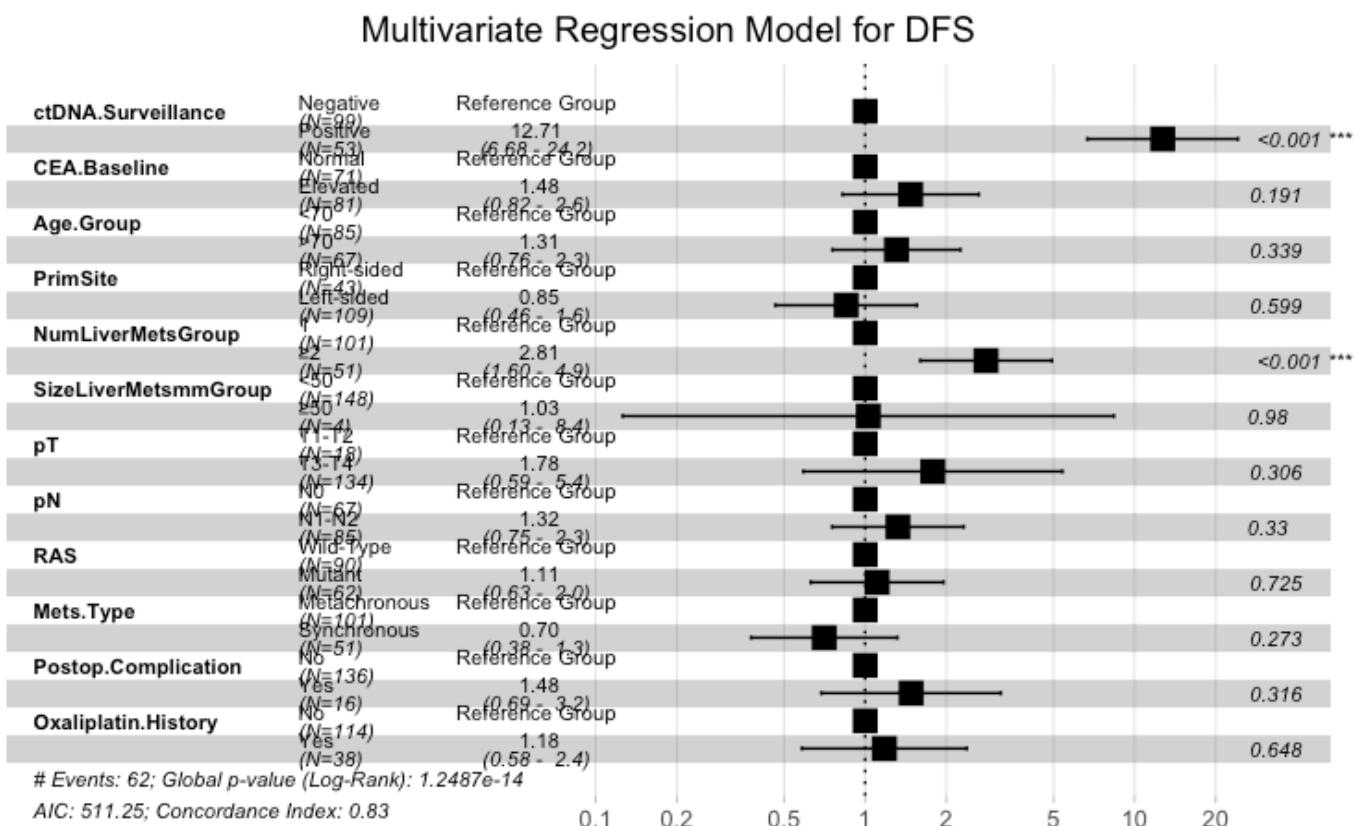
#Multivariate cox regression at Surveillance Window for DFS - Landmark 10 weeks

```

rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$ctDNA.Surveillance!="",]
circ_data$DFS.months=circ_data$DFS.months-2.5
circ_data <- circ_data[circ_data$DFS.months>=0,]
circ_dataadf <- as.data.frame(circ_data)

circ_data$ctDNA.Surveillance <- factor(circ_data$ctDNA.Surveillance, levels=c("NEGATIVE", "POSITIVE"), labels = c("Negative", "Positive"))
circ_data$CEA.Baseline <- factor(circ_data$CEA.Baseline, levels=c("NEGATIVE", "POSITIVE"), labels = c("Normal", "Elevated"))
circ_data$Gender <- factor(circ_data$Gender, levels = c("Female", "Male"))
circ_data$Age.Group <- factor(circ_data$Age.Group, levels = c("1", "2"), labels = c("<70", ">70"))
circ_data$PrimSite <- factor(circ_data$PrimSite, levels = c("Right-sided colon", "Left-sided colon"), labels = c("Right-sided", "Left-sided"))
circ_data$NumLiverMetsGroup <- factor(circ_data$NumLiverMetsGroup, levels = c("1", "≥ 2"))
circ_data$SizeLiverMetsmmGroup <- factor(circ_data$SizeLiverMetsmmGroup, levels = c("<50", "≥50"))
circ_data$ECOG <- factor(circ_data$ECOG, levels = c("0", "1"))
circ_data$pT <- factor(circ_data$pT, levels = c("T1-T2", "T3-T4"))
circ_data$pN <- factor(circ_data$pN, levels = c("N0", "N1-N2"))
circ_data$ACT <- factor(circ_data$ACT, levels = c("FALSE", "TRUE"), labels = c("Observation", "Chemotherapy"))
circ_data$Postop.ComPLICATION <- factor(circ_data$Postop.ComPLICATION, levels = c("No", "Yes"))
circ_data$Oxaliplatin.History <- factor(circ_data$Oxaliplatin.History, levels = c("No", "Yes"))
circ_data$Mets.Type <- factor(circ_data$Mets.Type, levels = c("Metachronous", "Synchronous"))
circ_data$MSI <- factor(circ_data$MSI, levels = c("MSS", "MSI-High"), labels = c("MSS", "MSI-High"))
circ_data$BRAF.V600E <- factor(circ_data$BRAF.V600E, levels = c("WT", "MUT"), labels = c("Wild-Type", "V600E"))
circ_data$RAS <- factor(circ_data$RAS, levels = c("WT", "MUT"), labels = c("Wild-Type", "Mutant"))
surv_object <- Surv(time = circ_data$DFS.MRD.months, event = circ_data$DFS.Event)
cox_fit <- coxph(surv_object ~ ctDNA.Surveillance + CEA.Baseline + Age.Group + PrimSite + NumLiverMetsGroup + SizeLiverMetsmmGroup + pT + pN + RAS + Mets.Type + Postop.ComPLICATION + Oxaliplatin.History, data=circ_data)
ggforest(cox_fit, data = circ_data, main = "Multivariate Regression Model for DFS", refLabel = "Reference Group")

```



Hide

```
test.ph <- cox.zph(cox_fit)
```

#OS by ctDNA at the MRD Window - pts with Radiological Recurrence

[Hide](#)

```
rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$RFS.Event=="TRUE",]
circ_data <- circ_data[circ_data$ctDNA.MRD!=""]
circ_data <- circ_data[circ_data$OS.MRD.months>=0,]
circ_datadf <- as.data.frame(circ_data)

survfit(Surv(time = circ_data$OS.MRD.months, event = circ_data$OS.Event)~ctDNA.MRD, data = circ_data)
```

Call: survfit(formula = Surv(time = circ_data\$OS.MRD.months, event = circ_data\$OS.Event)
~
ctDNA.MRD, data = circ_data)

	n	events	median	0.95LCL	0.95UCL
ctDNA.MRD=NEGATIVE	42	2	NA	NA	NA
ctDNA.MRD=POSITIVE	53	10	43.4	NA	NA

[Hide](#)

```
event_summary <- circ_data %>%
  group_by(ctDNA.MRD) %>%
  summarise(
    Total = n(),
    Events = sum(OS.Event),
    Fraction = Events / n(),
    Percentage = (Events / n()) * 100
  )
print(event_summary)
```

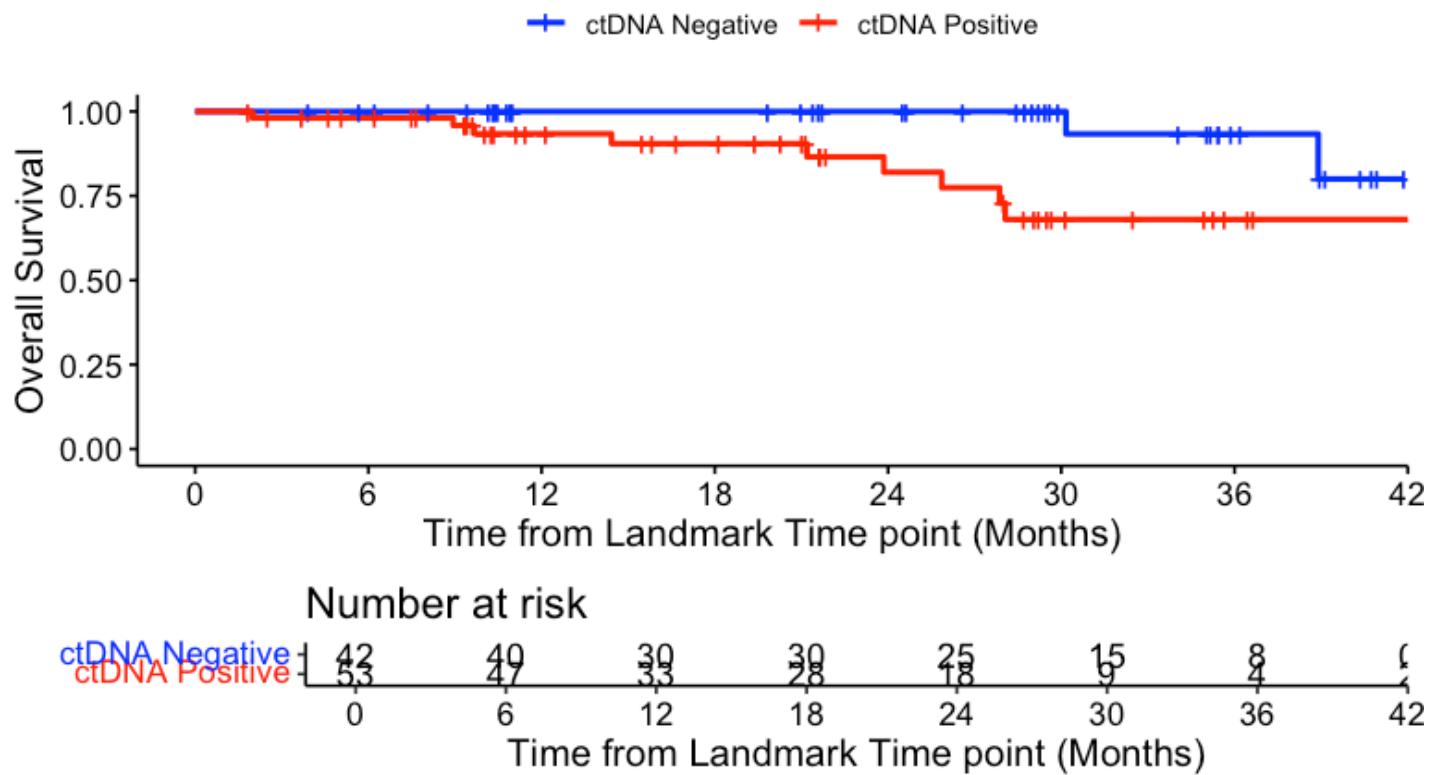
ctDNA.MRD	Total	Events	Fraction	Percentage
<chr>	<int>	<int>	<dbl>	<dbl>
NEGATIVE	42	2	0.04761905	4.761905
POSITIVE	53	10	0.18867925	18.867925

2 rows

[Hide](#)

```
surv_object <- Surv(time = circ_data$OS.MRD.months, event = circ_data$OS.Event)
KM_curve <- survfit(surv_object ~ ctDNA.MRD, data = circ_data, conf.int=0.95,conf.type="log-log")
ggsurvplot(KM_curve, data = circ_data, pval = FALSE, conf.int = FALSE, risk.table = TRUE,
break.time.by=6, palette=c("blue","red"), title="OS - Radiological Recurrence | ctDNA
MRD window", ylab= "Overall Survival", xlab="Time from Landmark Time point (Months)", legend.labs=c("ctDNA Negative", "ctDNA Positive"), legend.title="")
```

OS - Radiological Recurrence | ctDNA MRD window



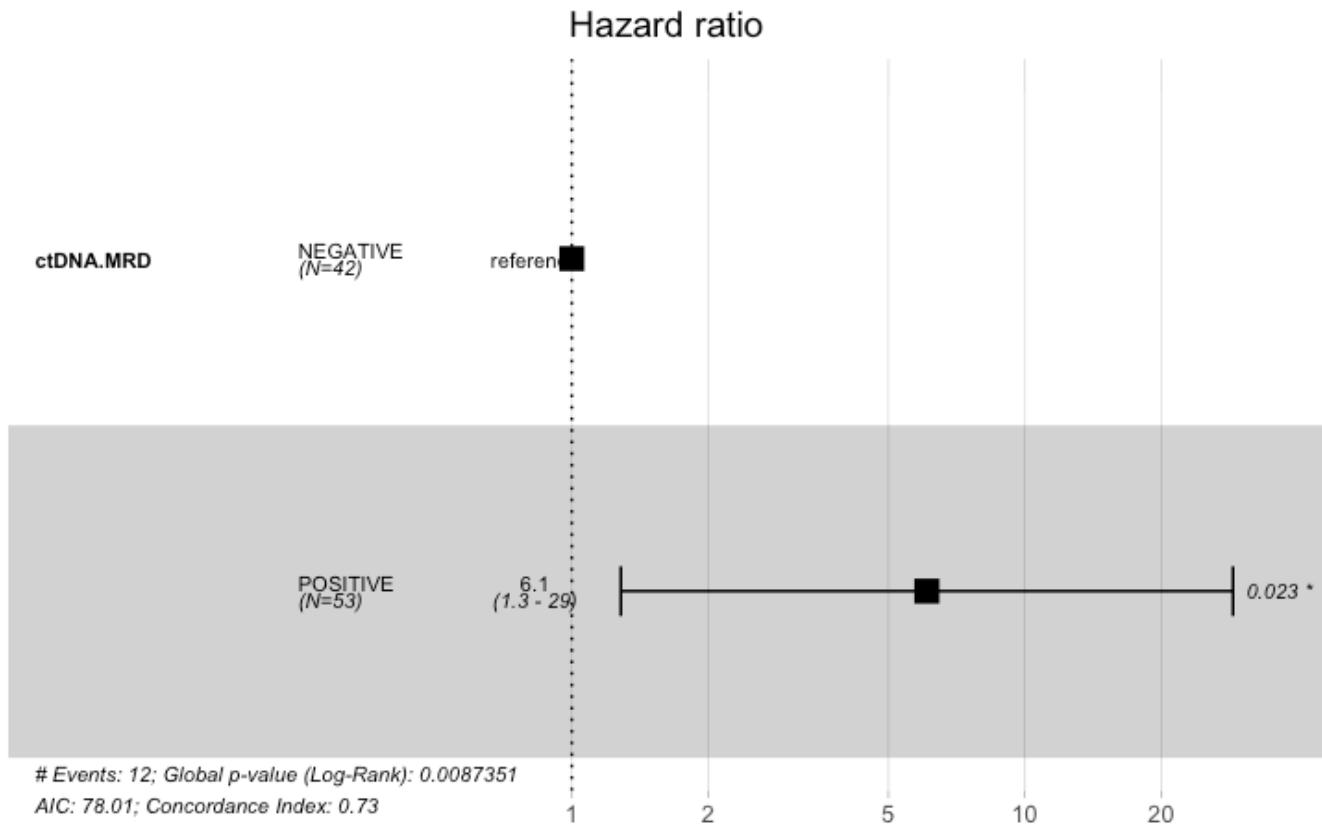
```
summary(KM_curve, times= c(24, 36))
```

```
Call: survfit(formula = surv_object ~ ctDNA.MRD, data = circ_data,
  conf.int = 0.95, conf.type = "log-log")
```

```
ctDNA.MRD=NEGATIVE
time n.risk n.event survival std.err lower 95% CI upper 95% CI
 24     25      0    1.000  0.0000      NA      NA
 36      8      1    0.933  0.0644    0.613    0.99
```

```
ctDNA.MRD=POSITIVE
time n.risk n.event survival std.err lower 95% CI upper 95% CI
 24     18      6    0.82   0.0709    0.627    0.919
 36      4      3    0.68   0.0942    0.459    0.827
```

```
circ_data$ctDNA.MRD <- factor(circ_data$ctDNA.MRD, levels=c("NEGATIVE","POSITIVE"))
cox_fit <- coxph(surv_object ~ ctDNA.MRD, data=circ_data)
ggforest(cox_fit, data = circ_data)
```



```
summary(cox_fit)
```

Call:
`coxph(formula = surv_object ~ ctDNA.MRD, data = circ_data)`

n= 95, number of events= 12

	coef	exp(coef)	se(coef)	z	Pr(> z)
ctDNA.MRDPOSITIVE	1.8047	6.0781	0.7937	2.274	0.023 *

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

	exp(coef)	exp(-coef)	lower .95	upper .95
ctDNA.MRDPOSITIVE	6.078	0.1645	1.283	28.8

Concordance= 0.735 (se = 0.031)
 Likelihood ratio test= 6.88 on 1 df, p=0.009
 Wald test = 5.17 on 1 df, p=0.02
 Score (logrank) test = 6.56 on 1 df, p=0.01

```
cox_fit_summary <- summary(cox_fit)

# Extract values for HR, 95% CI, and p-value
HR <- cox_fit_summary$coefficients[2]
lower_CI <- cox_fit_summary$conf.int[3]
upper_CI <- cox_fit_summary$conf.int[4]
p_value <- cox_fit_summary$coefficients[5]
label_text <- paste0("HR = ", round(HR, 2), " (", round(lower_CI, 2), "-", round(upper_CI, 2), "); p = ", round(p_value, 3))
print(label_text)
```

[1] "HR = 6.08 (1.28-28.8); p = 0.023"

[Hide](#)

```
#Fisher test for OS percentages at 24 and 36 months
os_times <- c(24, 36)
p_values <- sapply(os_times, function(time) {
  neg_count <- sum(circ_data$ctDNA.MRD == "NEGATIVE" & circ_data$OS.MRD.months >= time & circ_data$OS.Event == 0)
  pos_count <- sum(circ_data$ctDNA.MRD == "POSITIVE" & circ_data$OS.MRD.months >= time & circ_data$OS.Event == 0)
  neg_total <- sum(circ_data$ctDNA.MRD == "NEGATIVE")
  pos_total <- sum(circ_data$ctDNA.MRD == "POSITIVE")

  neg_surv <- neg_total - sum(circ_data$ctDNA.MRD == "NEGATIVE" & circ_data$OS.Event == 1 & circ_data$OS.MRD.months < time)
  pos_surv <- pos_total - sum(circ_data$ctDNA.MRD == "POSITIVE" & circ_data$OS.Event == 1 & circ_data$OS.MRD.months < time)

  surv_matrix <- matrix(c(neg_surv, pos_surv, neg_total - neg_surv, pos_total - pos_surv), nrow = 2)
  test_result <- fisher.test(surv_matrix)
  return(test_result$p.value)
})
names(p_values) <- paste0("p-value at ", os_times, " months")
print(p_values)
```

p-value at 24 months p-value at 36 months
 0.03245083 0.03892912

[Hide](#)

```
p_values_adjusted <- p.adjust(p_values, method = "bonferroni")
names(p_values_adjusted) <- paste0("Corrected p-value at ", os_times, " months")
print(p_values_adjusted)
```

Corrected p-value at 24 months Corrected p-value at 36 months
 0.06490165 0.07785824

#Percentage of ctDNA MRD Window positivity in pts with liver metastasis

Hide

```

rm(list = ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data %>%
  filter(LiverMets == "TRUE" & RFS.Event == "TRUE" & ctDNA.MRD != "")
circ_data$ctDNA.MRD <- factor(circ_data$ctDNA.MRD, levels = c("NEGATIVE", "POSITIVE"), labels = c("Negative", "Positive"))
positive_rate <- sum(circ_data$ctDNA.MRD == "Positive" & grepl("liver", circ_data$RelSite, ignore.case = TRUE)) / sum(circ_data$ctDNA.MRD == "Positive") * 100
positive_ci <- binconf(sum(circ_data$ctDNA.MRD == "Positive" & grepl("liver", circ_data$RelSite, ignore.case = TRUE)),
                       sum(circ_data$ctDNA.MRD == "Positive"),
                       alpha = 0.05)[c(2, 3)] * 100
negative_rate <- sum(circ_data$ctDNA.MRD == "Negative" & grepl("liver", circ_data$RelSite, ignore.case = TRUE)) / sum(circ_data$ctDNA.MRD == "Negative") * 100
negative_ci <- binconf(sum(circ_data$ctDNA.MRD == "Negative" & grepl("liver", circ_data$RelSite, ignore.case = TRUE)),
                       sum(circ_data$ctDNA.MRD == "Negative"),
                       alpha = 0.05)[c(2, 3)] * 100
data <- data.frame(
  ctDNA.MRD = c("Positive", "Negative"),
  percentage = c(positive_rate, negative_rate),
  lower_ci = c(positive_ci[1], negative_ci[1]),
  upper_ci = c(positive_ci[2], negative_ci[2]))
)
cross_tabs <- list(
  table(circ_data$ctDNA.MRD, grepl("liver", circ_data$RelSite, ignore.case = TRUE)),
  table(circ_data$ctDNA.MRD, grepl("lung", circ_data$RelSite, ignore.case = TRUE)) # Example for another site
)
chi_tests <- lapply(cross_tabs, chisq.test)
p_values <- sapply(chi_tests, function(test) test$p.value)
num_tests <- length(p_values) # Number of tests to be adjusted for
p_values_adjusted <- p.adjust(p_values, method = "bonferroni", n = num_tests)
print(data)

```

ctDNA.MRD	percentage	lower_ci	upper_ci
	<dbl>	<dbl>	<dbl>
Positive	73.58491	60.41934	83.56265
Negative	33.33333	21.01247	48.44749
2 rows			

Hide

```
print(cross_tabs)
```

[[1]]

	FALSE	TRUE
Negative	28	14
Positive	14	39

[[2]]

	FALSE	TRUE
Negative	16	26
Positive	39	14

Hide

print(chi_tests)

[[1]]

Pearson's Chi-squared test with Yates' continuity correction

data: X[[i]]
X-squared = 13.803, df = 1, p-value = 0.000203

[[2]]

Pearson's Chi-squared test with Yates' continuity correction

data: X[[i]]
X-squared = 10.695, df = 1, p-value = 0.001074

Hide

print(paste("Original p-values:", format.pval(p_values, digits = 4)))

[1] "Original p-values: 0.0002" "Original p-values: 0.0011"

Hide

print(paste("Adjusted p-values (Bonferroni):", format.pval(p_values_adjusted, digits = 4)))

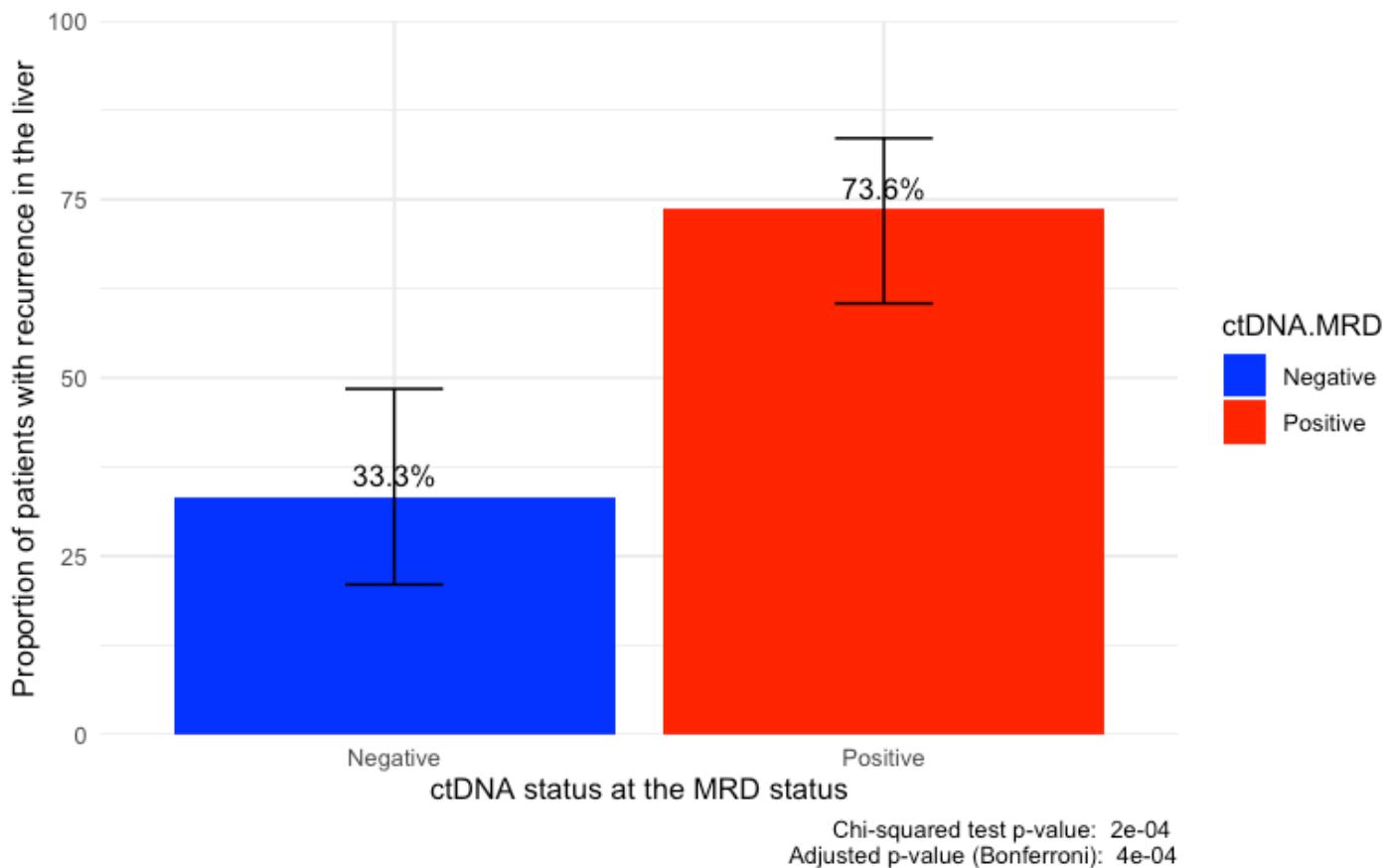
[1] "Adjusted p-values (Bonferroni): 0.0004" "Adjusted p-values (Bonferroni): 0.0021"

Hide

```

barplot <- ggplot(data, aes(x = ctDNA.MRD, y = percentage, fill = ctDNA.MRD)) +
  geom_bar(stat = "identity") +
  geom_errorbar(aes(ymin = lower_ci, ymax = upper_ci), width = 0.2) +
  geom_text(aes(label = paste0(round(percentage, 1), "%")), vjust = -0.5) +
  labs(
    x = "ctDNA status at the MRD status",
    y = "Proportion of patients with recurrence in the liver",
    caption = paste("Chi-squared test p-value: ", format.pval(p_values[1], digits = 4),
                    "\nAdjusted p-value (Bonferroni): ", format.pval(p_values_adjusted
    [1], digits = 4))
  ) +
  scale_y_continuous(expand = c(0, 0), limits = c(0, 100)) +
  scale_fill_manual(values = c("Negative" = "blue", "Positive" = "red")) +
  theme_minimal()
print(barplot)

```



#Percentage of ctDNA MRD Window positivity in pts with lung metastasis

Hide

```

rm(list = ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data %>%
  filter(LiverMets == "TRUE" & RFS.Event == "TRUE" & ctDNA.MRD != "")
circ_data$ctDNA.MRD <- factor(circ_data$ctDNA.MRD, levels = c("NEGATIVE", "POSITIVE"), labels = c("Negative", "Positive"))
positive_rate <- sum(circ_data$ctDNA.MRD == "Positive" & grepl("lung", circ_data$RelSite, ignore.case = TRUE)) / sum(circ_data$ctDNA.MRD == "Positive") * 100
positive_ci <- binconf(sum(circ_data$ctDNA.MRD == "Positive" & grepl("lung", circ_data$RelSite, ignore.case = TRUE)),
                       sum(circ_data$ctDNA.MRD == "Positive"),
                       alpha = 0.05)[c(2, 3)] * 100
negative_rate <- sum(circ_data$ctDNA.MRD == "Negative" & grepl("lung", circ_data$RelSite, ignore.case = TRUE)) / sum(circ_data$ctDNA.MRD == "Negative") * 100
negative_ci <- binconf(sum(circ_data$ctDNA.MRD == "Negative" & grepl("lung", circ_data$RelSite, ignore.case = TRUE)),
                       sum(circ_data$ctDNA.MRD == "Negative"),
                       alpha = 0.05)[c(2, 3)] * 100
data <- data.frame(
  ctDNA.MRD = c("Positive", "Negative"),
  percentage = c(positive_rate, negative_rate),
  lower_ci = c(positive_ci[1], negative_ci[1]),
  upper_ci = c(positive_ci[2], negative_ci[2])
)
cross_tabs <- list(
  table(circ_data$ctDNA.MRD, grepl("lung", circ_data$RelSite, ignore.case = TRUE)),
  table(circ_data$ctDNA.MRD, grepl("liver", circ_data$RelSite, ignore.case = TRUE))
)
chi_tests <- lapply(cross_tabs, chisq.test)
p_values <- sapply(chi_tests, function(test) test$p.value)
num_tests <- length(p_values) # Number of tests to be adjusted for
p_values_adjusted <- p.adjust(p_values, method = "bonferroni", n = num_tests)
print(data)

```

ctDNA.MRD	percentage	lower_ci	upper_ci
<chr>	<dbl>	<dbl>	<dbl>
Positive	26.41509	16.43735	39.58066
Negative	61.90476	46.81404	75.00027
2 rows			

Hide

```
print(cross_tabs)
```

[[1]]

	FALSE	TRUE
Negative	16	26
Positive	39	14

[[2]]

	FALSE	TRUE
Negative	28	14
Positive	14	39

Hide

print(chi_tests)

[[1]]

Pearson's Chi-squared test with Yates' continuity correction

data: X[[i]]
X-squared = 10.695, df = 1, p-value = 0.001074

[[2]]

Pearson's Chi-squared test with Yates' continuity correction

data: X[[i]]
X-squared = 13.803, df = 1, p-value = 0.000203

Hide

print(paste("Original p-values:", format.pval(p_values, digits = 4)))

[1] "Original p-values: 0.0011" "Original p-values: 0.0002"

Hide

print(paste("Adjusted p-values (Bonferroni):", format.pval(p_values_adjusted, digits = 4)))

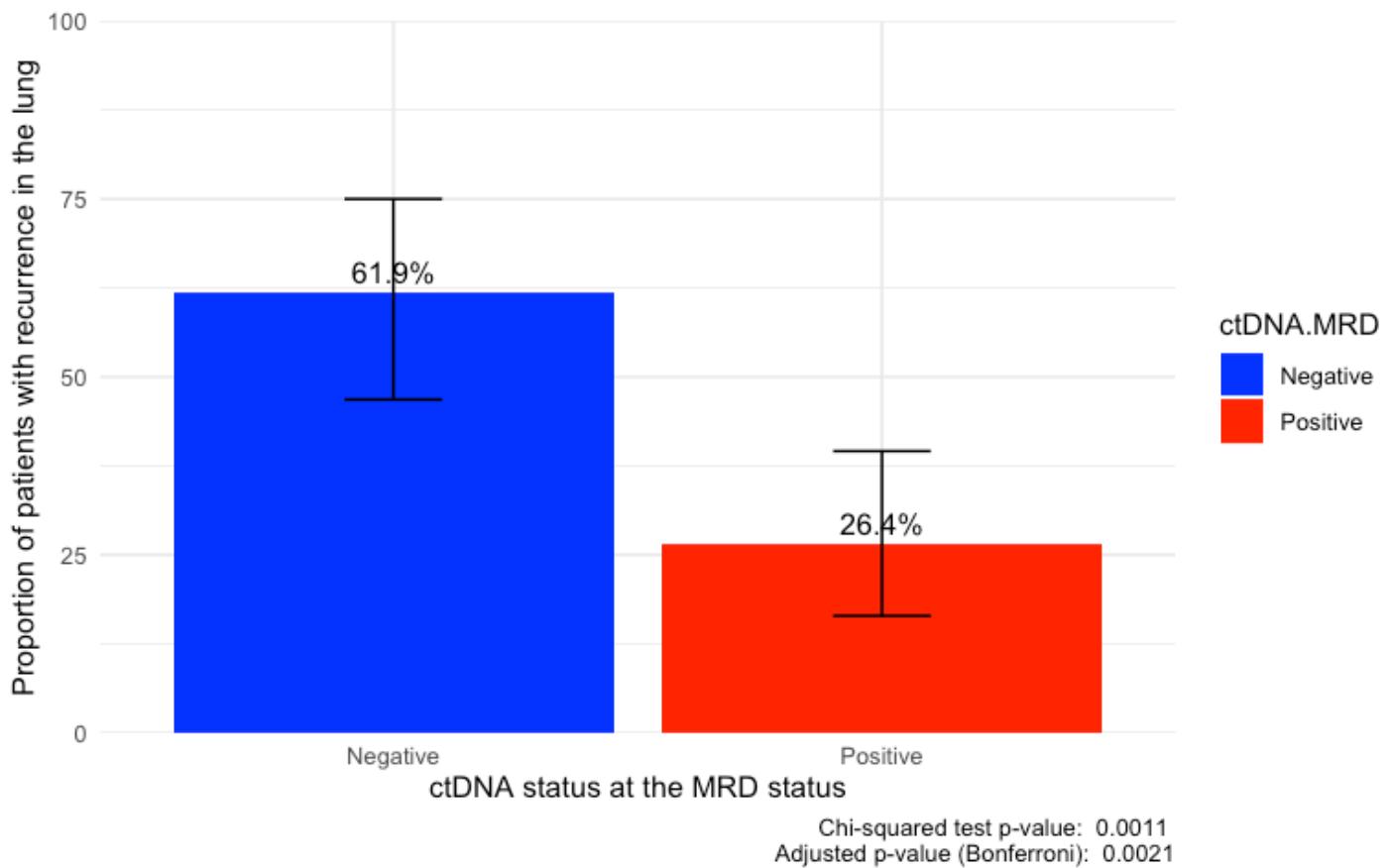
[1] "Adjusted p-values (Bonferroni): 0.0021" "Adjusted p-values (Bonferroni): 0.0004"

Hide

```

barplot <- ggplot(data, aes(x = ctDNA.MRD, y = percentage, fill = ctDNA.MRD)) +
  geom_bar(stat = "identity") +
  geom_errorbar(aes(ymin = lower_ci, ymax = upper_ci), width = 0.2) +
  geom_text(aes(label = paste0(round(percentage, 1), "%")), vjust = -0.5) +
  labs(
    x = "ctDNA status at the MRD status",
    y = "Proportion of patients with recurrence in the lung",
    caption = paste("Chi-squared test p-value: ", format.pval(p_values[1], digits = 4),
                    "\nAdjusted p-value (Bonferroni): ", format.pval(p_values_adjusted
    [1], digits = 4)))
  ) +
  scale_y_continuous(expand = c(0, 0), limits = c(0, 100)) +
  scale_fill_manual(values = c("Negative" = "blue", "Positive" = "red")) +
  theme_minimal()
print(barplot)

```



#Barplot with Recurrence Sites (Liver vs Others) by ctDNA at the MRD Window

Hide

```
rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$RFS.Event=="TRUE",]

circ_data$ctDNA.MRD <- factor(circ_data$ctDNA.MRD, levels = c("NEGATIVE", "POSITIVE"), labels = c("Negative", "Positive"))
circ_data$Liver <- factor(circ_data$Liver, levels = c("FALSE", "TRUE"), labels = c("Others", "Liver"))
contingency_table <- table(circ_data$ctDNA.MRD, circ_data$Liver)
chi_square_test <- chisq.test(contingency_table)
print(chi_square_test)
```

Pearson's Chi-squared test with Yates' continuity correction

```
data: contingency_table
X-squared = 18.352, df = 1, p-value = 1.836e-05
```

[Hide](#)

```
fisher_exact_test <- fisher.test(contingency_table)
print(fisher_exact_test)
```

Fisher's Exact Test for Count Data

```
data: contingency_table
p-value = 9.391e-06
alternative hypothesis: true odds ratio is not equal to 1
95 percent confidence interval:
 2.744866 20.863460
sample estimates:
odds ratio
 7.311049
```

[Hide](#)

```
print(contingency_table)
```

	Liver	
	Others	Liver
Negative	30	12
Positive	13	39

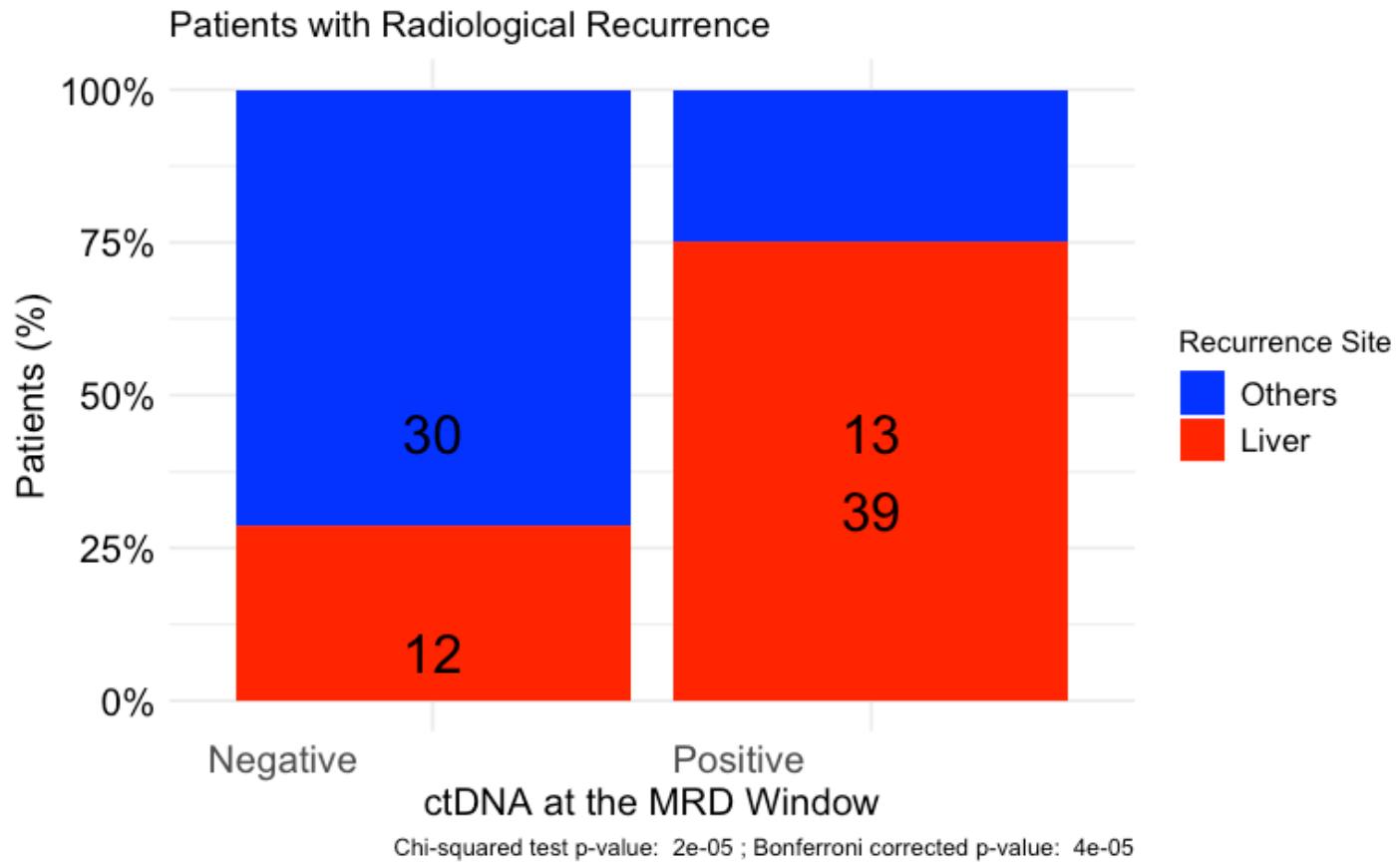
[Hide](#)

```
p_values <- c(chi_square_test$p.value, fisher_exact_test$p.value)
p_adjusted <- p.adjust(p_values, method = "bonferroni")
names(p_adjusted) <- c("Chi-Square Test", "Fisher's Exact Test")
print(p_adjusted)
```

```
Chi-Square Test Fisher's Exact Test
3.672023e-05      1.878209e-05
```

[Hide](#)

```
table_df <- as.data.frame(contingency_table)
table_df$Total <- ave(table_df$Freq, table_df$Var1, FUN = sum)
table_df$Percentage <- table_df$Freq / table_df$Total
table_df$MiddlePercentage <- table_df$Percentage / 2
ggplot(table_df, aes(x = Var1, y = Percentage, fill = Var2)) +
  geom_bar(stat = "identity") +
  geom_text(aes(y = MiddlePercentage, label = Freq), position = "stack", color = "black", vjust = 1.5, size = 7) +
  theme_minimal() +
  labs(title = "Patients with Radiological Recurrence",
       x = "ctDNA at the MRD Window",
       y = "Patients (%)",
       fill = "Recurrence Site",
       caption = paste("Chi-squared test p-value: ", format.pval(chi_square_test$p.value), "; Bonferroni corrected p-value: ", format.pval(p_adjusted["Chi-Square Test"]))) +
  scale_y_continuous(labels = scales::percent_format()) +
  scale_fill_manual(values = c("Others" = "blue", "Liver" = "red")) + # define custom colors
  theme(axis.text.x = element_text(angle = 0, hjust = 1.5, size = 14), # increase x-axis text size
        axis.text.y = element_text(size = 14, color = "black"), # increase y-axis text size
        axis.title.x = element_text(size = 14, color = "black"), # increase x-axis label size
        axis.title.y = element_text(size = 14, color = "black"), # increase y-axis label size
        legend.text = element_text(size = 12, color = "black")) # increase Recurrence label size
```



#Table with recurrence sites by ctDNA at the MRD Window

[Hide](#)

```

rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$RFS.Event=="TRUE",]

circ_data_subset1 <- circ_data %>%
  select(
    Rec.Site) %>%
  mutate(
    Rec.Site = factor(Rec.Site, levels = c("Local/LN", "Liver", "Lung", "Peritoneum", "Peritoneum & Others", "Brain")))

circ_data1 <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data1 <- circ_data1[circ_data1$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$RFS.Event=="TRUE",]

circ_data_subset2 <- circ_data %>%
  select(
    Rec.Site,
    ctDNA.MRD) %>%
  mutate(
    Rec.Site = factor(Rec.Site, levels = c("Local/LN", "Liver", "Lung", "Peritoneum", "Peritoneum & Others", "Brain")),
    ctDNA.MRD = factor(ctDNA.MRD, levels = c("NEGATIVE", "POSITIVE")))
Overall <- circ_data_subset1 %>%
 tbl_summary(
  statistic = list(
    all_continuous() ~ "{median} ({min} - {max})",
    all_categorical() ~ "{n} ({p}%)") %>%
bold_labels()
Overall

```

Characteristic	N = 95 ¹
Rec.Site	
Local/LN	4 (4.3%)
Liver	48 (51%)
Lung	30 (32%)
Peritoneum	4 (4.3%)
Peritoneum & Others	7 (7.4%)
Brain	1 (1.1%)
Unknown	1

Characteristic **N = 95¹**¹ n (%)

```
ByctDNA_MRД <- circ_data_subset2 %>%
 tbl_summary(
  by = ctDNA.MRD, # Subgroup by ctDNA.MRD
  statistic = list(
    all_continuous() ~ "{median} ({min} - {max})",
    all_categorical() ~ "{n} ({p}%)") %>%
  add_p(test = all_categorical() ~ "fisher.test", pvalue_fun = ~style_pvalue(p.adjust(.x, method = "bonferroni")))) %>%
  bold_labels()
ByctDNA_MRД
```

Characteristic	NEGATIVE, N = 42¹	POSITIVE, N = 53¹	p-value²
Rec.Site			<0.001
Local/LN	1 (2.4%)	3 (5.8%)	
Liver	12 (29%)	36 (69%)	
Lung	23 (55%)	7 (13%)	
Peritoneum	3 (7.1%)	1 (1.9%)	
Peritoneum & Others	3 (7.1%)	4 (7.7%)	
Brain	0 (0%)	1 (1.9%)	
Unknown	0	1	

¹ n (%)² Fisher's exact test

```
merged_table <-tbl_merge(tbls=list(Overall, ByctDNA_MRД))
merged_table
```

Characteristic	Table 1		Table 2	
	N = 95 ¹	NEGATIVE, N = 42 ¹	POSITIVE, N = 53 ¹	p-value ²
Rec.Site	<0.001			
Local/LN	4 (4.3%)	1 (2.4%)	3 (5.8%)	
Liver	48 (51%)	12 (29%)	36 (69%)	
Lung	30 (32%)	23 (55%)	7 (13%)	
Peritoneum	4 (4.3%)	3 (7.1%)	1 (1.9%)	
Peritoneum & Others	7 (7.4%)	3 (7.1%)	4 (7.7%)	
Brain	1 (1.1%)	0 (0%)	1 (1.9%)	
Unknown	1	0	1	

¹ n (%)² Fisher's exact test[Hide](#)

```
fit1 <- as_flex_table(
  merged_table,
  include = everything(),
  return_calls = FALSE,
  strip_md_bold = TRUE)
fit1
```

Characteristic	Table 1		Table 2	
	N = 95 ¹	NEGATIVE, N = 42 ¹	POSITIVE, N = 53 ¹	p-value ²
Rec.Site	<0.001			
Local/LN	4 (4.3%)	1 (2.4%)	3 (5.8%)	
Liver	48 (51%)	12 (29%)	36 (69%)	
Lung	30 (32%)	23 (55%)	7 (13%)	
Peritoneum	4 (4.3%)	3 (7.1%)	1 (1.9%)	
Peritoneum & Others	7 (7.4%)	3 (7.1%)	4 (7.7%)	
Brain	1 (1.1%)	0 (0%)	1 (1.9%)	
Unknown	1	0	1	

¹n (%)²Fisher's exact test

Hide

```
save_as_docx(fit1, path= "~/Downloads/merged_table.docx")
```

#Detection ctDNA rates based on sites of relapse

Hide

```
rm(list=ls())
setwd("~/Downloads")
circ_data <- read.csv("Galaxy Liver Mets Data_20240626 Complete Dataset.csv")
circ_data <- circ_data[circ_data$LiverMets=="TRUE",]
circ_data <- circ_data[circ_data$RFS.Event=="TRUE",]
circ_data <- subset(circ_data, !is.na(Rec.Site))

# Create a table of counts for the "Rec.Site" variable
relsite_counts <- table(circ_data$Rec.Site)
relsite_df <- as.data.frame(relsite_counts)
names(relsite_df) <- c("RelSite", "Count")
circ_data_pos_mrd <- circ_data[circ_data$ctDNA.MRD=="POSITIVE",]
circ_data_pos_anytime <- circ_data[circ_data$ctDNA.anytime=="POSITIVE",]
pos_counts_mrd <- table(circ_data_pos_mrd$Rec.Site)
pos_counts_anytime <- table(circ_data_pos_anytime$Rec.Site)
relsite_df$MRDPos_Count <- ifelse(is.na(match(relsite_df$RelSite, names(pos_counts_mrd))), 0, pos_counts_mrd[match(relsite_df$RelSite, names(pos_counts_mrd))])
relsite_df$MRDPos_Count[is.na(relsite_df$MRDPos_Count)] <- 0
relsite_df$AnytimePos_Count <- ifelse(is.na(match(relsite_df$RelSite, names(pos_counts_anytime))), 0, pos_counts_anytime[match(relsite_df$RelSite, names(pos_counts_anytime))])
relsite_df$AnytimePos_Count[is.na(relsite_df$AnytimePos_Count)] <- 0
relsite_df$Percent <- (relsite_df$Count / sum(relsite_df$Count)) * 100
relsite_df$MRDPos_Percent <- (relsite_df$MRDPos_Count / relsite_df$Count) * 100
relsite_df$AnytimePos_Percent <- (relsite_df$AnytimePos_Count / relsite_df$Count) * 100
total_observations <- sum(relsite_df$Count)
total_pos_mrd <- sum(relsite_df$MRDPos_Count)
total_pos_anytime <- sum(relsite_df$AnytimePos_Count)
total_row <- data.frame(RelSite = "Total", Count = total_observations, MRDPos_Count = total_pos_mrd, AnytimePos_Count = total_pos_anytime, Percent = 100, MRDPos_Percent = (total_pos_mrd / total_observations) * 100, AnytimePos_Percent = (total_pos_anytime / total_observations) * 100)
relsite_df <- rbind(relsite_df, total_row)
print(relsite_df)
```

RelSite	Co...	MRDPos_C...	AnytimePos_Count	Percent	MRDPos_Per...
<fctr>	<int>	<dbl>	<dbl>	<dbl>	<dbl>
	1	1	1	1.052632	100.00000
Brain	1	1	1	1.052632	100.00000
Liver	48	36	46	50.526316	75.00000
Local/LN	4	3	4	4.210526	75.00000

RelSite <fctr>	Co... <int>	MRDPos_C... <dbl>	AnytimePos_Count <dbl>	Percent <dbl>	MRDPos_Perc... <dbl>
Lung	30	7	14	31.578947	23.33333
Peritoneum	4	1	4	4.210526	25.00000
Peritoneum & Others	7	4	5	7.368421	57.14286
Total	95	53	75	100.000000	55.78947

8 rows | 1-6 of 7 columns

[Hide](#)

```
ft <- flextable(relsite_df)
doc <- read_docx() %>%
  body_add_flextable(value = ft)
print(doc, target = "relsite_df.docx")
```