

IntroSlides with focus on statistics

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Basic steps in workflow

1. *Define environment*
2. *Import*
3. *Transform*
4. *Explore (general/outlier/distribution) (go back to 3?)*
5. *Classify scale level / distribution (based on 3/4)*
6. *Describe*
7. *Test / Model (may include step 6)*
8. *Report*

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Define environment

- *Activate packages to use: library() / pacman::p_load()*
- *ggplot theme: theme_set() / theme_update()*
- *flextable settings: set_flextable_defaults()*
- *knitr::opts_chunk\$set()*

```
1 if(!requireNamespace("pacman")){install.packages("pacman")}
2 pacman::p_load(conflicted, tidyverse, wrappedtools, readxl, car, flextable,
3               ggbeeswarm, ggsignif, gggridges, patchwork, easystats)
4
5 conflicts_prefer(dplyr::filter, dplyr::select)
6 theme_set(theme_light(base_size = 20))
7 gdtools::register_gfont('Roboto') # Mono'
```

```
[1] TRUE
```

```
1 set_flextable_defaults(
2   theme_fun = theme_zebra, font.size = 18, font.family = 'Roboto',
3   table.layout = 'autofit',
4   padding.bottom = .2, padding.top = .2, padding.left = 2, padding.right = 2)
5
6 knitr::opts_chunk$set(message = FALSE, warning = FALSE, comment = NA, echo = TRUE)
```

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Import

- *read_xlsx() / read_csv() / read_csv2()*
- options related to separators, number formats, ranges etc.
- *rename() / rename_with()*

```
1 rawdata <- read_excel('Data/DOC-20230130-WA0000_.xlsx',
2                       sheet = 1, col_names = TRUE)
```

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Glimpse at data: Find the problems?

```
1 head(rawdata,n = 15) |> flextable()|>
2   theme_zebra(even_body = 'aquamarine',odd_body = 'antiquewhite')
```

CODE OF CUP	CODE OF SAMPLE	WEIGHT OF EMPTY ALUMINUM(wt)	WEIGHT OF ALMINIUM CUP + SAMPLE (Wt + s)	WEIGHT OF ALUMINIUM CAP + SAMPLE AFTER DRYING (Wt-AL +s+d)	weight of sample before drying (Wts)	weight of sample after drying (Wts+d)	MOISTURE CONTENT (%)
69	D	4.1974	9.3865	4.7000	5.1891	4.6865	90.31431
	D	4.1964	9.2734	4.4670	5.0770	4.8064	94.67008
A	D	4.2108	9.2653	4.6670	5.0545	4.5983	90.97438
114	D	4.2134	9.3146	4.6345	5.1012	4.6801	91.74508
M1	D	4.1856	9.3147	4.6171	5.1291	4.6976	91.58722
a/17	D	4.2090	9.3204	4.5661	5.1114	4.7543	93.01366
8	D	4.1894	9.2661	4.5778	5.0767	4.6883	92.34936
33	D	4.1968	9.2880	4.6057	5.0912	4.6823	91.96849
M	D	4.1535	9.2872	4.6350	5.1337	4.6522	90.62080
E/18/1	D	4.2534	9.2476	4.7403	4.9942	4.5073	90.25069
24/A2	D	4.2066	8.3463	4.5849	4.1397	3.7614	90.86166
13	A	4.1554	9.2384	4.7402	5.0830	4.4982	88.49498
Xp	A	4.1893	9.2495	4.7381	5.0602	4.5114	89.15458
2p/029	A	4.0654	9.2173	4.6940	5.1519	4.5233	87.79868
15	A	4.0641	9.2032	4.8124	5.1391	4.3908	85.43908

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Rename

```
1 colnames(rawdata)
```

```
[1] "CODE OF CUP"
[2] "CODE OF SAMPLE"
[3] "WEIGHT OF EMPTY ALUMINUM(wt)"
[4] "WEIGHT OF ALMINIUM CUP + SAMPLE (Wt + s)"
[5] "WEIGHT OF ALUMINIUM CAP + SAMPLE AFTER DRYING (Wt-AL +s+d)"
[6] "weight of sample before drying (Wts)"
[7] "weight of sample after drying (Wts+d)"
[8] "MOISTURE CONTENT (%)"
```

```
1 rawdata <- rawdata |>
2   rename(Region=`CODE OF SAMPLE`) |>
3   rename_with(.fn = ~str_replace_all(.,
4                                     c('AL.+UM'= 'Cup', 'C[UA]P' = 'Cup', '\\(\\w+.*\\)
5                                     'Cup Cup'='Cup', ' '= ' ')) |>
6     str_to_title() |> str_trim())
7   cn()
```

```
[1] "Code Of Cup"           "Region"
[3] "Weight Of Empty Cup"   "Weigth Of Cup + Sample"
[5] "Weigth Of Cup + Sample After Drying" "Weight Of Sample Before Drying"
[7] "Weight Of Sample After Drying"      "Moisture Content (%)"
```

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Transform

- *Change or create columns with mutate() / mutate(across())*
- *e.g. for log-transformation, creation of factors, text recoding*

```
1 rawdata <- rawdata |>
2   mutate(
3     `Weight Of Sample After Drying`=`Weigth Of Cup + Sample After Drying`-
4     `Weight Of Empty Cup`,
5     `Dry Content (%)`=`Weight Of Sample After Drying`*100/
6     `Weight Of Sample Before Drying`,
7     `Moisture Content (%)`=`100-`Dry Content (%)`)
```

Code Of Cup	Region	Weight Of Empty Cup	Weigth Of Cup + Sample	Weight Of Cup + Sample After Drying	Weight Of Sample Before Drying	Weight Of Sample After Drying	Moisture Content (%)	Dry Content (%)
69	D	4.1974	9.3865	4.7000	5.1891	0.5026	90.31431	9.685687
	D	4.1964	9.2734	4.4670	5.0770	0.2706	94.67008	5.329919
A	D	4.2108	9.2653	4.6670	5.0545	0.4562	90.97438	9.025621
114	D	4.2134	9.3146	4.6345	5.1012	0.4211	91.74508	8.254920
M1	D	4.1856	9.3147	4.6171	5.1291	0.4315	91.58722	8.412782

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Explore / group variables

Explore (general/outlier/distribution)

- *ggplot()+geom_boxplot() / geom_beeswarm() / geom_density()*
- *ks.test() / ksnormal() / shapiro.test()*

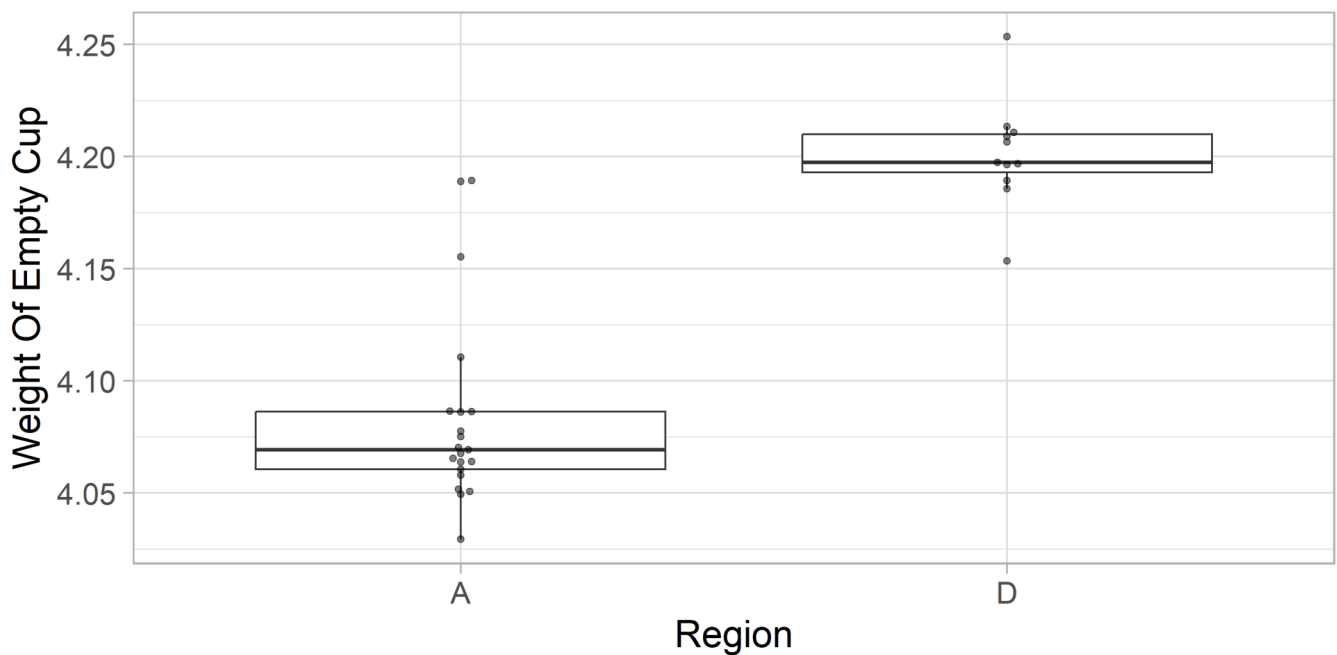
Classify scale level / distribution

- *gaussvars / ordvars / factvars, possibly more...*
- *Store variables accordingly, e.g. FindVars()*

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Explore: Outlier

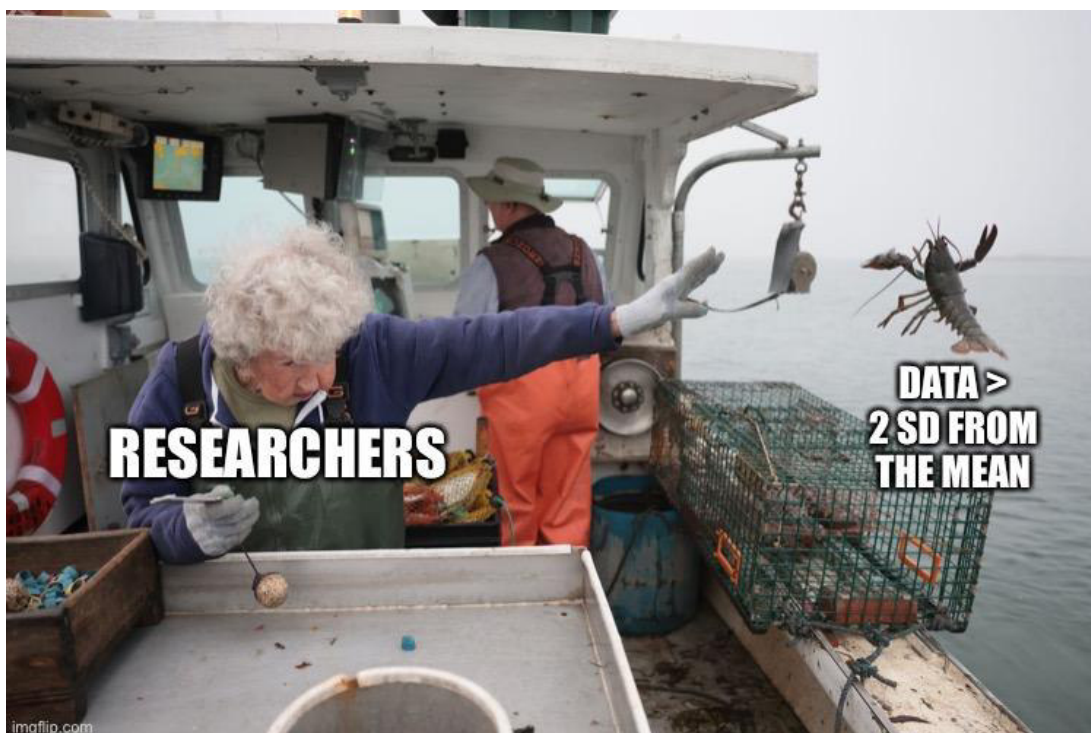
```
1 ggplot(data = rawdata,  
2       aes(x = `Region`,  
3           y = `Weight Of Empty Cup`))+  
4   geom_boxplot(outlier.alpha = 0) + #hide outliers, beeswarm will plot them  
5   geom_beeswarm(alpha=.5)
```



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Handle outliers?

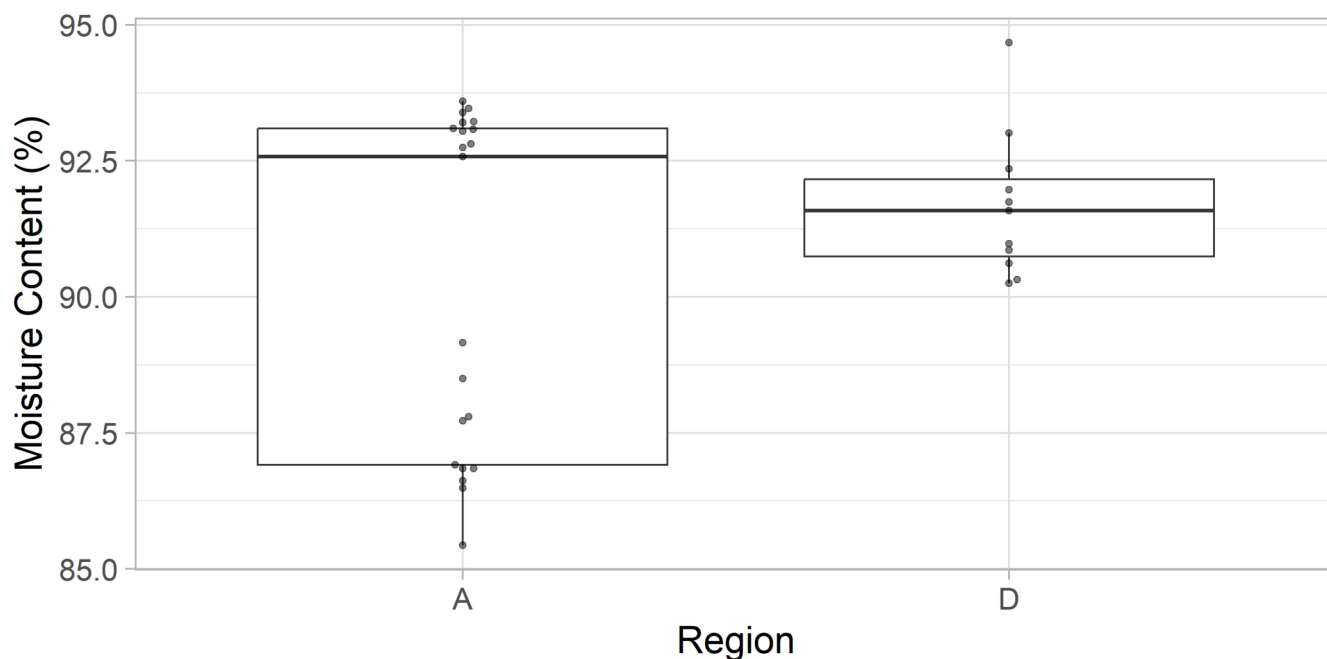
Removal is the worst strategy possible, correct errors, think about distributions, winsorize...



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Explore: Unexpected

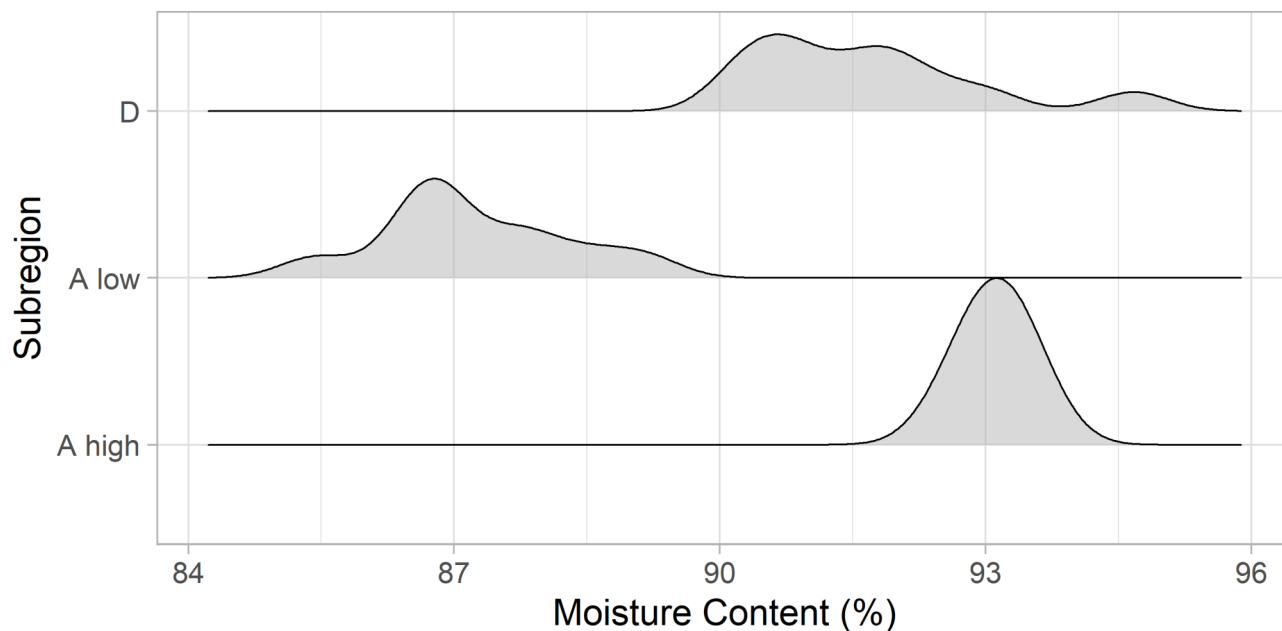
```
1 ggplot(data = rawdata,  
2       aes(x = `Region`,  
3           y = `Moisture Content (%)`))+  
4   geom_boxplot(outlier.alpha = 0) +  
5   geom_beeswarm(alpha=.5)
```



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Transform Subregions?

```
1 rawdata <- mutate(rawdata, Subregion = case_when(  
2   `Region`=='A' & `Moisture Content (%)` > 90 ~ 'A high',  
3   `Region`=='A' & `Moisture Content (%)` <= 90 ~ 'A low',  
4   `Region`=='D' ~ 'D') |> factor())  
5 ggplot(data = rawdata, aes(x = `Moisture Content (%)`, y=Subregion))+  
6   geom_density_ridges(alpha=.5, scale=1)
```



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Explore: Normal distribution 1

- Gaussian Normal distribution is required for many statistical procedures
- Common tests are graphical exploration, Shapiro-Wilk-test and Kolmogorov-Smirnov-test

```
1 p_normal <-  
2   shapiro.test(x = rawdata$`Moisture Content (%)`)  
3 p_normal
```

Shapiro-Wilk normality test

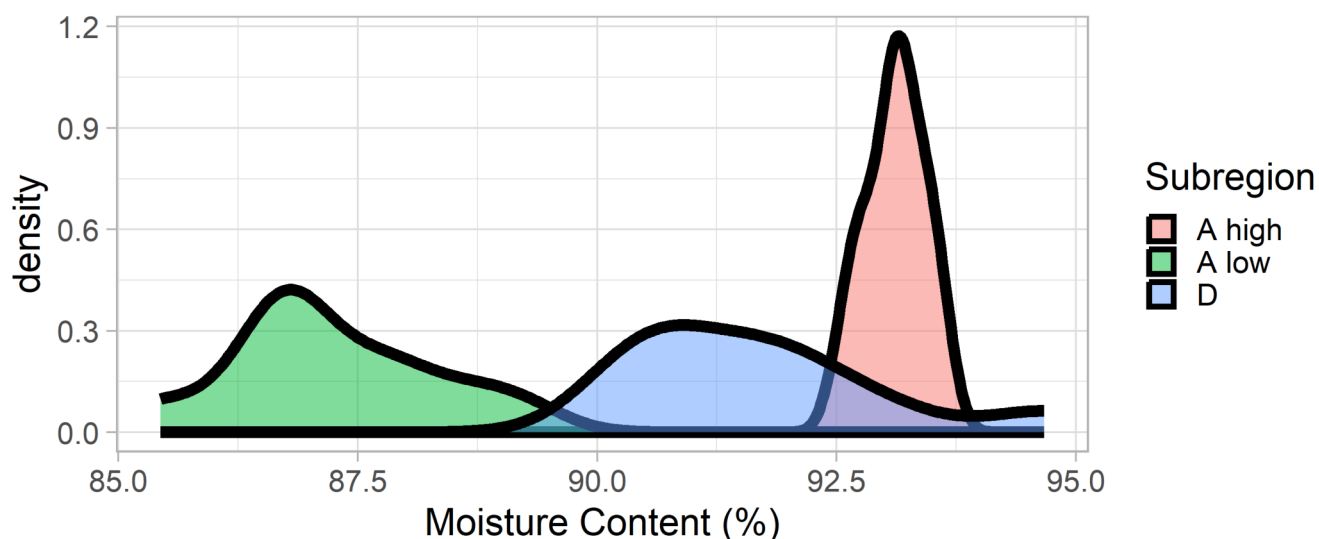
```
data: rawdata$`Moisture Content (%)`  
W = 0.89133, p-value = 0.003752
```

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```
1 ggplot(data = rawdata, aes(x = `Moisture Content (%)`, fill=`Subregion`))+  
2   geom_density(linewidth=3, alpha=.5)+  
3   labs(title = paste('p (Shapiro) global',  
4     formatP(pIn = p_normal$p.value, pretext = T)),  
5     subtitle = rawdata |> group_by(Subregion) |>  
6     summarize(pNormal=shapiro.test(`Moisture Content (%)`)$p.value |> formatP()) |>  
7     unite('p(Normal)', sep = ': p=') |> pull(1) |> paste(collapse = '; '))
```

p (Shapiro) global = 0.004

A high: p=0.923; A low: p=0.756; D: p=0.207



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Explore: Normal distribution 2

```
1 rawdata |>
2   group_by(`Subregion`) |>
3   summarize(across(.cols = where(is.numeric), .fns = ksnormal)) |>
4   pivot_longer(cols = -1, names_to = 'Variable', values_to = 'pKS') |>
5   pivot_wider(names_from = `Subregion`, values_from = pKS)
```

A tibble: 7 × 4

Variable	`A high`	`A low`	D
<chr>	<dbl>	<dbl>	<dbl>
1 Weight Of Empty Cup	0.989	0.444	0.697
2 Weigth Of Cup + Sample	0.785	0.980	0.0187
3 Weigth Of Cup + Sample After Drying	0.900	0.710	0.969
4 Weight Of Sample Before Drying	0.196	0.999	0.0716
5 Weight Of Sample After Drying	0.976	0.555	1.00
6 Moisture Content (%)	0.975	0.733	0.954
7 Dry Content (%)	0.975	0.733	0.954

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Explore: Group variables by type/distribution

Scale level determines what statistics are appropriate

Typical scale levels are

- nominal/categorical/factorial/qualitative: just different groups (species, eye color, genotype, treatment)
- ordered categories: few groups with inherent order (quality bad<medium<good, pain between 0 and 10)
- ordinal measures: many different values, natural order, no distribution assumption (satisfaction on a scale from 0 to 100)
- measures following a Normal distribution
- possibly measures from other known distributions (beta, log-normal, poisson...), often treated as ordinal

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Make type decision obvious/reproducible

```
1 gaussvars <- FindVars(varnames = c('Weight Of Sample','Content'),  
2                           allnames = cn(rawdata))  
3 gaussvars
```

\$index

```
[1] 6 7 8 9
```

\$names

```
[1] "Weight Of Sample Before Drying" "Weight Of Sample After Drying"  
[3] "Moisture Content (%)"           "Dry Content (%)"
```

\$bticked

```
[1] "`Weight Of Sample Before Drying`" "`Weight Of Sample After Drying`"  
[3] "`Moisture Content (%)`"           "`Dry Content (%)`"
```

\$count

```
[1] 4
```

```
1 ordvars <- FindVars(c('Cup'), exclude = 'Code')  
2 ordvars$names
```

```
[1] "Weight Of Empty Cup"           "Weigth Of Cup + Sample"  
[3] "Weigth Of Cup + Sample After Drying"
```

```
1 factvars <- FindVars('egion',casesensitive = FALSE)  
2 factvars$bticked
```

```
[1] "`Region`"           "`Subregion`"
```

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Model

Describe

- *mean() / sd() / meansd()*
- *median() / quantile() / median_quart()*
- *table() / prop.table() / cat_desc_stats()*

Test

- *t.test() / lm()+[Aa]nova() / compare2numvars()*
- *wilcox.test()*
- *fisher.test() / glm(family=binomial)*

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Model: Describe

Sample size n: per variable, if there are NAs

Mean: central tendency, the expected *typical* value

$$\frac{\sum x}{n}$$

Variance: measure for variability/heterogeneity of data

Standard deviation SD: the *typical* weighted deviation from the mean

Standard error of the mean SEM: how reliable is the mean *estimate*, what would be the expected SD of means from repeated experiments?

Median: Split between lower/upper 50% of data

Quartiles: Split at 25%/50%/75% of data (more general: **Quantiles**, e.g. **Percentiles**), used in boxplot

various computational approaches

```

1 desc_gauss <- rawdata |>
2   summarize(across(.cols = gaussvars$names,
3                     .fns = meansd))
4 desc_gauss

# A tibble: 1 × 4
  `Weight Of Sample Before Drying` Weight Of Sample Aft...1 `Moisture Content (%)`
  <chr>                                <chr>                                <chr>
1 5.1 ± 0.2                          0.47 ± 0.14                          91 ± 3
# i abbreviated name: 1`Weight Of Sample After Drying`
# i 1 more variable: `Dry Content (%)` <chr>

```

```

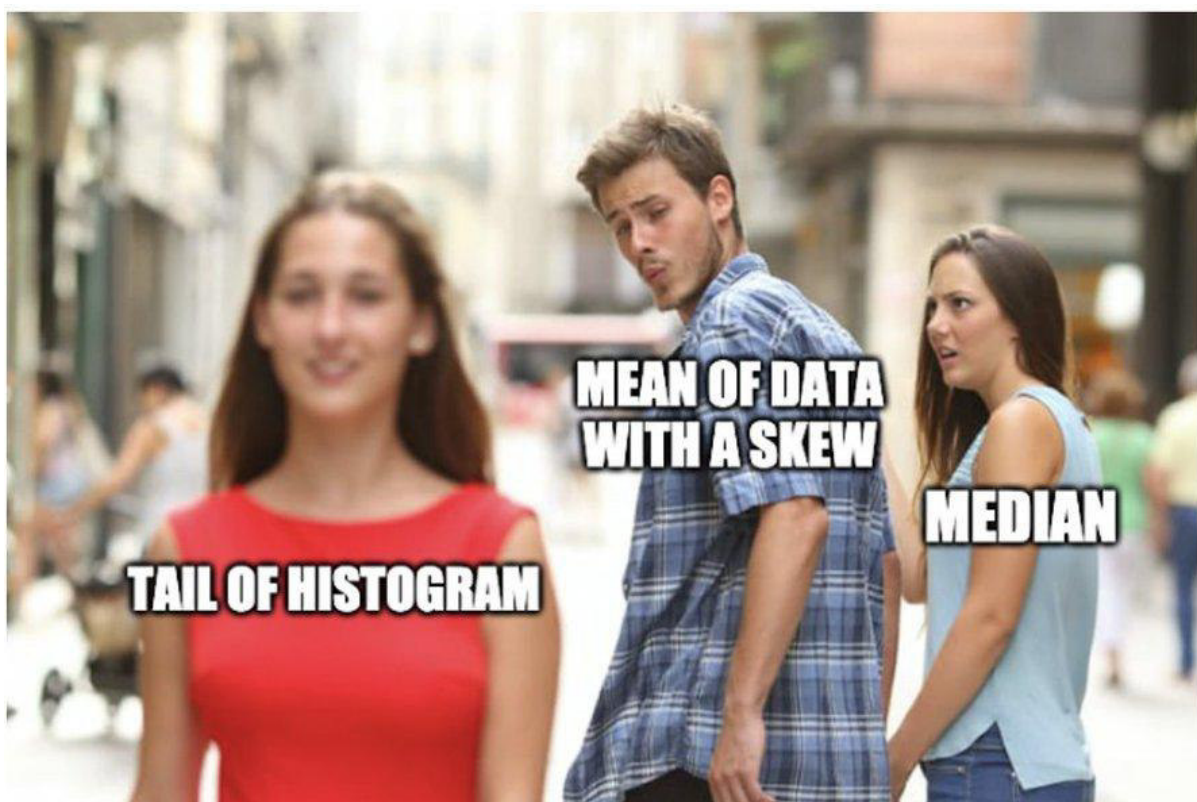
1 desc_ord <- rawdata |>
2   summarize(across(ordvars$names, .fns=~median_quart(.x, roundDig = 3))) |>
3   pivot_longer(everything(),
4                 names_to = 'Measure', values_to = 'Median[1Q/3Q]')
5 desc_ord

# A tibble: 3 × 2
  Measure                                `Median[1Q/3Q]`
  <chr>                                <chr>
1 Weight Of Empty Cup                  4.09 (4.06/4.19)
2 Weigth Of Cup + Sample               9.25 (9.22/9.29)
3 Weigth Of Cup + Sample After Drying 4.61 (4.45/4.72)

```

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Descriptive Stats should match distribution and data



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Model: Test

Tests require hypotheses



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Null hypothesis ?

- Working hypothesis: This is what you expect!
E.g. treatment is lowering blood pressure more than placebo, transgenic animals become obese, bio reactor A is more efficient than B, concentration of substance is correlated with speed of reaction ...
- Null hypothesis: This is what you test!
No difference / relation, BP under therapy = BP under placebo

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4 possibilities:

- Null hypothesis correct, test false positive (case A): alpha-error
- Null hypothesis correct, test correct negative (case B)
- Null hypothesis false, test false negative (case C): beta-error
- Null hypothesis false, test correct positive (case D)

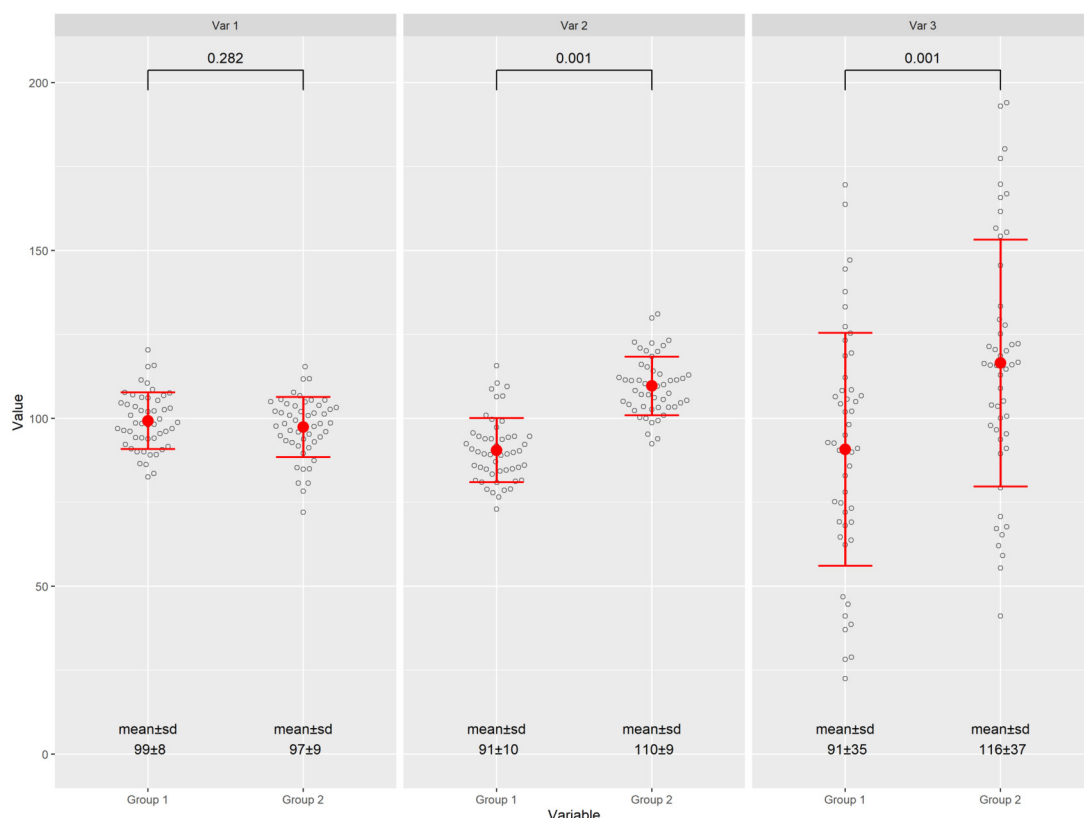
Significance: NOT probability of case A, but probability of your data given the NULL hypothesis, calculated from your data, conventionally <0.05

Power: Probability of case D, *estimated* based on assumptions about effects and sample size, *calculation* would require knowledge of true difference, conventionally set at 0.80

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Test functions

t-test / Wilcoxon-test (aka Mann-Whitney U-test)



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t-test

- Assumptions: Continuous data with Normal distribution
- 1 or 2 (independent or dependent) samples with/without equal variances
- how big is the mean difference relative to uncertainty?
 $t = (\text{mean}_1 - \text{mean}_2) / \text{SEM}$
- t follows a t-distribution, allows estimation of probability of t under the NULL hypothesis

Wilcoxon-test

- nonparametric, no distribution is assumed
- based on rank-transformed data
- insensitive to extreme values

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Test examples: *single variables*

```
1 #t-Test with test for equal variances
2 t_out <- t.test(formula='Moisture Content (%)'~'Region', data=rawdata,
3                 var.equal=var.test(
4                     formula='Moisture Content (%)'~'Region',
5                     data=rawdata)$p.value>.05)
6 t_out
```

Welch Two Sample t-test

```
data: Moisture Content (%) by Region
t = -1.7274, df = 29.239, p-value = 0.09465
alternative hypothesis: true difference in means between group A and group D is not equal to 0
95 percent confidence interval:
 -2.9624835  0.2490486
sample estimates:
mean in group A mean in group D
  90.31199      91.66870
```

```
1 #Wilcoxon-Test
2 wilcox.test('Moisture Content (%)'~'Region',
3             data = rawdata)
```

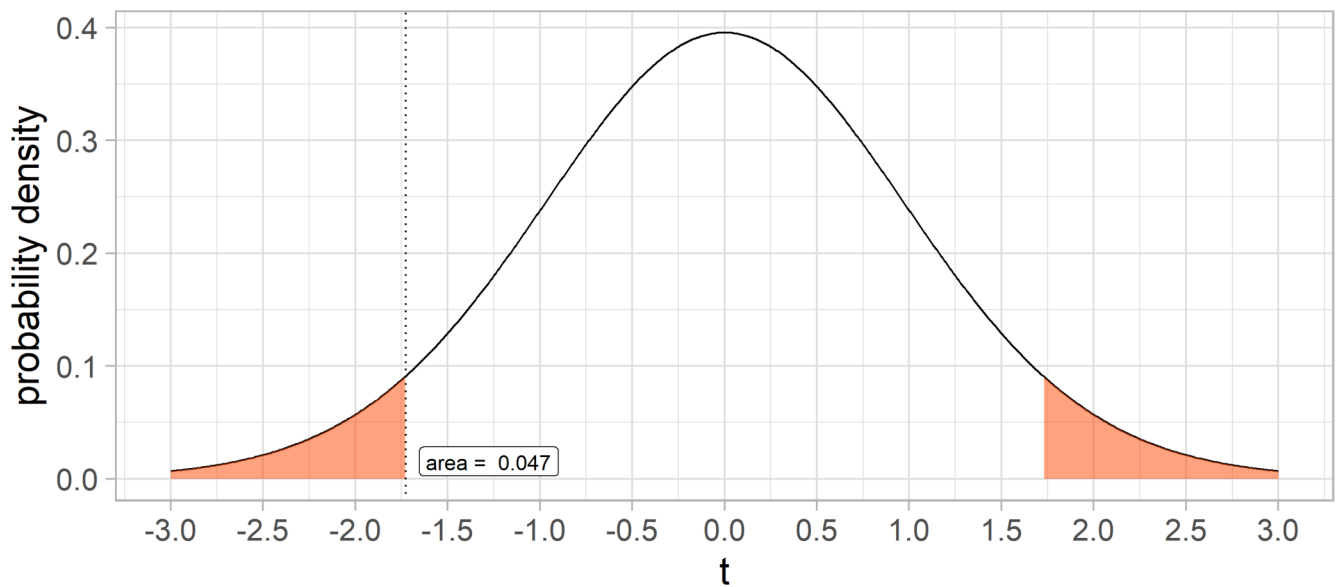
Wilcoxon rank sum exact test

```
data: Moisture Content (%) by Region
W = 107, p-value = 0.7547
alternative hypothesis: true location shift is not equal to 0
```

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From t to p

from t-test: $t = -1.7$, $p = 0.095$



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Model: Test 2 / multiple variables

```
1 test_gauss <- compare2numvars(data = rawdata,  
2                               dep_vars = gaussvars$names,  
3                               indep_var = 'Region',  
4                               gaussian = TRUE,  
5                               round_p = 5)  
6 test_gauss |> flextable()|>  
7   theme_zebra(even_body = 'aquamarine', odd_body = 'antiquewhite')
```

Variable	desc_all	Region A	Region D	p
Weight Of Sample Before Drying	5.1 ± 0.2	5.2 ± 0.1	5.0 ± 0.3	0.12830
Weight Of Sample After Drying	0.47 ± 0.14	0.50 ± 0.16	0.42 ± 0.07	0.04937
Moisture Content (%)	91 ± 3	90 ± 3	92 ± 1	0.09465
Dry Content (%)	9.2 ± 2.7	9.7 ± 3.1	8.3 ± 1.3	0.09465

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```

1 test_ord <- compare2numvars(data = rawdata,
2                             dep_vars = ordvars$names,
3                             indep_var = 'Region',
4                             gaussian = FALSE, round_desc = 3)
5 test_ord |> flectable() |>
6 theme_zebra(even_body = 'aquamarine', odd_body = 'antiquewhite')

```

Variable	desc_all	Region A	Region D	p
Weight Of Empty Cup	4.09 (4.06/4.19)	4.07 (4.06/4.09)	4.20 (4.19/4.21)	0.001
Weigh Of Cup + Sample	9.25 (9.22/9.29)	9.24 (9.22/9.25)	9.29 (9.27/9.31)	0.003
Weigh Of Cup + Sample After Drying	4.61 (4.45/4.72)	4.46 (4.42/4.74)	4.62 (4.58/4.66)	0.611

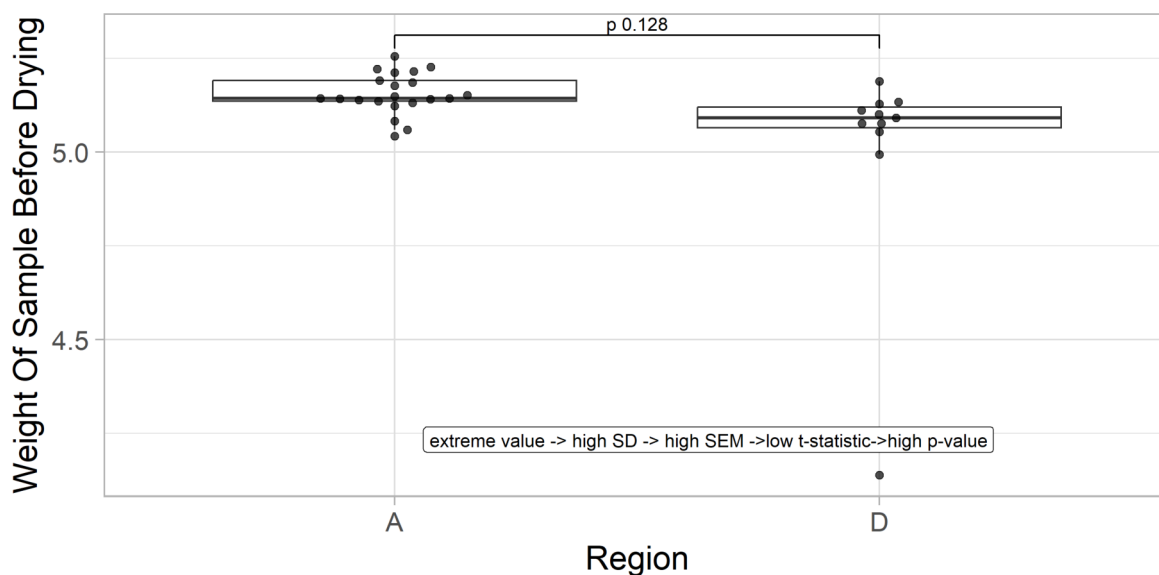
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Show results

```

1 ggplot(rawdata, aes(x = `Region`, y = `Weight Of Sample Before Drying`))+
2   geom_boxplot(outlier.alpha = 0)+
3   geom_beeswarm(alpha=.7, size=2, cex = 2)+
4   annotate(geom = 'label', x=2, y=4.2,
5            label='extreme value -> high SD -> high SEM ->low t-statistic->high p-value
6            hjust=0.8, vjust=0)+
7   geom_signif(comparisons = list(c(1,2)),
8               annotations = paste('p', formatP(t_out$p.value)))

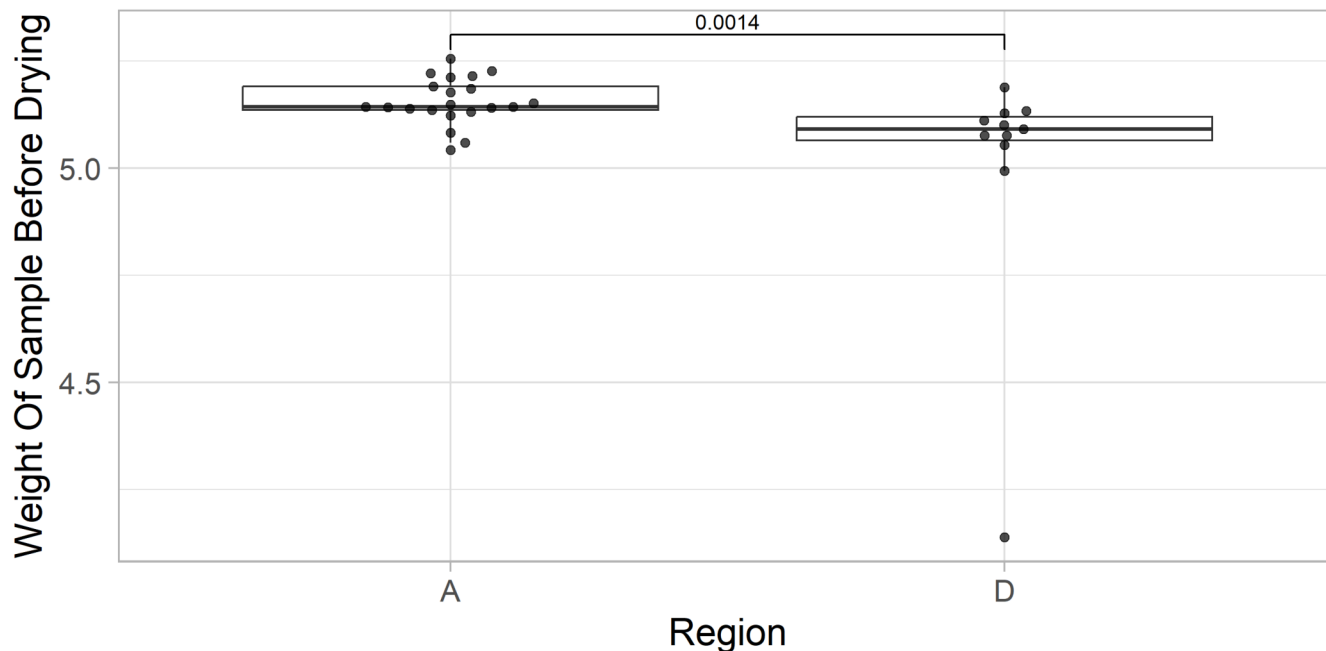
```



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Re-thinking test decision?

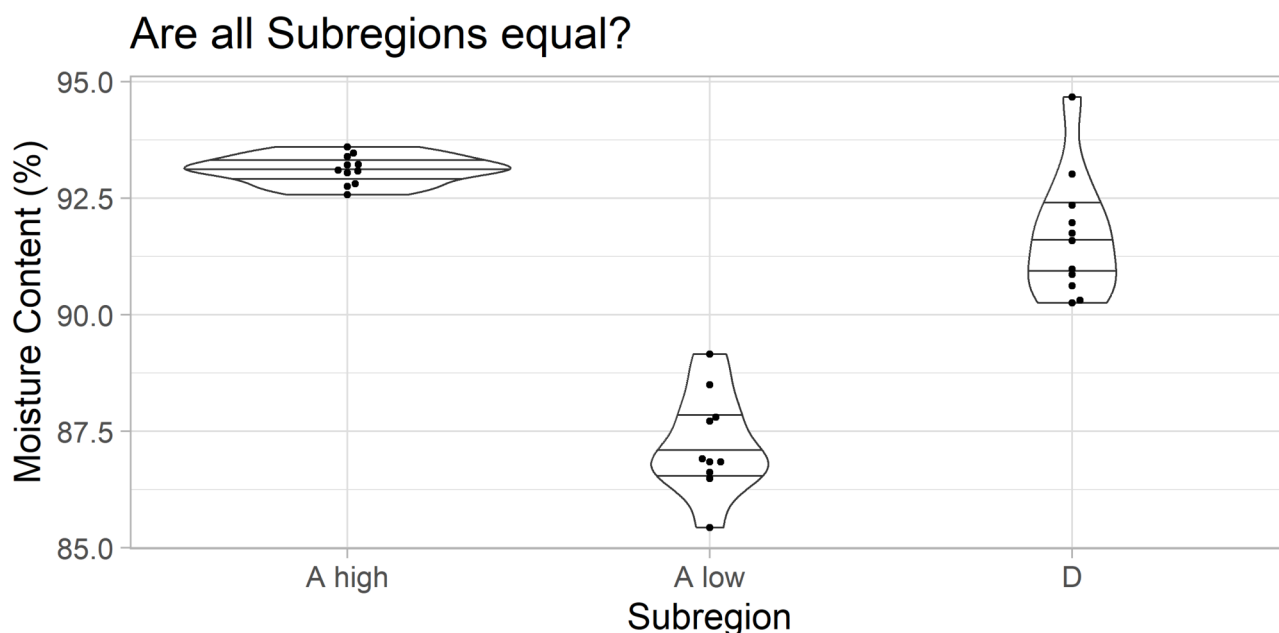
```
1 ggplot(rawdata, aes(x = `Region`, y = `Weight Of Sample Before Drying`))+  
2   geom_boxplot(outlier.alpha = 0)+  
3   geom_beeswarm(alpha=.7, size=2, cex = 2)+  
4   geom_signif(comparisons = list(c(1,2)), test = wilcox.test)
```



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Model: linear models 1 / univariable

```
1 plottmp <- ggplot(rawdata, aes(Subregion, `Moisture Content (%)`))+  
2   geom_violin(draw_quantiles = c(.25, .5, .75))+  
3   geom_beeswarm()+  
4  
5   ggtitle('Are all Subregions equal?')  
6   print(plottmp)
```



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ANOVA: build model

```
1 rawdata |> group_by(Subregion) |>
2   summarize(MeanMoisture=mean(`Moisture Content (%)`) |> roundR(4)) |>
3   pivot_wider(names_from = Subregion, values_from = MeanMoisture) |>
4   rename_with(~paste('Mean moisture %\n',.x)) |> flextable()|>
5   theme_zebra(even_body = 'aquamarine',odd_body = 'antiquewhite')
```

Mean moisture %	Mean moisture %	Mean moisture %
A high	A low	D
93.11	87.23	91.67

```
1 lm1<- lm(`Moisture Content (%)`~Subregion, data=rawdata)
2 lm1
```

Call:

```
lm(formula = `Moisture Content (%)` ~ Subregion, data = rawdata)
```

Coefficients:

(Intercept)	SubregionA low	SubregionD
93.112	-5.879	-1.443

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ANOVA: get p-values

```
1 anova(lm1) |> broom::tidy() |> flextable()|>
2   theme_zebra(even_body = 'aquamarine',odd_body = 'antiquewhite')
```

term	df	sumsq	meansq	statistic	p.value
Subregion	2	194.34239	97.1711969	97.76477	0.00000000000001292127
Residuals	29	28.82393	0.9939285		

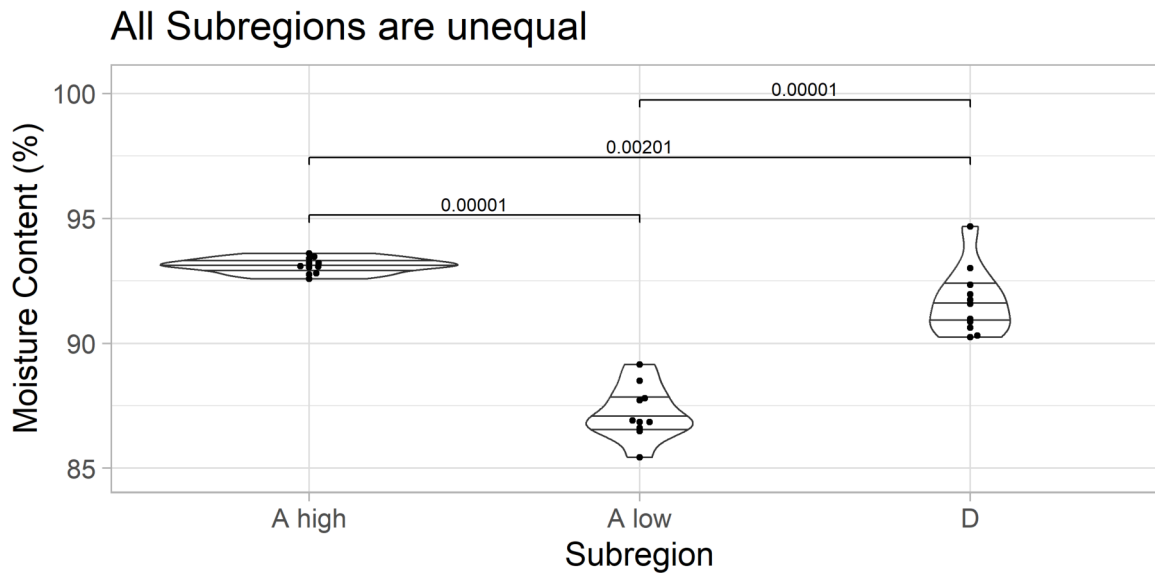
```
1 #post-hoc
2 (posthoc_out <- pairwise.t.test(x = rawdata$`Moisture Content (%)`,
3   g = rawdata$Subregion,
4   p.adjust.method = 'fdr')$p.value |>
5   formatP(ndigits = 5))
```

	A high	A low
A low	"0.00001"	"NA"
D	"0.00201"	"0.00001"

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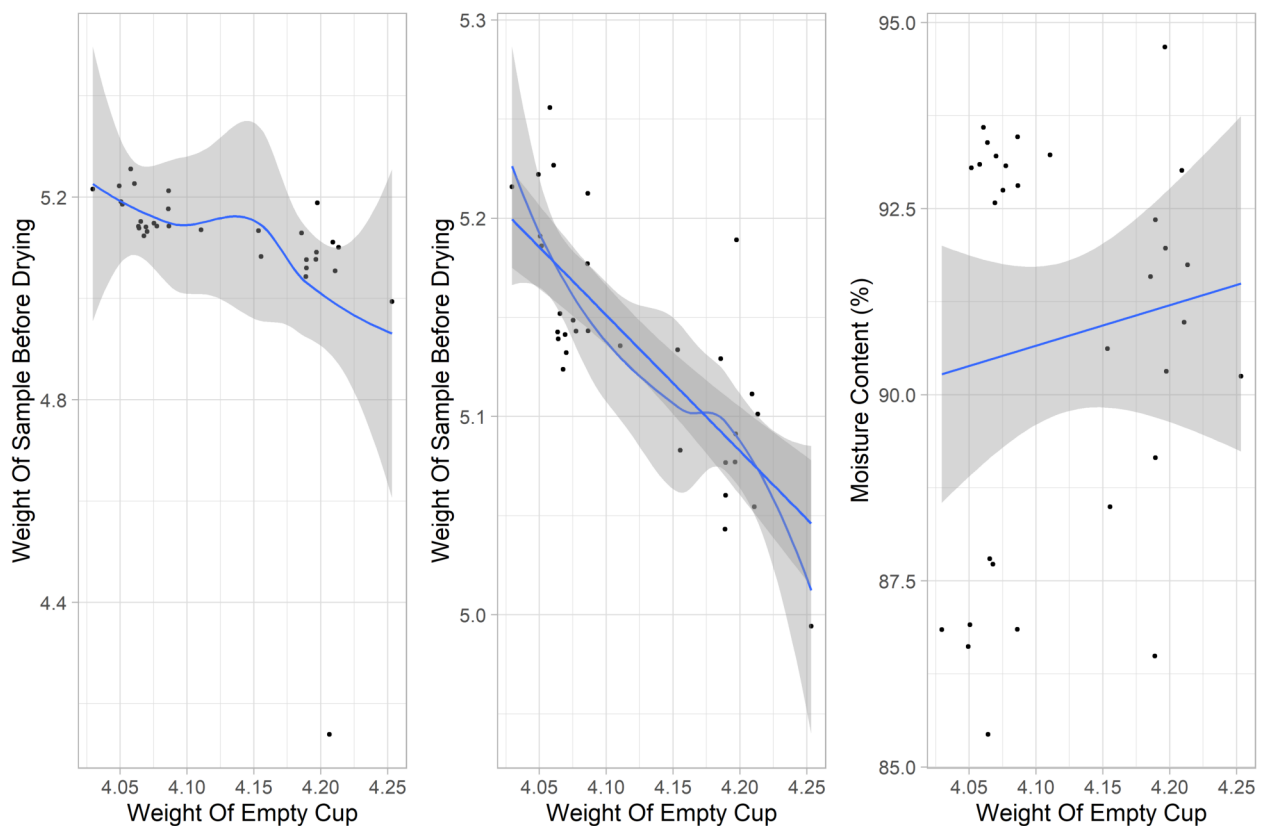
Visualize ANOVA

```
1 ggplot(rawdata, aes(Subregion, `Moisture Content (%)`)) +
2   geom_violin(draw_quantiles = c(.25, .5, .75)) +
3   geom_beeswarm() +
4   geom_signif(comparisons = list(c(1, 2), c(1, 3), c(2, 3)),
5               annotations = c(posthoc_out[, 1], posthoc_out[2, 2]),
6               step_increase = .25) +
7   scale_y_continuous(expand = expansion(mult = .1)) +
8   ggtitle('All Subregions are unequal')
```



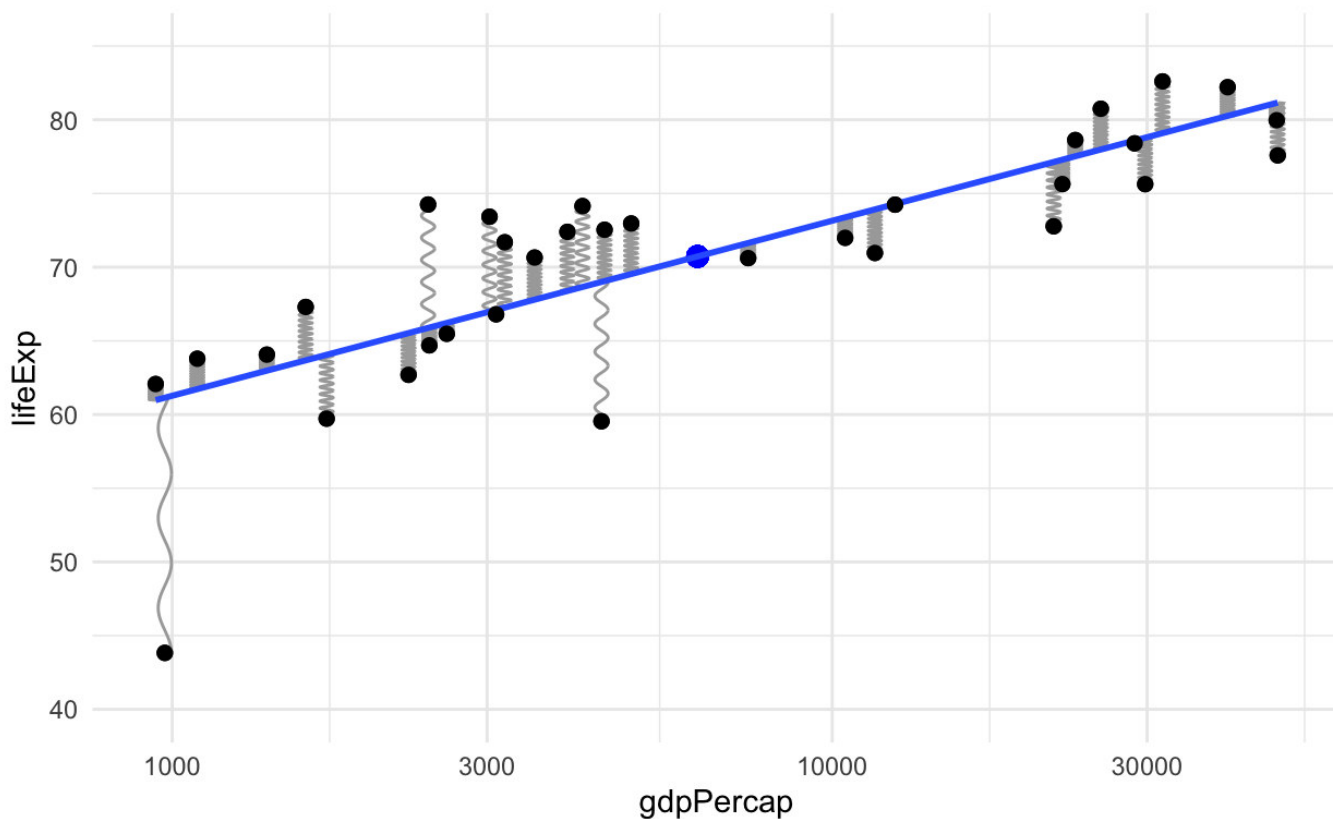
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Regression: Scatterplot



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Regression: Underlying *mechanics*



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Regression: Statistics

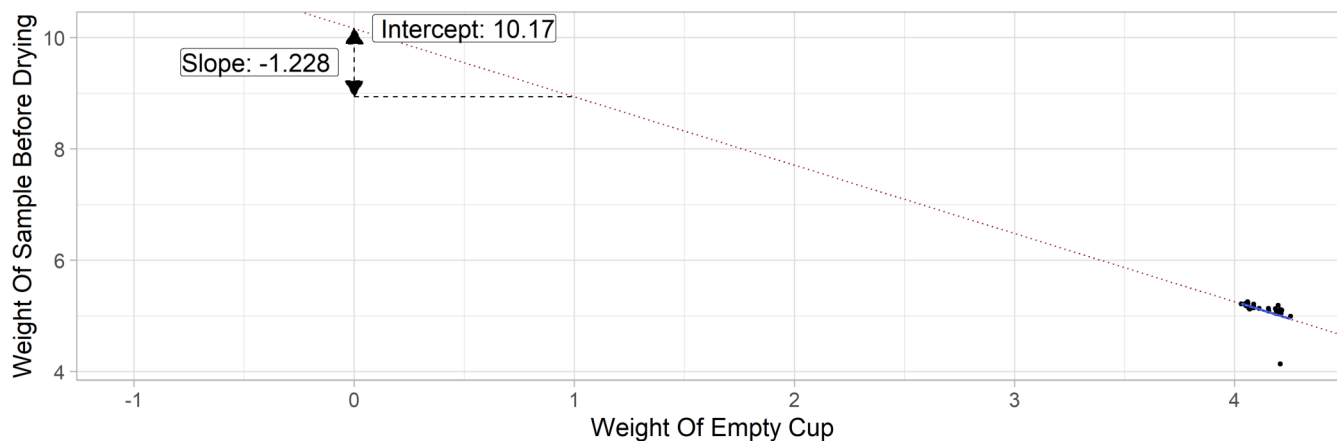
```
1 lm_out <- lm(`Weight Of Sample Before Drying` ~ `Weight Of Empty Cup`,
2             data=rawdata)
3 lm_out
```

Call:

```
lm(formula = `Weight Of Sample Before Drying` ~ `Weight Of Empty Cup`,
    data = rawdata)
```

Coefficients:

```
(Intercept)  `Weight Of Empty Cup`
      10.169             -1.228
```



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Regression: Significance

```
1 anova(lm_out) |> broom::tidy()
```

```
# A tibble: 2 × 6
```

	term <chr>	df <int>	sumsq <dbl>	meansq <dbl>	statistic <dbl>	p.value <dbl>
1	`Weight Of Empty Cup`	1	0.215	0.215	7.56	0.0100
2	Residuals	30	0.855	0.0285	NA	NA

```
1 model_parameters(lm_out)
```

Parameter	Coefficient	SE	95% CI	t(30)	p
(Intercept)	10.17	1.84	[6.41, 13.93]	5.52	< .001
Weight Of Empty Cup	-1.23	0.45	[-2.14, -0.32]	-2.75	0.010

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Report

- *RMarkdown and quarto are powerful tools to create reports and presentations*
- Export figures: `ggsave()` / `png()` / `pdf()`
- Export tables: `write_xlsx()`
- *Package flextable provides nice features for table formatting*

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Flextable example

```
1 test_ord |> select(-desc_all) |> rename_with(~str_remove(., 'Code Of ')) |>
2 flextable() |>
3 theme_zebra(even_body = 'aquamarine', odd_body = 'antiquewhite') |>
4 italic(~p<=0.05, j = 1) |> bg(~p<=0.05, j = 4, bg = 'yellow') |>
5 set_caption('Treatment effects, measures following a normal distribution') |>
6 add_footer_lines('Significance level is set at 0.05') |>
7 fontsize(size = 12, part = 'footer')
```

Variable	Region A	Region D	p
Weight Of Empty Cup	4.07 (4.06/4.09)	4.20 (4.19/4.21)	0.001
Weigth Of Cup + Sample	9.24 (9.22/9.25)	9.29 (9.27/9.31)	0.003
Weigth Of Cup + Sample After Drying	4.46 (4.42/4.74)	4.62 (4.58/4.66)	0.611

Significance level is set at 0.05

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Useful tools along the way

- Pick columns / rows: `select()` / `pull()` / `filter()` / `slice()`
- Change format of tibble wide <--> long (e.g. for repeated measures): `pivot_longer()`/`pivot_wider()`
- Regular expressions: `str_replace()` / `str_detect()` / `str_...`
- Merge text elements: `paste()` / `str_glue()`
- Apply functions: `purrr::map_xxx`

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