**Supplementary material S1 Testing the adsorption of FD4 to PS-NP**

**

*Figure S1:* Evaluation of the FD4 adsorption on PS-NP, by measuring FD4 fluorescence in the presence (dotted line- full circle; ⚫) and absence (solid line; ⭘) of NP (20 mg L-1, *i.e.* the highest concentration measured in the serous side). FD4+NP: y = 0.1853x - 2.4167; R2 = 0.9979; FD4: y = 0.1893x – 1.4333 ; R2 = 0.992.

Table S1: ANCOVA analyses testing the effect of treatment (FD4 or FD4 with NP added), and the concentration of FD4 show that within the range of 0 to 500 ng mL-1, fluorescence was not significantly changed by NP addition (F-value = 1.8857, p-value = 0.1814), whatever the FD4 concentration tested (F-value = 0.2779, p-value = 0.6026).

|  |
| --- |
| > anova(lm(Dataset3$Fluo~Dataset3$Test\*Dataset3$Conc) ) |
| Analysis of Variance Table |
|  |
| Response: Dataset3$Fluo |
| Df Sum Sq Mean Sq F value Pr(>F) |
| Dataset3$Test 1 18 18 1.8857 0.1814 |
| Dataset3$Conc 1 33010 33010 3429.9688 <2e-16 \*\*\* |
| Dataset3$Test:Dataset3$Conc 1 3 3 0.2779 0.6026 |
| Residuals 26 250 10 |
| --- |
| Signif. codes: 0 ‘\*\*\*’ 0.001 ‘\*\*’ 0.01 ‘\*’ 0.05 ‘.’ 0.1 ‘ ’ 1 |

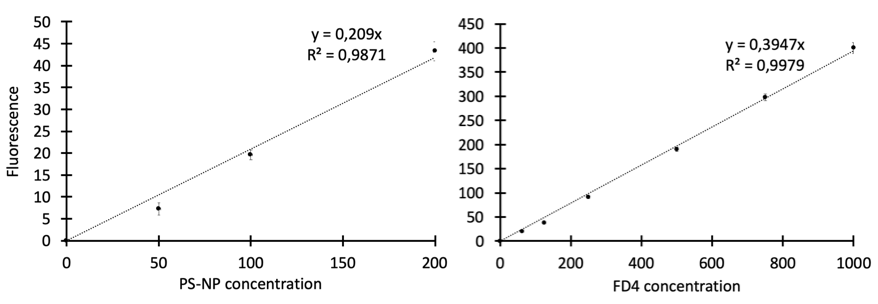
**Supplementary material S2: Control for leakage of dye by dialysis**

Table S2: Fluorescence of PS-NP measured in the total volume of each dialysis tube (n=5 tubes) at the PS-NP wavelength (360-460 nm), according to the methodology presented in Catarino et al. (2019). PS-NP fluorescence has been measured initially, and after 1:30 hour of incubation of PS-NP in each dialysis tubes. The difference between initial mean fluorescence and final fluorescence has been calculated for each tube and the mean calculated from the 5 tubes is indicated (in fluorescence and in percentage) in the third column. Data are presented as mean ± standard deviation of the mean.

|  |  |  |
| --- | --- | --- |
| Initial fluorescence (n=5) | Fluorescence after dialysis (n=5) | Difference (n=5) |
| 17 400.0 ± 4.0 | |  |  | | --- | --- | | |  | | --- | | 13 879.8 ± 1 273.9 | | | 3520.2 ± 1273.9  (20.2 % ± 7.3 %) |

**Supplementary material S3 : Calibration curves of PS-NP and FD4**

Fig. S2: Mean calibration curves of the PS-NP concentration (in mg L-1) and FD4 concentration (in ng mL-1). The PS-NP fluorescence was read at 360/460 nm (excitation/emission), and FD4 fluorescence was read at 485/528 nm (excitation/emission), with a gain of 50. The LOD for the PS-NP detection has been estimated to be 9.2 mg L-1 and that for FD4 is at 23.4 ng mL-1



**Supplementary material S4: Determination of the autofluorescence of the tissue**

Table S3: Measure of the autofluorescence of the tissue (n3 for the median and n=3 for the distal gut), ie. the fluorescence at the PS-NP wavelength (360/460 nm) and at the FD4 wavelength (485/528 nm) during 150 min, without any initial addition of substance. The concentrations measured are below the limit of detection which were estimated at 23.4 ng mL-1 for FD4 and 9.2 mg L-1 for PS-NP.

|  |  |  |
| --- | --- | --- |
| Median gut | FD4 concentration (ng mL-1) | PS-NP concentration (mg L-1) |
| 0 min | -63.34 ± 4.65 | -34.06 ± 4.78 |
| 30 min | -64.61 ± 7.76 | -38.93 ± 7.66 |
| 60 min | -68.41 ± 5.98 | -38.93 ± 4.77 |
| 150 min | -67.14 ± 6.57 | -34.06 ± 4.57 |
| Distal gut |  |  |
| 0 min | -72.21 ± 6.89 | -41.36 ± 3.66 |
| 30 min | -73.47 ± 8.97 | -43.80 ± 7.83 |
| 60 min | -72.21 ± 4.32 | -41.36 ± 9.49 |
| 150 min | -71.0 ± 6.89 | -38.93 ± 7.15 |

**Supplementary S5** **: Statistical analyses of the fluorescence dataset**

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# Treatment FD4

(n=14 median guts and n=15 distal guts)

## **1.1. Time effect, including both gut segments in one single analysis, and difference in FD4 concentration between gut segments**

### 1.1.1. Linear mixed effect model

*> datasetFD4<-subset(Dataset,Dataset$Treat=="FD4")*

*> Model2.lme<-lme(ConcFD4~Time2+Gut,datasetFD4,random=~1|NumFish)*

*> qqnorm(Model2.lme)*

*> summary(Model2.****lme)***

Linear mixed-effects model fit by REML

Data: datasetFD4

AIC BIC logLik

1797.213 1834.392 -887.6065

Random effects:

Formula: ~1 | NumFish

(Intercept) Residual

StdDev: 9.66619 12.5135

Fixed effects: ConcFD4 ~ Time2 + Gut

Value Std.Error DF t-value p-value

(Intercept) 7.956319 3.503617 203 2.2708874 0.0242

Time215 -2.572316 3.355243 203 -0.7666555 0.4442

Time230 -0.510422 3.286207 203 -0.1553225 0.8767

Time245 -2.053208 3.355243 203 -0.6119402 0.5413

Time260 -1.530103 3.286207 203 -0.4656137 0.6420

Time290 -2.069126 3.286207 203 -0.6296395 0.5296

Time2120 0.926490 3.286207 203 0.2819330 0.7783

Time2150 1.023563 3.354137 203 0.3051643 0.7606

GutMedian 2.700719 1.690010 203 1.5980494 0.1116

Correlation:

(Intr) Tim215 Tim230 Tim245 Tim260 Tim290 Tm2120 Tm2150

Time215 -0.460

Time230 -0.469 0.490

Time245 -0.460 0.485 0.490

Time260 -0.469 0.490 0.500 0.490

Time290 -0.469 0.490 0.500 0.490 0.500

Time2120 -0.469 0.490 0.500 0.490 0.500 0.500

Time2150 -0.460 0.479 0.490 0.479 0.490 0.490 0.490

GutMedian -0.226 0.000 0.000 0.000 0.000 0.000 0.000 0.000

Standardized Within-Group Residuals:

Min Q1 Med Q3 Max

-2.1485345 -0.5669434 -0.1591551 0.3215229 5.7102384

Number of Observations: 226

Number of Groups: 15

### 1.1.2. Pair-wise comparisons between sampling times

*> summary(glht(Model2.lme,linfct=mcp(Time2="Tukey")))*

Simultaneous Tests for General Linear Hypotheses

Multiple Comparisons of Means: Tukey Contrasts

Fit: lme.formula(fixed = ConcFD4 ~ Time2 + Gut, data = datasetFD4, random = ~1 | NumFish)

Linear Hypotheses:

Estimate Std. Error z value Pr(>|z|)

15 - 0 == 0 -2.57232 3.35524 -0.767 0.995

30 - 0 == 0 -0.51042 3.28621 -0.155 1.000

45 - 0 == 0 -2.05321 3.35524 -0.612 0.999

60 - 0 == 0 -1.53010 3.28621 -0.466 1.000

90 - 0 == 0 -2.06913 3.28621 -0.630 0.998

120 - 0 == 0 0.92649 3.28621 0.282 1.000

150 - 0 == 0 1.02356 3.35414 0.305 1.000

30 - 15 == 0 2.06189 3.35524 0.615 0.999

45 - 15 == 0 0.51911 3.40574 0.152 1.000

60 - 15 == 0 1.04221 3.35524 0.311 1.000

90 - 15 == 0 0.50319 3.35524 0.150 1.000

120 - 15 == 0 3.49881 3.35524 1.043 0.968

150 - 15 == 0 3.59588 3.42323 1.050 0.967

45 - 30 == 0 -1.54279 3.35524 -0.460 1.000

60 - 30 == 0 -1.01968 3.28621 -0.310 1.000

90 - 30 == 0 -1.55870 3.28621 -0.474 1.000

120 - 30 == 0 1.43691 3.28621 0.437 1.000

150 - 30 == 0 1.53398 3.35414 0.457 1.000

60 - 45 == 0 0.52310 3.35524 0.156 1.000

90 - 45 == 0 -0.01592 3.35524 -0.005 1.000

120 - 45 == 0 2.97970 3.35524 0.888 0.987

150 - 45 == 0 3.07677 3.42323 0.899 0.986

90 - 60 == 0 -0.53902 3.28621 -0.164 1.000

120 - 60 == 0 2.45659 3.28621 0.748 0.995

150 - 60 == 0 2.55367 3.35414 0.761 0.995

120 - 90 == 0 2.99562 3.28621 0.912 0.985

150 - 90 == 0 3.09269 3.35414 0.922 0.984

150 - 120 == 0 0.09707 3.35414 0.029 1.000

(Adjusted p values reported -- single-step method)

### 1.1.3. Pairwise comparisons between median and distal guts

*> summary(glht(Model2.lme,linfct=mcp(Gut="Tukey")))*

Simultaneous Tests for General Linear Hypotheses

Multiple Comparisons of Means: Tukey Contrasts

Fit: lme.formula(fixed = ConcFD4 ~ Time2 + Gut, data = datasetFD4, random = ~1 | NumFish)

Linear Hypotheses:

Estimate Std. Error z value Pr(>|z|)

Median - Distal == 0 2.701 1.690 1.598 0.11

(Adjusted p values reported -- single-step method)

## **1.2. Time effect concerning median segment**

### 1.2.1. Linear mixed effect model

> datasetFD4m<-subset(datasetFD4,datasetFD4$Gut=="Median")

> Model2m.lme<-lme(ConcFD4~Time2,datasetFD4m,random=~1|NumFish)

> qqnorm(Model2m.lme)

> summary(Model2m.lme)

Linear mixed-effects model fit by REML

Data: datasetFD4m

AIC BIC logLik

785.3047 811.4559 -382.6524

Random effects:

Formula: ~1 | NumFish

(Intercept) Residual

StdDev: 14.16424 7.810241

Fixed effects: ConcFD4 ~ Time2

Value Std.Error DF t-value p-value

(Intercept) 8.969244 4.322909 88 2.0748168 0.0409

Time215 -0.732879 3.017061 88 -0.2429116 0.8086

Time230 3.811467 2.951994 88 1.2911502 0.2000

Time245 1.281054 3.017061 88 0.4246033 0.6722

Time260 0.583951 2.951994 88 0.1978158 0.8436

Time290 0.275453 2.951994 88 0.0933109 0.9259

Time2120 2.929182 2.951994 88 0.9922725 0.3238

Time2150 3.153021 3.015861 88 1.0454796 0.2987

Correlation:

(Intr) Tim215 Tim230 Tim245 Tim260 Tim290 Tm2120

Time215 -0.334

Time230 -0.341 0.489

Time245 -0.334 0.485 0.489

Time260 -0.341 0.489 0.500 0.489

Time290 -0.341 0.489 0.500 0.489 0.500

Time2120 -0.341 0.489 0.500 0.489 0.500 0.500

Time2150 -0.334 0.478 0.489 0.478 0.489 0.489 0.489

Standardized Within-Group Residuals:

Min Q1 Med Q3 Max

-1.7264475 -0.4619005 -0.1081014 0.2479693 4.3404290

Number of Observations: 109

Number of Groups: 14

### 1.2.2. Pairwise comparisons between sample times concerning the median gut

*> summary(glht(Model2m.lme,linfct=mcp(Time2="Tukey")))*

Simultaneous Tests for General Linear Hypotheses

Multiple Comparisons of Means: Tukey Contrasts

Fit: lme.formula(fixed = ConcFD4 ~ Time2, data = datasetFD4m, random = ~1 | NumFish)

Linear Hypotheses:

Estimate Std. Error z value Pr(>|z|)

15 - 0 == 0 -0.7329 3.0171 -0.243 1.000

30 - 0 == 0 3.8115 2.9520 1.291 0.902

45 - 0 == 0 1.2811 3.0171 0.425 1.000

60 - 0 == 0 0.5840 2.9520 0.198 1.000

90 - 0 == 0 0.2755 2.9520 0.093 1.000

120 - 0 == 0 2.9292 2.9520 0.992 0.976

150 - 0 == 0 3.1530 3.0159 1.045 0.967

30 - 15 == 0 4.5443 3.0171 1.506 0.804

45 - 15 == 0 2.0139 3.0634 0.657 0.998

60 - 15 == 0 1.3168 3.0171 0.436 1.000

90 - 15 == 0 1.0083 3.0171 0.334 1.000

120 - 15 == 0 3.6621 3.0171 1.214 0.928

150 - 15 == 0 3.8859 3.0811 1.261 0.913

45 - 30 == 0 -2.5304 3.0171 -0.839 0.991

60 - 30 == 0 -3.2275 2.9520 -1.093 0.958

90 - 30 == 0 -3.5360 2.9520 -1.198 0.933

120 - 30 == 0 -0.8823 2.9520 -0.299 1.000

150 - 30 == 0 -0.6584 3.0159 -0.218 1.000

60 - 45 == 0 -0.6971 3.0171 -0.231 1.000

90 - 45 == 0 -1.0056 3.0171 -0.333 1.000

120 - 45 == 0 1.6481 3.0171 0.546 0.999

150 - 45 == 0 1.8720 3.0811 0.608 0.999

90 - 60 == 0 -0.3085 2.9520 -0.105 1.000

120 - 60 == 0 2.3452 2.9520 0.794 0.993

150 - 60 == 0 2.5691 3.0159 0.852 0.990

120 - 90 == 0 2.6537 2.9520 0.899 0.986

150 - 90 == 0 2.8776 3.0159 0.954 0.981

150 - 120 == 0 0.2238 3.0159 0.074 1.000

(Adjusted p values reported -- single-step method)

## **1.3. Time effect concerning the distal segment**

### 1.3.1. Linear mixed model

> datasetFD4d<-subset(datasetFD4,datasetFD4$Gut=="Distal")

> Model2d.lme<-lme(ConcFD4~Time2,datasetFD4d,random=~1|NumFish)

> qqnorm(Model2d.lme)

> summary(Model2d.lme)

Linear mixed-effects model fit by REML

Data: datasetFD4d

AIC BIC logLik

889.222 916.1355 -434.611

Random effects:

Formula: ~1 | NumFish

(Intercept) Residual

StdDev: 12.03534 10.07297

Fixed effects: ConcFD4 ~ Time2

Value Std.Error DF t-value p-value

(Intercept) 9.946499 4.052275 95 2.4545469 0.0159

Time215 -4.252470 3.752860 95 -1.1331278 0.2600

Time230 -4.544185 3.678127 95 -1.2354616 0.2197

Time245 -5.121415 3.752860 95 -1.3646699 0.1756

Time260 -3.503221 3.678127 95 -0.9524468 0.3433

Time290 -4.257400 3.678127 95 -1.1574912 0.2500

Time2120 -0.942689 3.678127 95 -0.2562959 0.7983

Time2150 -0.988217 3.751618 95 -0.2634108 0.7928

Correlation:

(Intr) Tim215 Tim230 Tim245 Tim260 Tim290 Tm2120

Time215 -0.445

Time230 -0.454 0.490

Time245 -0.445 0.485 0.490

Time260 -0.454 0.490 0.500 0.490

Time290 -0.454 0.490 0.500 0.490 0.500

Time2120 -0.454 0.490 0.500 0.490 0.500 0.500

Time2150 -0.445 0.480 0.490 0.480 0.490 0.490 0.490

Standardized Within-Group Residuals:

Min Q1 Med Q3 Max

-1.979555893 -0.366115935 -0.007029823 0.204568990 6.559398463

Number of Observations: 117

Number of Groups: 15

### 1.3.2. Pairwise comparisons between sample times concerning the distal gut

*> summary(glht(Model2d.lme,linfct=mcp(Time2="Tukey")))*

Simultaneous Tests for General Linear Hypotheses

Multiple Comparisons of Means: Tukey Contrasts

Fit: lme.formula(fixed = ConcFD4 ~ Time2, data = datasetFD4d, random = ~1 | NumFish)

Linear Hypotheses:

Estimate Std. Error z value Pr(>|z|)

15 - 0 == 0 -4.25247 3.75286 -1.133 0.950

30 - 0 == 0 -4.54418 3.67813 -1.235 0.921

45 - 0 == 0 -5.12141 3.75286 -1.365 0.873

60 - 0 == 0 -3.50322 3.67813 -0.952 0.981

90 - 0 == 0 -4.25740 3.67813 -1.157 0.944

120 - 0 == 0 -0.94269 3.67813 -0.256 1.000

150 - 0 == 0 -0.98822 3.75162 -0.263 1.000

30 - 15 == 0 -0.29172 3.75286 -0.078 1.000

45 - 15 == 0 -0.86894 3.80722 -0.228 1.000

60 - 15 == 0 0.74925 3.75286 0.200 1.000

90 - 15 == 0 -0.00493 3.75286 -0.001 1.000

120 - 15 == 0 3.30978 3.75286 0.882 0.988

150 - 15 == 0 3.26425 3.82644 0.853 0.990

45 - 30 == 0 -0.57723 3.75286 -0.154 1.000

60 - 30 == 0 1.04096 3.67813 0.283 1.000

90 - 30 == 0 0.28679 3.67813 0.078 1.000

120 - 30 == 0 3.60150 3.67813 0.979 0.977

150 - 30 == 0 3.55597 3.75162 0.948 0.981

60 - 45 == 0 1.61819 3.75286 0.431 1.000

90 - 45 == 0 0.86401 3.75286 0.230 1.000

120 - 45 == 0 4.17873 3.75286 1.113 0.954

150 - 45 == 0 4.13320 3.82644 1.080 0.961

90 - 60 == 0 -0.75418 3.67813 -0.205 1.000

120 - 60 == 0 2.56053 3.67813 0.696 0.997

150 - 60 == 0 2.51500 3.75162 0.670 0.998

120 - 90 == 0 3.31471 3.67813 0.901 0.986

150 - 90 == 0 3.26918 3.75162 0.871 0.989

150 - 120 == 0 -0.04553 3.75162 -0.012 1.000

(Adjusted p values reported -- single-step method)

# Treatment: FD4+NP 20

(n=6 per gut)

## 

## **2.1. Time effect, including both gut segments in one single analysis, and difference in FD4 concentration between gut segments**

### 2.1.1. Linear mixed model

> datasetFD4Nano20<-subset(Dataset,Dataset$Treat=="FD4Nano20")

>Model3.lme<-lme(ConcFD4~Time2+Gut,datasetFD4Nano20,random=~1|NumFish)

> qqnorm(Model3.lme)

> summary(Model3.lme)

Linear mixed-effects model fit by REML

Data: datasetFD4Nano20

AIC BIC logLik

794.729 821.3362 -386.3645

Random effects:

Formula: ~1 | NumFish

(Intercept) Residual

StdDev: 2.140598 22.10041

Fixed effects: ConcFD4 ~ Time2 + Gut

Value Std.Error DF t-value p-value

(Intercept) -9.903003 6.839228 78 -1.447971 0.1516

Time215 0.013772 9.470082 78 0.001454 0.9988

Time230 -0.579549 9.022455 78 -0.064234 0.9489

Time245 0.110649 9.470082 78 0.011684 0.9907

Time260 6.720807 9.022455 78 0.744898 0.4586

Time290 3.805355 9.022455 78 0.421765 0.6744

Time2120 2.717655 9.022455 78 0.301210 0.7641

Time2150 5.984875 9.022455 78 0.663331 0.5091

GutMedian 16.837797 4.608254 78 3.653834 0.0005

Correlation:

(Intr) Tim215 Tim230 Tim245 Tim260 Tim290 Tm2120 Tm2150

Time215 -0.628

Time230 -0.660 0.476

Time245 -0.628 0.455 0.476

Time260 -0.660 0.476 0.500 0.476

Time290 -0.660 0.476 0.500 0.476 0.500

Time2120 -0.660 0.476 0.500 0.476 0.500 0.500

Time2150 -0.660 0.476 0.500 0.476 0.500 0.500 0.500

GutMedian -0.337 0.000 0.000 0.000 0.000 0.000 0.000 0.000

Standardized Within-Group Residuals:

Min Q1 Med Q3 Max

-1.93419480 -0.63437666 0.08322873 0.40600243 2.21769191

Number of Observations: 92

Number of Groups: 6

### 2.1.2. Pairwise comparisons concerning the sampling times

*> summary(glht(Model3.lme,linfct=mcp(Time2="Tukey")))*

Simultaneous Tests for General Linear Hypotheses

Multiple Comparisons of Means: Tukey Contrasts

Fit: lme.formula(fixed = ConcFD4 ~ Time2 + Gut, data = datasetFD4Nano20, random = ~1 | NumFish)

Linear Hypotheses:

Estimate Std. Error z value Pr(>|z|)

15 - 0 == 0 0.01377 9.47008 0.001 1.000

30 - 0 == 0 -0.57955 9.02246 -0.064 1.000

45 - 0 == 0 0.11065 9.47008 0.012 1.000

60 - 0 == 0 6.72081 9.02246 0.745 0.996

90 - 0 == 0 3.80536 9.02246 0.422 1.000

120 - 0 == 0 2.71765 9.02246 0.301 1.000

150 - 0 == 0 5.98488 9.02246 0.663 0.998

30 - 15 == 0 -0.59332 9.47008 -0.063 1.000

45 - 15 == 0 0.09688 9.88360 0.010 1.000

60 - 15 == 0 6.70704 9.47008 0.708 0.997

90 - 15 == 0 3.79158 9.47008 0.400 1.000

120 - 15 == 0 2.70388 9.47008 0.286 1.000

150 - 15 == 0 5.97110 9.47008 0.631 0.998

45 - 30 == 0 0.69020 9.47008 0.073 1.000

60 - 30 == 0 7.30036 9.02246 0.809 0.993

90 - 30 == 0 4.38490 9.02246 0.486 1.000

120 - 30 == 0 3.29720 9.02246 0.365 1.000

150 - 30 == 0 6.56442 9.02246 0.728 0.996

60 - 45 == 0 6.61016 9.47008 0.698 0.997

90 - 45 == 0 3.69471 9.47008 0.390 1.000

120 - 45 == 0 2.60701 9.47008 0.275 1.000

150 - 45 == 0 5.87423 9.47008 0.620 0.999

90 - 60 == 0 -2.91545 9.02246 -0.323 1.000

120 - 60 == 0 -4.00315 9.02246 -0.444 1.000

150 - 60 == 0 -0.73593 9.02246 -0.082 1.000

120 - 90 == 0 -1.08770 9.02246 -0.121 1.000

150 - 90 == 0 2.17952 9.02246 0.242 1.000

150 - 120 == 0 3.26722 9.02246 0.362 1.000

(Adjusted p values reported -- single-step method)

### 2.1.3. Pairwise comparisons concerning the gut segment

*> summary(glht(Model3.lme,linfct=mcp(Gut="Tukey")))*

Simultaneous Tests for General Linear Hypotheses

Multiple Comparisons of Means: Tukey Contrasts

Fit: lme.formula(fixed = ConcFD4 ~ Time2 + Gut, data = datasetFD4Nano20, random = ~1 | NumFish)

Linear Hypotheses:

Estimate Std. Error z value Pr(>|z|)

Median - Distal == 0 16.838 4.608 3.654 0.000258 \*\*\*

---

Signif. codes: 0 ‘\*\*\*’ 0.001 ‘\*\*’ 0.01 ‘\*’ 0.05 ‘.’ 0.1 ‘ ’ 1

(Adjusted p values reported -- single-step method)

## **2.2. Time effect concerning the median segment**

### 2.2.1. Linear mixed model

*>*  *datasetFD4Nano20m<-subset(datasetFD4Nano20,datasetFD4Nano20$Gut=="Median")*

*> Model3m.lme<-lme(ConcFD4~Time2,datasetFD4Nano20m,random=~1|NumFish)*

*> qqnorm(Model3m.lme)*

*> summary(Model3m.lme)*

Linear mixed-effects model fit by REML

Data: datasetFD4Nano20m

AIC BIC logLik

348.6635 365.0394 -164.3318

Random effects:

Formula: ~1 | NumFish

(Intercept) Residual

StdDev: 23.72097 12.16577

Fixed effects: ConcFD4 ~ Time2

Value Std.Error DF t-value p-value

(Intercept) 6.947185 10.883402 33 0.6383285 0.5277

Time215 3.787840 7.420012 33 0.5104897 0.6131

Time230 -1.865953 7.023911 33 -0.2656572 0.7922

Time245 4.973170 7.420012 33 0.6702374 0.5074

Time260 6.689040 7.023911 33 0.9523242 0.3479

Time290 5.023709 7.023911 33 0.7152295 0.4795

Time2120 3.547136 7.023911 33 0.5050087 0.6169

Time2150 5.626835 7.023911 33 0.8010971 0.4288

Correlation:

(Intr) Tim215 Tim230 Tim245 Tim260 Tim290 Tm2120

Time215 -0.305

Time230 -0.323 0.473

Time245 -0.305 0.462 0.473

Time260 -0.323 0.473 0.500 0.473

Time290 -0.323 0.473 0.500 0.473 0.500

Time2120 -0.323 0.473 0.500 0.473 0.500 0.500

Time2150 -0.323 0.473 0.500 0.473 0.500 0.500 0.500

Standardized Within-Group Residuals:

Min Q1 Med Q3 Max

-2.90828483 -0.37473849 -0.04704859 0.34826700 1.85338207

Number of Observations: 46

Number of Groups: 6

### 2.2.2. Pairwise comparisons of sampling times concerning the median gut

*> summary(glht(Model3m.lme,linfct=mcp(Time2="Tukey")))*

Simultaneous Tests for General Linear Hypotheses

Multiple Comparisons of Means: Tukey Contrasts

Fit: lme.formula(fixed = ConcFD4 ~ Time2, data = datasetFD4Nano20m, random = ~1 | NumFish)

Linear Hypotheses:

Estimate Std. Error z value Pr(>|z|)

15 - 0 == 0 3.78784 7.42001 0.510 1.000

30 - 0 == 0 -1.86595 7.02391 -0.266 1.000

45 - 0 == 0 4.97317 7.42001 0.670 0.998

60 - 0 == 0 6.68904 7.02391 0.952 0.981

90 - 0 == 0 5.02371 7.02391 0.715 0.997

120 - 0 == 0 3.54714 7.02391 0.505 1.000

150 - 0 == 0 5.62683 7.02391 0.801 0.993

30 - 15 == 0 -5.65379 7.42001 -0.762 0.995

45 - 15 == 0 1.18533 7.69431 0.154 1.000

60 - 15 == 0 2.90120 7.42001 0.391 1.000

90 - 15 == 0 1.23587 7.42001 0.167 1.000

120 - 15 == 0 -0.24070 7.42001 -0.032 1.000

150 - 15 == 0 1.83900 7.42001 0.248 1.000

45 - 30 == 0 6.83912 7.42001 0.922 0.984

60 - 30 == 0 8.55499 7.02391 1.218 0.927

90 - 30 == 0 6.88966 7.02391 0.981 0.977

120 - 30 == 0 5.41309 7.02391 0.771 0.995

150 - 30 == 0 7.49279 7.02391 1.067 0.964

60 - 45 == 0 1.71587 7.42001 0.231 1.000

90 - 45 == 0 0.05054 7.42001 0.007 1.000

120 - 45 == 0 -1.42603 7.42001 -0.192 1.000

150 - 45 == 0 0.65367 7.42001 0.088 1.000

90 - 60 == 0 -1.66533 7.02391 -0.237 1.000

120 - 60 == 0 -3.14190 7.02391 -0.447 1.000

150 - 60 == 0 -1.06221 7.02391 -0.151 1.000

120 - 90 == 0 -1.47657 7.02391 -0.210 1.000

150 - 90 == 0 0.60313 7.02391 0.086 1.000

150 - 120 == 0 2.07970 7.02391 0.296 1.000

(Adjusted p values reported -- single-step method)

## **2.3. Time effect concerning the distal segment**

### 2.3.1. Linear mixed effect model

>datasetFD4Nano20d<-subset(datasetFD4Nano20,datasetFD4Nano20$Gut=="Distal")

>Model3d.lme<-lme(ConcFD4~Time2,datasetFD4Nano20d,random=~1|NumFish)

> qqnorm(Model3d.lme)

> summary(Model3d.lme)

Linear mixed-effects model fit by REML

Data: datasetFD4Nano20d

AIC BIC logLik

288.7249 305.1007 -134.3624

Random effects:

Formula: ~1 | NumFish

(Intercept) Residual

StdDev: 18.18177 5.117027

Fixed effects: ConcFD4 ~ Time2

Value Std.Error DF t-value p-value

(Intercept) -9.915394 7.711041 33 -1.2858697 0.2074

Time215 1.276064 3.121596 33 0.4087859 0.6853

Time230 0.706855 2.954317 33 0.2392617 0.8124

Time245 0.284489 3.121596 33 0.0911358 0.9279

Time260 6.752574 2.954317 33 2.2856635 0.0288

Time290 2.587002 2.954317 33 0.8756683 0.3875

Time2120 1.888174 2.954317 33 0.6391236 0.5272

Time2150 6.342916 2.954317 33 2.1469991 0.0392

Correlation:

(Intr) Tim215 Tim230 Tim245 Tim260 Tim290 Tm2120

Time215 -0.181

Time230 -0.192 0.473

Time245 -0.181 0.463 0.473

Time260 -0.192 0.473 0.500 0.473

Time290 -0.192 0.473 0.500 0.473 0.500

Time2120 -0.192 0.473 0.500 0.473 0.500 0.500

Time2150 -0.192 0.473 0.500 0.473 0.500 0.500 0.500

Standardized Within-Group Residuals:

Min Q1 Med Q3 Max

-1.43971546 -0.39533164 0.07654207 0.32072966 3.99627156

Number of Observations: 46

Number of Groups: 6

### 2.3.2. Pairwise comparisons concerning the sampling time for the distal gut

*> summary(glht(Model3d.lme,linfct=mcp(Time2="Tukey")))*

Simultaneous Tests for General Linear Hypotheses

Multiple Comparisons of Means: Tukey Contrasts

Fit: lme.formula(fixed = ConcFD4 ~ Time2, data = datasetFD4Nano20d, random = ~1 | NumFish)

Linear Hypotheses:

Estimate Std. Error z value Pr(>|z|)

15 - 0 == 0 1.2761 3.1216 0.409 1.000

30 - 0 == 0 0.7069 2.9543 0.239 1.000

45 - 0 == 0 0.2845 3.1216 0.091 1.000

60 - 0 == 0 6.7526 2.9543 2.286 0.301

90 - 0 == 0 2.5870 2.9543 0.876 0.988

120 - 0 == 0 1.8882 2.9543 0.639 0.998

150 - 0 == 0 6.3429 2.9543 2.147 0.384

30 - 15 == 0 -0.5692 3.1216 -0.182 1.000

45 - 15 == 0 -0.9916 3.2363 -0.306 1.000

60 - 15 == 0 5.4765 3.1216 1.754 0.651

90 - 15 == 0 1.3109 3.1216 0.420 1.000

120 - 15 == 0 0.6121 3.1216 0.196 1.000

150 - 15 == 0 5.0669 3.1216 1.623 0.736

45 - 30 == 0 -0.4224 3.1216 -0.135 1.000

60 - 30 == 0 6.0457 2.9543 2.046 0.450

90 - 30 == 0 1.8801 2.9543 0.636 0.998

120 - 30 == 0 1.1813 2.9543 0.400 1.000

150 - 30 == 0 5.6361 2.9543 1.908 0.545

60 - 45 == 0 6.4681 3.1216 2.072 0.433

90 - 45 == 0 2.3025 3.1216 0.738 0.996

120 - 45 == 0 1.6037 3.1216 0.514 1.000

150 - 45 == 0 6.0584 3.1216 1.941 0.522

90 - 60 == 0 -4.1656 2.9543 -1.410 0.853

120 - 60 == 0 -4.8644 2.9543 -1.647 0.721

150 - 60 == 0 -0.4097 2.9543 -0.139 1.000

120 - 90 == 0 -0.6988 2.9543 -0.237 1.000

150 - 90 == 0 3.7559 2.9543 1.271 0.909

150 - 120 == 0 4.4547 2.9543 1.508 0.803

(Adjusted p values reported -- single-step method)

# Treatment: FD4+NP 200

(n=7 per gut)

## **3.1. Time effect, including both gut segments in one single analysis, and difference in FD4 concentration between gut segments**

### 3.1.1. Linear mixed effect models

>datasetFD4Nano200<-subset(Dataset,Dataset$Treat=="FD4Nano200")

>Model3.lme<-lme(ConcFD4~Time2+Gut,datasetFD4Nano200,random=~1|NumFish)

> qqnorm(Model3.lme)

> summary(Model3.lme)

Linear mixed-effects model fit by REML

Data: datasetFD4Nano200

AIC BIC logLik

849.4872 877.9219 -413.7436

Random effects:

Formula: ~1 | NumFish

(Intercept) Residual

StdDev: 10.60079 13.58864

Fixed effects: ConcFD4 ~ Time2 + Gut

Value Std.Error DF t-value p-value

(Intercept) 5.043724 5.566190 92 0.9061358 0.3672

Time215 7.578521 5.136023 92 1.4755620 0.1435

Time230 -0.173212 5.136023 92 -0.0337250 0.9732

Time245 0.101644 5.241281 92 0.0193929 0.9846

Time260 0.953745 5.241281 92 0.1819679 0.8560

Time290 3.245271 5.241281 92 0.6191752 0.5373

Time2120 15.185851 5.136023 92 2.9567333 0.0040

Time2150 7.774185 5.372123 92 1.4471345 0.1513

GutMedian 3.286552 2.637636 92 1.2460218 0.2159

Correlation:

(Intr) Tim215 Tim230 Tim245 Tim260 Tim290 Tm2120 Tm2150

Time215 -0.461

Time230 -0.461 0.500

Time245 -0.458 0.490 0.490

Time260 -0.458 0.490 0.490 0.483

Time290 -0.458 0.490 0.490 0.483 0.483

Time2120 -0.461 0.500 0.500 0.490 0.490 0.490

Time2150 -0.441 0.478 0.478 0.467 0.467 0.467 0.478

GutMedian -0.237 0.000 0.000 0.023 0.023 0.023 0.000 -0.001

Standardized Within-Group Residuals:

Min Q1 Med Q3 Max

-1.28119008 -0.51323177 -0.07486702 0.26495566 5.74321830

Number of Observations: 107

Number of Groups: 7

### 3.1.2. Pairwise comparisons concerning the sampling times

*> summary(glht(Model3.lme,linfct=mcp(Time2="Tukey")))*

Simultaneous Tests for General Linear Hypotheses

Multiple Comparisons of Means: Tukey Contrasts

Fit: lme.formula(fixed = ConcFD4 ~ Time2 + Gut, data = datasetFD4Nano200, random = ~1 | NumFish)

Linear Hypotheses:

Estimate Std. Error z value Pr(>|z|)

15 - 0 == 0 7.5785 5.1360 1.476 0.8206

30 - 0 == 0 -0.1732 5.1360 -0.034 1.0000

45 - 0 == 0 0.1016 5.2413 0.019 1.0000

60 - 0 == 0 0.9537 5.2413 0.182 1.0000

90 - 0 == 0 3.2453 5.2413 0.619 0.9986

120 - 0 == 0 15.1859 5.1360 2.957 0.0621 .

150 - 0 == 0 7.7742 5.3721 1.447 0.8350

30 - 15 == 0 -7.7517 5.1360 -1.509 0.8027

45 - 15 == 0 -7.4769 5.2413 -1.427 0.8451

60 - 15 == 0 -6.6248 5.2413 -1.264 0.9119

90 - 15 == 0 -4.3332 5.2413 -0.827 0.9916

120 - 15 == 0 7.6073 5.1360 1.481 0.8178

150 - 15 == 0 0.1957 5.3721 0.036 1.0000

45 - 30 == 0 0.2749 5.2413 0.052 1.0000

60 - 30 == 0 1.1270 5.2413 0.215 1.0000

90 - 30 == 0 3.4185 5.2413 0.652 0.9981

120 - 30 == 0 15.3591 5.1360 2.990 0.0566 .

150 - 30 == 0 7.9474 5.3721 1.479 0.8187

60 - 45 == 0 0.8521 5.3299 0.160 1.0000

90 - 45 == 0 3.1436 5.3299 0.590 0.9990

120 - 45 == 0 15.0842 5.2413 2.878 0.0768 .

150 - 45 == 0 7.6725 5.4777 1.401 0.8574

90 - 60 == 0 2.2915 5.3299 0.430 0.9999

120 - 60 == 0 14.2321 5.2413 2.715 0.1176

150 - 60 == 0 6.8204 5.4777 1.245 0.9183

120 - 90 == 0 11.9406 5.2413 2.278 0.3051

150 - 90 == 0 4.5289 5.4777 0.827 0.9916

150 - 120 == 0 -7.4117 5.3721 -1.380 0.8667

---

Signif. codes: 0 ‘\*\*\*’ 0.001 ‘\*\*’ 0.01 ‘\*’ 0.05 ‘.’ 0.1 ‘ ’ 1

(Adjusted p values reported -- single-step method)

### 3.1.3. Pairwise comparisons concerning the gut segments

*> summary(glht(Model3.lme,linfct=mcp(Gut="Tukey")))*

Simultaneous Tests for General Linear Hypotheses

Multiple Comparisons of Means: Tukey Contrasts

Fit: lme.formula(fixed = ConcFD4 ~ Time2 + Gut, data = datasetFD4Nano200, random = ~1 | NumFish)

Linear Hypotheses:

Estimate Std. Error z value Pr(>|z|)

Median - Distal == 0 3.287 2.638 1.246 0.213

(Adjusted p values reported -- single-step method)

## **Time effect concerning the median gut**

### 3.2.1. Linear mixed effect models

>datasetFD4Nano200m<-subset(datasetFD4Nano200,datasetFD4Nano200$Gut=="Median")

>Model3m.lme<-lme(ConcFD4~Time2,datasetFD4Nano200m,random=~1|NumFish)

> qqnorm(Model3m.lme)

> summary(Model3m.lme)

Linear mixed-effects model fit by REML

Data: datasetFD4Nano200m

AIC BIC logLik

389.013 406.8549 -184.5065

Random effects:

Formula: ~1 | NumFish

(Intercept) Residual

StdDev: 14.87897 11.3255

Fixed effects: ConcFD4 ~ Time2

Value Std.Error DF t-value p-value

(Intercept) 8.656741 7.067539 38 1.2248593 0.2282

Time215 2.102520 6.053737 38 0.3473095 0.7303

Time230 -0.702931 6.053737 38 -0.1161152 0.9082

Time245 1.763785 6.344459 38 0.2780040 0.7825

Time260 2.233387 6.344459 38 0.3520217 0.7268

Time290 5.431649 6.344459 38 0.8561248 0.3973

Time2120 17.914457 6.053737 38 2.9592395 0.0053

Time2150 10.491411 6.333361 38 1.6565313 0.1058

Correlation:

(Intr) Tim215 Tim230 Tim245 Tim260 Tim290 Tm2120

Time215 -0.428

Time230 -0.428 0.500

Time245 -0.409 0.477 0.477

Time260 -0.409 0.477 0.477 0.469

Time290 -0.409 0.477 0.477 0.469 0.469

Time2120 -0.428 0.500 0.500 0.477 0.477 0.477

Time2150 -0.409 0.478 0.478 0.453 0.453 0.453 0.478

Standardized Within-Group Residuals:

Min Q1 Med Q3 Max

-1.70350212 -0.67223758 0.04982426 0.30089424 3.57867707

Number of Observations: 52

Number of Groups: 7

### 3.2.2. Pairwise comparisons of the sampling times concerning the median gut

*> summary(glht(Model3m.lme,linfct=mcp(Time2="Tukey")))*

Simultaneous Tests for General Linear Hypotheses

Multiple Comparisons of Means: Tukey Contrasts

Fit: lme.formula(fixed = ConcFD4 ~ Time2, data = datasetFD4Nano200m, random = ~1 | NumFish)

Linear Hypotheses:

Estimate Std. Error z value Pr(>|z|)

15 - 0 == 0 2.1025 6.0537 0.347 1.0000

30 - 0 == 0 -0.7029 6.0537 -0.116 1.0000

45 - 0 == 0 1.7638 6.3445 0.278 1.0000

60 - 0 == 0 2.2334 6.3445 0.352 1.0000

90 - 0 == 0 5.4316 6.3445 0.856 0.9897

120 - 0 == 0 17.9145 6.0537 2.959 0.0612 .

150 - 0 == 0 10.4914 6.3334 1.657 0.7152

30 - 15 == 0 -2.8055 6.0537 -0.463 0.9998

45 - 15 == 0 -0.3387 6.3445 -0.053 1.0000

60 - 15 == 0 0.1309 6.3445 0.021 1.0000

90 - 15 == 0 3.3291 6.3445 0.525 0.9995

120 - 15 == 0 15.8119 6.0537 2.612 0.1512

150 - 15 == 0 8.3889 6.3334 1.325 0.8896

45 - 30 == 0 2.4667 6.3445 0.389 0.9999

60 - 30 == 0 2.9363 6.3445 0.463 0.9998

90 - 30 == 0 6.1346 6.3445 0.967 0.9790

120 - 30 == 0 18.6174 6.0537 3.075 0.0439 \*

150 - 30 == 0 11.1943 6.3334 1.768 0.6419

60 - 45 == 0 0.4696 6.5388 0.072 1.0000

90 - 45 == 0 3.6679 6.5388 0.561 0.9993

120 - 45 == 0 16.1507 6.3445 2.546 0.1761

150 - 45 == 0 8.7276 6.6274 1.317 0.8926

90 - 60 == 0 3.1983 6.5388 0.489 0.9997

120 - 60 == 0 15.6811 6.3445 2.472 0.2075

150 - 60 == 0 8.2580 6.6274 1.246 0.9179

120 - 90 == 0 12.4828 6.3445 1.968 0.5039

150 - 90 == 0 5.0598 6.6274 0.763 0.9949

150 - 120 == 0 -7.4230 6.3334 -1.172 0.9398

---

Signif. codes: 0 ‘\*\*\*’ 0.001 ‘\*\*’ 0.01 ‘\*’ 0.05 ‘.’ 0.1 ‘ ’ 1

(Adjusted p values reported -- single-step method)

## 

## **Time effect concerning the distal gut**

### 3.3.1. Linear mixed effect model

> datasetFD4Nano200d<-subset(datasetFD4Nano200,datasetFD4Nano200$Gut=="Distal")

> Model3d.lme<-lme(ConcFD4~Time2,datasetFD4Nano200d,random=~1|NumFish)

> qqnorm(Model3d.lme)

> summary(Model3d.lme)

Linear mixed-effects model fit by REML

Data: datasetFD4Nano200d

AIC BIC logLik

427.192 445.6935 -203.596

Random effects:

Formula: ~1 | NumFish

(Intercept) Residual

StdDev: 9.358055 14.21774

Fixed effects: ConcFD4 ~ Time2

Value Std.Error DF t-value p-value

(Intercept) 4.717260 6.433364 41 0.7332493 0.4676

Time215 13.054521 7.599700 41 1.7177680 0.0934

Time230 0.356506 7.599700 41 0.0469106 0.9628

Time245 0.638505 7.599700 41 0.0840171 0.9335

Time260 1.818462 7.599700 41 0.2392808 0.8121

Time290 3.332786 7.599700 41 0.4385418 0.6633

Time2120 12.457245 7.599700 41 1.6391758 0.1088

Time2150 4.943376 7.942635 41 0.6223849 0.5371

Correlation:

(Intr) Tim215 Tim230 Tim245 Tim260 Tim290 Tm2120

Time215 -0.591

Time230 -0.591 0.500

Time245 -0.591 0.500 0.500

Time260 -0.591 0.500 0.500 0.500

Time290 -0.591 0.500 0.500 0.500 0.500

Time2120 -0.591 0.500 0.500 0.500 0.500 0.500

Time2150 -0.565 0.478 0.478 0.478 0.478 0.478 0.478

Standardized Within-Group Residuals:

Min Q1 Med Q3 Max

-1.28412193 -0.47850360 -0.02818227 0.27474730 4.63289949

Number of Observations: 55

Number of Groups: 7

### 3.3.2. Pairwise comparisons of the sampling times concerning the distal gut

*> summary(glht(Model3d.lme,linfct=mcp(Time2="Tukey")))*

Simultaneous Tests for General Linear Hypotheses

Multiple Comparisons of Means: Tukey Contrasts

Fit: lme.formula(fixed = ConcFD4 ~ Time2, data = datasetFD4Nano200d,

random = ~1 | NumFish)

Linear Hypotheses:

Estimate Std. Error z value Pr(>|z|)

15 - 0 == 0 13.0545 7.5997 1.718 0.676

30 - 0 == 0 0.3565 7.5997 0.047 1.000

45 - 0 == 0 0.6385 7.5997 0.084 1.000

60 - 0 == 0 1.8185 7.5997 0.239 1.000

90 - 0 == 0 3.3328 7.5997 0.439 1.000

120 - 0 == 0 12.4572 7.5997 1.639 0.726

150 - 0 == 0 4.9434 7.9426 0.622 0.999

30 - 15 == 0 -12.6980 7.5997 -1.671 0.706

45 - 15 == 0 -12.4160 7.5997 -1.634 0.730

60 - 15 == 0 -11.2361 7.5997 -1.478 0.819

90 - 15 == 0 -9.7217 7.5997 -1.279 0.907

120 - 15 == 0 -0.5973 7.5997 -0.079 1.000

150 - 15 == 0 -8.1111 7.9426 -1.021 0.971

45 - 30 == 0 0.2820 7.5997 0.037 1.000

60 - 30 == 0 1.4620 7.5997 0.192 1.000

90 - 30 == 0 2.9763 7.5997 0.392 1.000

120 - 30 == 0 12.1007 7.5997 1.592 0.755

150 - 30 == 0 4.5869 7.9426 0.577 0.999

60 - 45 == 0 1.1800 7.5997 0.155 1.000

90 - 45 == 0 2.6943 7.5997 0.355 1.000

120 - 45 == 0 11.8187 7.5997 1.555 0.777

150 - 45 == 0 4.3049 7.9426 0.542 0.999

90 - 60 == 0 1.5143 7.5997 0.199 1.000

120 - 60 == 0 10.6388 7.5997 1.400 0.858

150 - 60 == 0 3.1249 7.9426 0.393 1.000

120 - 90 == 0 9.1245 7.5997 1.201 0.932

150 - 90 == 0 1.6106 7.9426 0.203 1.000

150 - 120 == 0 -7.5139 7.9426 -0.946 0.981

(Adjusted p values reported -- single-step method)

# Treatment: NP 200

(n=6 per gut distal and proximal; n=3 per gut proximal)

## **4.1. Time effect, including both gut segments in one single analysis, and difference in nano concentration between gut segments**

### 4.1.1. Linear mixed effect models

> dataset200<-subset(Dataset,Dataset$Treat=="Nano200")

> Model2.lme<-lme(ConcNano~Time2+Gut,dataset200,random=~1|NumFish)

> qqnorm(Model2.lme)

> summary(Model2.lme)

Linear mixed-effects model fit by REML

Data: dataset200

AIC BIC logLik

815.1315 847.5373 -395.5657

Random effects:

Formula: ~1 | NumFish

(Intercept) Residual

StdDev: 5.476505 7.354786

Fixed effects: ConcNano ~ Time2 + Gut

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
|  | Value | Std.Error | DF | t-value | p-value |
| (Intercept) | 8.045210 | 3.046480 | 105 | 2.640822 | 0.0095 |
| Time215 | 10.523949 | 2.685588 | 105 | 3.918676 | 0.0002 |
| Time230 | 8.288154 | 2.685588 | 105 | 3.086160 | 0.0026 |
| Time245 | 9.453491 | 2.685588 | 105 | 3.520082 | 0.0006 |
| Time260 | 9.980815 | 2.685588 | 105 | 3.716436 | 0.0003 |
| Time290 | 8.289693 | 2.685588 | 105 | 3.086733 | 0.0026 |
| Time2120 | 8.833167 | 2.685588 | 105 | 3.289100 | 0.0014 |
| Time2150 | 7.603058 | 2.685588 | 105 | 2.831059 | 0.0056 |
| GutMedian | -6.690835 | 1.501289 | 105 | -4.456726 | 0.0000 |
| GutProximal | -4.219129 | 1.971596 | 105 | -2.139956 | 0.0347 |

Correlation:

(Intr) Tim215 Tim230 Tim245 Tim260 Tim290 Tm2120 Tm2150 GutMdn

Time215 -0.441

Time230 -0.441 0.500

Time245 -0.441 0.500 0.500

Time260 -0.441 0.500 0.500 0.500

Time290 -0.441 0.500 0.500 0.500 0.500

Time2120 -0.441 0.500 0.500 0.500 0.500 0.500

Time2150 -0.441 0.500 0.500 0.500 0.500 0.500 0.500

GutMedian -0.246 0.000 0.000 0.000 0.000 0.000 0.000 0.000

Standardized Within-Group Residuals:

Min Q1 Med Q3 Max

-2.2230306 -0.5776741 0.0110387 0.4662640 3.5229171

Number of Observations: 120

Number of Groups: 6

### 4.1.2. Pairwise comparisons concerning the sampling times

*> summary(glht(Model2.lme,linfct=mcp(Time2="Tukey")))*

Simultaneous Tests for General Linear Hypotheses

Multiple Comparisons of Means: Tukey Contrasts

Fit: lme.formula(fixed = ConcNano ~ Time2 + Gut, data = dataset200, random = ~1 | NumFish)

Linear Hypotheses:

Estimate Std. Error z value Pr(>|z|)

15 - 0 == 0 10.523949 2.685588 3.919 0.00243 \*\*

30 - 0 == 0 8.288154 2.685588 3.086 0.04239 \*

45 - 0 == 0 9.453491 2.685588 3.520 0.01033 \*

60 - 0 == 0 9.980815 2.685588 3.716 0.00495 \*\*

90 - 0 == 0 8.289693 2.685588 3.087 0.04249 \*

120 - 0 == 0 8.833167 2.685588 3.289 0.02230 \*

150 - 0 == 0 7.603058 2.685588 2.831 0.08748 .

30 - 15 == 0 -2.235795 2.685588 -0.833 0.99130

45 - 15 == 0 -1.070458 2.685588 -0.399 0.99993

60 - 15 == 0 -0.543134 2.685588 -0.202 1.00000

90 - 15 == 0 -2.234256 2.685588 -0.832 0.99133

120 - 15 == 0 -1.690782 2.685588 -0.630 0.99848

150 - 15 == 0 -2.920891 2.685588 -1.088 0.95952

45 - 30 == 0 1.165337 2.685588 0.434 0.99987

60 - 30 == 0 1.692661 2.685588 0.630 0.99847

90 - 30 == 0 0.001539 2.685588 0.001 1.00000

120 - 30 == 0 0.545013 2.685588 0.203 1.00000

150 - 30 == 0 -0.685096 2.685588 -0.255 1.00000

60 - 45 == 0 0.527325 2.685588 0.196 1.00000

90 - 45 == 0 -1.163798 2.685588 -0.433 0.99987

120 - 45 == 0 -0.620324 2.685588 -0.231 1.00000

150 - 45 == 0 -1.850433 2.685588 -0.689 0.99730

90 - 60 == 0 -1.691122 2.685588 -0.630 0.99848

120 - 60 == 0 -1.147648 2.685588 -0.427 0.99988

150 - 60 == 0 -2.377757 2.685588 -0.885 0.98742

120 - 90 == 0 0.543474 2.685588 0.202 1.00000

150 - 90 == 0 -0.686635 2.685588 -0.256 1.00000

150 - 120 == 0 -1.230109 2.685588 -0.458 0.99981

---

Signif. codes: 0 ‘\*\*\*’ 0.001 ‘\*\*’ 0.01 ‘\*’ 0.05 ‘.’ 0.1 ‘ ’ 1

(Adjusted p values reported -- single-step method)

### 4.1.3. Pairwise comparisons concerning the segment guts

*> summary(glht(Model2.lme,linfct=mcp(Gut="Tukey")))*

Simultaneous Tests for General Linear Hypotheses

Multiple Comparisons of Means: Tukey Contrasts

Fit: lme.formula(fixed = ConcNano ~ Time2 + Gut, data = dataset200, random = ~1 | NumFish)

Linear Hypotheses:

Estimate Std. Error z value Pr(>|z|)

Median - Distal == 0 -6.691 1.501 -4.457 <0.001 \*\*\*

---

Signif. codes: 0 ‘\*\*\*’ 0.001 ‘\*\*’ 0.01 ‘\*’ 0.05 ‘.’ 0.1 ‘ ’ 1

(Adjusted p values reported -- single-step method)

## **Time effect concerning the median gut**

### 4.2.1. Linear mixed effect model

*> dataset200m<-subset(dataset200,dataset200$Gut=="Median")*

*> Model2m.lme<-lme(ConcNano~Time2,dataset200m,random=~1|NumFish)*

*> qqnorm(Model2m.lme)*

*> summary(Model2m.lme)*

Linear mixed-effects model fit by REML

Data: dataset200m

AIC BIC logLik

264.9689 281.8577 -122.4845

Random effects:

Formula: ~1 | NumFish

(Intercept) Residual

StdDev: 1.269057 4.176302

Fixed effects: ConcNano ~ Time2

Value Std.Error DF t-value p-value

(Intercept) 2.846427 1.781947 35 1.597369 0.1192

Time215 5.261503 2.411189 35 2.182119 0.0359

Time230 6.191002 2.411189 35 2.567614 0.0147

Time245 6.596143 2.411189 35 2.735639 0.0097

Time260 8.683390 2.411189 35 3.601289 0.0010

Time290 8.271116 2.411189 35 3.430306 0.0016

Time2120 9.248370 2.411189 35 3.835605 0.0005

Time2150 6.784390 2.411189 35 2.813711 0.0080

Correlation:

(Intr) Tim215 Tim230 Tim245 Tim260 Tim290 Tm2120

Time215 -0.677

Time230 -0.677 0.500

Time245 -0.677 0.500 0.500

Time260 -0.677 0.500 0.500 0.500

Time290 -0.677 0.500 0.500 0.500 0.500

Time2120 -0.677 0.500 0.500 0.500 0.500 0.500

Time2150 -0.677 0.500 0.500 0.500 0.500 0.500 0.500

Standardized Within-Group Residuals:

Min Q1 Med Q3 Max

-1.84251424 -0.48239860 -0.09223824 0.30607902 2.97040602

Number of Observations: 48

Number of Groups: 6

### 4.2.2. Pairwise comparisons of sampling times concerning the median gut

*> summary(glht(Model2m.lme,linfct=mcp(Time2="Tukey")))*

Simultaneous Tests for General Linear Hypotheses

Multiple Comparisons of Means: Tukey Contrasts

Fit: lme.formula(fixed = ConcNano ~ Time2, data = dataset200m, random = ~1 | NumFish)

Linear Hypotheses:

Estimate Std. Error z value Pr(>|z|)

15 - 0 == 0 5.2615 2.4112 2.182 0.36264

30 - 0 == 0 6.1910 2.4112 2.568 0.16775

45 - 0 == 0 6.5961 2.4112 2.736 0.11243

60 - 0 == 0 8.6834 2.4112 3.601 0.00764 \*\*

90 - 0 == 0 8.2711 2.4112 3.430 0.01390 \*

120 - 0 == 0 9.2484 2.4112 3.836 0.00309 \*\*

150 - 0 == 0 6.7844 2.4112 2.814 0.09187 .

30 - 15 == 0 0.9295 2.4112 0.385 0.99994

45 - 15 == 0 1.3346 2.4112 0.554 0.99934

60 - 15 == 0 3.4219 2.4112 1.419 0.84872

90 - 15 == 0 3.0096 2.4112 1.248 0.91732

120 - 15 == 0 3.9869 2.4112 1.653 0.71741

150 - 15 == 0 1.5229 2.4112 0.632 0.99845

45 - 30 == 0 0.4051 2.4112 0.168 1.00000

60 - 30 == 0 2.4924 2.4112 1.034 0.96941

90 - 30 == 0 2.0801 2.4112 0.863 0.98922

120 - 30 == 0 3.0574 2.4112 1.268 0.91072

150 - 30 == 0 0.5934 2.4112 0.246 1.00000

60 - 45 == 0 2.0872 2.4112 0.866 0.98900

90 - 45 == 0 1.6750 2.4112 0.695 0.99716

120 - 45 == 0 2.6522 2.4112 1.100 0.95702

150 - 45 == 0 0.1882 2.4112 0.078 1.00000

90 - 60 == 0 -0.4123 2.4112 -0.171 1.00000

120 - 60 == 0 0.5650 2.4112 0.234 1.00000

150 - 60 == 0 -1.8990 2.4112 -0.788 0.99379

120 - 90 == 0 0.9773 2.4112 0.405 0.99992

150 - 90 == 0 -1.4867 2.4112 -0.617 0.99868

150 - 120 == 0 -2.4640 2.4112 -1.022 0.97128

---

Signif. codes: 0 ‘\*\*\*’ 0.001 ‘\*\*’ 0.01 ‘\*’ 0.05 ‘.’ 0.1 ‘ ’ 1

(Adjusted p values reported -- single-step method)

## **Time effect concerning the distal gut**

### 4.3.1. Linear mixed effect model

> dataset200d<-subset(dataset200,dataset200$Gut=="Distal")

> Model2d.lme<-lme(ConcNano~Time2,dataset200d,random=~1|NumFish)

> qqnorm(Model2d.lme)

> summary(Model2d.lme)

Linear mixed-effects model fit by REML

Data: dataset200d

AIC BIC logLik

304.66 321.5488 -142.33

Random effects:

Formula: ~1 | NumFish

(Intercept) Residual

StdDev: 11.64604 5.690589

Fixed effects: ConcNano ~ Time2

Value Std.Error DF t-value p-value

(Intercept) 8.634723 5.291708 35 1.631746 0.1117

Time215 12.983281 3.285463 35 3.951736 0.0004

Time230 7.594492 3.285463 35 2.311544 0.0268

Time245 10.875372 3.285463 35 3.310149 0.0022

Time260 10.106436 3.285463 35 3.076107 0.0041

Time290 5.904565 3.285463 35 1.797179 0.0809

Time2120 6.662729 3.285463 35 2.027942 0.0502

Time2150 4.129347 3.285463 35 1.256854 0.2171

Correlation:

(Intr) Tim215 Tim230 Tim245 Tim260 Tim290 Tm2120

Time215 -0.31

Time230 -0.31 0.50

Time245 -0.31 0.50 0.50

Time260 -0.31 0.50 0.50 0.50

Time290 -0.31 0.50 0.50 0.50 0.50

Time2120 -0.31 0.50 0.50 0.50 0.50 0.50

Time2150 -0.31 0.50 0.50 0.50 0.50 0.50 0.50

Standardized Within-Group Residuals:

Min Q1 Med Q3 Max

-1.59867755 -0.47738845 0.01805587 0.42460534 2.82355549

Number of Observations: 48

Number of Groups: 6

### 4.3.2.Pairwise comparisons of the sampling times for the distal gut

*> summary(glht(Model2d.lme,linfct=mcp(Time2="Tukey")))*

Simultaneous Tests for General Linear Hypotheses

Multiple Comparisons of Means: Tukey Contrasts

Fit: lme.formula(fixed = ConcNano ~ Time2, data = dataset200d, random = ~1 | NumFish)

Linear Hypotheses:

Estimate Std. Error z value Pr(>|z|)

15 - 0 == 0 12.9833 3.2855 3.952 <0.01 \*\*

30 - 0 == 0 7.5945 3.2855 2.312 0.2877

45 - 0 == 0 10.8754 3.2855 3.310 0.0208 \*

60 - 0 == 0 10.1064 3.2855 3.076 0.0438 \*

90 - 0 == 0 5.9046 3.2855 1.797 0.6221

120 - 0 == 0 6.6627 3.2855 2.028 0.4625

150 - 0 == 0 4.1293 3.2855 1.257 0.9145

30 - 15 == 0 -5.3888 3.2855 -1.640 0.7261

45 - 15 == 0 -2.1079 3.2855 -0.642 0.9983

60 - 15 == 0 -2.8768 3.2855 -0.876 0.9882

90 - 15 == 0 -7.0787 3.2855 -2.155 0.3796

120 - 15 == 0 -6.3206 3.2855 -1.924 0.5343

150 - 15 == 0 -8.8539 3.2855 -2.695 0.1243

45 - 30 == 0 3.2809 3.2855 0.999 0.9748

60 - 30 == 0 2.5119 3.2855 0.765 0.9948

90 - 30 == 0 -1.6899 3.2855 -0.514 0.9996

120 - 30 == 0 -0.9318 3.2855 -0.284 1.0000

150 - 30 == 0 -3.4651 3.2855 -1.055 0.9658

60 - 45 == 0 -0.7689 3.2855 -0.234 1.0000

90 - 45 == 0 -4.9708 3.2855 -1.513 0.8007

120 - 45 == 0 -4.2126 3.2855 -1.282 0.9057

150 - 45 == 0 -6.7460 3.2855 -2.053 0.4457

90 - 60 == 0 -4.2019 3.2855 -1.279 0.9069

120 - 60 == 0 -3.4437 3.2855 -1.048 0.9670

150 - 60 == 0 -5.9771 3.2855 -1.819 0.6068

120 - 90 == 0 0.7582 3.2855 0.231 1.0000

150 - 90 == 0 -1.7752 3.2855 -0.540 0.9994

150 - 120 == 0 -2.5334 3.2855 -0.771 0.9945

---

Signif. codes: 0 ‘\*\*\*’ 0.001 ‘\*\*’ 0.01 ‘\*’ 0.05 ‘.’ 0.1 ‘ ’ 1

(Adjusted p values reported -- single-step method)

# Treatment NP 20

(n=6 per gut)

## **5.1. Time effect, including both gut segments in one single analysis, and difference in nano concentration between gut segments**

### 5.1.1. Linear mixed effect model

*> dataset20<-subset(Dataset,Dataset$Treat=="Nano20")*

*> Model3.lme<-lme(ConcNano~Time2+Gut,dataset20,random=~1|NumFish)*

*> qqnorm(Model3.lme)*

*> summary(Model3.lme)*

Linear mixed-effects model fit by REML

Data: dataset20

AIC BIC logLik

921.3331 956.1071 -448.6665

Random effects:

Formula: ~1 | NumFish

(Intercept) Residual

StdDev: 2.499827 5.974401

Fixed effects: ConcNano ~ Time2 + Gut

Value Std.Error DF t-value p-value

(Intercept) 1.035768 1.876229 129 0.552048 0.5819

Time215 9.374920 1.991467 129 4.707545 0.0000

Time230 8.002268 1.991467 129 4.018278 0.0001

Time245 10.287234 1.991467 129 5.165656 0.0000

Time260 11.798169 1.991467 129 5.924360 0.0000

Time290 14.840695 1.991467 129 7.452142 0.0000

Time2120 12.758620 1.991467 129 6.406644 0.0000

Time2150 16.962053 1.991467 129 8.517366 0.0000

GutMedian -0.005869 1.219520 129 -0.004813 0.9962

Correlation:

(Intr) Tim215 Tim230 Tim245 Tim260 Tim290 Tm2120 Tm2150 GutMdn

Time215 -0.531

Time230 -0.531 0.500

Time245 -0.531 0.500 0.500

Time260 -0.531 0.500 0.500 0.500

Time290 -0.531 0.500 0.500 0.500 0.500

Time2120 -0.531 0.500 0.500 0.500 0.500 0.500

Time2150 -0.531 0.500 0.500 0.500 0.500 0.500 0.500

GutMedian -0.325 0.000 0.000 0.000 0.000 0.000 0.000 0.000

Standardized Within-Group Residuals:

Min Q1 Med Q3 Max

-1.8669421 -0.5945511 -0.1130959 0.3783212 4.6712345

Number of Observations: 144

Number of Groups: 6

### 5.1.2. Pairwise comparisons concerning the sampling time

*> summary(glht(Model3.lme,linfct=mcp(Time2="Tukey")))*

Simultaneous Tests for General Linear Hypotheses

Multiple Comparisons of Means: Tukey Contrasts

Fit: lme.formula(fixed = ConcNano ~ Time2 + Gut, data = dataset20, random = ~1 | NumFish)

Linear Hypotheses:

Estimate Std. Error z value Pr(>|z|)

15 - 0 == 0 9.3749 1.9915 4.708 <0.01 \*\*\*

30 - 0 == 0 8.0023 1.9915 4.018 <0.01 \*\*

45 - 0 == 0 10.2872 1.9915 5.166 <0.01 \*\*\*

60 - 0 == 0 11.7982 1.9915 5.924 <0.01 \*\*\*

90 - 0 == 0 14.8407 1.9915 7.452 <0.01 \*\*\*

120 - 0 == 0 12.7586 1.9915 6.407 <0.01 \*\*\*

150 - 0 == 0 16.9621 1.9915 8.517 <0.01 \*\*\*

30 - 15 == 0 -1.3727 1.9915 -0.689 0.9973

45 - 15 == 0 0.9123 1.9915 0.458 0.9998

60 - 15 == 0 2.4232 1.9915 1.217 0.9272

90 - 15 == 0 5.4658 1.9915 2.745 0.1093

120 - 15 == 0 3.3837 1.9915 1.699 0.6879

150 - 15 == 0 7.5871 1.9915 3.810 <0.01 \*\*

45 - 30 == 0 2.2850 1.9915 1.147 0.9462

60 - 30 == 0 3.7959 1.9915 1.906 0.5466

90 - 30 == 0 6.8384 1.9915 3.434 0.0141 \*

120 - 30 == 0 4.7564 1.9915 2.388 0.2473

150 - 30 == 0 8.9598 1.9915 4.499 <0.01 \*\*\*

60 - 45 == 0 1.5109 1.9915 0.759 0.9951

90 - 45 == 0 4.5535 1.9915 2.286 0.3011

120 - 45 == 0 2.4714 1.9915 1.241 0.9198

150 - 45 == 0 6.6748 1.9915 3.352 0.0180 \*

90 - 60 == 0 3.0425 1.9915 1.528 0.7925

120 - 60 == 0 0.9605 1.9915 0.482 0.9997

150 - 60 == 0 5.1639 1.9915 2.593 0.1574

120 - 90 == 0 -2.0821 1.9915 -1.045 0.9674

150 - 90 == 0 2.1214 1.9915 1.065 0.9639

150 - 120 == 0 4.2034 1.9915 2.111 0.4079

---

Signif. codes: 0 ‘\*\*\*’ 0.001 ‘\*\*’ 0.01 ‘\*’ 0.05 ‘.’ 0.1 ‘ ’ 1

(Adjusted p values reported -- single-step method)

### 5.1.3. Pairwise comparisons concerning the gut segment

*> summary(glht(Model3.lme,linfct=mcp(Gut="Tukey")))*

Simultaneous Tests for General Linear Hypotheses

Multiple Comparisons of Means: Tukey Contrasts

Fit: lme.formula(fixed = ConcNano ~ Time2 + Gut, data = dataset20, random = ~1 | NumFish)

Linear Hypotheses:

Estimate Std. Error z value Pr(>|z|)

Median - Distal == 0 -0.005869 1.219520 -0.005 1.0000

---

Signif. codes: 0 ‘\*\*\*’ 0.001 ‘\*\*’ 0.01 ‘\*’ 0.05 ‘.’ 0.1 ‘ ’ 1

(Adjusted p values reported -- single-step method)

## **5.2. Time effect concerning the median gut**

### 

### 5.2.1. Linear mixed effect models

*> dataset20m<-subset(dataset20,dataset20$Gut=="Median")*

*> Model3m.lme<-lme(ConcNano~Time2,dataset20m,random=~1|NumFish)*

*> qqnorm(Model3m.lme)*

*> summary(Model3m.lme****)***

Linear mixed-effects model fit by REML

Data: dataset20m

AIC BIC logLik

277.3358 294.2246 -128.6679

Random effects:

Formula: ~1 | NumFish

(Intercept) Residual

StdDev: 5.969755 4.227838

Fixed effects: ConcNano ~ Time2

Value Std.Error DF t-value p-value

(Intercept) 0.757361 2.986430 35 0.253601 0.8013

Time215 8.476137 2.440943 35 3.472484 0.0014

Time230 7.582776 2.440943 35 3.106494 0.0037

Time245 10.272125 2.440943 35 4.208260 0.0002

Time260 11.069560 2.440943 35 4.534952 0.0001

Time290 14.701999 2.440943 35 6.023081 0.0000

Time2120 15.073651 2.440943 35 6.175339 0.0000

Time2150 19.028014 2.440943 35 7.795353 0.0000

Correlation:

(Intr) Tim215 Tim230 Tim245 Tim260 Tim290 Tm2120

Time215 -0.409

Time230 -0.409 0.500

Time245 -0.409 0.500 0.500

Time260 -0.409 0.500 0.500 0.500

Time290 -0.409 0.500 0.500 0.500 0.500

Time2120 -0.409 0.500 0.500 0.500 0.500 0.500

Time2150 -0.409 0.500 0.500 0.500 0.500 0.500 0.500

Standardized Within-Group Residuals:

Min Q1 Med Q3 Max

-1.89126168 -0.58496841 0.02988815 0.30377603 2.60938731

Number of Observations: 48

Number of Groups: 6

### 5.2.2. Pairwise comparisons of sampling times for the median gut

*> summary(glht(Model3m.lme,linfct=mcp(Time2="Tukey")))*

Simultaneous Tests for General Linear Hypotheses

Multiple Comparisons of Means: Tukey Contrasts

Fit: lme.formula(fixed = ConcNano ~ Time2, data = dataset20m, random = ~1 | NumFish)

Linear Hypotheses:

Estimate Std. Error z value Pr(>|z|)

15 - 0 == 0 8.4761 2.4409 3.472 0.01194 \*

30 - 0 == 0 7.5828 2.4409 3.106 0.04026 \*

45 - 0 == 0 10.2721 2.4409 4.208 < 0.001 \*\*\*

60 - 0 == 0 11.0696 2.4409 4.535 < 0.001 \*\*\*

90 - 0 == 0 14.7020 2.4409 6.023 < 0.001 \*\*\*

120 - 0 == 0 15.0737 2.4409 6.175 < 0.001 \*\*\*

150 - 0 == 0 19.0280 2.4409 7.795 < 0.001 \*\*\*

30 - 15 == 0 -0.8934 2.4409 -0.366 0.99996

45 - 15 == 0 1.7960 2.4409 0.736 0.99592

60 - 15 == 0 2.5934 2.4409 1.062 0.96438

90 - 15 == 0 6.2259 2.4409 2.551 0.17453

120 - 15 == 0 6.5975 2.4409 2.703 0.12184

150 - 15 == 0 10.5519 2.4409 4.323 < 0.001 \*\*\*

45 - 30 == 0 2.6893 2.4409 1.102 0.95663

60 - 30 == 0 3.4868 2.4409 1.428 0.84426

90 - 30 == 0 7.1192 2.4409 2.917 0.06936 .

120 - 30 == 0 7.4909 2.4409 3.069 0.04483 \*

150 - 30 == 0 11.4452 2.4409 4.689 < 0.001 \*\*\*

60 - 45 == 0 0.7974 2.4409 0.327 0.99998

90 - 45 == 0 4.4299 2.4409 1.815 0.61011

120 - 45 == 0 4.8015 2.4409 1.967 0.50495

150 - 45 == 0 8.7559 2.4409 3.587 0.00798 \*\*

90 - 60 == 0 3.6324 2.4409 1.488 0.81411

120 - 60 == 0 4.0041 2.4409 1.640 0.72560

150 - 60 == 0 7.9585 2.4409 3.260 0.02443 \*

120 - 90 == 0 0.3717 2.4409 0.152 1.00000

150 - 90 == 0 4.3260 2.4409 1.772 0.63884

150 - 120 == 0 3.9544 2.4409 1.620 0.73820

---

Signif. codes: 0 ‘\*\*\*’ 0.001 ‘\*\*’ 0.01 ‘\*’ 0.05 ‘.’ 0.1 ‘ ’ 1

(Adjusted p values reported -- single-step method)

## 

## **5.3. Time effect concerning the distal gut**

### 5.3.1. Linear mixed effect model

> dataset20d<-subset(dataset20,dataset20$Gut=="Distal")

> Model3d.lme<-lme(ConcNano~Time2,dataset20d,random=~1|NumFish)

> qqnorm(Model3d.lme)

> summary(Model3d.lme)

Linear mixed-effects model fit by REML

Data: dataset20d

AIC BIC logLik

288.6877 305.5765 -134.3439

Random effects:

Formula: ~1 | NumFish

(Intercept) Residual

StdDev: 2.718542 5.428643

Fixed effects: ConcNano ~ Time2

Value Std.Error DF t-value p-value

(Intercept) 2.826474 2.478596 35 1.140353 0.2619

Time215 7.749529 3.134229 35 2.472548 0.0184

Time230 6.741281 3.134229 35 2.150858 0.0385

Time245 8.237312 3.134229 35 2.628178 0.0127

Time260 9.189916 3.134229 35 2.932114 0.0059

Time290 9.915902 3.134229 35 3.163745 0.0032

Time2120 10.388903 3.134229 35 3.314660 0.0021

Time2150 17.475468 3.134229 35 5.575684 0.0000

Correlation:

(Intr) Tim215 Tim230 Tim245 Tim260 Tim290 Tm2120

Time215 -0.632

Time230 -0.632 0.500

Time245 -0.632 0.500 0.500

Time260 -0.632 0.500 0.500 0.500

Time290 -0.632 0.500 0.500 0.500 0.500

Time2120 -0.632 0.500 0.500 0.500 0.500 0.500

Time2150 -0.632 0.500 0.500 0.500 0.500 0.500 0.500

Standardized Within-Group Residuals:

Min Q1 Med Q3 Max

-1.7880577 -0.4819966 -0.1331791 0.4865105 3.6971864

Number of Observations: 48

Number of Groups: 6

### 5.3.2. Pairwise comparisons of the sampling times for the distal gut

*> summary(glht(Model3d.lme,linfct=mcp(Time2="Tukey")))*

Simultaneous Tests for General Linear Hypotheses

Multiple Comparisons of Means: Tukey Contrasts

Fit: lme.formula(fixed = ConcNano ~ Time2, data = dataset20d, random = ~1 |

NumFish)

Linear Hypotheses:

Estimate Std. Error z value Pr(>|z|)

15 - 0 == 0 7.7495 3.1342 2.473 0.2078

30 - 0 == 0 6.7413 3.1342 2.151 0.3823

45 - 0 == 0 8.2373 3.1342 2.628 0.1464

60 - 0 == 0 9.1899 3.1342 2.932 0.0662 .

90 - 0 == 0 9.9159 3.1342 3.164 0.0336 \*

120 - 0 == 0 10.3889 3.1342 3.315 0.0208 \*

150 - 0 == 0 17.4755 3.1342 5.576 <0.001 \*\*\*

30 - 15 == 0 -1.0082 3.1342 -0.322 1.0000

45 - 15 == 0 0.4878 3.1342 0.156 1.0000

60 - 15 == 0 1.4404 3.1342 0.460 0.9998

90 - 15 == 0 2.1664 3.1342 0.691 0.9972

120 - 15 == 0 2.6394 3.1342 0.842 0.9907

150 - 15 == 0 9.7259 3.1342 3.103 0.0404 \*

45 - 30 == 0 1.4960 3.1342 0.477 0.9998

60 - 30 == 0 2.4486 3.1342 0.781 0.9941

90 - 30 == 0 3.1746 3.1342 1.013 0.9727

120 - 30 == 0 3.6476 3.1342 1.164 0.9421

150 - 30 == 0 10.7342 3.1342 3.425 0.0140 \*

60 - 45 == 0 0.9526 3.1342 0.304 1.0000

90 - 45 == 0 1.6786 3.1342 0.536 0.9995

120 - 45 == 0 2.1516 3.1342 0.686 0.9974

150 - 45 == 0 9.2382 3.1342 2.948 0.0638 .

90 - 60 == 0 0.7260 3.1342 0.232 1.0000

120 - 60 == 0 1.1990 3.1342 0.383 0.9999

150 - 60 == 0 8.2856 3.1342 2.644 0.1405

120 - 90 == 0 0.4730 3.1342 0.151 1.0000

150 - 90 == 0 7.5596 3.1342 2.412 0.2356

150 - 120 == 0 7.0866 3.1342 2.261 0.3155

---

Signif. codes: 0 ‘\*\*\*’ 0.001 ‘\*\*’ 0.01 ‘\*’ 0.05 ‘.’ 0.1 ‘ ’ 1

(Adjusted p values reported -- single-step method)

**Reference**

Catarino, A.I., Frutos, A., Henry, T.B., 2019. Use of fluorescent-labelled nanoplastics (NPs) to demonstrate NP absorption is inconclusive without adequate controls. Science of The Total Environment 670, 915–920. https://doi.org/10.1016/j.scitotenv.2019.03.194