

Use of non-ablation in Shrimp hatcheries: production and animal welfare

Simão Zacarias, PhD

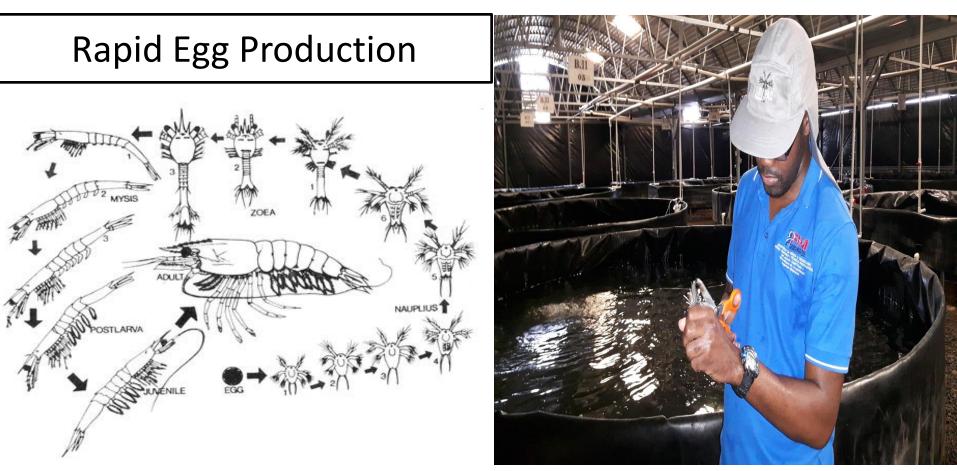
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BE THE DIFFERENCE

Shrimp Hatcheries: Unilateral eyestalk ablation



Eyestalk ablation X Welfare Issues



Non-ablation PROJECT part 1



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Reproductive performance and offspring quality of non-ablated Pacific white shrimp (*Litopenaeus vannamei*) under intensive commercial scale conditions

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Experimental design

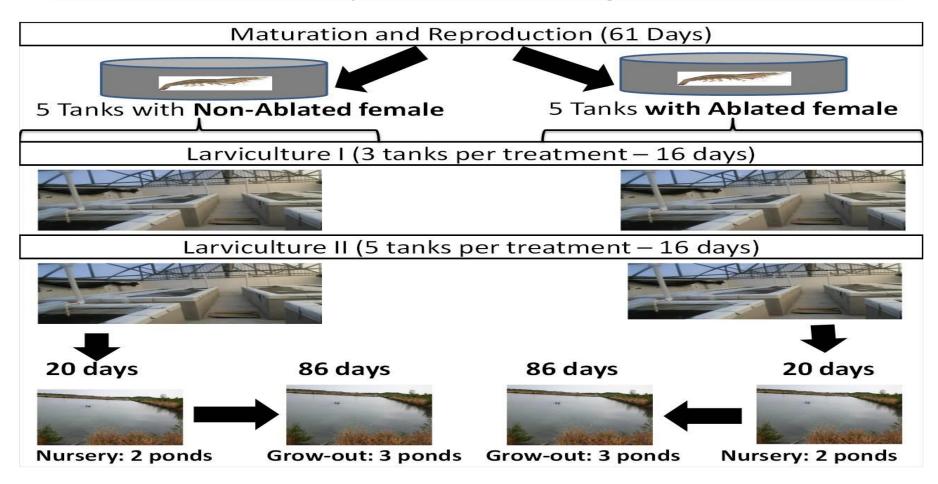


Table 1: Reproductive performance of non-ablated and ablated female

| Reproductive Performance | | | | | | | |
|--------------------------------------|-------------------------------|-------------------------------------|--|--|--|--|--|
| Study I | NAF | AF | | | | | |
| Mating success per day (%) | 3.2 ± 0.0^{b} | 7.6 ± 0.0^{a} Sex ratio 1:2 | | | | | |
| Spawning event day (%) | 90.1± 0.1 ^a | 95.5 ± 0.0^{a} | | | | | |
| Hatching rate per day (%) | 78.0 ± 0.0^{a} | 81.7 ± 0.0^{a} | | | | | |
| Mortality of female per day (%)*** | 1.3 ± 0.0^{b} | 2.3 ± 0.0^{a} | | | | | |
| Number of eggs/spawned female/day | $142\;413\pm4558\;{}^{a}$ | $116\ 752 \pm 3568\ ^{b}$ | | | | | |
| Number of nauplii/spawned female/day | $112\ 610\pm4923\ ^{a}$ | 95 127 ± 2954 ^b | | | | | |
| Number of eggs/tank/day | 811 004 ± 86 858 ^b | 1440 285.7 \pm 116 344 $^{\rm a}$ | | | | | |
| Number of nauplii/tank/day | $653\ 004 \pm 73\ 466\ ^{b}$ | 1186 450 \pm 103 853 $^{\rm a}$ | | | | | |

NAF - non-ablated; **AF** - ablated female

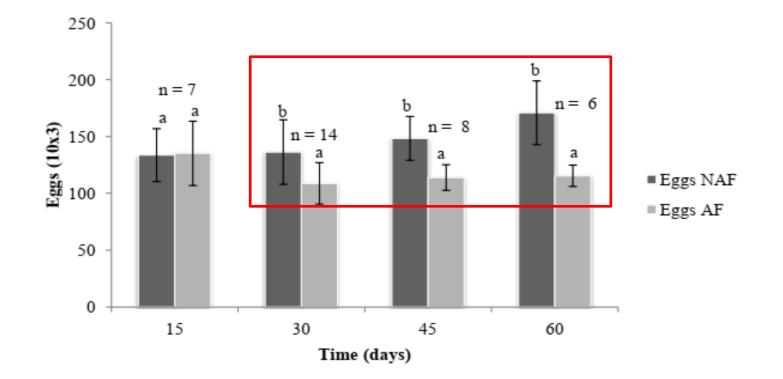


Fig. 1: Egg production per non-ablated (NAF) and ablated female (AF) over time

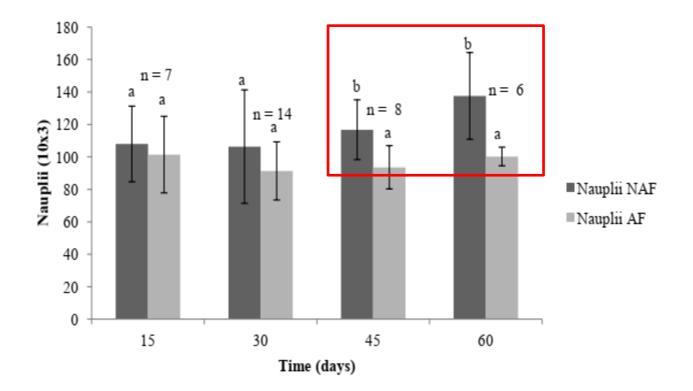


Fig. 2: Nauplii production per non-ablated (NAF) and ablated female (AF) over time

Larval growth and development of non-ablated and ablated female

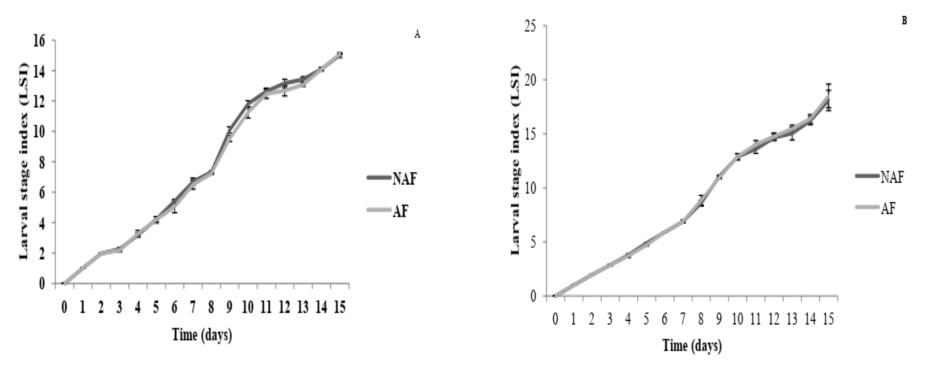


Fig. 3: Larval stage index from non-ablated (NAF) and ablated female (AF)

Table 3: Growth performance, final survival and survival of PLs from nonablated (NAF) and ablated female (AF)

| Study I | Larviculture I | | Larviculture II | | | |
|----------------------|--------------------------|--------------------------|--------------------------|---------------------|--|--|
| Parameters | NAF | AF | NAF | AF | | |
| Survival to SST (%) | 94.7 ± 2.6^{a} | 85.7 ± 6.1^{a} | 97.4 ± 0.2^{a} | 94.9 ± 0.5^{b} | | |
| Final survival (%) | $48.0\pm8.7^{\text{ a}}$ | 41.7 ± 7.0^{a} | 48.8 ± 2.6^{a} | 43.9 ± 5.6^{a} | | |
| Final weight (mg) | $3.4\pm0.3~^a$ | $3.7\pm0.2^{\text{ a}}$ | 6.5 ± 0.6^{a} | $6.7\pm0.8^{\ a}$ | | |
| Study II | Larvice | ulture III | Larviculture IV | | | |
| Parameters | NAF | AF | NAF | AF | | |
| Eggs diameter (µm)* | 269.6 ± 8.5^{a} | 264.3 ± 1.3^{a} | 282.4 ± 1.1^{a} | 282.9 ± 2.0^{a} | | |
| Nauplii length (µm)* | 437.7 ± 5.4^{a} | 451.0 ± 2.8^{a} | 449.4 ± 3.1^{a} | 445.2 ± 2.9^{a} | | |
| Survival to SST (%) | $99.0\pm0.0~^a$ | 96.0 ± 1.0^{b} | 90.0 ± 0.6^{a} | 87.7 ± 0.9^{b} | | |
| Final survival (%) | $42.6\pm3.7^{\text{ a}}$ | $42.1\pm3.8~^{a}$ | 43.8 ± 3.3^{a} | $41.0\pm6.0~^{a}$ | | |
| Final weight (mg) | $6.7\pm0.4~^a$ | 6.2 ± 0.1 ^a | 5.0 ± 0.6 ^a | $5.3\pm0.9^{\ a}$ | | |
| PLs length (mm) | 8.9 ± 0.3 ª | 8.7 ± 0.0^{a} | 8.6 ± 0.4 ª | 8.6 ± 0.4 ª | | |

Table 4: Growth performance and final survival of offspring from nonablated (NAF) and ablated female (NF)

| Nursery | Stu | dy I | Study II | | | |
|--------------------------------|-----------------|-----------------|------------------|----------------|--|--|
| Parameters | NAF | AF | NAF | AF | | |
| Final Weight (g) | 0.5 ± 0.0 | 0.6 ± 0.0 | 0.8 ± 0.0 | 0.8 ± 0.1 | | |
| Weekly Growth (g) | 0.2 ± 0.0 | 0.2 ± 0.0 | 0.2 ± 0.0 | 0.2 ± 0.0 | | |
| Final survival (%) | 40.2 ± 7.5 | 45.4 ± 3.0 | 89.2 ± 2.3 | 92.2 ± 1.8 | | |
| FCR* | 0.9 ± 0.1 | 0.7 ± 0.1 | 0.9 ± 0.1 | 0.8 ± 0.1 | | |
| Yield (Kg/ha)/ (g/m3) | 273.7 ± 32.8 | 340.6 ± 41.1 | 165.4 ± 14.6 | 188.4 ± 22.0 | | |
| Grow-out | | | | | | |
| Parameters | NAF | AF | NAF | AF | | |
| Initial weight (g) | 0.6 ± 0.0 | 0.6 ± 0.0 | 0.8 ± 0.0 | 0.8 ± 0.0 | | |
| Final weight (g) | 14.7 ± 0.5 | 14.9 ± 0.2 | 14.3 ± 0.0 | 14.5 ± 0.3 | | |
| weight gain (g) | 14.1 ± 0.5 | 14.2 ± 0.2 | 13.5 ± 0.0 | 13.7 ± 0.3 | | |
| Weekly Growth (g) | 1.2 ± 0.0 | 1.2 ± 0.0 | 3.1 ± 0.0 | 3.2 ± 0.01 | | |
| SGR (%)** | 3.7 ± 0.0 | 3.7 ± 0.0 | 8.9 ± 0.0 | 8.9 ± 0.1 | | |
| Final survival (%) | 51.7 ± 0.9 | 47.7 ± 2.5 | 93.0 ± 1.4 | 95.9 ± 0.8 | | |
| FCR | 1.3 ± 0.1 | 1.3 ± 0.1 | 0.7 ± 0.0 | 0.8 ± 0.0 | | |
| Yield (Kg/ha)/ $(g/m^3)^{***}$ | 1875.2 ± 27.6 | 1776.6 ± 82.8 | 591.6 ± 8.6 | 617.7 ± 7.5 | | |

Key points

1. Egg commercial production with non-ablated female can depend on SEX RATIO change

2. Non-ablation does not affect larval, post-larlavae and juveniles growth performance and survival

3. Offsping of non-ablated animals might be more resistant to environmental stress

Non-ablation PROJECT part 2



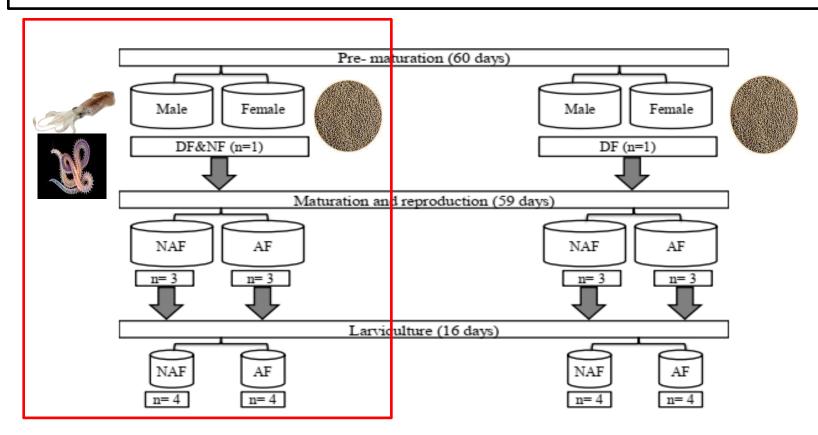
Literature Review





Effect of pre-maturation conditioning on **broodstock reproductive performance** and **offspring quality** of nonablated Pacific white shrimp (*Litopenaeus vannamei*)

Experimental design



DF – Dry feed; **NF** – Natural Feed; **NAF** - Non-ablated female; **AF** - Ablated female

Results

Table 1: Sperm quality during pre-maturationDF – Dry feed; NF – Natural Feed

| Time (Days) | | 1 | | 22 | | 45 | Signficance | | |
|----------------------------------|-------------------|-------------------|-----------------|-------------------|-------------------|-------------------|-------------|--|--|
| Parameters | DF&NF | DF | DF&NF | DF | DF&NF | DF | F T FxT | | |
| GSI (%) (n= 10) | $1.5\pm~0.3^{ab}$ | $1.7\pm~0.6^{ab}$ | $0.9\pm\ 0.1^b$ | 2.4 ± 0.2^{a} | $1.7\pm~0.4^{ab}$ | $1.5\pm~0.3^{ab}$ | ns ns * | | |
| HPSI (%) (n= 10) | $3.5\pm\ 0.2$ | 3.5 ± 0.3 | 3.4± 0.1 | $4.2\pm\ 0.2$ | 3.1 ± 0.3 | 3.1 ± 0.3 | ns * ns | | |
| Spermatophore weight (mg) (n= 7) | 35.2 ± 2.5 | 30.4 ± 2.9 | $38.5\pm~4.6$ | $46.0\pm~4.6$ | $43.0\pm~3.3$ | $50.6\pm~5.9$ | ns * ns | | |
| Sperm Count (10^6) (n= 7) | 17.5 ± 2.5 | 11.8 ± 1.9 | 24.1 ± 4.5 | 28.9 ± 5.2 | 35.6 ± 3.2 | 24.1 ± 4.0 | ns * ns | | |
| Dead Sperm (%) $(n=7)$ | 21.1 ± 7.4 | $22.8\pm~7.4$ | $28.2\pm\ 8.7$ | $17.2\pm~3.8$ | $28.3\pm\ 6.0$ | 32.2 ± 2.7 | ns ns ns | | |



| | DF&NF DF | | Sign | nific | cance | | |
|--------------------------------------|-----------------------|---------------------|--------------------|-----------------------|-------|----|-----|
| Parameters | NAF | AF | NAF | AF | F | A | FxA |
| Mating success per day (%) | $5.8\pm~0.5$ | $6.9\pm~0.6$ | 3.8 ± 0.1 | $7.0\pm~0.7$ | ns | * | ns |
| Spawning event per day (%) | 92.1 ± 1.9 | $90.0\pm\ 0.9$ | $92.3\pm~0.7$ | 91.8 ± 1.9 | ns | ns | ns |
| Hatching rate per day (%) | 55.5 ± 1.0 | 53.1 ± 1.4 | 50.6 ± 1.4 | 51.3 ± 1.0 | * | ns | ns |
| Fertilization rate (%) | $78.4\pm~0.5^{\rm a}$ | 70.5 ± 1.4^{b} | 69.7 ± 2.1^{b} | $68.6\pm~0.5^{\rm b}$ | * | * | * |
| Number of eggs/spawned female/day | 158090 ± 8212 | 140364 ± 2351 | 169938 ± 2341 | 146582 ± 3372 | ns | * | ns |
| Number of nauplii/spawned female/day | 85708.1 ± 1984.1 | 71942 ± 2261 | 85049 ± 3180 | 74474 ± 2109 | ns | * | ns |
| Number of eggs/tank/day | 924598 ± 70823 | 1142248 ± 66564 | 851764 ± 46620 | 1165342 ± 24034 | ns | * | ns |
| Number of nauplii/tank/day | 506501 ± 34489 | 590934 ± 35189 | 453077 ± 25441 | 594259 ± 13703 | ns | * | ns |
| Mortality of female per day (%) | 5.0 ± 1.2 | 3.8 ± 0.3 | 3.5 ± 0.1 | $4.9\pm~0.9$ | ns | ns | ns |

DF – Dry feed; **NF** – Natural Feed; **NAF** - Non-ablated female; **AF** - Ablated female

| Table 3. | | Parameters (mg/g)* | Squid | Polychaete | Dry Feed |
|-------------|---|-----------------------|-----------------------|----------------------|-------------------------|
| | ר | Proteins | 907.2 ± 23.8^{a} | 414.0 ± 10.3^{b} | $357.7 \pm 1.5^{\circ}$ |
| | | Carbohydrates | 30.3 ± 4.4^{b} | 46.6 ± 3.7^{b} | 274.4 ± 11.0^{a} |
| io_ | | Total lipids | 121.9 ± 1.3^{b} | 137.7 ± 1.8^{a} | 117.4 ± 0.1^{b} |
| sit | | Total saturated | $16.6\pm\ 0.4^a$ | 15.3 ± 1.3^{a} | 17.7 ± 0.3^{a} |
| composition | | Total monounsaturated | 6.3 ± 0.0^{b} | 3.6 ± 0.4^{c} | $23.5\pm~0.3^{a}$ |
| | | Total n-6 PUFA | 1.3 ± 0.0^{b} | $0.9\pm\ 0.0^c$ | $25.5\pm~0.4^a$ |
| | | Total n-3 PUFA | 31.5 ± 1.3^{a} | 20.2 ± 0.4^{b} | $9.5\pm0.1^{\circ}$ |
| Feed | | 18:2n-6 | $0.2 \pm 0.0^{\circ}$ | 0.9 ± 0.0^{b} | 24.8 ± 0.4^{a} |
| Ľ۳ | | 20:4n-6 | 0.4 ± 0.0^{a} | $0.4\pm~0.0^a$ | 0.3 ± 0.0^{a} |
| | | 20:5n-3 | 8.1 ± 0.4^{a} | 8.1 ± 0.4^{a} | 3.7 ± 0.1^{b} |
| | - | 22:6n-3 | 22.5 ± 0.9^{a} | 3.1 ± 0.4^{b} | $0.2 \pm 0.1^{\circ}$ |

Table 4: Larval growth performance and survival

Zacarias et al. Under prep.

| | DF&NF | |] | DF | | Significance | | |
|-----------------------------|----------------|----------------|----------------|----------------|----|--------------|-----|--|
| Parameters | NAF | AF | NAF | AF | F | Α | FxA | |
| LSI at Z1 | 1.0 ± 0.0 | 1.0 ± 0.0 | 1.0 ± 0.0 | 1.0 ± 0.0 | ns | ns | ns | |
| LSI at M1 | 3.9 ± 0.0 | 3.7 ± 0.2 | 3.7 ± 0.1 | 3.9 ± 0.1 | ns | ns | ns | |
| LSI at PL1 | 6.7 ± 0.1 | 6.6 ± 0.1 | 6.6 ± 0.1 | 6.7 ± 0.09 | ns | ns | ns | |
| LSI at PL10 | 15.4 ± 0.2 | 15.3 ± 0.2 | 15.6 ± 0.1 | 15.6 ± 0.1 | ns | ns | ns | |
| Survival to SST PL10-11 (%) | 88.5 ± 2.9 | 82.7 ± 5.1 | 90.0 ± 2.5 | 80.0 ± 1.2 | ns | * | ns | |
| Final weight (mg) | 2.2 ± 0.0 | 2.2 ± 0.3 | 2.4 ± 0.3 | 2.2 ± 0.0 | ns | ns | ns | |
| Final Survival (%) | 37.1 ± 3.9 | 34.8 ± 1.6 | 41.0 ± 3.7 | 30.4 ± 2.2 | ns | ns | ns | |
| Parameters (mg/g) | NAF | AF | NAF | AF | F | Α | FxA | |
| Total lipids | 22.9 ± 1.4 | 20.0 ± 0.4 | 19.3 ± 0.8 | 19.7 ± 0.9 | ns | ns | ns | |
| Total saturated | 2.4 ± 0.1 | 2.3 ± 0.0 | 1.8 ± 0.4 | 2.4 ± 0.1 | ns | ns | ns | |
| Total monounsaturated | 2.0 ± 0.1 | $1.9\pm\ 0.0$ | 1.5 ± 0.3 | 2.2 ± 0.1 | ns | ns | ns | |
| Total n-6 PUFA | 1.5 ± 0.3 | 1.5 ± 0.0 | 1.2 ± 0.2 | 1.7 ± 0.1 | ns | ns | ns | |
| Total n-3 PUFA | 1.9 ± 0.2 | 1.9 ± 0.0 | 1.4 ± 0.3 | 1.9 ± 0.1 | ns | ns | ns | |
| 18:2n-6 | $1.0\pm~0.0$ | $1.1\pm\ 0.0$ | 0.8 ± 0.2 | 1.2 ± 0.1 | ns | ns | ns | |
| 20:4n-6 | 0.3 ± 0.0 | $0.3\pm\ 0.0$ | 0.2 ± 0.0 | 0.3 ± 0.0 | * | ns | ns | |
| 20:5n-3 | $0.7\pm\ 0.1$ | $0.8\pm\ 0.1$ | 0.6 ± 0.1 | $0.8\pm~0.0$ | ns | ns | ns | |
| 22:6n-3 | $0.9\pm~0.0$ | $0.9\pm~0.0$ | 0.7 ± 0.0 | 0.9 ± 0.0 | ns | ns | ns | |
| | | | | | - | | | |

DF – Dry feed; NF – Natural Feed; NAF - Non-ablated female; AF - Ablated female

Key points

1. Supplementing squid and polychaete in pre-maturation is a potential key step to success of non-ablated shrimp female

2. Non-ablation does not affect larval and post-larlavae growth performance and survival

3. Offspring of non-ablated animals might be more resistant to environmental stress

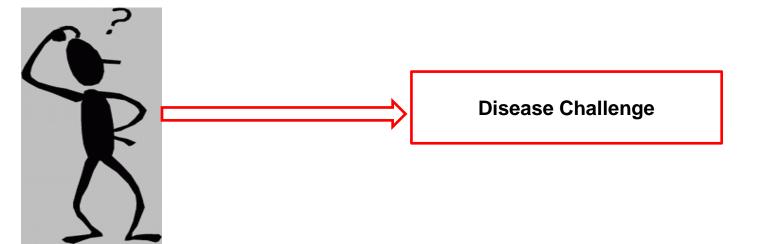
Non-ablation PROJECT part 3



Salinity stress test: Non-ablated shrimp broodstock produced ROBUST OFFSPRING

Focus :

Is there a link between non-ablation and the "fitness" of the offspring?





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Increased robustness of postlarvae and juveniles from non-ablated Pacific whiteleg shrimp, *Penaeus vannamei*, broodstock post-challenged with pathogenic isolates of *Vibrio parahaemolyticus* (Vp_{AHPND}) and white spot disease (WSD)

Simão Zacarias^{a,*}, Daniel Fegan^b, Siriroj Wangsoontorn^b, Nitrada Yamuen^c, Tarinee Limakom^c, Stefano Carboni^a, Andrew Davie^a, Matthijs Metselaar^d, David C. Little^a, Andrew P. Shinn^{a,c,d}





Experimental Set Up

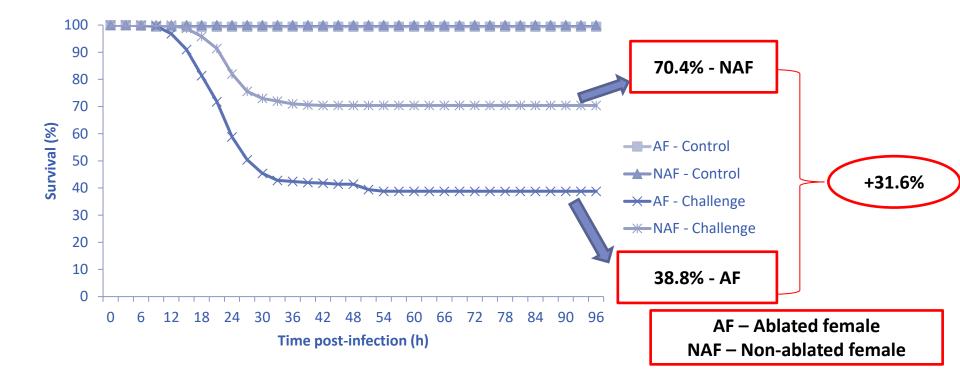


1. Challenge *P. Vannamei* Postlarvae with AHPND/EMS

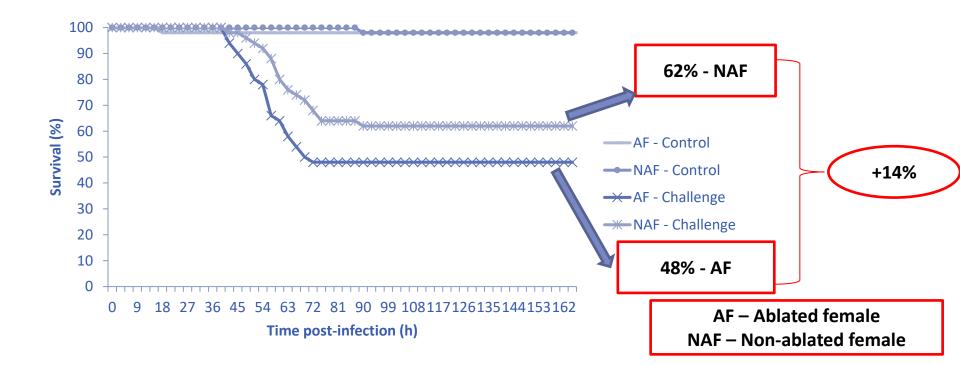
2. Challenge *P. Vannamei* Juveniles with WSD



Results 1. Survival of *P. vannamei* Postlarvae challenged with and without AHPND/EMS



2. Survival of *P. vannamei* Juveniles challenged with and without WSSV



Key points

1. Offspring of non-ablated female are robust and more resistant to typical diseases

2. Validation of the data under commercial conditions are still required

3. Understanding mechanistic factors contributing to robustness improvement of offspring from non-ablated is also required

Final considerations

1. Adopting Non-ablation will require change in some practices including SEX RATIO MANIPULATION and/or include or improve pre-maturation conditioning

2. Non-ablation does not affect larvae, post-larlavae and juvenile growth performance and survival under normal conditions

3. Offspring of non-ablated female are more resistant to diseases

Other Key Considerations

1. Broodstock Genetic or Strain: different response to non-ablation

2. Selective breeding program: Increase selection frequency

3. Maturation system: RAS or flowthrough

Global use of Non-Ablation





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THANK YOU!



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