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1 **Potential combinations of mabé, keshi and cultured pearl production from**
2 **colourful hatchery-produced *Pinctada margaritifera***

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17

1 **Abstract**

2 Aquaculture of nacreous gems, such as cultured pearls, mabé or keshi, is done mostly using
3 different mollusc species grown in countries of the Indo-Pacific region. To date, no single
4 species has been exploited for the simultaneous generation of more than one of these
5 bioproducts, but all require animals with colourful shells. Historically, *Pinctada* species have
6 mainly been used for nucleated pearl production, selecting the rarer colourful individuals to
7 be used as graft donors. By contrast, colourful *Pteria* species have mostly been used for mabé
8 production, as the grafting operation for pearl production is associated with low yield. In this
9 study, we report the potential for cumulating cultured pearl and mabé (MP), or keshi and
10 mabé (MK) production, using a colourful hatchery-produced G2 family of *P. margaritifera*.
11 For these trials, MP and MK combinations were compared with the operations to produce
12 pearls (P), mabé (M) or keshi (K) alone in an experimental design using groups of small and
13 large recipients from the G2 family. Results showed no significant impact of combining
14 operation types on subsequent pearl weight, keshi weight, or mabé thickness within recipient
15 oyster size group. By contrast, significant differences were observed between the large and
16 small recipients. The small group produced the thickest mabé, while the large group produced
17 the heaviest pearls and keshi. These contrasting results revealed: 1) the relative independence
18 between the two tissues capable of biomineralisation activities, the mantle (shell and mabé
19 growth) and the pearl sac (pearl or keshi growth); 2) the potential compensatory growth of the
20 small recipient oyster group, which had the highest shell growth performance; and 3) the
21 regulation capacity of the larger oyster group of pearl sac activity. With the same growing
22 area and number of cultured oysters, it would be possible for the *P. margaritifera* pearl
23 industry to benefit from hatchery propagation of selected colourful shell and produce valuable
24 keshi and mabé together with cultured pearls.

25

- 1 **Keywords**

- 2 Pearl oyster; *Pinctada margaritifera*; Mabé; Keshi; Pearls

1 Introduction

2 Mollusc shell is produced by the mantle tissue, which has biomineralisation capacities
3 that carry out shell biosynthesis. The pearl oyster industry exploits this property of the mantle
4 to artificially produce unique gems from living organisms by nucleated pearl production in
5 species of the *Pinctada* and *Pteria* genera. In this process, nacre secretion from mantle tissue
6 covers a foreign element. In the case of cultured pearl production, two animals are required: a
7 donor, from which a small piece of mantle tissue (the *saibo*) is dissected and a recipient, into
8 which this tissue is inserted with a round bead of nacre (a nucleus, made of mussel shell in
9 French Polynesia) positioning the nucleus and graft in the gonad (Gervis and Sims, 1992;
10 Taylor and Strack, 2008). This graft process is commonly called "seeding" or "grafting" and
11 concerns mainly three species: *P. fucata*, *P. margaritifera* and *P. maxima*. As a result of
12 nucleus rejection during the culturing process, a by-product, called *keshi* (small non-nucleated
13 pearls entirely composed of nacre) can also be harvested from these species. In half-pearl
14 (mabé) production, only one animal is required: a hemispherical nucleus of varying shape
15 (usually made of plastic) is pasted onto the interior surface of the shell of a recipient pearl
16 oyster. This technique has mainly been used to produce mabé from the winged pearl oyster,
17 *Pteria penguin*, which is not used for mass cultured nucleated pearl production (Matlins,
18 1996; Gordon *et al.*, 2018). Mabé are not as valuable as cultured pearls, but are easier to
19 produce. They require less time to grow and less specialized skills to implant multiple nuclei
20 in the same recipient. In contrast to cultured pearls, however, mabé require processing after
21 harvest (Kishore *et al.*, 2015).

22 Cultured pearls produced from the black-lipped pearl oyster *P. margaritifera* mostly
23 come from French Polynesia (around 90% of world production). This unique expansion is
24 mostly due to the reliable, continuous and adequate supply of wild spat collected from several
25 specific lagoons where it settles at high density (Ky *et al.*, 2015). Other countries of the

1 Pacific region, such as the Cook Islands, Fiji or Micronesia, do not have this advantage of
2 wild settlement, which constrains the development of *P. margaritifera* pearl farming
3 industries in these areas (Cartier *et al.* 2013). Like species from the *Pteria* genus (*P. penguin*
4 and *P. sterna*) (Acosta-Salmón, 2003), *P. margaritifera* has one of the largest ranges of nacre
5 and pearl colours of any pearl oyster species across all its developmental stages (Ky *et al.*,
6 2017a, Ky *et al.*, 2018a and 2018b). Some countries and companies produce both cultured
7 pearls and mabé, mainly using different species. This is the case in Fiji for the tandem species
8 *P. margaritifera* and *P. penguin* and in southern Japan (Amami Island), for *P. maxima* and *P.*
9 *penguin* (Kishore *et al.*, 2018). In French Polynesia, mabé production is not established, as
10 *Pteria* species are not present and highly colourful individuals of *P. margaritifera* are rare and
11 mostly used as donors for cultured pearl production (Ky *et al.*, 2017a). As the Polynesian
12 pearl industry matures and larval culture ceases to be a bottleneck, massive hatchery
13 production, begun in 2015, will assume a role in the supply of colourful pearl oysters to the
14 industry (Ky and Devaux, 2016a). Hatchery development has contributed to a great increase
15 in the frequency of previously rare colourful phenotypes, which could also now be used as
16 recipient oysters to also produce mabé (Ky *et al.*, 2016b).

17 This study is the first to report the potential of combining graft and mabé
18 implantations in a double operation on the same animal. Such an approach should allow the
19 simultaneous production of cultured pearls and mabé (MP) or valuable keshi and mabé (MK),
20 but would require selection of a colourful *P. margaritifera* recipient. By comparison with
21 single operations for cultured pearl production only (P), specific keshi production (K), or
22 mabé implantations only (M), the experiment was designed to examine the possible impact of
23 double *vs.* single operation combinations, on cultured pearl/ keshi weight and mabé thickness.
24 The experimental culture of these five graft/ implantation combinations (M, P, K, MP and
25 MK) was performed under pearl farm conditions, using two groups, large and small, of 200

- 1 individual recipient oysters each, providing from a single multi-parental second generation of
- 2 hatchery-produced oysters. The information generated is intended to show the potential for
- 3 diversification of pearl products from *P. margaritifera*, without the need to increase the
- 4 number of cultured animals or farming area.

1 **Materials and methods**

2

3 **Pearl oysters**

4 Pearl oysters used for this experiment were hatchery-produced and cultured on long
5 lines prior to the experiment, which was performed at a commercial pearl farm (Regahiga
6 Pearl Farm & Hatchery), on the island of Mangareva, Gambier archipelago, French Polynesia
7 (23°07'S, 133°58'W). A single colourful multi-parental family was selected for the
8 experiment. This family was a second hatchery generation (G2) and bred from a cross
9 between males and females originating from two distinct multi-parental families (G1) selected
10 for the colour and lustre of their inner shell colour bands. The G1 families were produced
11 from wild individuals (also by multi-parental breeding), which had been also selected for their
12 colourful inner shell colour bands. The G2 pearl oysters were reared on spat collectors, on
13 which they had settled at 20 days post-fertilisation. All breeding and larval rearing procedures
14 were performed as described in Ky *et al.* (2015). The spat collectors were made of black
15 plastic tinsel of 60 cm length, attached at roughly one meter intervals along a 200 meters rope,
16 commonly used in French Polynesia (Ky *et al.*, 2014). Spat collectors were attached in threes
17 to obtain a length of 1.80 m and protected using plastic mesh to prevent predation in the
18 lagoon. Every six months, the spat collectors were washed and rid of their parasites (mainly
19 epibionts) with a high sea water pressure spray. At the start of the experiment, at 2 years old,
20 the oysters were removed from the spat collector, then cleaned and divided visually into two
21 groups according to their shell size/ area: 1) the small shell size group (N = 200), and 2) the
22 large shell size group (N = 200) (see picture on Table 1). The oysters were placed in baskets
23 in groups of 20 according to their size. A plastic wedge was inserted between the shell valves
24 to prevent closure and the baskets were placed in a pond with continuous sea water flow.

25

1 **Experimental design**

2 Operations were performed to produce five treatments consisting of mabé (M), keshi
3 (K), cultured pearl (P), mabé and keshi (MK) and mabé and pearl (MP) production
4 combinations, using the small and large recipient oyster groups, as illustrated in Figure 1. The
5 operations for each combination were performed by the same expert, in batches of 20 oysters
6 of a size/ production combination in a random order to limit time-related effects (for example,
7 20 small oysters for keshi production, then 20 large oysters for mabé implantation etc.). The
8 entire experiment took two days, with a total of 400 oysters (200 small oysters, 200 large
9 oysters; 40 oysters for each of the 5 production combinations).

10 For K and MK, small strips of epithelium were prepared before being transplanted into
11 the recipient oysters; such grafts are pieces measuring approximately 4 mm². For each group
12 of 20 recipient oysters, one donor pearl oyster was used to provide 20 excised grafts from the
13 two valves (10 grafts per valve). All donors came from the same G2 families. The grafter used
14 a speculum to open the oyster valves. The grafter first incised the recipient oyster gonad in
15 which he placed the graft for the K combination, followed by two opposite mabé
16 implantations (one per valve, see Figure 1) for the MK combination. The hemispherical nuclei
17 used for mabé production (M, MK, MP combinations) were made from clear plastic with a
18 base diameter of 9 mm. Polycyanoacrylate gel glue (Super Glue, Duro) was used to attach the
19 hemispherical nuclei to the shell under the mantle. The hemispherical nuclei were attached in
20 two positions; one per valve (see Figure 1). Each nucleus was pressed to the inner shell
21 surface for 5–10 s for complete adhesion (Haws *et al.*, 2006). They were positioned to allow
22 the oysters to be able to close their shells normally (Saucedo *et al.*, 1998). For P and MP, the
23 grafter first incised the recipient oyster gonad into which he placed the nucleus and then the
24 graft (P), followed for MP by the two hemispherical nucleus implantations. The nuclei used
25 for the graft purpose were made from the shells of freshwater mussels (1.8 BU size,

1 equivalent to 5.45 mm diameter, 0.26 g weight; Imai Seikaku Co. Ltd., Japan) which consist
2 of nacreous layers with thickness and hardness of the nacreous layers offering a specific
3 gravity and thermal conductivity (Gervis and Sims, 1992).

4 Oysters from the different treatments were placed in separate subdivisions in
5 transparent retention bags (10 oysters per retention bag) with their hinges facing upwards so
6 that the nucleus could not slip out of place due to the pull of gravity. Traceability and
7 correspondence between oysters (small and large) and the treatment combinations was
8 maintained using coloured plastic labels attached to the retention bags of the rearing system.
9 All oysters were cultured in these retention bags for a period of 9 months and washed every 3
10 months (3 times in all) with a high pressure seawater spray. Mortality was assessed visually
11 and counted at harvest time by assessing the number of oysters with open shells or the
12 presence of shell fragments inside the bags.

13

14 **Quantitative traits measurements of shell, keshi, pearls and mabé**

15 The following four biometric measurements were taken on the recipient oysters at the
16 beginning of the experiments: 1) dorso-ventral measurements (DVM) or shell height, 2) the
17 antero-posterior measurements (APM) or width, 3) thickness, and 4) the total weight (shells +
18 soft tissues) of the recipient oysters. All size measurements were recorded to the nearest 0.01
19 mm using a Vernier calliper and weight recorded using an electronic balance.

20 Harvested cultured pearls and keshi were cleaned by ultrasonication in soapy water
21 (hand washing) with a LEO 801 laboratory cleaner (2-L capacity, 80 W, 46 kHz), then they
22 were rinsed in distilled water. The weight of the nacre on the cultured pearls was measured
23 using a digital balance and the following formula, nacre weight = (cultured pearl weight -
24 nucleus weight). The weight of keshi was directly assessed using the digital balance.

1 Measurement of mabé thickness was realised by cutting each recipient valve through
2 the centre of each mabé using a fixed band saw. Cross sections of the mabé were
3 photographed with a Motic® binocular loupe. As nacre thickness covering the mabé was not
4 equal over its whole surface, the thickness was then measured with ImageJ at the top of each
5 mabé (Figure 2) (Gordon *et al.*, 2018).

6 **Statistical analysis**

7 All analysis were performed using R© version 3.2.3 software (R foundation for
8 Statistical Computing). The significance threshold was set at $p \leq 0.05$. All measures are given
9 as the mean and variability as the standard deviation.

10 Survival rate between size of host oysters and between mabé, keshi and/or pearl graft
11 oyster were tested using proportions test and if significant the pairwise test associated with
12 Bonferroni correction. Quantitative parameters of mabé, keshi and pearls were tested with t-
13 tests when conditions of normality and homoscedasticity were respected, otherwise Mann-
14 Whitney test were performed. Normality and homoscedasticity were verified with Shapiro
15 and Bartlett tests, respectively.

1 **Results**

2 The overall *P. margaritifera* recipient survival rate from the experiments was 81.7 %
3 (81.2 % for the small pearl oyster group and 82.2 % for the large oyster group). No significant
4 differences in nucleus retention rates were found between recipient oysters from the small and
5 large size groups, whatever the P or MP combinations.

6 **Shell size differences between small and large recipient pearl oyster groups**

7 Biometry of the recipient pearl oysters at the beginning of the experiments showed
8 significant differences ($p < 0.001$) between groups for all the four shell biometric traits
9 recorded (Table 1). Differences between small and large oyster groups for DVM, APM, shell
10 thickness and total weight were respectively: +21.0%, +22.2%, +78.4% and +52.7%.

11 After nine months of culture, all the recipient oysters were again measured for the
12 dorso-ventral variable. The large recipient oysters were still significantly bigger (DVM :
13 108.88 ± 36.51 mm) than the small recipient oyster group (93.60 ± 11.33 mm; $p < 0.001$), but,
14 the rate of DVM shell growth between the measurements was 22.56% for the small oysters
15 and only 11.59% for the large ones.

16 **Mabé and cultured pearl production**

17 Mabé and cultured pearls produced by using hatchery selected *P. margaritifera* are
18 illustrated in Figure 3 (a & b).

19 Mabé thickness showed no significant difference between treatments M (mabé
20 implantation only) and MP (mabé implantation + grafting operation) in either large or small
21 recipient oysters. By contrast, mabé thickness produced from the small recipient oyster group
22 was 41% thicker on average ($p < 0.001$) than that produced from the large oyster group
23 overall: 0.53 ± 0.18 mm vs. 0.31 ± 0.10 mm, respectively (Figure 4A).

1 Cultured pearl nacre weight showed no significant difference between treatments P
2 (grating operation) and MP (grafting operation followed by mabé implantation) in either
3 recipient oyster size group. By contrast, the large recipient oyster group produced
4 significantly heavier ($p = 0.003$) cultured pearls (+26%) on average than the small recipient
5 group: $0.62 \text{ g} \pm 0.20 \text{ g}$. vs. $0.46 \pm 0.15 \text{ g}$, respectively (Figure 4B).

6 **Mabé and keshi production**

7 Keshi produced using hatchery selected *P. margaritifera* are illustrated in Figure 3c.

8 Mabé thickness showed no significant difference between treatments M (only mabé
9 implantations) and MK (mabé implantation and keshi production) for either small or large
10 recipient pearl oysters. By contrast, mabé thickness produced from recipient oysters in the
11 small group was significantly ($p < 0.001$) thicker on average (43%) than that produced from
12 recipient oysters in the large oyster group: $0.54 \pm 0.19 \text{ mm}$ vs. $0.31 \pm 0.11 \text{ mm}$, respectively
13 (Figure 5A).

14 Keshi nacre weight showed no significant difference between treatments K (keshi
15 production) and MK (keshi production followed by mabé implantation) in either of the
16 recipient oyster size groups. By contrast, the large recipient oyster group produced keshi with
17 on average twice the weight of those from the small recipient group ($p < 0.001$): 0.14 ± 0.10
18 g. vs. $0.07 \pm 0.04 \text{ g}$ respectively (Figure 5B).

19

1 Discussion

2 The present study is the first to report the possibility of simultaneously producing both
3 valuable mabé and cultured pearls, or valuable mabé and keshi within the same *P.*
4 *margaritifera* recipient oysters. This was done by using selected hatchery-produced oysters to
5 maximise the occurrence of colourful individuals that could be used both as donor and
6 recipient. In French Polynesia, occurrence of colourful oysters from wild are rare (Ky *et al.*,
7 2017a). Production of cultured round pearls and mabé are usually dissociated in terms of
8 mollusc species and genus (*i.e.*: mabé and pearls are not produced from the same species). For
9 mabé production, the genera *Pteria* and *Pinctada* (Taylor and Strack, 2008) and even abalone
10 (Matlins, 1996) have been used. However, traditionally, the term mabé refers to the product
11 from the winged pearl oyster, *Pteria penguin* (Röding, 1758) or "mabé gai" (Southgate *et al.*,
12 2008). *Pteria penguin* is cultured for mabé production in Japan, Australia, the Philippines,
13 Indonesia, Thailand, Vietnam and Tonga (Southgate *et al.*, 2008; Gordon *et al.*, 2019). Other
14 species found along the Gulf of California and the Pacific coast of Mexico also used for mabé
15 production are the related rainbow-lip pearl oyster, *Pteria sterna* (Gould 1851) (Ruíz-Rubio
16 *et al.*, 2006) and *Pinctada mazatlanica* (Hanley, 1856) (Saucedo *et al.*, 1998). Cultured round
17 pearl production concerns mainly three species of the *Pinctada* genus: *P. fucata*, *P. maxima*
18 and *P. margaritifera*. The first two of these species produces lighter coloured pearls than *P.*
19 *margaritifera*. In fact, the "Akoya pearls" produced by *P. fucata*, can be pink, white, silver,
20 cream or yellow (Tong & Shen 2001). In the case of *P. maxima*, golden or silver-white
21 cultured pearls are regarded as superior to yellow or cream ones (Taylor 2002). *P.*
22 *margaritifera* produces a very wide range of pearl colours, from the purest white to the
23 deepest black, and these can have different combinations of main bodycolor and secondary
24 colour (Karampelas *et al.* 2011), passing through every shade of silver, peacock, green,
25 aubergine, purple, golden brown and even rainbow, which can be used to make characteristic

1 multicolour necklaces. By contrast, keshi are not an intentionally cultured product, but a
2 derived/ secondary nacreous product secreted by these three species. At recent auctions in
3 Tahiti (year 2017) rainbow peacock keshi lots (Figure 3c) were sold for a trader price of
4 around 40 euros per gram, which is more expensive than the price of gold per gram. There
5 would be therefore an economic interest in producing these gems. This potential to
6 simultaneously produce both valuable mabé and cultured pearls, or valuable mabé and keshi
7 within the same *P. margaritifera* recipient, is reinforced by the fact that mabé thickness was
8 not affected and did not affect the other product being cultured simultaneously in the same
9 recipient animal; *i.e.* weight of cultured pearls or keshi, regardless of the size class of the
10 recipient at the time of implantation. As the mabé and pearl or mabé and keshi were produced
11 in the same recipient individuals, but by two distinct biomineralizing tissues – the mantle for
12 shell and mabé formations and the pearl sac (formed by proliferation of the mantle tissue of a
13 donor oyster) for pearl or keshi formations – our results reveal the relative independence of
14 these two tissues from a physiological point of view. Indeed, shell and cultured pearl
15 formations are respectively the result of the biomineralization activities of two distinct tissues:
16 the recipient's own mantle and the pearl sac formed from the graft, respectively (Ellis and
17 Haws 1999). Nacre thickness and weight are directly correlated with the nacre
18 biomineralization process in *P. margaritifera*. The epithelial cells from the outer surface of
19 the mantle tissue (lining the inner surface of the shell) are capable of synthesizing different
20 calcium carbonate polymorphs (Wilbur, 1964; Watabe, 1988), which cover the mabé implant
21 or the nucleus (in the case of a mantle tissue graft), as observed by electron microscopy
22 (Zhang and Xu, 2013).

23 Shell size and shape is one of the criteria for multiple mabé implantations. Indeed,
24 efforts have been made for the study of the relationship between the shell dimensions with the
25 optimal number, size, shape and location of hemispherical nucleus implantations for mabé,

1 such as in the works of Saucedo *et al.* (1998). Our results showed that the small *P.*
2 *margaritifera* recipient group, produced up to 40% thicker mabé than their larger
3 counterparts. Although this seems contradictory, it is consistent with the higher shell growth
4 performance observed in the small recipient group, which was double that of the larger oyster
5 group. This greater growth in the smaller group could be attributed to the rearing system
6 transition, between high density rearing on spat collectors to low density rearing in individual
7 retention bags. During their growth on the spat collectors, differences in shell size could be
8 related to unfavourable rearing conditions, that could attributed to food access; particularly
9 considering the common genetic background of all the animals used in this study. The small
10 oysters found on spat collector rearing system would therefore correspond to individuals that
11 experienced unfavourable conditions (growth depression), whereas the large oysters would
12 correspond to individuals that had not experienced such unfavourable conditions, as a result
13 of their physical position on the spat collector. Faster growth from the small oyster group after
14 the transition to the retention bag rearing system could be attributed to compensatory growth,
15 which can be defined as a physiological process whereby an organism accelerates its growth
16 after a period of restricted development to then reach the same weight as animals whose
17 growth was never restricted (Hornick *et al.*, 2000; Jobling, 2010). In a previous study on *P.*
18 *margaritifera* aging 3.5 months old, Pit and Southgate (2003) showed that, given appropriate
19 conditions, small spat (<5 mm) are capable of similar growth rates as larger spat. This
20 phenomenon occurs in a wide range of aquatic animals such as crayfish (*Cherax*
21 *quadricarinatus*) (Stumpf *et al.*, 2010), *Pangasius bocourti* (Jiwyam, 2010), Atlantic halibut
22 (*Hippoglossus hippoglossus*) (Foss *et al.*, 2009) and Chinese shrimp (*Fenneropenaeus*
23 *chinensis*) (Wu & Dong 2001), as well as in *L. vannamei* (Lin *et al.*, 2008). The degree of
24 recovery (catch-up) growth is dependent on the intensity of daily feeding post-restriction and
25 such a compensatory response can be obtained with increased feed utilization efficiency

1 (Stumpf *et al.*, 2010 & 2014). In the present study, as growth of shell and mabé thickness
2 were intimately linked by the biomineralisation activity of the mantle tissue, it is easy to see
3 how the compensatory growth of the small oyster group contributed to both shell and mabé
4 growth performances. The mechanisms of compensatory growth could be (i) an improvement
5 in feed conversion efficiency or (ii) an increase in food intake upon reestablishment of an
6 abundant food supply; both may work simultaneously in some species (Foss *et al.*, 2009). For
7 *P. margaritifera* shell growth, Linard *et al.* (2011) used microscopy to show that the thickness
8 of newly formed aragonite tables was thinnest for oysters that had been treated under a low
9 trophic regime (800 cells/mL *vs.* 15 000 cells/mL).

10 Cultured pearls or keshi weight were not affected by the presence or absence of mabé
11 implants. Indeed, no significant weight difference was observed between individuals with or
12 without mabé implants within either of the recipient size groups. This again underlines the
13 independence of the two biomineralisation tissues: the mantle and the pearl sac. By contrast,
14 pearls and keshi produced by the pearl sac of the recipient oysters were significantly heavier
15 in the large group, than the small one. This was the opposite of what was observed with mabé
16 thickness, which was possibly driven by compensatory growth. Pearl sac biomineralisation
17 activities seemed greater in larger oysters than smaller ones. This same pattern was already
18 noticed in a previous study on *P. margaritifera*, where shell weight was correlated with pearl
19 size (nacre weight and thickness). Indeed, several positive correlations were observed
20 between cultured pearl size and shell biometric parameters, with cultured pearl nacre
21 thickness showing a significant positive correlation with recipient oyster shell thickness,
22 height, and width, and nacre weight correlated with shell thickness (Le Pabic *et al.*, 2016).
23 The recipient oyster can affect pearl development in three key ways, as the nucleus had no
24 direct contact with the recipient oyster (it was enveloped within the pearl sac). First, the

1 recipient oyster regulates the metabolism of the pearl sac, which is dependent upon nutrient
2 supply throughout the culture period. In an appropriate environment, the recipient oyster can
3 supply a high level of nutrients for the pearl sac, and then promote nacre secretion rates for
4 the formation of the cultured pearl. Secondly, the filtration capacity of the recipient oysters
5 could differ according to size, thus impacting the nutrient supply (Yukihira *et al.*, 1998;
6 Pouveau *et al.*, 2000). Thirdly, recipient oysters can regulate the expression of the
7 biomineralization genes in the pearl sac tissue.

8 **Conclusions**

9 For years, commercial pearl culture with *P. margaritifera* has relied mainly on spat
10 collection and/or controlled extractions of wild adults, but in French Polynesia attention has
11 recently shifted towards hatchery research. An increase in the frequency of pearl oysters with
12 colourful shells could be achieved through hatchery propagation, and corresponding
13 individuals could be used as either donors or recipient. This preliminary study opens the way
14 for: 1) simultaneous culture of different nacreous products within a common recipient oyster,
15 and 2) diversification of nacreous products from *P. margaritifera*, with production of
16 colourful and valuable mabé and keshi. The simplicity of mabé and keshi cultures provides
17 opportunities for the development of these alternative products and will allow producers to
18 optimise and maximize the use of their farming areas and stock exploitation. For mabé
19 production, if recipient oysters can be prepared to stimulate compensatory growth prior to
20 implantation; this may offer advantages for the pearling industry. The combination of genetic
21 selection for a fast growing recipient line with physiological preparation/ conditioning (to
22 recover compensatory growth) could benefit combined mabé implantation, together with
23 cultured pearl or keshi production.

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Figure 1. Experimental design showing the five treatment combinations examined in this study for production of mabé (M), keshi (K), cultured pearls (P), mabé and keshi (MK) and mabé and pearl (MP) in *Pinctada margaritifera*. These combinations were applied to groups of small (N = 200) and large (N = 200) recipient pearl oysters.

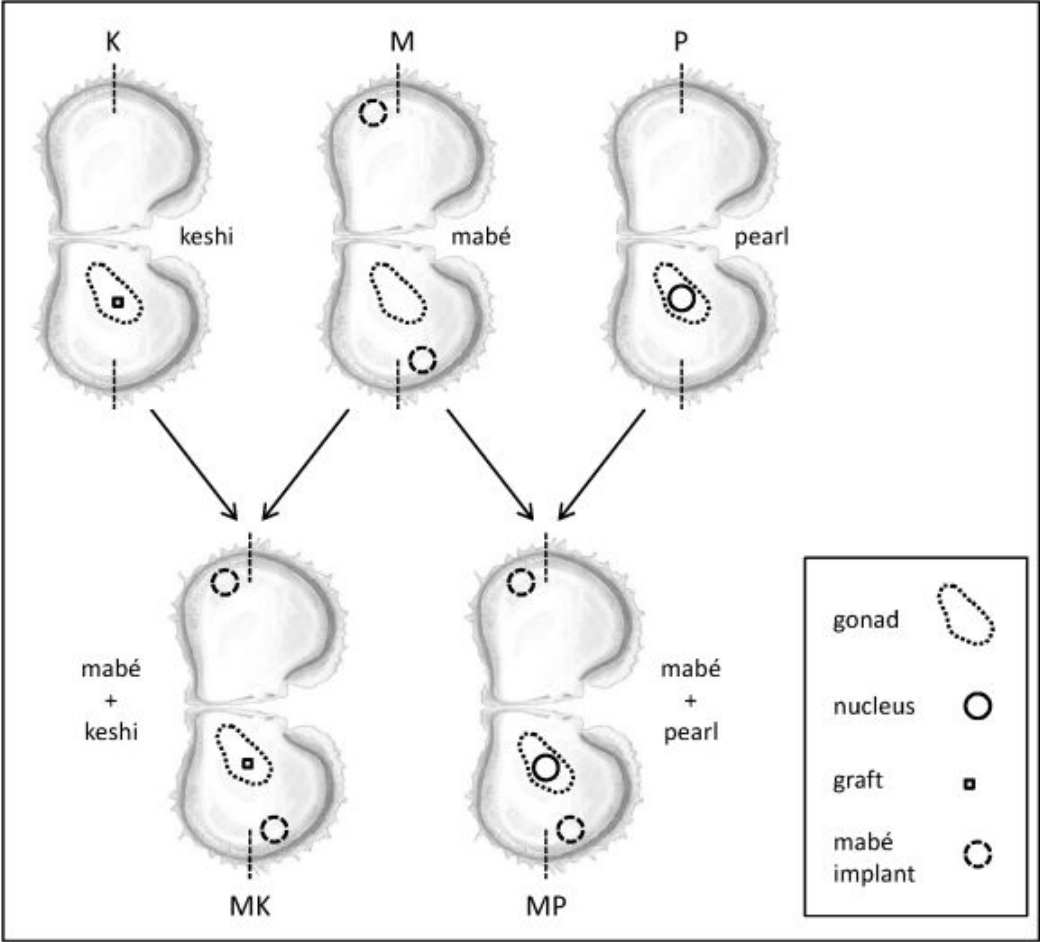


Figure 2. Measurement of nacre thickness at the top of mabé implanted on the inner shell valve of *Pinctada margaritifera*.

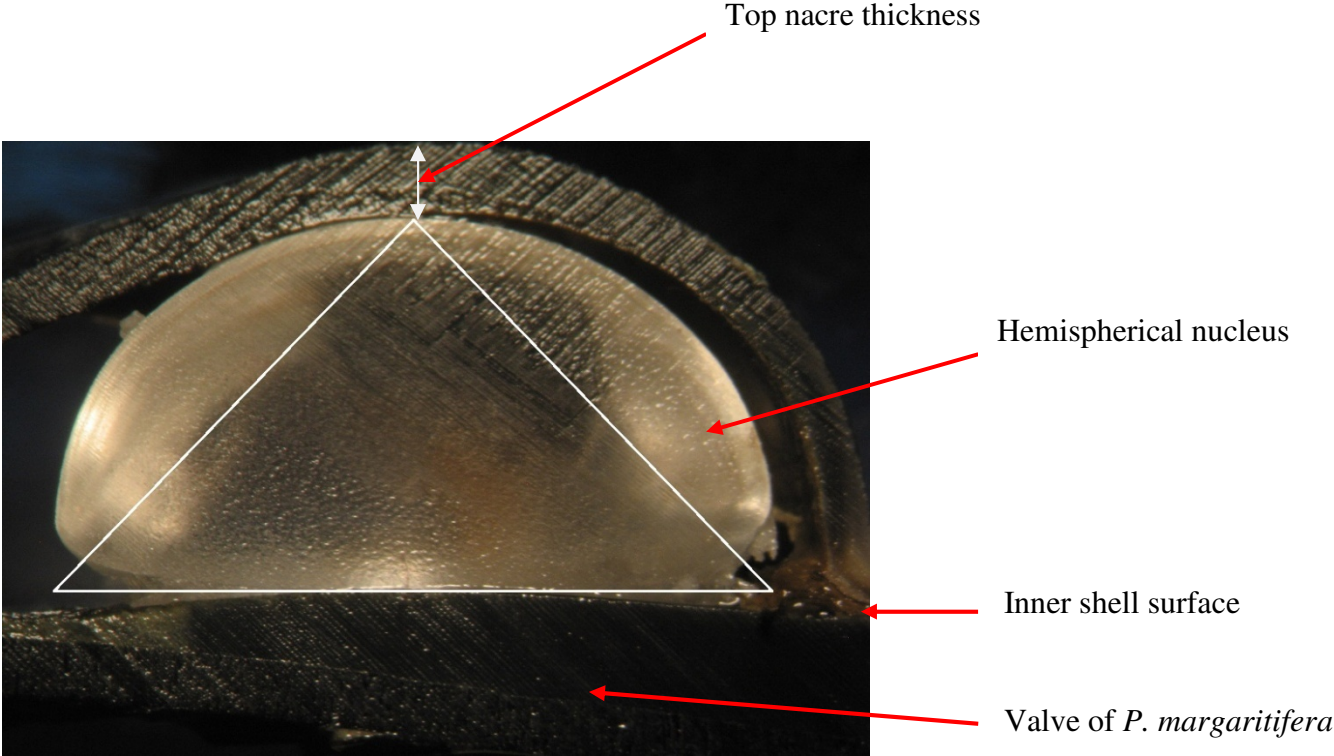
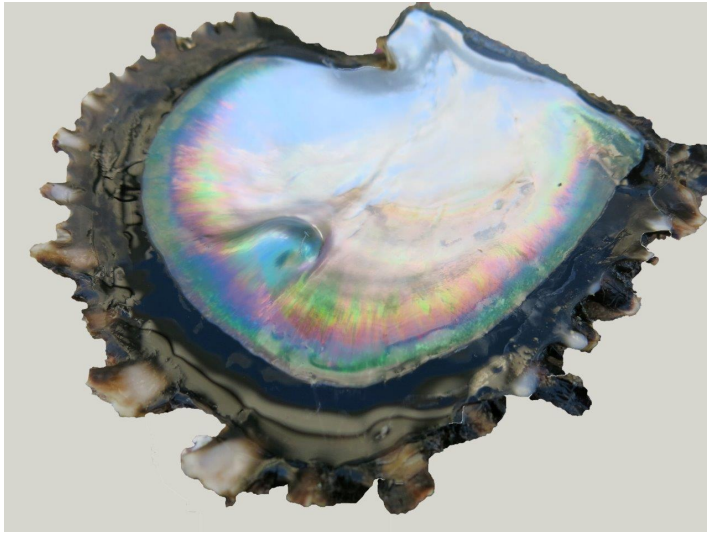


Figure 3. Mabé (half-pearl) (a), cultured pearls (b) and keshi (c) produced from selected hatchery-produced *P. margaritifera*.

a) Mabé



b) Cultured pearls



c) Keshi



Figure 4. Mabé thickness (A) and cultured pearl nacre weight (B) from recipient *Pinctada margaritifera* selected for their small or large shell size, following M, P and MP operations. Letters indicate significant difference between small and large recipient pearl oyster (n = 40 oysters per treatment) groups.

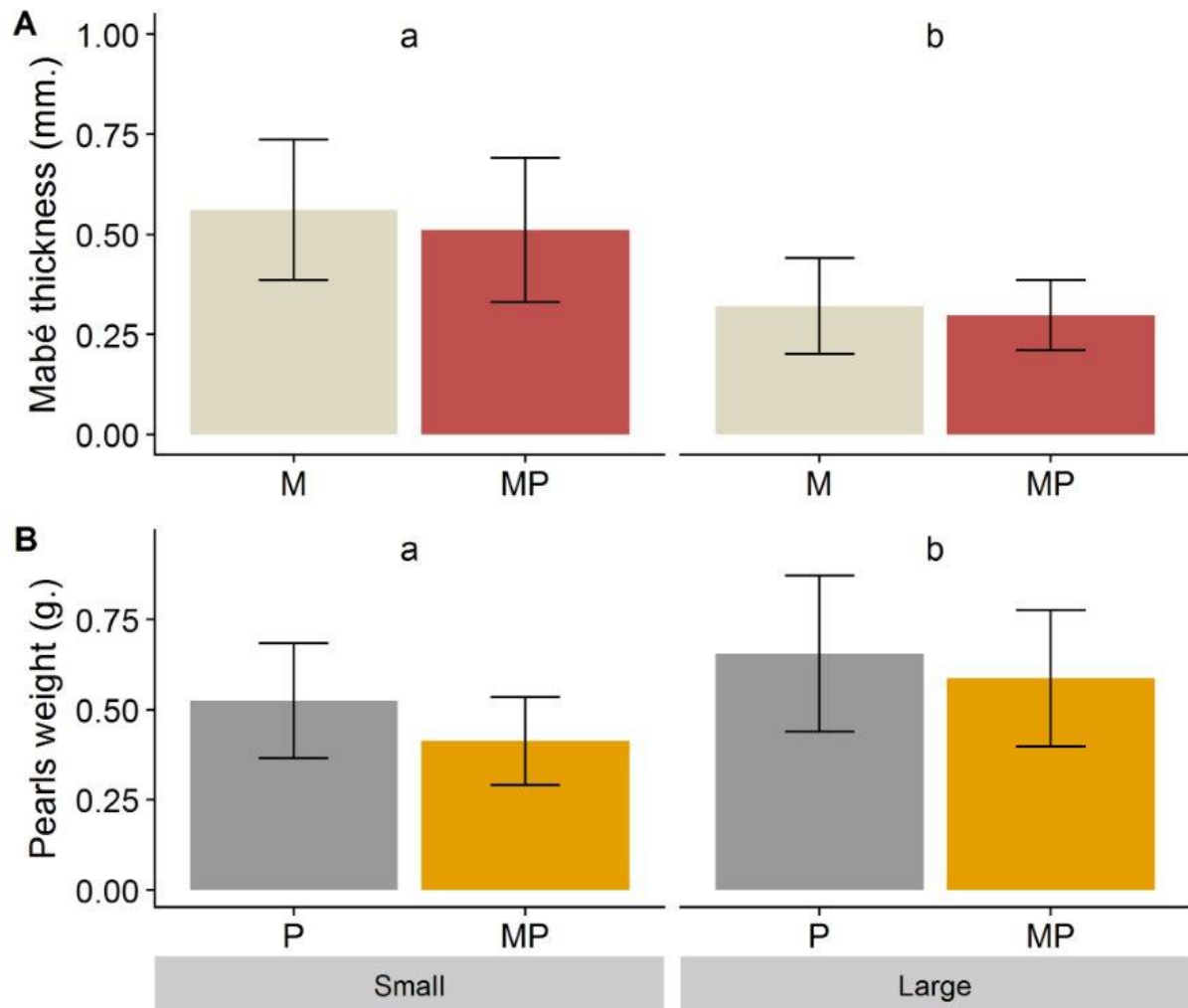


Figure 5. Mabé thickness (A) and keshi weight (B) from recipient *Pinctada margaritifera* selected for their small or large shell size, following M, K and MK operations. Letters indicate significant differences between small and large recipient pearl oyster groups (n = 40 oysters per treatment).

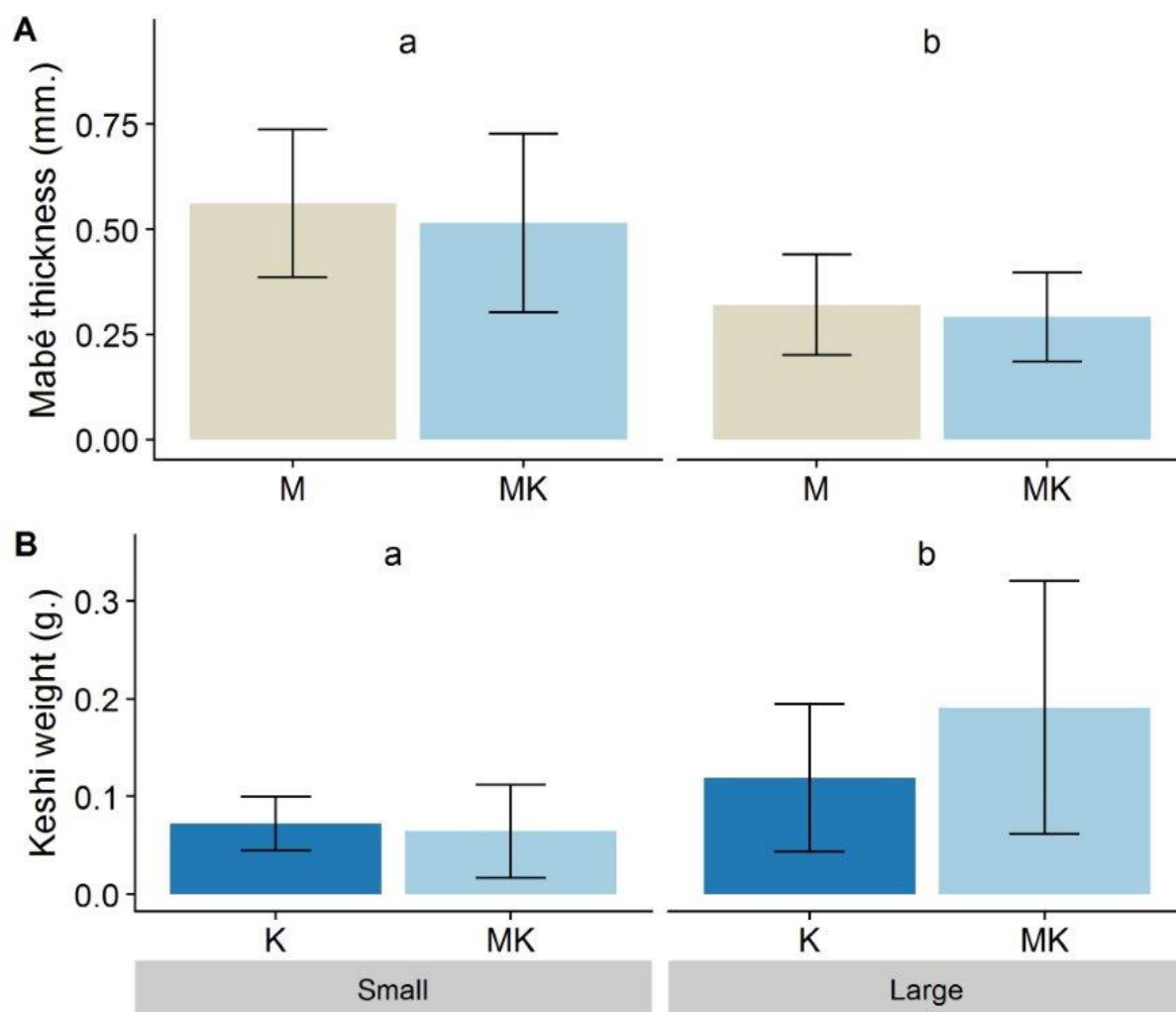


Table 1. Biometric measurements made on hatchery selected *Pinctada margaritifera* at the beginning of the experiments. Four growth traits were recorded: 1) dorso-ventral measurement (DVM), 2) antero-posterior measurement (APM), 3) shell thickness (all of each were recorded to the nearest 0.01 mm using a Vernier caliper), and 4) total weight (shells + soft tissues) of the recipient oysters (using an electronic balance).

	Small	Large
Dorso-ventral \pm SD (mm)	77.02 \pm 4.04	97.52 \pm 7.09
Antero-posterior \pm SD (mm)	76.57 \pm 4.52	98.39 \pm 7.71
Shell thickness \pm SD (mm)	21.20 \pm 2.02	98.39 \pm 7.71
Total weight (shells + soft tissues) \pm SD (g)	55.47 \pm 8.50	117.35 \pm 23.49

Pictures of recipient oysters
(scale of shell size maintained between
small and large recipient oysters)

