



Review

A meta-analysis of growth rate in diploid and triploid oysters

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A B S T R A C T

Around 34 years ago, the first reports on the performance of triploid oysters were published. Since then, triploid oysters have offered many benefits to the oyster industry, such as faster growth, improved meat quality, partial sterility, and increased survival due to disease resistance. However, the extent of a triploid growth advantage, in particular, can vary across studies, measurement parameters, environmental conditions and husbandry practices. To quantitatively compare diploid and triploid oyster growth rates, a meta-analysis was performed with 29 published studies using triploid oysters produced by chemical induction or by crossing diploid and tetraploid oysters (i.e., “mated triploids”). The difference in growth rate between ploidy was evaluated using natural log transformed response ratios ($\ln [3n/2n]$) in a random-effects model weighted by sample size. The positive response ratios in 126 of the 148 independent experiments showed a significant growth advantage of the triploid over the diploid. On average, mated triploids grew 20% faster than diploids in shell height and 49% faster in whole wet weight. While chemically induced triploids had marginally less growth advantage than mated triploids, growing on average 8% faster than diploids in shell height and 31% faster in whole wet weight. Response ratios for experiments using mated triploids and measuring whole wet weight was significantly affected by species and length of study, while response ratios for experiments using chemically induced triploids and measuring whole wet weight was significantly affected by initial size at deployment. Conversely, response ratios for experiments using mated triploids or chemically induced triploids and measuring shell height were not affected by any tested moderator. The lack of a triploid growth advantage in 15% of the experiments (22/148) could have been influenced by a variety of factors including intraspecific variation, differences in sampling, husbandry practices, and environmental conditions.

1. Introduction

Research into genetic improvements (e.g., polyploidy) correlates with the rise in hatchery produced oysters and a dependence on hatchery produced seed in areas such as, the Pacific Northwest of the U.S. (Clark and Langmo, 1979). Groundbreaking experiments by Stanley et al. (1984) with *Crassostrea virginica* and Allen and Downing (1986) with *C. gigas*, were some of the first to quantify the performance of triploids relative to diploids. By the 1999–2000 season, triploid *C. gigas* accounted for one-third of aquaculture production in Washington and Oregon (Nell, 2002). Soon after, areas on the east coast of the U.S. (Chesapeake Bay) adopted triploid aquaculture for *C. virginica* in response to the collapse of wild oyster stocks. Since 2008, triploids consistently make up around 80 to 95% of total oysters grown in Virginia (Murray and Hudson, 2015; Callam et al., 2016). Nowadays, triploid production for the half-shell market has been adopted worldwide (S. Allen, pers. comm.).

There are two primary techniques to produce triploids: chemical induction or through the mating of a diploid and a tetraploid (i.e., mated triploids). Chemical induction involves either 6-dimethyl-aminopurine (6-DMAP) or cytochalasin B (CB) to retain the first or second polar body thereby blocking meiosis I or meiosis II respectively. CB is a

known carcinogen and a more dangerous chemical to work with than 6-DMAP (Gérard et al., 1999). However, CB has been found to produce a higher percentage of triploids than 6-DMAP (Gérard et al., 1999) and is often the most effective chemical used. Treatment of the eggs at meiosis I must be done within the first 15 min after fertilization as to block the first chromosome division and retain 2N chromosomes. Treatment at meiosis II is done between 15 and 30 min after fertilization to block the second chromosome division and retain 1N chromosomes that are genetically identical, except in cases where there has been recombination. In this way, meiosis I treatment can result in increased heterozygosity (genetic variation) compared to oysters treated at meiosis II. However, treatment at meiosis I is not common commercially as the triploids are harder to induce, have lower survival and the treatment is more likely to produce aneuploids (Gérard et al., 1999; Hand et al., 1999; Guo et al., 1992).

Chemical induction is not reliable in producing a 100% triploid population, while crossing a tetraploid with a diploid achieves very close to pure triploid (Guo and Allen, 1994b). Since its inception in 1994, tetraploids are now produced in North America (e.g. Stone et al., 2013; Dégrement et al., 2012; Ibarra et al., 2017), Europe (e.g. Buestel et al., 2009), Australia (e.g. Nell and Perkins, 2005), Chile (Cultimar; Tongoy, Chile), China (Guo, 2004) and Korea (Guo et al., 2008).

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Tetraploid production is achieved by finding the very small percentage of fecund female triploids, and fertilizing these eggs with sperm from a diploid and blocking the first polar body (Guo and Allen, 1994b). Mated triploids, in turn, are produced by fertilizing female diploid eggs with sperm from male tetraploids. These mated triploids, therefore, receive two sets of chromosomes from the male tetraploid, while chemically induced triploids receive two sets of chromosomes from the female diploid and the two chromosomes are either genetically different (retaining the first polar body) or identical (retaining the second polar body). The distinction in the origin of the extra set of chromosomes is important to note as it could influence differences in triploid performance (Callam et al., 2016; Wang et al., 2002).

Depending on the species and culture conditions, the advantages of triploids can vary from faster growth (Walton et al., 2013; Nell and Perkins 2005), improved meat condition (Garnier-Géré et al., 2002; Barber and Mann, 1991), greater disease resistance (Dégremont et al., 2015) and population control (Guo, 2009). The enhanced performance in triploids can be explained by their partial sterility while faster growth, in particular, is also related to energy reallocation, polyploid gigantism, and increased heterozygosity. Energy reallocation is apparent once oysters reach sexual maturation, where triploids reallocate energy from gametogenesis to somatic growth (Allen and Downing, 1986). Polyploid gigantism helps to explain increased growth prior to sexual maturity, where faster growth in triploids is a result of the increase in cell volume and lack of cell-number compensation (Guo and Allen, 1994a). Only heterozygosity can explain differences in growth between chemically induced and mated triploids. Increased heterozygosity is commonly correlated with faster growth among diploid individuals (Zouros et al., 1988; Alvarez et al., 1989) and is thought to contribute to faster growth in meiosis I triploids compared to meiosis II triploids (Stanley et al., 1984; Hawkins et al., 1994; Mallia et al., 2006) and faster growth in mated triploids compared to meiosis II triploids (Wang et al., 2002). However, studies have found no correlation between increased heterozygosity and triploid growth in other bivalves; *Pinctada martensii* (Jiang et al., 1993), *Mytilus edulis* (Beaumont et al., 1995) and *Mya arenaria* (Allen et al., 1982).

The comparison between triploid and diploid growth and mortality has been found to depend on environmental conditions. Under unfavorable growth conditions (low salinity, low dissolved oxygen, high disease pressure, poor food quality and availability) several studies have cited faster growth in triploids and similar survival to diploids (Garnier-Géré et al., 2002; Smith et al., 2000), faster growth in triploids and lower survival than diploids (Gouletquer et al., 1996; Stanley et al., 1984), or similar growth and lower survival than diploids (Callam, 2013; Cheney et al., 2000). Part of the variation in triploid growth and mortality comes from the difficulty in comparing vastly different waterbodies with “poor” water quality given the complexities of acute (e.g., disease and parasites) and chronic stressors (e.g., low dissolved oxygen, high temperature, low salinity, and harmful algal blooms).

Additionally, measuring oyster growth through morphology and biomass is highly influenced by environmental conditions and poses challenges unlike isodiametric shellfish, such as clams or scallops. Shell morphometry (i.e., shell height, length and width) is influenced by habitat, how the oyster settles on a substrate, how densely packed the oysters are, or, in aquaculture, how they are handled (Harding, 2007; Stone et al., 2013). Whereas biomass (i.e., whole, tissue, and shell weight) is an indicator of food quality, food availability, oyster filtration rate, and fecundity (Chávez-Villalba et al., 2010; Davis, 1994; Li et al., 2009; Cox and Mann, 1992). When determining growth, it is therefore important to use both shell morphometry and biomass to account for effects of the environmental conditions. This study uses meta-analysis to determine whether there is a significant growth advantage of triploid oysters over diploid oysters across a wide range of studies, species, and environmental and physical conditions.

2. Methods

2.1. Literature search

Studies were obtained from several literature databases, including Web of Science, Google Scholar, and Aquatic Sciences and Fisheries Abstracts (ASFA), using combinations of the following relevant keywords: “diploid”, “triploid”, “oysters” and “growth”. Out of 100 results in Web of Science, 2540 results in Google Scholar, and 214 results in ASFA, the list was reduced to 29 independent studies that directly reported growth rates of both diploids and triploids or reported initial and final measurements in shell height, whole wet weight, or both to allow the calculation of an average oyster growth rate. Studies were separated by whether triploids were chemically induced (blocking either polar body I or polar body II during fertilization) or produced through mating a tetraploid and a diploid. When calculating response ratios (described in detail below in Section 2.2), only diploids and triploids of the same species were compared. Comparisons were not made across species, such as between diploid *Crassostrea virginica* and triploid *C. ariakensis*. Six species were included in the dataset: *Crassostrea gigas*, *Crassostrea hongkongensis*, *Crassostrea madrasensis*, *Crassostrea virginica*, *Ostrea edulis*, and *Sassostrea glomerata* (formerly *S. commercialis*).

For the meta-analysis, more than one response ratio was calculated from a single publication if the experiments took place in a unique body of water with different environmental parameters, such as temperature and salinity, given their effects on growth. Initial and final shell height, whole wet weight, or both measurements of diploid and triploid oysters were extracted from each study to calculate the growth rate per day (Eq. (1)). Shell height was defined as the length from the hinge to growing edge. When data were presented in figures, the relevant information was extracted using ImageJ software (Rasband, 2014). In studies with significantly different growth between selectively bred diploid or triploid lines, the selected lines were compared separately. At a minimum, data associated with sample size and species were collected. When possible, other parameters, such as temperature, salinity, tidal height, and grow out gear (e.g., cage, floating bag, and lantern net) were noted.

$$\text{Growth rate} = \frac{\text{Final size} - \text{Initial size}}{\text{Days deployed}} \quad (1)$$

where final and initial size were in grams (whole wet weight) or millimeters (shell height).

2.2. Effect size calculations

All calculations were conducted using the *metafor* package (Viechtbauer, 2010) in the statistical software program, R (R Core Team, 2014). The natural log-transformed ratio of means, also called the response ratio (Eq. 2; Hedges et al., 1999), was chosen to quantify the magnitude of the difference between triploid and diploid growth rates. Response ratios compare the mean difference between an experimental treatment (triploid) and a control treatment (diploid) in a unitless ratio and are commonly used in ecology due to the ease of interpretation and strong statistical properties. A natural log-transformed response ratio of zero would indicate no difference in growth between triploids and diploids. Response ratios greater than zero (lower 95% confidence interval is greater than zero) would indicate that triploids grow faster than diploids.

$$\text{Response ratio} = \ln \left(\frac{X_T}{X_D} \right) \quad (2)$$

where X_T and X_D are the growth rate means of the triploid and diploid oysters, respectively.

As most studies included only initial and final measurements, error estimates for growth rate were not available. Instead, studies were

weighted based on an estimated sampling variance using the equation described by Adams et al. (1997) (Eq. 3).

$$v_i = \frac{N^T + N^D}{N^T N^D} + \frac{RR^2}{2(N^T + N^D)} \quad (3)$$

where N^T and N^D are the sample sizes for the triploid and diploid groups, respectively, and RR is the natural log-transformed response ratio.

A total of 148 response ratios were calculated across 29 studies, consisting of 87 experiments using chemical induction (46 measuring shell height growth; 41 measuring whole wet weight growth) and 61 experiments using diploid x tetraploid mating (38 measuring shell height growth; 23 measuring whole wet weight growth). Out of 87 experiments using chemical induction, 81 experiments used CB and the remaining experiments used 6-DMAP (Mallia et al., 2006) or did not specify (Garnier-Géré et al., 2002). There were only 7 experiments that produced meiosis I triploids (Stanley et al., 1984; Hawkins et al., 1994; Mallia et al., 2006).

A sample size weighted, random-effects model was used to calculate the mean response ratio for both growth rate responses. The random-effect model was chosen because it assumes that each study has a unique response ratio and takes into account biological and environmental variation across studies (Nakagawa and Santos, 2012). To determine whether heterogeneity was due to the influence of moderators, a mixed-effects model was used for the following variables: species, initial size, or length of experiment. Initial size refers to the size of the oyster in shell height or whole wet weight at the start of the experiment and was grouped into small seed, large seed, and juvenile, where small seed ≤ 25 mm (< 10 g), large seed = 26–50 mm (10–20 g), and juvenile = 51–75 mm (21–50 g). Given the lack of replicated experiments ($n = 1$) deploying juvenile oysters for all experiments using chemically induced triploids and experiments using mated triploids and measuring whole wet weight, this size class was excluded from statistical analysis. Length of experiment was compared by grouping experiments into 0, 0.5, 1, or 2 years, where 0 years ≤ 181 days, 0.5 years = 182–364 days, 1 year = 365–729 days, 2 years = 730–1095 days.

To determine whether there was significant publication bias, the Rosenberg method was used to calculate a fail-safe number for each triploid production method (chemical induction or mating) and growth measure. A fail-safe number measures the number of unpublished or nonsignificant studies that would make a significant outcome nonsignificant if added to the meta-analysis. The Rosenberg method was a more practical method to use compared to original methods laid out by Rosenthal (1979) and Orwin (1983) because the calculations take into account random-effects models (Rosenberg, 2005). If the resulting fail-safe numbers were large relative to the number of observations, then publication bias likely did not have a significant influence.

3. Results

3.1. Comparison of response ratios between diploid and triploid growth rates

There was a significant growth advantage (both height and weight growth measurements; $p < .01$; Table 1) for chemically induced and mated triploids over diploids. For experiments using chemically induced triploids, the mean response ratio for growth in whole wet weight was 0.27, which was more than three times that of the response ratio for growth in shell height (0.08, Fig. 1). A similar pattern was observed for experiments using mated triploids, where the mean response ratio for growth in whole wet weight was 0.40 and the mean response ratio for growth in shell height was 0.18 (Fig. 1). Back transformed to a linear scale, this suggests that, on average, chemically induced triploids grow 31% faster than diploids in whole wet weight and mated triploids grow 49% faster than diploids. In terms of shell length, chemically induced triploids grow 8% faster than diploids and

mated triploids grow 20% faster than diploids.

A Pearson correlation was used to compare the two growth measures, shell height (mm/day) and whole wet weight (g/day), for experiments using chemically induced triploids or mated triploids (Fig. 2). To compare these growth measures, only experiments that reported growth in both shell height and whole wet weight could be used. This consisted of 21 experiments using mated triploids and 28 experiments using chemically induced triploids out a total of 148 experiments. There was a similar positive correlation ($r = 0.78$, $p < .01$) for both experiments using chemically induced triploids and experiments using mated triploids.

Growth rates in shell height and whole wet weight were highly variable across studies with greater variation in whole wet weight than shell height and greater variation among experiments using chemically induced triploids than experiments using mated triploids (Table 1). The measure of heterogeneity (I^2), as stated by Higgins and Thompson (2002), indicated considerable between-study variation for experiments using mated triploids, with 85% variation for whole wet weight measurements and 55% variation for shell height measurements that could be explained by moderators. Conversely, for experiments using chemically induced triploids, the low I^2 values (17% and 0% for whole wet weight and shell height respectively) suggested that most of the variability is due to sampling error within the experiments (Table 1).

In terms of the effect of publication bias, the likelihood that the selected experiments showed only a certain result (i.e., publishing only statistically significant results) was measured by the fail-safe number. It is generally considered that a fail-safe number less than $5n + 10$ (where n is the number of experiments) suggests publication bias, or more specifically, an overestimation of the number of significant results (Rosenthal, 1991; Rosenberg, 2005). The response ratios for experiments using chemically induced triploids as measured by shell height, had a fail-safe number of 55 and suggested presence of publication bias as the number was less than $5n + 10$, where n is equal to 46 experiments. While response ratios for experiments using chemically induced triploids as measured by whole wet weight, and response ratios for experiments using mated triploids as measured by whole wet weight and shell height suggested that the experiments lacked bias, with fail-safe numbers of 853, 1212, and 801, respectively.

3.2. Effect of species on response ratios

The influence of species on the difference in growth rate between triploids and diploids varied depending on the growth measurement, triploid production method, and the number of studies published on each species (Fig. 3). The mixed effect model showed no significant difference ($p > .05$) in the response ratio between species for experiments with chemically induced triploids (measuring growth in shell height and whole wet weight) and experiments with mated triploids (measuring growth in shell height). However, there was a significant difference ($p = .02$) for experiments with mated triploids and measuring growth as whole wet weight, with *C. gigas* having a greater response ratio (0.59 ± 0.12) than *C. virginica* (0.28 ± 0.07) (Fig. 3). Across all experiments, the most commonly used species was *C. virginica*, which was used in 31 out of 38 experiments with mated triploids and measuring growth in shell height, 14 out of 23 experiments with mated triploids and measuring growth in whole wet weight, and 13 out of 46 experiments with chemically induced triploids and measuring growth in shell height. In terms of experiments using chemically induced triploids and measuring growth in whole wet weight, only 5 out of 41 experiments used *C. virginica*, while 25 out of the 41 experiments used *S. glomerata*. Overall, there was no clear relationship between the response ratio and species, likely due to the limited number of studies per species.

Table 1

Mean growth rates measured in whole wet weight (g/day) and shell height (mm/day) for diploid and triploid oysters as cited in 29 studies (148 experiments) and random effects model results using the natural log-transformed response ratio (RR) measured as $\ln(3n/2n)$. Experiments were separated based on whether triploids were produced through chemical induction (i.e., “Chemical”) or mating a tetraploid x diploid (i.e., “Mated”). Triploid advantage refers to improved growth in triploids relative to diploids, calculated by back transforming the response ratio to a linear scale (i.e., $\exp[\ln(3n/2n)]$). I^2 denotes the measure of heterogeneity, where by a high value indicates greater between-study variation and a low value indicates greater within-study variation (Higgins & Thompson).

Triploid	Measure	Diploid growth		Triploid growth		Mean RR	Triploid Advantage	I^2	P-value
		Mean ± SD	Range _{95%}	Mean ± SD	Range _{95%}				
Chemical	g/day	0.133 ± 0.184	0.006–0.781	0.167 ± 0.107	0.006–1.229	0.27	31%	17%	< 0.01
	mm/day	0.167 ± 0.107	0.056–0.456	0.189 ± 0.122	0.057–0.490	0.08	8%	0%	< 0.01
Mated	g/day	0.103 ± 0.076	0.010–0.278	0.158 ± 0.116	0.013–0.480	0.40	49%	85%	< 0.01
	mm/day	0.148 ± 0.048	0.052–0.243	0.178 ± 0.032	0.075–0.273	0.18	20%	55%	< 0.01

* Range values are cited as follows, starting at chemical induction g/day for diploids and triploids unless specified: Troup et al., 2005, Shpigel et al., 1992, Smith et al., 2000, Walton and Murphy, 2011, Guo et al., 1996, Walton et al., 2013, Harding et al. 2007, Ibarra et al., 2017 (diploid only), Stone et al., 2013 (triploid only).

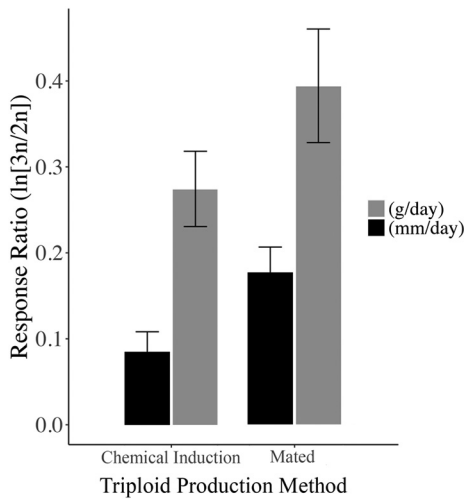


Fig. 1. Mean ($n = 148$) response ratios ($\ln[\text{triploid}/\text{diploid growth}]$) and 95% confidence intervals of growth rate (g/day and mm/day) for experiments using chemically induced triploids and mated triploids. All mean response ratios were significantly different ($p < .05$).

3.3. Effect of moderators on response ratios

The length of study and size of oyster at initial deployment did not significantly influence the response ratio ($p > .05$) for experiments measuring growth in shell height and using chemically induced triploids or mated triploids. For experiments with chemically induced triploids and measuring growth in whole wet weight, there was a

significant difference in the response ratios based on initial size ($p < .001$). Experiments initially deploying large seed (10–20 g) resulted in a greater mean response ratio (0.53 ± 0.09) than experiments initially deploying small seed (< 10 g, response ratio of 0.22 ± 0.4) (Fig. 4). For experiments with mated triploids and measuring growth in whole wet weight, there was a significant difference in the response ratios based on length of study. Experiments lasting one year (365–729 days) had a greater mean response ratio (0.68 ± 0.12) than experiments lasting < 182 days (0.38 ± 0.09) and 182 to 364 days (0.25 ± 0.08) (Fig. 5).

4. Discussion

This study is the first quantitative synthesis to show that triploids significantly grow faster than diploids in whole wet weight and shell height. Mated triploids outperformed chemically induced triploids, with a 14% greater triploid advantage in whole wet weight and 11% greater triploid advantage in shell height. The lack of equivalency in triploid advantage could be due to a number of differences between chemically induced and mated triploids, including the variable percentage of chemically induced triploids and low larval survival in experiments using chemical induction.

The percentage of triploid in chemical induction experiments ranged from $> 99\%$ (Matthiessen and Davis, 1992; Zhang et al., 2017) to 30% (shell height only, Shatkin, 1992) with an average 88% triploid population in whole wet weight experiments and an average 81% triploid population in shell height experiments. The experiments with the highest level of triploidy had some of the highest response ratios in shell height, 0.24 (Matthiessen and Davis, 1992) and 0.18 (Zhang et al., 2017), while the experiments with the lowest level of triploidy had

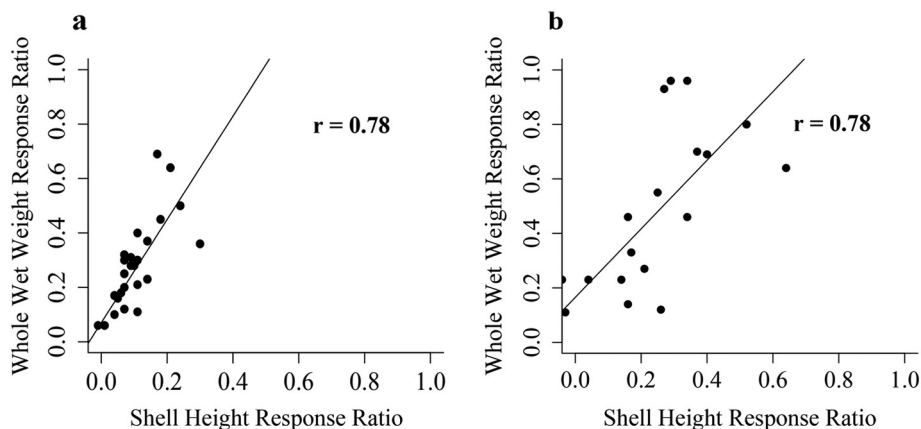


Fig. 2. Scatterplots for response ratios ($\ln[\text{triploid}/\text{diploid growth}]$) when growth is measured in whole wet weight and shell height for (A) experiments using chemically induced triploids ($r = 0.78$) and (B) experiments using mated triploids ($r = 0.78$).

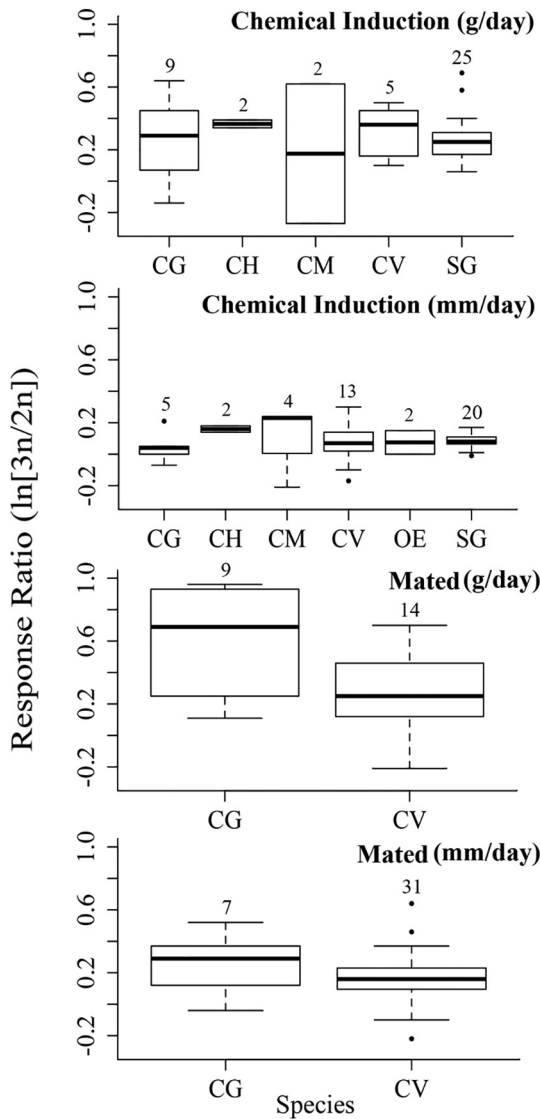


Fig. 3. Comparison of response ratios (ln[triploid/diploid growth]) for different oyster species from experiments using chemically induced triploids and mated triploids and measuring growth in whole wet weight (g/day) or shell height (mm/day). The number of experiments per species taken from a total of 29 studies is displayed above each box and whisker plot. Oyster species abbreviations: CG = *Crassostrea gigas*, CH = *Crassostrea hongkongensis*, CM = *Crassostrea madrasensis*, CV = *Crassostrea virginica*, OE = *Ostrea edulis*, SG = *Sassostrea glomerata*.

some of the lowest response ratios, -0.17 and -0.1 (Shatkin, 1992). The lower percentage of triploids in chemical induction experiments might have contributed to the lower triploid advantage compared to experiments using mated triploids if diploids were unintentionally included in the putative triploid treatments. Only three studies using chemically induced oysters determined oyster ploidy individually prior to measurements of response variables. Ideally, individual determination is the preferred technique for an accurate comparison to diploids if the triploid population is known to be $< 99\%$ triploid.

Chemically induced triploids are also known to have lower larval survival than mated triploids and diploids due in part to the toxicity of CB as well as the poor viability of tetraploids and aneuploids that are unintentionally produced. Likewise, meiosis I triploids often have a higher mortality than meiosis II triploids due to the higher proportion of aneuploids and tetraploids produced (Yamamoto et al., 1988; Guo et al., 1992). Across the studies analyzed, only a few studies explicitly

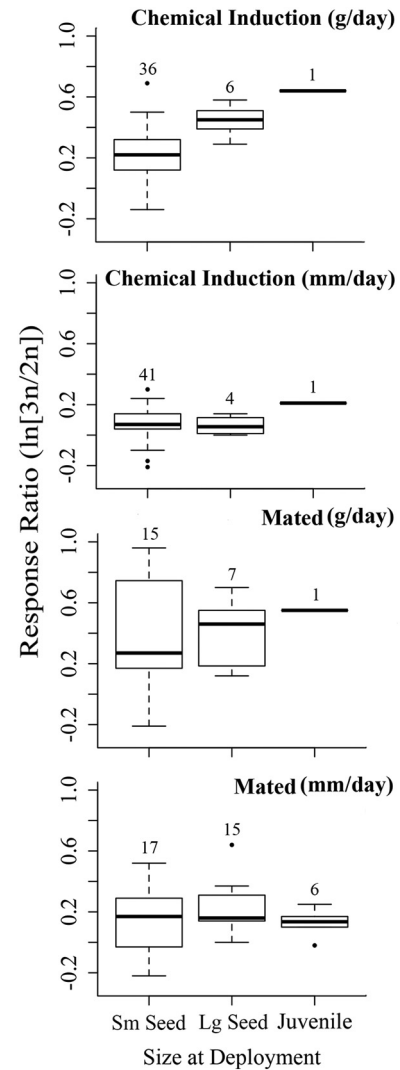


Fig. 4. Response ratios (ln[triploid/diploid growth]) compared based on initial oyster size at deployment from experiments using chemically induced triploids and mated triploids and measuring growth in whole wet weight (g/day) or shell height (mm/day). The number of experiments taken from a total of 29 studies is displayed above each box and whisker plot. Initial size at deployment was defined as follows: small (sm) seed ≤ 25 mm (< 10 g), large (lg) seed = 26–50 mm (10–20 g), and juvenile = 51–75 mm (21–50 g).

mentioned larval survival due to chemical induction and therefore we are unable to state with confidence overall differences in larval survival between chemically induced triploids and mated triploids. Furthermore, field survival for seed and juvenile oysters was highly variable across all studies and suggests that the technique of chemical induction compared to mating, does not increase the vulnerability of triploids to environmental stressors post-settlement.

Experiments with mated triploids and measuring whole wet weight resulted in a significant difference in the triploid advantage between species (greater mean response ratio in *C. gigas*) and length of experiment (greater mean response ratio in experiments lasting between 365 and 729 days). The significance is likely due to the study by Nell and Perkins (2005) which involved three experiments using *C. gigas* and observed exceptionally fast triploid growth, with response ratios > 0.90 over a 579-day experiment. Conversely, experiments with chemically induced triploids and measuring whole wet weight resulted in a significant difference in the triploid advantage between oysters initially deployed as small seed and as large seed, with a greater mean response ratio for large seed (10–20 g). The greater triploid advantage in large

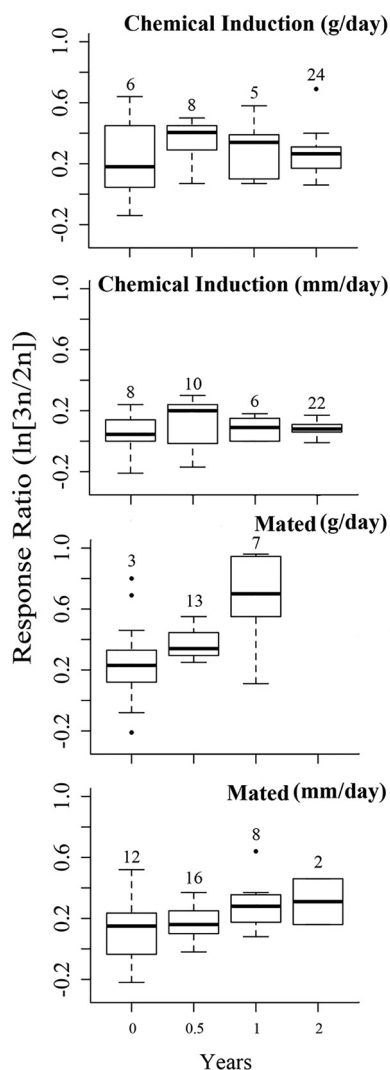


Fig. 5. Response ratios ($\ln[\text{triploid}/\text{diploid growth}]$) compared based on length of experiment (years deployed) from experiments using chemically induced triploids and mated triploids and measuring growth in whole wet weight (g/day) or shell height (mm/day). The number of experiments taken from a total of 29 studies is displayed above each box and whisker plot. Length of experiment was defined as follows: 0 years = 0–181 days, 0.5 years = 182–364 days, 1 year = 365–729 days, 2 years = 730–1095 days.

seed compared to small seed could be correlated with the size at which gametogenesis and spawning starts to affect the growth of diploids. Hand et al. (1998) noted that an advantage in chemically induced triploid Sydney rock oysters (*S. glomerata*) only became apparent when the oysters reached a whole weight of 5–10 g or a shell height of 30–40 mm and attributed the growth to the onset of gametogenesis. However, the minimal effect of moderators (species, length of study, size at initial deployment) in experiments measuring shell height suggests that triploid oysters (chemically induced and mated) can grow faster than diploids in shell height throughout all stages of development. Several studies showed increased shell height growth in triploid oysters starting at an initial size < 25 mm (< 10 g) and a length of study less than a year (Shatkin, 1992; Matthiessen and Davis, 1992; Callam, 2013).

The similar response ratios across species, with the exception of experiments using mated triploids and measuring whole wet weight, suggests that the advantages of triploids can apply to a variety of oyster species. When comparing the range of species used for both growth measurements and triploid induction techniques, there was a greater

variety of species across experiments using chemically induced triploids than experiments using mated triploids. Experiments using mated triploids only involved two species, *C. gigas* and *C. virginica*, and reflects the current production of tetraploids which focuses on only the few species with high commercial potential. As tetraploid production increases, the range of species used in experiments with mated triploids should also increase.

The results of this study support current oyster aquaculture practices and assumptions with triploid and diploid oyster growth. Consumer acceptance is dependent on the weight and meat quality of oysters (e.g. Nell et al., 1994), and it is generally accepted that triploid oysters can have improved marketability relative to diploid oysters (Dégremont et al., 2012; Nell, 2002), particularly during the reproductive season (Nell, 2002). The significant triploid advantage in whole wet weight relative to diploids found in this study supports the market goals of superior meat quality. In addition, the significantly faster growth in whole wet weight and shell height, as well as, the correlation between the measures supports the assumption that triploids can have a shorter growing season and can reach market size faster than diploids.

It is important to note that not all studies observed a significant growth advantage in triploids. At sites with unfavorable environmental conditions for oyster growth or with increased husbandry practices, the advantage of triploids was not as noticeable, if at all. For experiments with mated triploids, 9% (2/23) of the experiments reported equal or lower whole wet weight growth than diploids and 21% (8/38) of the experiments reported equal or lower shell height growth than diploids. For experiments with chemically induced triploids, 5% (2/41) of the experiments reported equal or lower whole wet weight growth than diploids and 30% (14/46) of the experiments reported equal or lower shell height growth than diploids. When looking specifically at the experiments with poor triploid growth, it is clear that triploids can respond differently than diploids to environment conditions such as, low salinity and high temperature. Callam et al. (2016) found that under low salinity (6–13 ppt), growth in shell height of *C. virginica* triploids was not significantly different from diploids for all wild broodstock lines and hatchery bred “Superlines” and growth in wet tissue weight was similarly not significant, with the exception of significantly slower growth in one of the triploid Superlines. In terms of temperature, Ibarra et al. (2017) found that at the temperate site (17.2–26.5 °C) triploid *C. gigas* had 4% slower growth in shell height and a 6% greater whole weight gain compared to diploids. Whereas, at the two tropical sites (18.8–31.5 °C), the authors found that the oysters had slower growth overall, but triploids gained 64–70% more whole weight per month than diploids (Ibarra et al., 2017). Similarly, in the temperate climate of Maine, triploid *C. virginica* had a minimal growth advantage and the lack of growth was attributed to a very limited spawning period, which allowed diploids to behave as sterile triploids for the majority of the growing season (Shatkin, 1992). Like temperature and salinity, how the oysters are handled can also influence relative growth rates of triploids and diploids. Increased husbandry, such as regular tumbling and removing biofouling off of culture bags, can reduce the difference in shell height growth between ploidy (Stone et al., 2013). Different gear types can also influence growth; for example, Walton et al. (2013) compared the change in dry tissue weight of diploids and triploids and reported a similar increase between ploidy when using floating bags, but a greater increase in triploids when using other gear types (i.e., bottom cages, floating cages and adjustable long-lines).

The experiments with slower triploid growth than diploids (negative response ratios) were removed from the dataset to determine whether these experiments influenced the effect of the moderators (species, length of study and initial size) on the response ratio. Removing these negative response ratios only influenced the analysis of experiments using mated triploids and measuring growth in shell height. Similar to the analysis of experiments using mated triploids and measuring growth

in whole wet weight, response ratios for experiments measuring growth in shell height was significantly different ($p = .03$) across species (greater in *C. gigas*) and length of study (greater after two years). This further supports that *C. gigas* mated triploids might grow faster compared to their diploid counterpart than *C. virginica* mated triploids, especially under favorable growing conditions. Additionally, for mated triploids, it might take longer for the triploid advantage to become apparent, as longer studies (at least one year) resulted in a greater average triploid advantage.

The influence of temperature and salinity on triploid and diploid growth rates stresses the impact of water quality on oyster growth and the importance of taking these measurements during growth studies. Close to a third of the studies included in this meta-analysis did not report temperature or salinity averages. Lack of information on the environmental conditions made it difficult to accurately compare the growth measurements and could account for the high observed heterogeneity, as well as the lack of a relationship between moderators. Other potential influencing factors, such as tidal height and culture gear, could not be compared due to lack of data. While culture gear was typically mentioned, the gear types were too varied (e.g., upweller, floating bags, fixed bag on rack, cage, lantern net, and bottom tray) with an insufficient number of studies using each gear for an accurate comparison.

In conclusion, this study validates the clear growth advantage of triploid oysters as compared to diploid oysters in both shell height and whole wet weight, and supports the commercial production of triploids for the half shell market. There was a measurable advantage of mated triploids over chemically induced triploids, which suggests that these two techniques are not equivalent and must be treated as such when discussing triploid performance. Another valuable finding of this study was the number of experiments which expressed similar or slower growth in triploids, especially in terms of shell height growth. Within the experiments which compared both shell height and whole wet weight (21 experiments using mated triploids and 28 experiments using chemically induced triploids), only half of the experiments having equal or slower shell height growth in triploids also had equal or slower whole wet weight growth, suggesting that triploids might perform better in terms of whole wet weight growth than shell height growth. While temperature and salinity appear to have the most significant impact on triploid vs. diploid growth, the effect of aquaculture practices, as cited in a few studies, may be similarly important when comparing growth rates.

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