

Nutritive Value of Golden Apple Snail (*Pomacea canaliculata*) as Animal and Aquaculture Feed

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ABSTRACT

Feed is a crucial input for terrestrial animal and aquaculture production, but these sectors face the same feed availability and sustainability challenges. Despite their reputation as rice pests, causing economic loss in agriculture, golden apple snails could be used as an alternative animal feed. This study reviews the nutritional value, including bioactive compounds, constraints, and future utilization of golden apple snails as animal and aquaculture feed. An integrative literature review was conducted on data retrieved from publications available on Google Scholar, Scopus, PubMed, Web of Science, and official websites. The golden apple snail is rich in protein in their meat and calcium in the shell, representing 39.11 to 68.67% and 41.38% (dry matter basis), respectively. The inclusion of golden apple snails in the diet resulted in good growth performance in monogastric animals and fish due to their nutritive value. Golden apple snails may be available as feed resources, supplied from the wilds and heliciculture. The astaxanthin of the eggs of golden apple snails and chitosan derived from their shell are interesting due to their bioactivities, thereby opening new avenues for future research in functional feed additives.

Keywords

bioactive, feed, golden apple snail, nutritional value

1. Introduction

The demand for food, including animal products, is on the rise concomitant with the global increase increasing the world population and food consumption. According to the United Nations, the world population is projected to be 9.7 billion in 2050 [1], whereas the total food demand of 35% from 2010 is expected to reach 55% in 2050 [2]. Similarly, the need for animal-source food is expected to increase by 70% in 2050 compared with the year 2000 [3]. Livestock and aquaculture sector play a significant role in the provision of global food protein, contributing 21.58 – 39.90% of the total supply, depending on region [4].

Feeding is one of the determinants of terrestrial and aquatic animal production, specifically in the intensive system where high-quality feed is a vital input to produce better animal growth. The profitability of livestock and aquaculture enterprises also depends on feed costs, accounting for 60–70% of the total production cost. Before the COVID-19 pandemic, global feed industries produced a total of 1,187.1 million tons (grew 1% per year) of compound feed to supply farm animals, aquaculture, and pet animals [5]. Due to the impact of COVID-19 blockade measures in feed-producer countries, feed production was reduced by 0.42%. Nevertheless, global feed production may be recovered by the countermeasures after the COVID-19 pandemic that elaborate movement and importation of agricultural commodities, provision of subsidies, and stabilization of agricultural sectors in top compound feed producers, such as China [6, 7] and India [8].

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The growing human population intensifies the burden of land utilization for inhabitant settlement and agriculture for food and feed, termed feed-food competition [9, 10]. Therefore, it significantly affected the availability of natural resources and animal feedstuffs. A previous study showed that the production of biofuels (ethanol and biodiesel) derived from agricultural commodities elevates feed prices [11]. During the last decade, the average price of several protein source feedstuffs, such as soybean meal and corn gluten meal, experienced an annual increase of 10.0 and 9.1%, respectively [12]. On the other hand, fishmeal price increased by 14.1% due to declining capture [13], and this circumstance was worse in importing countries, such as Malaysia and Indonesia [14, 15]. The same challenge is faced in feed sustainability and price by land-based and aquatic animal production [16], necessitating many efforts to explore and investigate alternative feedstuffs.

The golden apple snail (*Pomacea canaliculata*), found in freshwater, belongs to the Ampullariidae family and is native to Argentina. According to Naylor [17], golden apple snail (GAS) was introduced to Asia in early 1980 by the Philippines with the commercial purpose of protein food sources. It was later introduced to Taiwan, Japan, China, and some Southeast Asian countries. GAS spreads through waterways and agricultural irrigation systems and becomes a rice pest by grazing on the paddy field at the vulnerable stage when the seeds grow as young plants (4 weeks after being seeded). In a study on the mitigation of *Pomacea* spp, Azmi [18] showed that the top 25 rice producers such as China, Indonesia, Vietnam, and Japan, suffered economic losses due to reduced rice yield, cost of pest control, and replanting due to GAS invasion. Several approaches have been reported to cope with the GAS infestation on paddy fields, such as crop rotation [19], chemical and botanical agents, biological and mechanical controls [18, 20], unfortunately, GAS still appears to be a persistent rice pest and is becoming a problem of environmental pollution.

Despite their reputation as a pest, GAS contains considerable nutrients that could be used as a potential feedstuff. The use of GAS as animal feed stems from the mitigation of its invasion in paddy fields, using natural predators as biological control. Several workers reported that duck and fish (common carp) efficiently reduced the density of GAS and provided benefits from rice-fish integrated farming [21, 22, 23]. Even though these findings suggested that GAS could be used as a feed, other essential factors, such as bioactive potentiality and future perspectives in utilizing GAS have not been addressed. Additionally, GAS might serve as an available feed resource due to its high density and biomass on paddy fields. The density of adult GAS in the conventional paddy field is 15.71 snails/m² [24], and the GAS biomass was estimated at up to 31.5 g/m² [25], depending on environmental conditions [26].

Based on the background above, this study reviewed the GAS nutritional value as animal and aquaculture feed. We used an integrative literature review approach on the data retrieved from publications available on Google Scholar, Scopus, PubMed, and Web of Science. We also examined relevant information from official websites. The sections of the paper discuss the nutrient composition, nutritive value, and constraints of GAS. It further evaluates the bioactive potentiality and future perspective in utilizing GAS.

2. Nutrient composition of GAS

The nutritional composition of feedstuff is a fundamental factor in evaluating the quality and value of animal feed. It served as essential data for feed formulation, enabling the creation of a balanced diet. The parts of GAS consist of meat, whole body (meat and shell), eggs, and shell (Table 1). The dry matter content (DM) varies within a species, between 15.00 and 99.30%, depending on the parts of GAS and sample preparation. Fresh meat contains considerable moisture, ranging from 60 to 85%.

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Table 1: Chemical composition and energy content of meat, whole body, shell, and egg of golden apple snail (*Pomacea canaliculata*)

Parts of GAS	Sample preparation	Chemical composition (%)							GE (MJ/kg)
		DM	CP	EE	CH	NFE	Ash	CF	
Meat	Boiled, sun-dried, hammer-milled [27]	93.00	67.00	7.21	n.a.	n.a.	4.12	2.13	n.a.
	Sun-dried, meal [28]	n.a.	56.40	1.60	n.a.	n.a.	11.80	1.00	12.29
	Fresh, without viscera [29]	15.00	68.67	3.33	17.33	n.a.	12.00	n.a.	15.48
	Fresh [30]	22.40	54.46	1.79	29.46	n.a.	14.29	n.a.	n.a.
	Boiled & Minced [31]	33.99	49.54	0.83	n.a.	n.a.	13.98	n.a.	n.a.
	Fermented after boiled & minced [31]	24.77	39.11	0.75	n.a.	n.a.	3.62	n.a.	n.a.
Whole body (meat & shell)	Boiled, crushed [33]	47.06	15.02	0.91	n.a.	15.53	68.36	0.08	n.a.
	Fresh, crushed [30]	24.45	13.58	0.78	29.12	n.a.	56.48	n.a.	n.a.
Egg	Fresh, homogenated [34]	18.07	18.70	1.25	76.40	n.a.	57.30	n.a.	16.90
	Drying, Powdered [35]	94.45	15.93	2.08	0.43	n.a.	60.62	n.a.	n.a.
Shell	Dried [36]	99.30	0.50	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.

Notes: values are based on dry matter (DM); CP – crude protein; EE–ether extract; CH–carbohydrate; NFE–nitrogen free extract, CF: crude fiber; GE–gross energy; n.a.–not available

The crude protein (CP) content of meat and eggs of GAS is in the range of 39.11–68.67% and 13.58–18.70% (DM basis), respectively. The CP content of the whole body is 15.02% lower than their meat. Furthermore, the ash content of GAS meat is about 2.62–21.90%, but the whole body and egg are much higher, accounting for 68.36% and 56.48–60.62%, respectively, attributed to the mineral in their shells. High ash content could reduce the feed protein quality (amino acids per unit of protein) [37]. GAS contains lower ether extract, ranging from 0.75 to 7.21%, compared to fishmeal (9.20%) [38], while the gross energy (GE) ranges between 12.29 and 16.90 MJ/kg.

The variation in the chemical composition of GAS is affected by the sample type, preparation, processing, and analytical methods. In terms of processing methods, drying increases the protein content of GAS [22, 23] compared to boiled [24, 25], while boiling reduces its fat content. Amongst macronutrient composition, the CP of the meat is commonly higher than other parts, and it is adequate for poultry and fish dietary protein sources.

Amino acids are functional and structural protein units, emphasizing that the quality is based on their composition. The amino acid composition of GAS is presented in Table 2. Lysin and methionine are vital as they are the most limiting essential amino acids (EAA) in animal and fish feeding [29, 28]. Compared to low-grade fish meal and soybean meal [38], GAS contains lower lysin (7.5% CP in fishmeal) and methionine (2.7% CP in fishmeal), but it is relatively comparable to soybean meal with 6.2% CP of lysine and 1.4% CP of methionine. Therefore, the inclusion of GAS in feed formulation needs either an addition to an EAA supplement or a combination with other feedstuffs to achieve a balanced feed.

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Table 2: Amino acids profile of *Pomacea canaliculata* meat and egg

Amino acids	Meat (% of protein, DM basis)				Egg, fresh (mole %) [39]
	Fresh without viscera [29]	Oven-dried, powdered [35]	Sun-dried meal [28]	Freeze drying [32]	
Essential:					
Histidine	1.80	1.60	2.13	1.60	1.90
Isoleucine	3.90	4.10	3.36	3.20	5.40
Leucine	8.10	8.20	6.46	7.00	9.00
Lysine	6.00	6.80	2.91	9.70	6.60
Methionine	1.80	1.00	1.95	2.10	2.50
Phenylalanine	3.80	4.30	2.93	3.30	3.90
Threonine	4.50	4.70	1.98	4.00	5.50
Tryptophan	1.10	0.20	1.95	4.00	n.a.
Valine	4.60	4.30	3.71	3.80	8.10
Non essential:					
Arginine*	8.50	9.10	5.73	6.60	4.50
Alanine	6.10	6.00	5.59	6.10	5.70
Aspartic acid	10.30	8.50	6.27	9.30	11.60
Cysteine	1.10	0.80	n.a.	n.a.	0.80
Glutamic acid	16.10	17.30	12.48	13.60	11.60
Glycine	7.00	5.80	4.30	5.50	6.50
Proline	4.90	4.70	2.91	3.70	5.70
Serine	4.70	4.90	3.20	4.30	7.50
Tyrosine	3.30	4.50	3.20	1.90	4.10

Notes: n.a. – not available; *essential amino acids in fish.

The functions of minerals include skeleton formation, osmotic balance, and enzyme cofactor. Based on the amount required in diets, minerals are grouped into macro- and microminerals (Table 3). Oyster shell and bone meal (MBM) are animal-based calcium sources for poultry diets [42]. The GAS is rich in calcium, which is attributed to the calcareous structure of their shells and egg capsules. The calcium content in the GAS shell (41.31%) is slightly higher than that of oyster and bone meal (34–36%) [39]. In addition, the GAS shell is composed of a substantial amount of iron and manganese, making it essential in animal and fish feed.

Table 3: Minerals composition of *Pomacea canaliculata*

Minerals	Meat, steamed [35]	Meat, boiled [32]	Shell [36]	Egg, fresh [30]
Macrominerals (%):				
Calcium	5.16	6.20	41.38	21.11
Phosphorus	0.55	1.20	0.01	2.25
Potassium	0.36	n.a.	0.07	1.49
Magnesium	0.06	n.a.	0.00	0.23
Sodium	0.09	n.a.	0.22	0.38
Micromineral (ppm):				
Iron	455.00	n.a.	1580.00	32.00
Copper	71.00	n.a.	81.00	41.60
Zinc	101.00	n.a.	46.00	21.60
Manganese	20.00	n.a.	124.00	n.a.
Molybdenum	n.a.	n.a.	0.10	n.a.
Chloride	n.a.	n.a.	300.00	n.a.

Notes: Values are on dry matter basis. n.a.–not available

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3. Nutritive value of GAS

3.1 The meat

GAS meat is suitable as an alternative protein source for duck, fish, and crustacea. The nutritive value of GAS in animals and fish is summarized in Table 4, while the GAS feature is presented in Figure 1. The function of GAS meat as the primary protein source for duck is confirmed, with a dietary inclusion level of up to 40%. However, the recommended level to achieve excellent growth performance is 30% [22, 34]. This result showed that GAS meat can replace fishmeal without compromising fish growth. Previous studies on GAS meat digestibility and the EAA index in terrestrial animals are limited, and the EAA index in shrimp is 0.84. The digestibility of GAS meat in fish is high, resulting in 84.9, 88.1, 80.6, and 86.3% for DM, CP, OM and GE, respectively [44]. The CP digestibility of raw GAS for catfish is 92.6% but it experiences a slight increase of 93.6% when boiled.

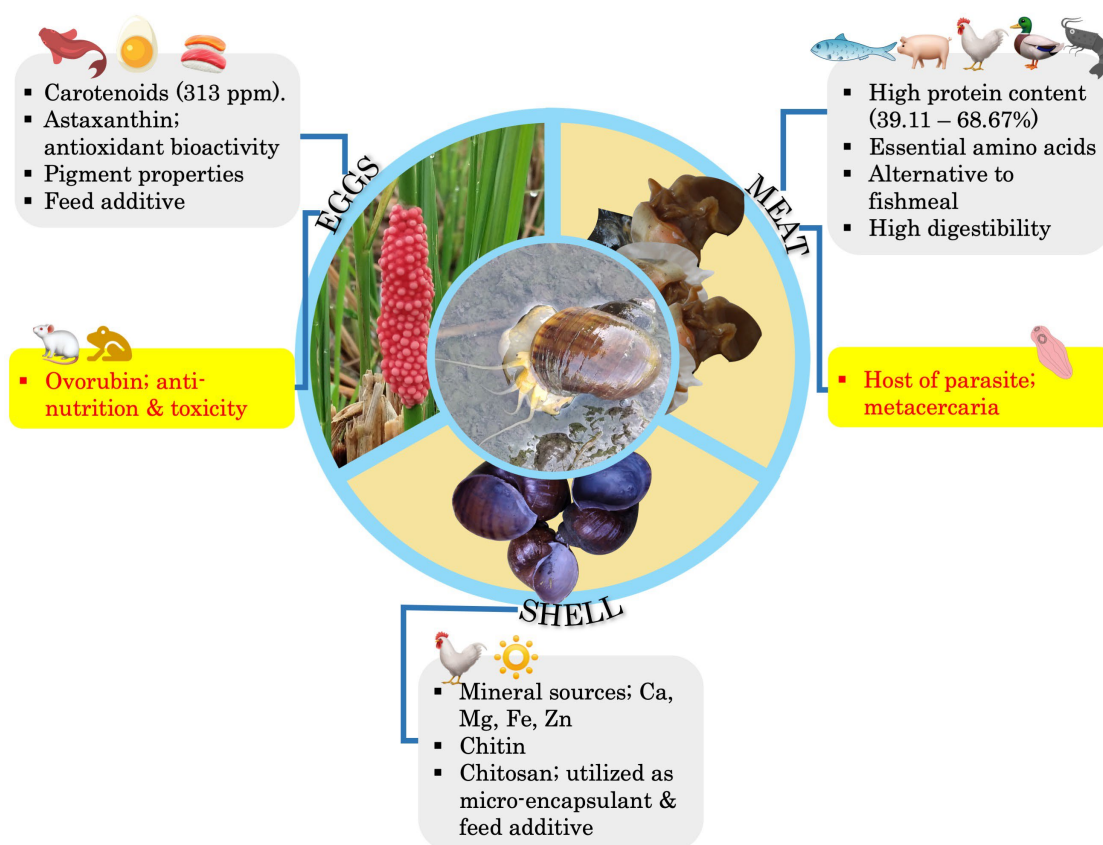


Figure 1: Utilization of different parts of golden apple snail, featuring nutrition, constraint, and bioactivity potential in animals

3.2 The shell

Literatures reporting the nutritional value of GAS shells in animals and fish are scarce. A study to examine the effect of GAS shells on growth performance, carcass quality, tibial bone strength, and small intestinal histology was

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conducted using Thai native chickens [45]. In this study, chickens were fed 0.35% GAS shell, replacing limestone as a calcium source, for six weeks, followed by 0.70% until 16 weeks. The treatment includes particle size of the GAS shell ranging from 0.50–1.00, 1.00–1.70, and 1.70–2.80 mm, as well as a control diet (using limestone). Overall, no difference was found in the growth rate, carcass quality, or bone strength of Thai chicken. The result showed that the chicken fed with the GAS shell of 1.00 to 1.70 mm particle size from the 13 to 16 weeks improved in weight gain and feed efficiency due to the increased villus surface of the duodenum and the number of crypt cells in the jejunum. Merit [36] examined the wild-caught captive raised and captive-born Chacoan peccary, and offered dried GAS shells, by placing clean dry snail shells in three groups of animals (approximately 50 g of GAS shells each). Only one group of wild-caught captive Chacoan peccary consumed the GAS shells.

3.3 The eggs

The eggs of GAS can be distinguished by their cohort of 14 to 500 eggs, laid on pants or substrates above the waterline, with pink and reddish color owing to carotenoid (313,48 ppm of total carotenoids)[30]. As a comparison, maize contains a total carotenoid of 16–156.14 ppm [46], shrimp 14.86–68.86 ppm [47], and carrot 58.15–64.94 ppm [48]. Until recently, only a few studies report the utilization of GAS eggs as a feedstuff.

Due to expensive synthetic carotenoid pigment, astaxanthin was extracted from fresh GAS eggs, yielding an amount of 16.8 g of astaxanthin mixture (containing free, mono-, and diester astaxanthin) from 1000 g of eggs [49]. The extract was supplemented for ornamental carp (*Cyprinus carpio*) feed at levels of 0, 25, 50, 100, and 200 mg/kg to obtain better skin pigmentation, and it also used synthetic astaxanthin at the level of 50 mg/kg feed. After a 25-day feeding trial, the astaxanthin supplementation significantly increased the skin redness along the levels used. The experiment found that 50 mg/kg of natural astaxanthin from GAS eggs was comparable to synthetic astaxanthin and could be used to improve the skin color of the carp.

Another experiment studying the effect of GAS eggs on coloration, antioxidant capacity, and survival in blood parrot fish was conducted by Yang *et al.* [50]. In this study, GAS eggs, in powdered form, were formulated at 0, 5, and 15% in the diet and then given to fish for 60 days. The redness coloration of fish skin was significantly enhanced by supplementing GAS egg powder. Compared with the control, skin carotenoids increased to 62.6% and 102.2% in those fed with 5% and 15% egg powder, respectively. Similarly, the total number of scale chromatophores (i.e., pigment-bearing cells) of fish given 5% and 15% increased to 105.8% and 145.9% relative to control. The experiment found that GAS egg powder exhibited an antioxidant defense system, evident from the increasing parameter of super oxidase dismutase (SOD) and catalase (CAT) activity in the fish liver. It also showed that astaxanthin derived from GAS eggs serve as bioactive compounds. However, there was no significant difference in mortality, suggesting that GAS eggs did not affect fish survival.

One of the determinants of internal chicken egg quality is egg yolk color, which depends on the carotenoid in the body. However, animals, including chicken cannot endogenously synthesize carotenoids [51] and should be supplied through the diets. The effect of dietary GAS egg powder as a natural carotenoid source on the quality of native chicken eggs resulted in an improvement in yolk color [52]. The dietary treatments consisted of varying GAS egg powder levels, ranging from 0 to 12% in maize-based rations. After four weeks, egg yolk color and total carotenoid increased in line with the treatment level but had no significant effect on Haugh unit and yolk index. The 4% inclusion level in the diet resulted in a scale of 12 of yolk color fans, which indicated that the yolk color matched consumer preference.

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Table 4: Nutritional value of *Pomacea canaliculata* meat in animal and aquaculture species

Animal/fish, age	Objective	Treatment	Principal finding	Reference
Mallard duck, 5 weeks old	Substitution of GAS meal (GASM) for commercial feed	Control (100% commercial feed) + 0% GASM, 90% control + 10% GASM, 80% control + 20% GASM, 70% control + 30% GASM, 60% control + 40% GASM	Duck performance (FI, WG, FC) increased due to increasing feed palatability and protein content. The best substitution level of GAS was 30%.	[27]
Laying duck, 20 weeks	Effect of GAS and cassava on lipid and cholesterol of carcass and blood	0, 5% GAS inclusion combined with 0, 5 or 10% cassava leaf meal	No interaction between GAS and cassava. The inclusion of 5% GAS resulted in high FI. Increased cassava lowering FI of duck. Increasing GAS and cassava reduced cholesterol in blood, egg, and meat in duck.	[53]
Muscovy duck, 8 weeks	Evaluation of dietary GAS on carcass composition	Inclusion of 0, 10, 20, and 30% of GAS	FBW increased along the inclusion level of GAS. Slaughtered weight, carcass weight, and yield were affected by treatment. 30% GAS was the best treatment.	[43]
Hybrid Peking x Mojosari duck, 23 days old	Effect of dietary GAS Supplementation on growth performance and carcass traits	0, 10, 15, 20% of GAS	Ducks given 10% GAS showed the highest performance (FBW=1415.55 and BWG=868.62, but 15% GAS showed the highest carcass yield and lowest abdominal fat.	[33]
Alabio, Mojosari, and Raja duck (hybrid Alabio x Mojosari), 22 weeks	Effect of replacing yellow corn with steamed sago and GAS on growth performance and carcass traits	Factorial feeding trial consists of 0–39% sago with 0–6% GAS to replace 0, 15, 30, and 45% yellow corn	No interaction between feeding treatment and duck strains. FBW, BWG, and FC were affected by the strain of the duck. 6% GAS+39% sago resulted in the highest FI. The carcass, breast, leg, and back percentage were not affected by feed and duck strain.	[54]
Japanese quail	Fishmeal substitution	Substitution levels of 0, 25, 50, 75, and 100%	Quail egg production was similar, with up to 50% substitution. The substitution level of 50% was the best, considering quail performance and return of investment after 15 months of laying.	[55]
Catfish (<i>Pangasius</i> sp), fingerling	Substitution of FM with GAS meat meal for growth of catfish	0,10,20,30, & 40% GAS substitution for FM	10% GAS substitution was well balanced of protein to energy ratio and amino acid, resulting in the highest growth, and better FC.	[56]
Red tilapia	Utilization of fermented or raw GAS as a protein source alternative to FM	0, 25, 50, 75, 100% substitution fermented GAS to FM, and 50% raw GAS	Fermented GAS can substitute FM up to 100%, but 75% substitution of fermented GAS was recommended. 50% raw GAS is beneficial for tilapia's growth, feed intake, feed	[31]

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			conversion, protein efficiency ratio, and protein digestibility.	
Striped catfish (<i>Pangasianodon hypophthalmus</i>), fingerling	Digestibility study of animal and plant protein feed	Reference diet (26% fishmeal), 30% inclusion of shrimp head meal, GAS, earthworm meal, and catfish by-product.	The GAS meat digestibility is high, being 84.9, 88.1, 80.6, 86.3% for DM, CP, OM, and GE, respectively.	[44]
Striped catfish (<i>Pangasianodon hypophthalmus</i>), fingerling	Replacement of FM with other animal and plant protein sources, including GAS meal	Reference diet (26% FM) and six other rations, including shrimp head, GAS, earthworm, catfish by-product, groundnut cake, and rice. Each ration replaced 30% FM.	FBW, BWG, growth rate, and protein intake GAS-fed fish fed GAS are similar to others but better than in the control. The sarcass trait was similar to those of FM. GAS can totally replace FM without compromising growth and carcass traits.	[28]
African catfish (<i>Clarias gariepinus</i>), juveniles	Digestibility study of some feedstuffs, including GAS	Basal diet+ 5% BW raw GAS, Basal diet + 5% BW boiled GAS, Basal diet + others feedstuffs	The apparent digestibility of OM, CP, EE, NFE, CH, and GE was 80.7, 87.8, 65.7, 65.6, 62.0, 65.0, and 86.6%, respectively (raw GAS), and boiled GAS was 75.8, 90.5, 53.3, 44.7, 57.6, and 78.3%, respectively.	[57]
Red tilapia, kerok (<i>Anabas testudineus</i>), jalak (<i>Ophicephalus striatus</i>), African catfish and common carp	Evaluation of 5 species of fish for biological control of GAS in rice	Individual predation in aquaria, prey-predation in aquaria, field trial on rice plantation	Carp showed the ability to consume GAS meat, and the density of 10 fish/plot (2041 fish/ha.) results in a 2.14 kg fish yield/plot. Catfish were not adaptable in rice field conditions.	[58]
Tiger shrimp (<i>Panaeus monodon</i>)	Study the utilization of GAS on shrimp growth and its economic aspect.	4 dietary treatments: 60% maize+ 40% GAS, 60% cassava+ 40% GAS, 100% maize, and 100% GAS.	Essential amino acids index of GAS was 0.84. Shrimp fed cassava GAS showed the highest carapace length (40.3 mm) with a total production of 276 kg/ha, resulting in the best net income and return on investment (206%). GAS utilized with either cassava or maize improved shrimp production and was more profitable compared to GAS alone.	[32]
Piglet	Access the digestibility of ensiled and fresh GAS in piglets	Basal diet, basal + 30% ensiled GAS, basal + 30% fresh GAS	The apparent digestibility of DM is 58.6% and 55.1% of ensiled GAS and fresh, respectively. The apparent digestibility of CP is 82.8% and 80.9% for ensiled GAS and fresh, respectively. The apparent digestibility of OM is 61.6% and 60.9% for ensiled GAS and fresh GAS, respectively.	[59]

Notes: GAS–golden apple snail; FI–feed intake; WG–weight gain; FC–feed conversion; FBW–final body weight; FM–fish meal; DM – dry matter; CP–crude protein; OM–organic matter; GE–gross energy; BW–body weight; BWG–body weight gain; EE– ether extract; NFE–nitrogen-free extract; CH–carbohydrate

4. Constraints in using GAS

4.1 Antinutritive compounds

Feed ingredients have limiting factors in their use, and in this context, GAS eggs contain a specific perivitelline fluid (PV) composed of glycol-lipo-carotenoid protein, also termed ovorubin [46]. PV of *Pomacea* species includes three lipoprotein fractions, namely perilipovitellins 1, 2, 3 (PV1, PV2, and PV3), representing 6.7, 10.0 and 53.2%, respectively, of the egg total lipids [60]. PV1 and P2 are very high density lipoproteins with 300 and 400 kDa molecular weight, respectively, while PV3 is a high density lipoprotein with 164 kDa molecular weight [48, 49]. Another fraction that has similar properties to PV1 is PsSC. It is an oligomeric of carotenoprotein, with a molecular weight of 380 kDa, from the eggs of *Pomacea scalaris* [62]. In the context of GAS biology, the PV serves as a nutrient and energy storage for embryos. and it has protective functions from sun radiation and egg desiccation and is one of the defense systems against egg predators [63].

Pure ovorubin, isolated from the fresh egg of GAS, is toxic in animals. In amphibians, toxicity tests showed that perivitelline was not lethal to frogs even though the high dose of PV2 (170 mg/kg body weight) was intraperitoneally injected [64]. Observations after 24 hours showed inflammation and morphological changes in the small intestine of the frog but showed recovery (not different from the control) after 48 hours. The result showed that frogs also exhibit adaptability to the negative effect of perivitelline fluid, and the alterations of the small intestine by perivitelline were reversible. Egg extract was lethal (LD₅₀; 96 hours 2.3 mg/kg body) for rats due to its neuro- and enterotoxicity [65].

In a feeding trial using rats [66], oral administration of 100 uL/day purified ovorubin reduced the standard growth rate during the first three days (of 16 days experiment) compared to control. Interestingly, the growth of rats recovered after the fourth day of treatment, and there was no significant effect on feed intake. In rats, the mode of action of proteinase inhibition is characterized by hindering trypsin activity (anti-digestive role). There were no antimicrobial properties when ovorubin was tested on Gram (+) as well as Gram (-) bacteria, particularly against *Escherichia coli*, *Salmonella typhimurium*, *Bacillus subtilis*, and *Lactobacillus casei*. Ituarte *et al.* [62] reported that Wistar rats given PsSC (5 mg/day) for 48 hours showed narrowing and inflammation of the intestinal villi. After exposure to perivitelline for 72 days, there was a tendency for intestinal morphology to return to normal, indicating the ability of rats to adapt and cope with the adverse effects of PsSC [62].

Ovorubin is categorized as a small Kunitz-type trypsin inhibitor (KTI) family [66]. Several methods are available to overcome KTI activity, such as thermal inactivation, ultrasonic application, acidic condition, and zinc (Zn) application [53, 54, 55, 56, 57]. Amongst these methods, thermal treatment and acidic conditions were shown to reduce the antinutritional of GAS eggs. A temperature of 100 °C for 15 minutes led to the loss of PV2 toxicity in rats [65], while pre-incubation at pH 2.0 for 48 hours and heating at 100 °C for 40 minutes led to the loss of almost all inhibitory activity of ovorubin [66]. The above literature shows that antinutritionals in GAS may negatively influence animal performance, and this is an open opportunity to study mitigating the negative impact of antinutritionals when GAS eggs are used as a feed additive.

4.2 Bioaccumulation of heavy metals

Gastropods including GAS absorb heavy metals, such as Hg, Cu, and U, from their habitats. The ability to bioaccumulate metals is attributed to pigmented corpuscles within cells of the midgut gland of snails and the level of bioconcentrated elements in the GAS body depends on the degree and duration of exposure, as well as environmental conditions [72]. As an illustration, when GAS was exposed to 164 µg/L Cu for 96 hours, an accumulation of 187 µg/L was observed [73]. In contrast, exposure to a concentration of 2 µg/L of Hg for eight

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Nusantoro *et al.* Reviews in Agricultural Science, 12: 147–164, 2024

https://doi.org/10.7831/ras.12.0_147

weeks accumulated 5.05 mg/kg dry mass of the digestive gland [74]. This phenomenon showed that the use of GAS as feed, specifically when collected from the wild, might be exposed to the pollutants, suggesting pre-treatment.

Some techniques of heavy metal alleviation in food products are available in previous studies and can be adapted for GAS, such as acid soaking and sorbents. A previous report showed that Japan is ranked 5th in world seafood consumption, and one of their primary product is scallops (member of Mollusca, together with GAS) [75]. However, the internal organs of scallops contain high Cd (up to 80 ppm), ranging between 4 to 5 tons of Cd from scallop mariculture transferred to land. Ren *et al.* [76] used 2% acetic acid and 2% citric acid solution to wash scallop hepatopancreas. After washing four times, the concentration of Cd declined from 38.9 ppm to less than 0.6 ppm.

4.3 Parasitic hosts

GAS is known as a host for zoonotic parasites. The infection rate was 30.6% for metacercaria in those collected from canals, whereas infected snails from ponds showed a lower rate of 4%. Yang *et al.* [77] reported eosinophilic meningitis outbreak, caused by *Angiostrongylus cantonensis* in China, from 1997 to 2008. *Angiostrongylus cantonensis* is a rat lungworm hosted by GAS, with the infection rate in GAS at 68.4%. The study showed that the risk of infection in humans was higher when undercooked GAS was consumed.

5. Future use of GAS

This section covers the potential application of GAS, particularly as a functional feed additive. The availability, sustainability, and possible future GAS research are also outlined. In recent years, many studies have investigated various new alternative feedstuffs including insects and the results showed that new feedstuffs were suitable for animal and fish feed and acceptable in the perspective of farmers, stakeholders, and consumers [55, 56, 57]. The main perceived benefits were that using insects in animal feed lowered dependence on foreign protein sources, allowing better valorization of organic waste [81].

Considering the information on alternative feedstuffs used in meat production, a study investigating consumer perceptions, demand, and preferences showed that alternative feedstuffs are highly acceptable to environmentally aware meat [51] and chicken eggs consumers [82]. Consequently, it was concluded that from the perspective of farmers and consumers, the acceptance of novel alternative feedstuffs is improving. Likewise, accompanied by the dissemination of scientific information, GAS is expected to be an interesting alternative feedstuff.

5.1 Bioactive and pigment components of GAS

Astaxanthin content derived from the eggs of GAS has a high value and could be of interest due to its bioactive properties. It is a lipophilic keto-carotenoid, a bio-pigment with an orange-red color, and acts as an antioxidant. Compared to α -tocopherol, lycopene, lutein, and β -carotene, astaxanthin has the highest antioxidant activity against peroxy radical [83]. In poultry industries, commercial synthetic carotenoids play a significant role as feed additives to achieve desired yolk coloration as well as skin carcasses of chicken [71, 72]. However, natural carotenoids are preferable due to the increase in organic farming and the potential health risks posed by synthetic carotenoids [84].

Many workers reported the beneficial effect of incorporating other natural astaxanthin sources, such as microalgae, plants, and fruits. In laying hens, astaxanthin increased antioxidant enzyme activity and decreased malondialdehyde (MDA) of ovaries, in which 60 mg/kg astaxanthin supplementation was more effective than 100 mg/g vitamin E [86]. The concentration of astaxanthin in laying hen increased with increasing levels of dietary astaxanthin (63.34 μ g/g), leading to a rise in yolk color. The addition of astaxanthin upheld reproductive hormones and reduced apoptosis of ovarian cells by upregulating steroid synthesis gene (FSHR gene) and inhibiting pro-

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Nusantoro *et al.* Reviews in Agricultural Science, 12: 147–164, 2024

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apoptosis genes (BAX gene). These mechanisms also involve other genes, such as LHR, CYP family, and NRF2. As a result, laying hens' ovarian aging is lessened by astaxanthin, which may also improve their productivity and egg quality.

In aquaculture, muscle pigmentation is an important criterion for the selection of quality salmonid fillets, in which high-intensity red color is classified as superior [87]. The muscle color of cultured salmonids is considered paler than wild captured fish, which led to the reduction of the product's economic value. To deal with this problem, carotenoids, commonly astaxanthin and canthaxanthin, were added to the ration to increase its deposition in the fish muscle and improve color intensity. For example, 100 mg/kg astaxanthin supplementation increased the chroma value of b^* (redness) rainbow trout fillet by 2.4 times [88].

The GAS eggs could be an alternative animal source of natural astaxanthin, next to crustaceans, such as krill meal. However, the features of GAS eggs have not been extensively investigated, and only three publications reported the bioactivity and pigment of GAS eggs in fish [44, 45] and native chicken [52]. Over the current decade, interest in bioactive natural compounds has been increasing due to their benefits for animals' and consumers' health [89]. The incorporation of GAS eggs in feed is expected to be an essential way to produce innovative functional animal and aquaculture products, as well as achieve better growth performance.

5.2 Chitosan source

Chitosan is derived from chitin, a polymer of N-acetyl-D-glucosamine, by N-deacetylation. Chitin is abundant in nature and commonly found in crustacea, insects, and fungi [90]. A previous study showed that the chemical and biochemical properties of chitosan, such as solubility in organic and inorganic solution as well as its reactivity, are better than chitin [91]. The various applications of chitosan in industries comprise antimicrobials, pharmaceutical materials, cosmetics, food additives, separators, and sewage disposal [91].

Chitosan is used as a feed additive in animal husbandry. A meta-analysis showed that the addition of chitosan decreased ruminal acetate proportion and blood cholesterol, as well as increased propionate proportion, DM, and CP digestibility [92]. It modifies rumen fermentation towards a favorable direction but limitedly affects the performance of ruminants. Similarly, dietary chitosan improves poultry performance and reduces pathogenic bacteria in pigs [93].

Some bioactive compounds, such as essential oils and polyphenols, are vulnerable to oxidation and less bioavailable when passing through the digestive tract. A study showed that the constraint can be dealt with using encapsulation [94]. Saez *et al.* [95] examined the effect of alginate and chitosan as encapsulants on the delivery of bovine serum albumin (BSA) protein for fish. The encapsulant efficiency of chitosan (100%) was higher than alginate (80%) after 30 minutes hardening period. Although this study was limited in methodology as it was unable to precisely quantify the protein within beads, through in vitro and in vivo examination, a combination of alginate and chitosan polymers (30 g/L and 1 g/L, respectively) was found to be the best balance for BSA protection from gastrointestinal proteolytic enzymes, and sustained release of protein in the gut lumen.

Chitosan can be extracted from the shell of GAS, using the following steps, namely powdering, deproteination, demineralization, and deacetylation [83, 84]. This process yielded 53.91% of chitosan, which had low moisture (1.68%) and fair solubility (95.53%) but high ash content (12.31%) due to less effective demineralization. While the processing of chitosan from GAS shell is well studied, the beneficial applications of chitosan from this shell have not been examined in animals and fish.

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Nusantoro *et al.* Reviews in Agricultural Science, 12: 147–164, 2024
https://doi.org/10.7831/ras.12.0_147

5.3 Availability and sustainability of GAS

As described in the previous section, in addition to nutrient content, the factors included in the consideration for feedstuffs are availability and sustainability. Though accurate data on the yield of total biomass from the wild is unavailable, the population of GAS is abundant in paddy fields, specifically in rice-producing countries. This condition can be attributed to the ecological resilience of GAS, which withstands heat stress and cold tolerance [98]. Additionally, it displays a high reproductive rate, with females averaging 1.4 times per week and rapid growth [99].

Pomacea canaliculata has been accepted as edible food in China, Taiwan, and Southeast Asia, whereas the land snail is more prevalent in Southern Europe, specifically Italy [100]. According to Ghosh *et al.* [101], snail gathering is an important food source of livelihood for rural inhabitants but it can affect snail community of a region and it does not sustainably supply edible snails. Therefore, developing snail culture (heliculture) ensures a regular GAS supply. The cultivation of this snail resulted in an edible snail yield of 6.3 kg/m²/cycle, including shell, and the production cycle occurs up to four times a year. In contrast, land snail culture has the potential to yield 6.3 tons/ha/year [100].

The culture of GAS must strictly apply measures to avoid GAS proliferation from cultivating areas to paddy fields or other ecosystems. Invasion of GAS on the ecosystems poses ecological risks, such as threatening native snail (*Cipangopaludina chinensis*) survival, changing community structure due to occupation of the same ecological niche, and changing macrophyte dominance to planktonic algae [86, 87]. Thus, barriers can be installed surrounding the cultivating areas. A set-net barrier could hold up to approximately 4000 adults of GAS [104]. In an experimental setup [105], horizontal electric fencing with a minimum of 0.35 A/m² for less than 10 seconds could inactivate GAS. Other options, such as closed areas using the construction of polythene tunnels [101], might be adopted to deal with it.

6. Conclusions

The nutritional composition, nutritive value, constraints, and future use of GAS as animal and fish feed have been reviewed. The GAS is rich in protein and calcium, representing 39.11 to 68.67% (DM) and 41.38% (DM) from their meat and shell, respectively. The nutritive value of GAS is well examined, and its utilization as a feed alternative to fishmeal protein shows good performance results and benefits for farm animals (particularly monogastric) and fish. The constraints in using GAS as a feed include antinutritional, heavy metals absorption, and parasitic risks. However, they can be reduced using some techniques, such as acidic conditions and heat treatment to inactivate antinutritional, and some animals can adapt to the antinutritional of GAS. The availability of GAS as a feed resource can be obtained from the wilds (paddy fields, swamps, and canals) and heliculture, which strictly apply measures to prevent escaping GAS from cultivating areas. Amongst the parts of GAS, the utilization of the eggs and shells as animal and fish feed is studied to a lesser extent. The astaxanthin of the eggs of GAS and chitosan derived from their shell are interesting due to their bioactivities, opening new avenues for further research in functional feed additives.

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