



Determination of reference data for the
water/protein ratio
in farmed shrimp
of the species *Litopenaeus vannamei*
in relevant countries of origin,
taking into account the processing stages

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Introduction

The water/protein ratio for shrimp is used to assess the extraneous water content. The currently available values vary considerably and there is uncertainty as to what is to be regarded as "natural", i.e. what may be expected in the sense of a commercial perception. In the opinion of the authors, this study is the first of its kind which, with a view to representativeness, is based on a relevant number of comparable samples, depending on the provenance, and living conditions, and provides reliable and valid reference data. For this purpose, farmed shrimp of the species *Litopenaeus vannamei* were sampled in Ecuador, India, and Vietnam under the usual real technological conditions. The present study shows, that technological processing steps after killing, such as storing, transporting, heading, peeling, deveining or rinsing, result in absolute crude protein loss. Therefore, the degree of processing of shrimp must be considered when assessing the water/protein ratio.

Background

There is considerable uncertainty as to which water/protein ratio for shrimps is to be regarded as "natural", i.e. if products are manufactured without water-binding substances, what kind of water/protein ratio may be expected in the sense of a commercial perception. Reliable reference data are required to assess the quality of imported goods or to identify shrimp reliably that have been treated but not labelled correctly.

Almost all known previous scientific studies refer to relatively few samples or products already traded commercially, which were taken in retail or wholesale trade. In the opinion of the authors, the present study is the first of its kind to be based on a relevant number of comparable samples with a view to representativeness and, above all, to document the exact history of each individual sample. In other words, the "biographies" of the individual samples are fully known, which is not the case with samples taken at trading level.

Furthermore, as far as known to the authors, industrial processing of farmed shrimp of the species *Litopenaeus vannamei* has not yet been scientifically investigated. This is another focus and innovation in the present study, because it was taken into account that the water/protein ratio changes in the course of the processing procedure of a shrimp simply by the fact, that shrimp are killed, stored, transported, decapitated, peeled and deveined. The aim is therefore also to determine and evaluate the development of the water/protein ratio throughout the entire process. Sampling was carried out concomitantly during the usual working hours of the processing steps harvesting, storage, transport, production or processing in the respective countries. Thus, they corresponded to the usual real technological conditions.

By examining shrimp of different provenance and season, the available data can help to understand and evaluate the diversity of the water/protein ratio of untreated products. It remains to be taken into account that shrimp are individuals, that are not homogeneous in their chemical composition.

Structure of the study

Sampling of farmed shrimp of the species *Litopenaeus vannamei* took place in the countries of origin Ecuador, Vietnam and India, which are mainly relevant for the German and neighbouring European market. Only shrimp originating from farms and processing plants, that have passed a certification procedure of the Aquaculture Stewardship Council (ASC) and were marketed under the ASC seal, were examined. The breeding tanks (ponds) in Ecuador were filled with seawater, in India with freshwater and in Vietnam with brackish water.

In preparation for the study, a test plan including working instructions for the investigating laboratories was drawn up so that comparable sampling and investigations could be carried out in all countries. A total of six or seven processing stages were sampled individually:

- Sample 1: Farm (Shrimp HOSO¹), unfrozen
Analysis: shrimp complete, with head and carapace
- Sample 2: Farm (Shrimp HOSO), unfrozen
Analysis: shrimp peeled in the laboratory, only edible part, with intestine
- Sample 3: Arrival factory (Shrimp HOSO), unfrozen
Analysis: shrimp peeled in the laboratory, only edible part, with intestine
- Sample 4: Shrimp HLSO², frozen
Analysis: shrimp peeled, edible part only, with intestine
- Sample 5: HL Easy Peel³, frozen
Analysis: shrimp peeled, edible part only
- Sample 6: PPV⁴ / PCD⁵, frozen
Analysis: shrimp peeled, edible part only
- Sample 7: PPV / PCD with salt, frozen
Analysis: shrimp peeled, edible part only

Processing stage 7 represents a permitted use of table salt, that is declared in the list of ingredients. The addition of extraneous water was less than 5 % in this processing stage and was adjusted by differential weighing during production.

The sampling was carried out by Eurofins or independent partners in the respective countries and documented in writing and photographically. Sampling protocols were drawn up and the geographical location of the respective ponds and processing plants was recorded by means of digital map display. The samples were sealed directly during sampling, so that subsequent manipulation was precluded.

In all three countries of origin samples 1 to 3 were analysed immediately on site when unfrozen in order to obtain through immediate analysis the most unbiased results possible during harvest and transport to the processing factories. Any technological step, such as freezing or thawing, would have distorted the actual chemical state of the farmed shrimp through water absorption or release.

The other samples 4 to 7 were stored in the factories, internally transported, headed, peeled and deveined analogous to the usual production conditions and market-standard customer requirements. According to the variants, the shrimp were individually frozen (IQF, individually quick frozen) after the processing step had been carried out. The samples taken at the respective processing stages were sealed directly at the time of sampling by the independent sampler, exported by the processing plants to Germany with the ordered standard goods in the container and provided by the importer in Germany

¹ „head on / shell on“ - with head and carapace

² „headless / shell on“ – decapitated, with carapace

³ „headless Easy Peel“ - decapitated, carapace incised for easy peeling

⁴ „peeled pin deveined“ - decapitated, peeled, intestine pulled

⁵ „peeled cut deveined“ - decapitated, peeled, intestine cut out

or France. Subsequently, the samples were delivered to Eurofins. Thus, the samples were exposed to the same transport conditions as the standard traded goods.

For each processing stage, eight samples in two sizes (small/large) were to be analysed as single determinations. Thus, six production stages were planned for Ecuadorian goods (the producer in Ecuador did not carry out production stage 7) and seven production stages in Vietnam and India with 16 samples each in two sizes and three countries, in total 320 planned individual examinations.

In addition, three water samples were planned to be taken during the harvest in one pond each in order to obtain data on the main habitat of the shrimp and to take into account, that the ponds are filled with freshwater, brackish water or seawater, depending on the geographical location (inland, coastal areas). Likewise, this should be used to investigate whether the living environment has a significant influence on the chemical composition of the shrimp.

Due to the difficulty in finding appropriate farms and processing plants in India, sampling was delayed for over a year. Initially the Indian trading partners were very reluctant to sampling and did not want to allow it. After a supplier was found, one sampling could be carried out. The second sampling in India could no longer be realised due to the global impact caused by the appearance of the Coronavirus SARS-CoV-2. Hence this investigation is only based on the results from the first sampling.

Table 1 describes the parameters recorded from the shrimp sampling and the water sampling. A total of 264 individual samples were examined.

Table 1: Parameters recorded from Ecuador, Vietnam and India

Shrimp samples (Ecuador, first sampling)	E_A1 to E_A6
Sampling date	12/13 March 2019
Stocking density [shrimp per m ³]	38
Product temperature at harvest [°C]	32
Count [Piece per lb] decapitated, peeled, deveined	41-50
Samples	E_A1.1 to E_A6.8
Number of individual samples	6 x 8 = 48
Shrimp samples (Ecuador, second sampling)	E_B1 to E_B6
Sampling date	12/13 April 2019
Stocking density [shrimp per m ³]	28
Product temperature at harvest [°C]	31
Count [Piece per lb] decapitated, peeled, deveined	31-35
Samples	E_B1.1 to E_B6.8
Number of individual samples	6 x 8 = 48
Shrimp samples (Vietnam, first sampling)	V_A1 to V_A7
Sampling date	29 May 2019
Stocking density [shrimp per m ³]	180
Product temperature at harvest [°C]	29
Count [Piece per lb] decapitated, peeled, deveined	61/70
Samples	V_A1.1 to V_A7.8
Number of individual samples	7 x 8 = 56
Shrimp samples (Vietnam, second sampling)	V_B1 to V_B7
Sampling date	29 May 2019
Stocking density [shrimp per m ³]	153
Product temperature at harvest [°C]	29
Count [Piece per lb] decapitated, peeled, deveined	41/50
Samples	V_B1.1 to V_B7.8
Number of individual samples	7 x 8 = 56

Paul et al. (2021), Reference data for the water/protein ratio in farmed shrimp of the species *Litopenaeus vannamei*.

Shrimp samples (India, first sampling)	I_A1 to I_A7
Sampling date	10/11 January 2020
Stocking density [shrimp per m ³]	60
Product temperature at harvest [°C]	28
Count [Piece per lb] decapitated, peeled, deveined	61/70
Samples	I_A1.1 to I_A7.8
Number of individual samples	7 x 8 = 56
Water samples (Ecuador)	E_W1 to E_W3
Sampling date	12 April 2019
Product temperature at harvest [°C]	No indication
Number of individual samples	3
Water samples (Vietnam)	V_W1 to V_W3
Sampling date	29 May 2019
Product temperature at harvest [°C]	30,2
Number of individual samples	3
Water samples (India)	I_W1 to I_W3
Sampling date	10 January 2020
Product temperature at harvest [°C]	27,5
Number of individual samples	3

Investigated Parameters

Table 2 lists the chemical parameters studied and methods used for each country. A total of 1,848 individual values could be generated from the 264 individual samples with the seven parameters listed. If the shrimp were glazed, they were deglazed before analysis by removing the ice glaze in accordance with the Codex STAN 921981 method (as of 2014).

Table 2: Chemical parameters investigated

Parameter	Methods Ecuador	Methods Vietnam	Methods India	Methods Germany
Water	INEN 464:2013	Internal method (EHC-TP2-048) (Ref. 79.7 FAO Food 14/7-1986)	AOAC 952.08	§64 LFGB L 06.00-3: 2004-07, mod., PV1100, Gravimetry
Crude protein	Kjeldahl	Internal method (EHC-TP2-047) (Ref. FAO Food 14/7-1986)	IS 7219:1973	§64 LFGB L 06.00-7: 2014-07, mod., PV1402, Kjeldahl (titrimetric)
Sodium	AOAC 985.35: 19th 2012	AOAC 969.23	AOAC 2011.14	DIN EN ISO 11885, mod., CON-PV00006 (2017-08), ICP-OES
Chloride	AOAC 937.09: 19th 2012	Internal method (EHC-TP2-061) (Ref. FAO Food 14/7-1986)	AOAC 937.09	Internal method PV1501:2016-04, Potentiometry

Paul et al. (2021), Reference data for the water/protein ratio in farmed shrimp of the species *Litopenaeus vannamei*.

Parameter	Methods Ecuador	Methods Vietnam	Methods India	Methods Germany
Phosphorus	AOAC 965.17: 19th 2012	AOAC 995.11	AOAC 2011.14	DIN EN ISO 11885, mod., CON-PV00006 (2017-08), ICP-OES
Potassium	-	AOAC 969.23	AOAC 2011.14	DIN EN ISO 11885, mod., CON-PV00006 (2017-08), ICP-OES
pH value	INEN 783:1985	TVCN 7806:2007 (ISO 1842:1991)	Internal method (pH meter)	§ 64 LFGB L06.00-2: 1980-09, PV 1602, Potentiometry

Individual results

Table 3 shows the arithmetic mean (AM) and standard deviation (SD) of the individual results per sampling from the countries Ecuador, Vietnam and India as well as the results of the water samples.

Table 3: Sampling from Ecuador, Vietnam and India - mean values with standard deviation

		Water	Crude protein	Chloride	pH value	Phosphorus	Phosphate calculated as P ₂ O ₅	Potassium	Sodium
		g/100 g				mg/kg			
Individual results from Ecuador, first sampling									
E_A1	AM	76,0	20,3	0,797	5,88	7575	17481	-	3355
	SD	±0,5	±1,6	±0,475	±0,10	±817	±1886	-	±469
E_A2	AM	75,0	22,8	0,903	5,92	3463	7990	-	3418
	SD	±2,2	±1,4	±0,358	±0,06	±795	±1833	-	±255
E_A3	AM	77,1	22,6	0,343	6,35	3429	7912	-	1213
	SD	±0,7	±1,0	±0,044	±0,10	±442	±1021	-	±161
E_A4	AM	77,1	20,3	0,203	6,50	2200	5025	3188	1238
	SD	±0,3	±0,1	±0,020	±0,00	±107	±255	±113	±52
E_A5	AM	78,0	20,0	0,198	6,44	2125	4850	3050	1200
	SD	±0,1	±0,1	±0,004	±0,05	±89	±177	±214	±76
E_A6	AM	79,0	18,7	0,169	6,24	2000	4600	2650	979
	SD	±0,6	±0,2	±0,012	±0,05	±0	±0	±76	±22
Individual results from Ecuador, second sampling									
E_B1	AM	74,4	20,6	1,698	5,96	2733	6308	-	3396
	SD	±0,9	±3,6	±0,106	±0,05	±225	±519	-	±226
E_B2	AM	73,5	22,2	0,969	6,00	2825	6519	-	3899
	SD	±2,0	±1,4	±0,086	±0,13	±1412	±3258	-	±328
E_B3	AM	75,6	26,7	0,361	6,30	3613	8337	-	1808
	SD	±1,2	±2,8	±0,049	±0,10	±494	±1140	-	±483

Paul et al. (2021), Reference data for the water/protein ratio in farmed shrimp of the species *Litopenaeus vannamei*.

		Water	Crude protein	Chloride	pH value	Phosphorus	Phosphate calculated as P ₂ O ₅	Potassium	Sodium
		g/100 g				mg/kg			
E_B4	AM	75,9	21,3	0,204	6,44	2500	5700	3575	1238
	SD	±0,2	±0,2	±0,009	±0,05	±0	±0	±104	±52
E_B5	AM	76,4	21,0	0,205	6,50	2400	5486	3275	1263
	SD	±0,2	±0,2	±0,014	±0,00	±115	±261	±139	±106
E_B6	AM	78,0	19,8	0,180	6,25	2357	5414	2925	957
	SD	±0,2	±0,2	±0,000	±0,05	±53	±107	±183	±27
Individual results from Vietnam, first sampling									
V_A1	MW	78,2	20,1	0,229	7,44	2500	5729	3789	1815
	SD	±0,3	±0,5	±0,007	±0,05	±82	±187	±72	±16
V_A2	MW	78,0	19,9	0,236	7,34	2375	5442	3848	1844
	SD	±0,3	±0,3	±0,008	±0,16	±116	±267	±93	±65
V_A3	MW	78,1	20,0	0,224	7,20	2525	5786	3874	1773
	SD	±0,3	±0,3	±0,009	±0,15	±183	±420	±40	±59
V_A4	MW	78,6	19,3	0,349	6,90	2500	5725	3714	1613
	SD	±0,3	±0,2	±0,012	±0,00	±227	±518	±157	±155
V_A5	MW	78,9	19,7	0,249	7,00	2500	5700	3625	1463
	SD	±0,3	±0,3	±0,022	±0,00	±0	±0	±128	±92
V_A6	MW	80,6	17,8	0,176	6,80	2325	5338	3063	924
	SD	±0,1	±0,2	±0,005	±0,00	±71	±169	±74	±49
V_A7	MW	81,8	16,3	0,729	6,80	2000	4600	2313	3613
	SD	±0,3	±0,4	±0,043	±0,00	±0	±0	±83	±83
Individual results from Vietnam, second sampling									
V_B1	MW	79,2	19,2	0,266	7,43	2275	5213	3697	2075
	SD	±0,2	±0,3	±0,009	±0,03	±139	±318	±42	±64
V_B2	MW	79,2	19,1	0,266	7,42	2263	5184	3719	2109
	SD	±0,3	±0,2	±0,007	±0,05	±119	±272	±101	±34
V_B3	MW	79,1	19,3	0,271	7,18	2413	5528	3713	2093
	SD	±0,4	±0,3	±0,021	±0,06	±113	±258	±74	±134
V_B4	MW	77,8	20,1	0,294	6,90	2600	6000	3900	1500
	SD	±0,2	±0,3	±0,026	±0,00	±0	±0	±0	±0
V_B5	MW	77,8	20,3	0,262	6,80	2538	5825	3650	1338
	SD	±0,3	±0,1	±0,004	±0,00	±106	±260	±107	±74
V_B6	MW	80,2	18,2	0,161	6,80	2300	5300	3086	856
	SD	±0,1	±0,1	±0,008	±0,00	±0	±0	±69	±42
V_B7	MW	80,1	18,1	0,584	6,74	2275	5213	2800	2888
	SD	±0,4	±0,4	±0,017	±0,05	±71	±189	±107	±136
Individual results from India, first sampling									
I_A1	MW	78,2	19,2	0,253	7,13	2033	4684	2880	1472
	SD	±0,2	±0,3	±0,028	±0,22	±77	±176	±247	±127
I_A2	MW	78,6	18,9	0,217	6,76	1956	4508	2655	1410
	SD	±0,6	±0,4	±0,025	±0,13	±105	±242	±227	±109

Paul et al. (2021), Reference data for the water/protein ratio in farmed shrimp of the species *Litopenaeus vannamei*.

	Water	Crude protein	Chloride	pH value	Phosphorus	Phosphate calculated as P ₂ O ₅	Potassium	Sodium	
	g/100 g				mg/kg				
I_A3	MW	78,1	19,1	0,226	7,11	2438	5617	3151	1272
	SD	±0,3	±0,4	±0,020	±0,11	±179	±413	±114	±54
I_A4	MW	78,6	19,1	0,210	6,65	2138	4900	2829	1375
	SD	±0,2	±0,3	±0,095	±0,05	±151	±334	±125	±158
I_A5	MW	80,3	17,4	0,280	6,71	2113	4838	2043	2525
	SD	±0,2	±0,2	±0,168	±0,12	±136	±292	±127	±225
I_A6	MW	79,8	17,9	0,218	6,63	1850	4238	2088	1386
	SD	±0,6	±0,4	±0,098	±0,05	±107	±262	±136	±69
I_A7	MW	82,4	15,5	0,510	6,80	2086	4771	1388	3000
	SD	±0,5	±0,5	±0,019	±0,00	±69	±138	±83	±160
Mean values from the two samplings for Ecuador, Vietnam and one sampling for India for processing steps 4-6									
E_4		76,5	20,8	0,204	6,5	2350	5363	3382	1238
E_5		77,2	20,5	0,202	6,5	2263	5168	3163	1232
E_6		78,5	19,3	0,175	6,2	2179	5007	2788	968
V_4		78,2	19,7	0,322	6,9	2550	5863	3807	1557
V_5		78,4	20,0	0,256	6,9	2519	5763	3638	1401
V_6		80,4	18,0	0,169	6,8	2313	5319	3075	890
I_4		78,6	19,1	0,210	6,7	2138	4900	2829	1375
I_5		80,3	17,4	0,280	6,7	2113	4838	2043	2525
I_6		79,8	17,9	0,218	6,6	1850	4238	2088	1386
Mean values with standard deviation of the individual results of the water samples									
		Country		Table salt		pH value		Chloride	
				g/100 g				g/100 g	
E_B_W1-W2	MW	Ecuador		3,31		8,05		2,02	
	SD			±0,06		±0,09		±0,03	
V_A_W1-W3	MW	Vietnam		2,44		7,13		1,49	
	SD	SD		±0,05		±0,06		±0,03	
I_A_W1-W3	MW	India		0,77		7,43		0,47	
	SD	SD		±0,03		±0,05		±0,02	

Statistics

Simple statistical methods were used to evaluate the data. Upper and lower outliers were eliminated, if they were more than 1.5 interquartile range above the upper quartile Q3 or below the lower quartile Q1. The mean was then calculated for graphical representation.

Paul et al. (2021), Reference data for the water/protein ratio in farmed shrimp of the species *Litopenaeus vannamei*.

Evaluation of the water content

After harvesting and killing the shrimp they were transported to the processing factories in containers filled with ice water. There, if necessary, cold storage on ice was continued until the next processing steps began.

Looking at sample no. 2, the sampling at the farm (shrimp HOSO, shrimp peeled, only edible part, with intestine), we can already see a large variance with 73.5 % to 79.2 % water content. The individual values vary widely from 71.6 % (minimum) to 79.7 % (maximum). Tawade et al. (2019) describe values of 73.2 % to 77.9 % in their studies. The samples presented in that study were taken in India, so that the maximum values from India presented here are confirmed.

Due to the use of ice or water for cooling and rinsing, which is necessary during the technological processing steps, an increase in water takes place over the process without this being caused by the use of additives. This increase varies depending on the region: In Ecuador, an increase of approx. 4% was analysed from harvesting (sample 2) to the headed, peeled and de-shelled shrimp (sample 6), in Vietnam and India only approx. 1 - 2% at the comparable processing stages 2 to 6.

Figures 1a and 1b below show the water contents at the respective processing stages.

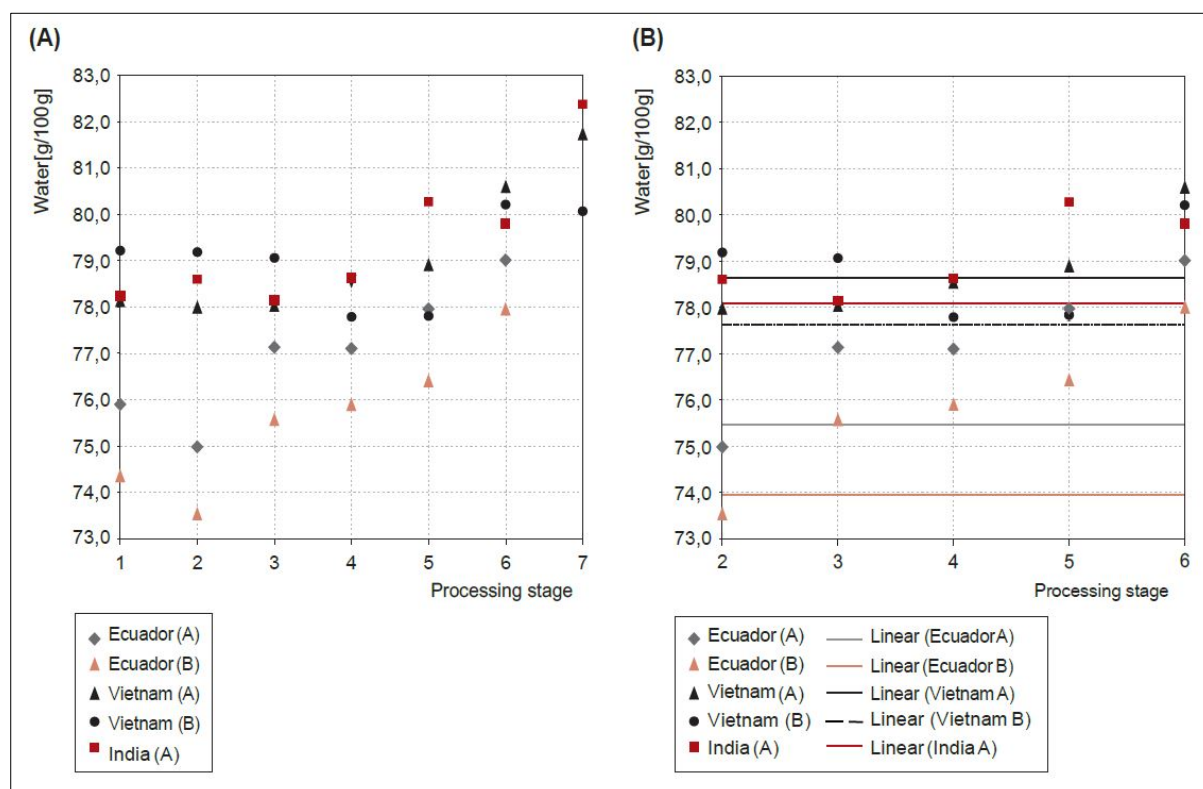


Figure 1 Water (g/100 g), mean values per country, sampling and processing stages 1-7 (A); water (g/100 g), mean values per country, sampling and processing stages 2-6 with linear distribution (B)

Paul et al. (2021), Reference data for the water/protein ratio in farmed shrimp of the species *Litopenaeus vannamei*.

Evaluation of the crude protein content

A loss of crude protein was documented throughout the technological processing steps. Mechanical stress and the use of water for rinsing flush the crude protein out of the shrimp.

The samples from processing stage 2 have a relatively high crude protein content of 18.9 % to 22.8 % in unprocessed shrimp. However, a clear decrease of approx. 1-3 % can be seen with the untreated samples of processing stage 6, which are comparable across all origins.

Müller-Hohe et al. (2019) define mean crude protein values for the species *Litopenaeus vannamei* of 21.4 % in their study. In Tawade et al. (2019) values between 16.8 % and 17.6 % are found.

As the individual results (see Table 3) and the following graphical evaluations (see Figures 2a and 2b) show in this study, the lowest crude protein content within the technological necessary processing steps up to the untreated sample 6 is a minimum of 17.4 % (India sample A5, shrimp headed, shell incised for easy peeling, with intestine), which again is in line with the values from the study by Tawade et al. (2019). The mean crude protein content of the three provenances determined here was 19.2 % (from 17.9 % to 20.8 %).

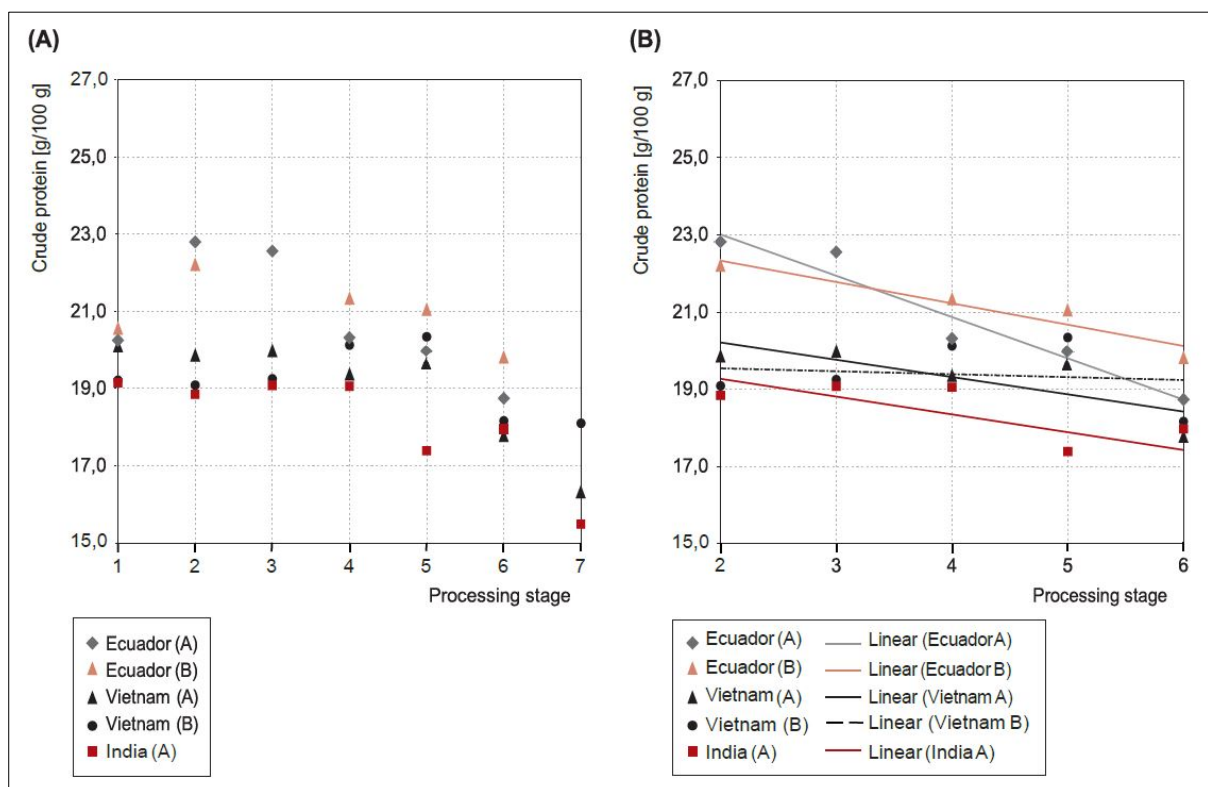


Figure 2 Crude protein (g/100 g), mean values per country, sampling and processing stages 1-7 (A); crudeprotein (g/100 g), mean values per country, sampling and processing stages 2-6 with linear distribution (B)

Paul et al. (2021), Reference data for the water/protein ratio in farmed shrimp of the species *Litopenaeus vannamei*.

Evaluation of the water/protein ratio

Correlating to the increasing water and the decreasing crude protein contents, the water/protein quotient in the observation of the linear trend lines increased in the course of the technologically necessary processing steps up to the untreated sample 6.

As the individual results (see Table 3) and their graphical evaluations (see Figures 3a and 3b) show, the highest water/protein ratio within the technological necessary processing steps up to untreated sample 6 is a maximum of 4.42 (Vietnam sample B6, shrimp headed, peeled, without intestine). For the preceding processing steps 4 (shrimp headed, with intestine) and 5 (shrimp headed, shell incised for easy peeling, with intestine), the maximum water/protein ratios are around 4.

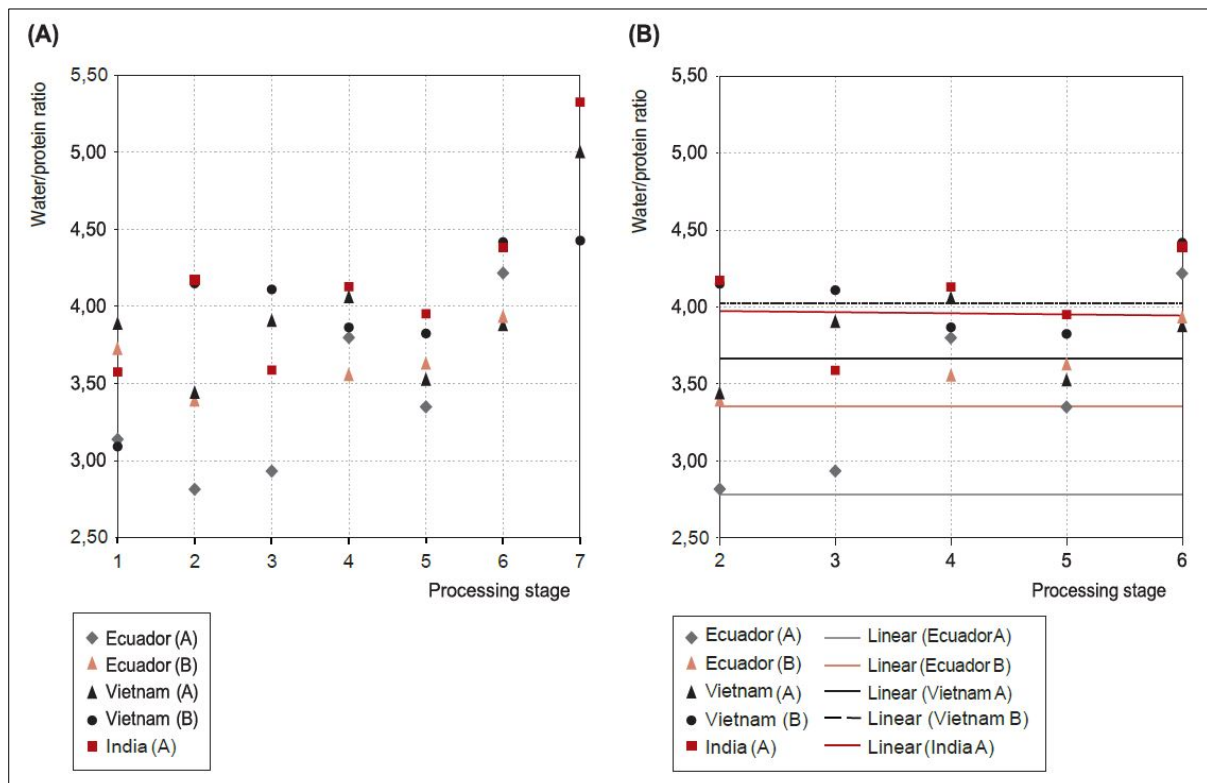


Figure 3 Water/protein ratio, mean values per country, sampling and processing stages 1-7 (A); pH values, mean values per country, sampling and processing stages 2-6 with linear distribution (B)

Evaluation of the pH values of the shrimp and water samples

Figures 4a and 4b show the mean values of the pH values of the shrimp at the respective processing stages and of the ambient water.

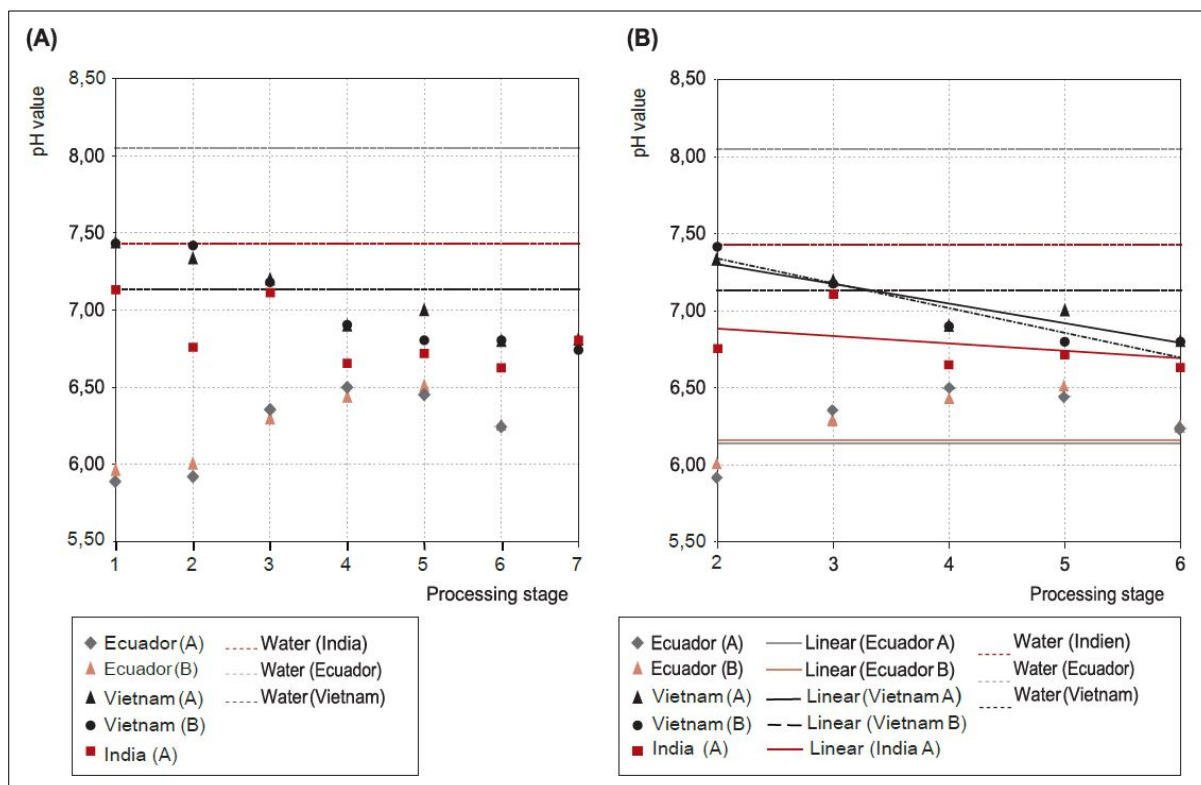


Figure 4 pH values, mean values per country, sampling and processing stages 1-7 (A); pH values, mean values per country, sampling and processing stages 2-6 with linear distribution (B)

As these two figures and the individual results (see Table 3) show, the pH value trend in the species *Litopenaeus vannamei* differs depending on the origin. In Ecuador, the pH values increase from approx. 6 to approx. 6.5, whereas in Vietnam and India a decrease in pH values from 7.4 to approx. 6.7 is evident. The pH values of the water do not show any direct connection to the pH values of the individual shrimp, so that no conclusions can be drawn here. In Liang et al. (2008), pH values of raw shrimp of 7.0 are described as a function of the salt content of the culture tanks. When the salinity is 3 g/100 g, the water and protein contents of the shrimp are lower than in animals living in water with a lower salinity (0.005-0.15 g/100 g). This is consistent with the results presented here.

In the minutes of the 79th working meeting of the ALTS (*Arbeitskreis der auf dem Gebiet der Lebensmittelhygiene und der Lebensmittel tierischer Herkunft tätigen Sachverständigen*; "working group of experts for food hygiene and food of animal origin") the guideline: "Water addition in unprocessed fishery products - detection and possibilities of assessment" was published. The ALTS defines that raw, unprocessed fish fillets usually have pH values below 7.0. This value can be confirmed from the present results for the harvested samples from Ecuador and India and for the frozen samples from Vietnam.

Evaluation of the chloride values

Figures 5a and 5b show the chloride levels of the shrimp at each stage of processing and the chloride content of the ambient water.

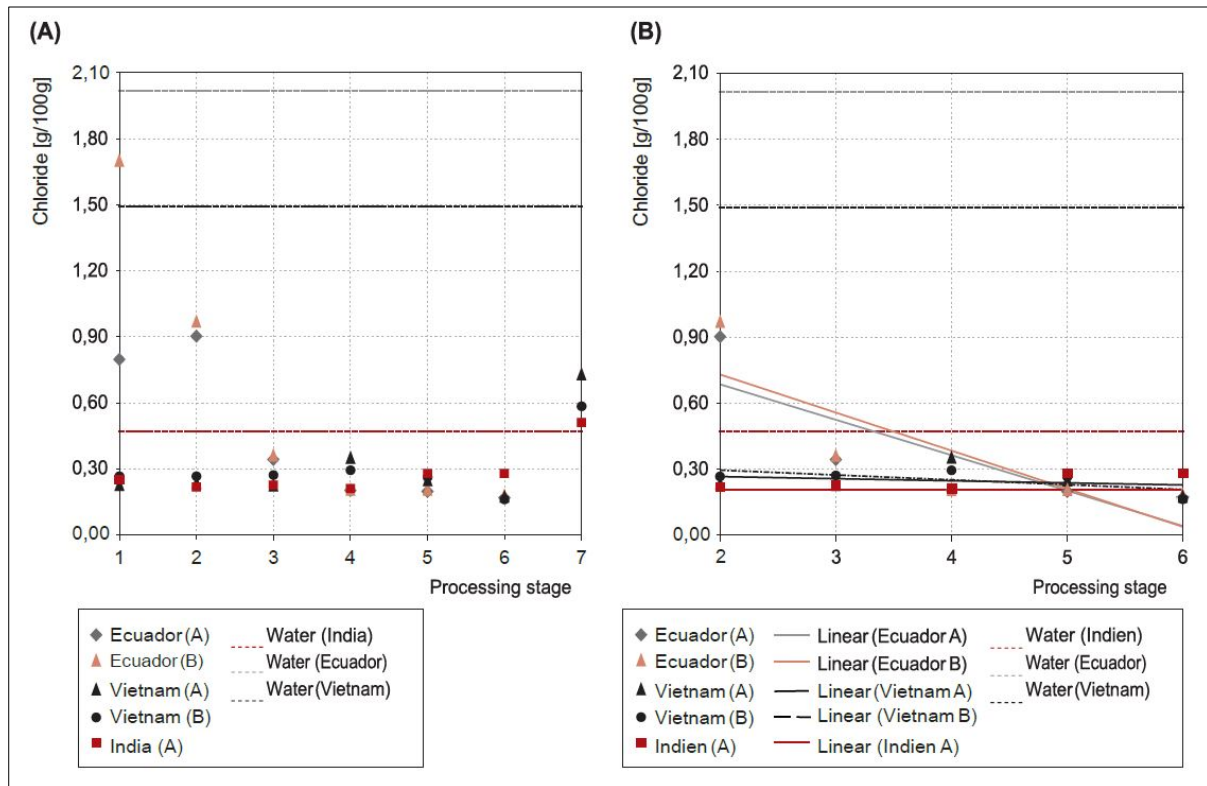


Figure 5 Chloride (g/100 g), mean values per country, sampling and processing stages 1-7, and water sample contents, mean (A); Chloride (g/100 g), mean values per country, sampling and processing stages 2-6 with linear distribution, and water sample contents, mean (B)

The chloride levels in the pond water have an influence on the chloride content in the shrimp. The higher the content in the water, the higher the value in the shrimp. This can be shown with the example of Ecuador, where the chloride content in the water is 2.10 g/100 g and in the shrimp 1.70 g/100 g. The chloride content in the water increases over the course of the processing stages. The values decrease significantly in the course of the processing stages, up to the addition of salt. Similarly, the chloride values analysed reflect the local conditions: in Vietnam, where the shrimps are kept in brackish water, 1.49 g/100 g, and in India, where the shrimps are kept in fresh water, 0.47 g/100 g. The chloride values in the shrimps are also lower than in the brackish water. Müller-Hohe et al. (2019) also mention chloride contents in this order of magnitude (mean value 0.21 g/100 g, maximum value 0.39 g/100 g).

Paul et al. (2021), Reference data for the water/protein ratio in farmed shrimp of the species *Litopenaeus vannamei*.

Evaluation of the elements sodium/potassium/ phosphorus

Figures 6-8 show the elements sodium, potassium and phosphorus for the individual countries and processing stages.

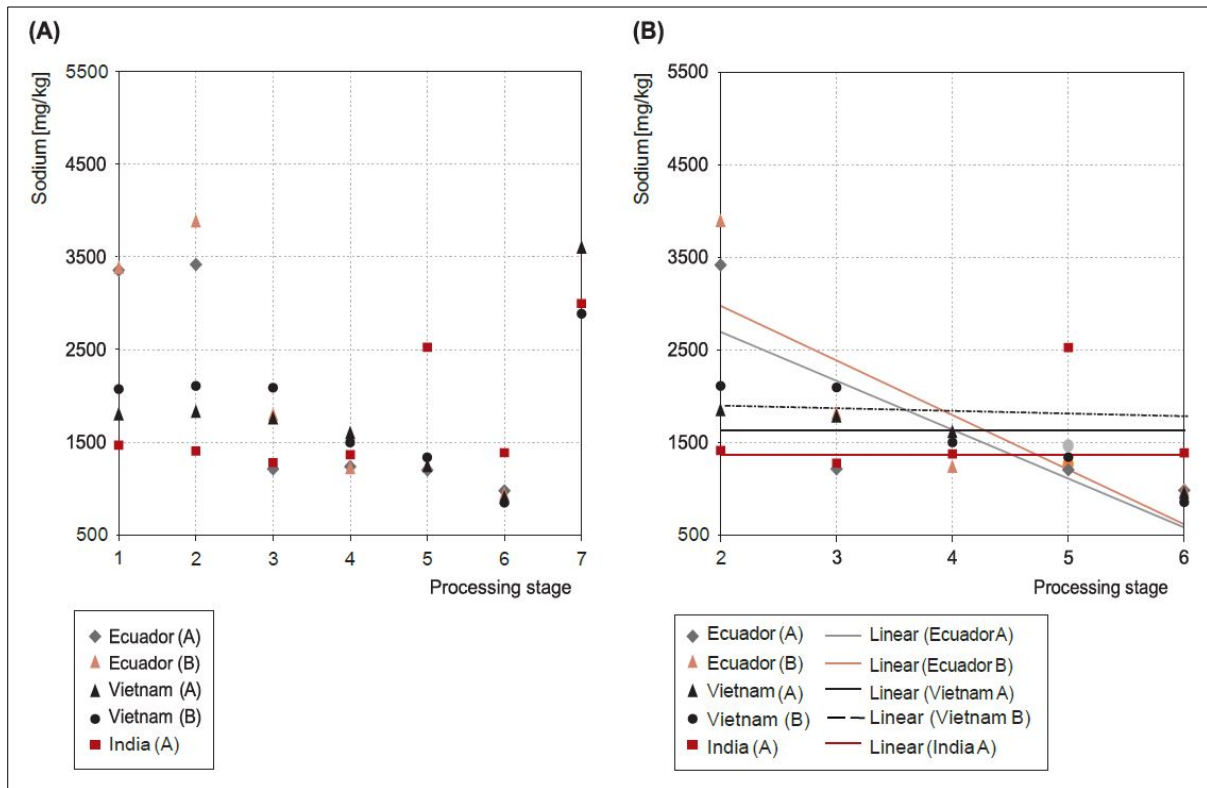


Figure 6 Sodium (mg/kg), mean values per country, sampling and processing levels 1-7 (A); sodium (mg/kg), mean values per country, sampling and processing levels 2-6 with linear distribution (B)

Paul et al. (2021), Reference data for the water/protein ratio in farmed shrimp of the species *Litopenaeus vannamei*.

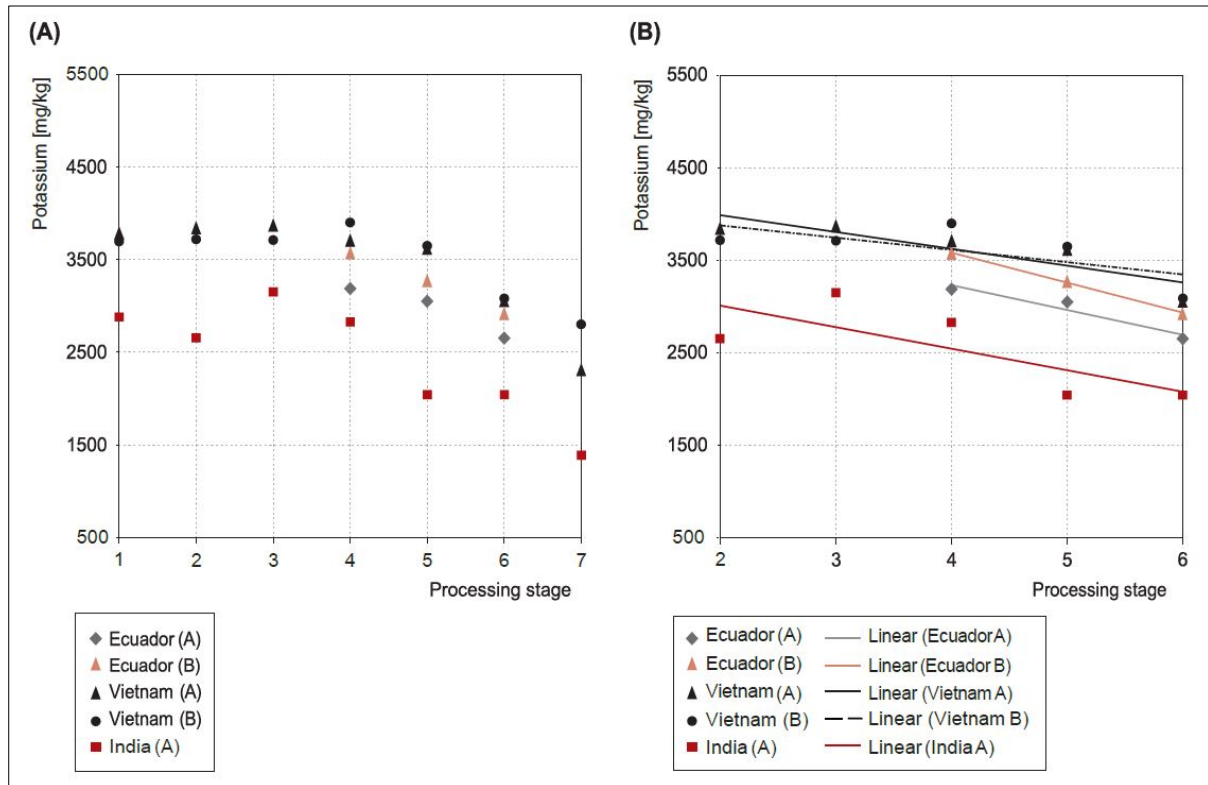


Figure 7 Potassium (mg/kg), mean values per country, sampling and processing stages 1-7 (A); potassium (mg/kg), mean values per country, sampling and processing stages 2-6 with linear distribution (B)

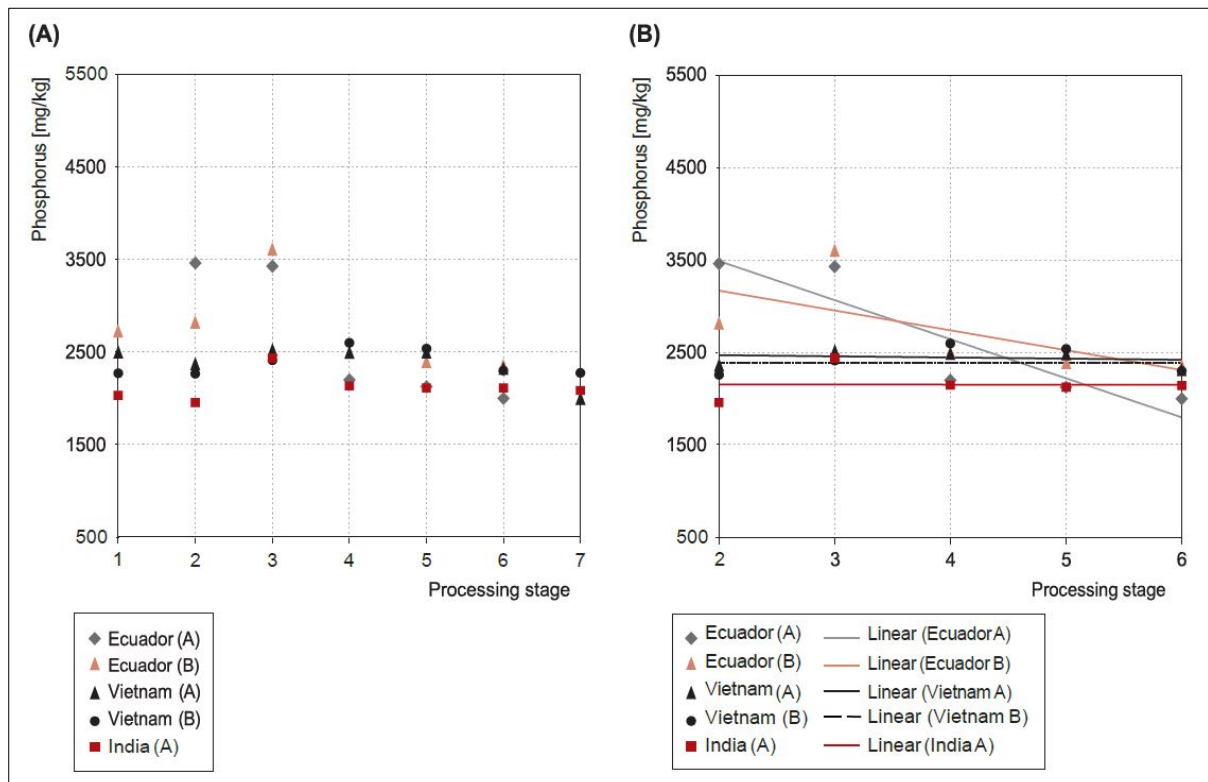


Figure 8 Phosphorus (mg/kg), mean values per country, sampling and processing stages 1-7 (A); phosphorus (mg/kg), mean values per country, sampling and processing stages 2-6 with linear distribution (B)

Paul et al. (2021), Reference data for the water/protein ratio in farmed shrimp of the species *Litopenaeus vannamei*.

It can be seen that all values decrease during the processing stages. Only in the Indian samples the sodium values increase. Unfortunately, due to the omission of the second sampling in India, this phenomenon cannot be verified. Miller et al. (2018) show that the processing steps of glazing, freezing and deglazing can lead to a loss of sodium and potassium, among other things. This is confirmed by the present results.

Major factors influencing the growth of farmed shrimps

When assessing commercial farmed shrimps, it must be taken into account, that various factors influence the chemical composition. First and foremost, the breeding tanks differ, as they may be filled with freshwater, brackish water or seawater depending on the geographical location (inland, coastal areas). Depending on the country of origin, breeding tanks are limed for animal health reasons before the introduction of new larvae and the associated refilling with water in order to prevent diseases, which can slightly change the pH value of the water (Mahesh Aqua, 2020).

In addition, the feedstuff used plays a significant role, as the biochemical utilisation of the various plant or animal crude proteins differs in addition to the crude protein content (Wang et al., 2014).

Likewise, different climatic conditions, especially temperature differences or flooding, lead to variances in metabolism and thus also in crude protein and fat formation (Mahesh Aqua, 2020).

Technological limits or water uptake versus crude protein loss

Regularly, official sources calculate extraneous water contents on the basis of chemical data available to date, which are in the mid two-digit range. However, there are technological limits to the water uptake of farmed shrimps. The "inflation" of shrimps with water is limited. Thus, based on technological experience, up to approx. 25 % water addition can be achieved by pH shift and/or water-binding additives (e.g. diphosphates).

Therefore, the analysed water/protein ratio does not reflect the water absorption. The evaluation of the extraneous water content on the basis of the water/protein ratio can as a consequence not be used as a valid and decisive criterion, but must be evaluated as an indication.

Conclusion

The examination of a large number of samples subjected to comparable treatment and analysis shows the high variability in terms of shrimp composition, depending on provenance and living conditions.

The pH values determined in this investigation are in accordance with the ALTS Guideline from 2017 "Water addition in unprocessed fishery products - detection and possibilities of assessment", according to which pH values above 7.5 in the final product are evaluated as an indication of the use of alkalisating substances in the species *Litopenaeus vannamei*.

The decrease of the elements sodium, potassium and phosphorus in the course of the processing steps is recognisable. The steps 2-6 are processing steps, which lead to products, that are on the market in different forms of presentation. However, there is an exception if the addition of salt is declared in the ingredients and the product is marketed with the corresponding declaration. Therefore, this process step was included in the investigations with processing step 7. In all other processing steps, no ingredients or additives were used. The increase in sodium in India is not assessable.

The absolute loss of crude protein, that occurs during the technological processing steps, plays an essential role in the evaluation of the water/protein ratio and the associated extraneous water calculations. Crude protein is washed out through mechanical stress and the use of ice or water for cooling and rinsing or also transporting the shrimps to be processed in tanks. The duration of the respective processing step must also be taken into account. An "increased" water/protein ratio, compared to the shrimps in the pond, therefore does not necessarily mean, that extraneous water was

Paul et al. (2021), Reference data for the water/protein ratio in farmed shrimp of the species *Litopenaeus vannamei*.

added. Rather, the degree of processing of the shrimps must be considered, and the crude protein content evaluated or the crude protein loss taken into account. The mean crude protein content of the three provenances determined here was 19.2 % (from 17.9 % to 20.8 %) over the processing stages 4-6.

For the evaluation of products that commercially traded (retail or whole sale) the question arises the question whether the water/protein ratio should be defined for each processing step. This poses the challenge of generalising a quotient, since the origin and size of the animals lead to significant variances. As the evaluations according to Figure 3a and 3b show, the water/protein ratio can vary greatly within a processing step. Whilst in the relatively untreated processing stages 2 and 3 the dispersion of the water/protein ratio is very large, a certain consolidation can be observed at processing stage 6.

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