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Effect of fermentation and drying practices on the chemical and physical profiles of Ghana cocoa

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A series of 12 cocoa fermentations were carried out at the Cocoa Research Institute of Ghana in 1987. The cocoa fermentations were varied by cultivar (Amelonado, Amazon and Hybrid), post-harvest pod storage time (1 and 7 days) and fermentation method (heap and box). During each fermentation, chemical (sugars, ethanol, organic acids and pH) and physical (moisture, temperature and pulp/cotyledon ratio) changes were monitored. A large number (104) of statistically significant differences were found, indicating that the choice of cultivar, method of fermentation and duration of post-harvest pod storage, affect the chemical and physical profiles of cocoa fermentations. However, after the fermented beans were either sun or mechanically dried, the quality of the beans as measured by the cut-test and organic acid concentration, revealed only one significant difference, that of a higher proportion of mouldy beans when the beans had been subjected to the longer 7-day pod-storage period prior to splitting and fermenting.

INTRODUCTION

Ghanian cocoa is considered to be the standard for bulk cocoa and is characterised by a high chocolate flavour, and low acidity and astringency. Cocoa from some other regions tends to have a weak chocolate flavour, high acidity and astringency, and occasionally undesirable flavours (Baigrie & Rumbelow, 1987; Lewis, 1987). The consequence is that such cocoa may be sold at a discount. Ghanaian cocoa fermentation practices (Adomako, 1983; Duncan, 1984; Duncan *et al.*, 1989) differ from other regions, principally in the cocoa cultivar used (Amelonado/Amazon), the use of a longer post-harvest pod-storage step prior to pod splitting, the method of fermentation (the heap method which is typically shorter and turned less frequently), and the method of drying (sun drying as opposed to mechanical).

Investigations of the cocoa fermentation practices which influence the perceived flavour of cocoa have not been conclusive. A comparison of Amelonado and Amazon cultivars (Dougan, 1979) in Ghana revealed that Amelonado has a lower concentration of pulp sugars, less pulp on a per bean basis, and produces less lactic acid during the fermentation. However, the effect of perceived cocoa quality was ambiguous since the cocoa was acidic compared to commercial Ghana cocoa. In comparisons of the heap and box fermentation methods, it was noted that the pH, acidity, temperature and aeration were more consistent for the heap method, although there was no difference in perceived cocoa quality (Carr *et al.*, 1979; Anon, 1981).

Some studies (Carr *et al.*, 1981; Lewis & Lee, 1986; Duncan *et al.*, 1989; Meyer *et al.*, 1989) have recommended changes in the processing of the cocoa which reflect differences between Ghanaian practices and those of other cocoa-producing regions. This involves the use of post-harvest storage of the cocoa pods, a shortened fermentation period and less frequent turning.

The method of drying or curing of the fermented beans can also affect cocoa quality. A number of studies (Howat *et al.*, 1957; Carr & Dougan, 1977; Anon, 1981, 1982; Duncan *et al.*, 1989) have reported that sun-dried beans were less acid than mechanically-dried beans and that this is principally due to reductions in acetic acid levels whereas levels of lactic acid appear little changed. However, Carr *et al.* (1979) reported no

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differences between sun and mechanical drying. Duncan *et al.* (1989) recommended that when mechanical driers are employed, the quality of the cocoa can be improved by reducing the drying rate to balance the speed of evaporation of liquid from the surface of the bean with its rate of diffusion from the cotyledon.

The aim of this study was to monitor the chemical and physical profiles of cocoa fermented by different methods and to determine how this affects the chemical and physical quality of cocoa dried by sun or mechanical methods.

MATERIALS AND METHODS

Experimental cocoa fermentations

A total of 12 experimental cocoa fermentations were carried out at the Cocoa Research Institute of Ghana (CRIG) between September and December 1987. The fermentations were varied by cultivar (Amelonado, Upper Amazon, Tafo Series II Hybrid) post-harvest pod storage (1, 7 days) fermentation method (heap, box) and drying method (sun, mechanical).

Traditional plantain leaf-covered heap fermentations were carried out in the field close to where the pods were harvested. The box fermentations were accomplished at the fermentary at CRIG in wooden boxes (length 140 cm, breadth 125 cm, height 86 cm with holes of 1·4 cm diameter at 4 cm apart) with the top layer of beans covered with plantain leaves. Each fermentation comprised between 5000 and 8000 pods.

The duration of each fermentation was 6 days, the beans being turned to achieve thorough mixing, on the second and fourth days. At the completion of each fermentation, the beans were divided into two portions, one of which was sun dried, and the other mechanically dried.

Sampling

Cocoa fermentations
During each fermentation, the temperature of the heap or box (top, centre and bottom) was recorded at hourly intervals. Samples of fermenting beans (400 g; top, centre and bottom) were collected daily and immediately analysed.

Dried beans
Samples of dried cocoa (1 kg) were collected from throughout each batch after either sun or mechanical drying.

Physical and chemical analysis of the fermenting and dried cocoa beans

Standards and solvents
All chemicals were of analytical grade (AR, BDH Chemicals Ltd, Poole, UK). Sulphuric acid and water (HPLC grade) were obtained from FSA Laboratory Supplies, Loughborough, UK.

Sample preparation and the pH and moisture determinations were carried out in the laboratories at CRIG. Further analyses of the extracts (sugars, acids and ethanol) and dried beans (acids, cut-test) were carried out at the Natural Resources Institute, UK.

pH determination

Cotyledons (10 g) or pulp and testa (5 g) were weighed into a 1000 ml blender jar followed by the addition of 90 ml boiling distilled water. After blending for 2 min, the resultant homogenate was filtered through a Whatman No. 1 filter paper (18·5 cm) and *c.* 50 ml were collected. Immediately after cooling, the pH was measured using a meter accurate to within 0·1 pH unit.

Moisture determination

The moisture content of the whole bean and cotyledon was determined by an oven method (Carr *et al.*, 1979; Dougan, 1981).

Preparation of cocoa extracts from the fermenting bean (pulp and cotyledon)

Extracts of cocoa beans can be successfully preserved in solutions of benzoic acid (0·2% w/v) (Carr *et al.*, 1980; Dougan, 1981; Tomlins *et al.*, 1990). A total of 20 whole beans was weighed (+/− 0·01 g) and homogenised in 200 ml benzoic acid (0·2% w/v) for 3 min. A further 20 whole beans were weighed and the pulp and testa removed by a scalpel. The cotyledons were weighed and the weight of the pulp calculated by difference. The cotyledons were homogenised in benzoic acid as above. The whole bean and cotyledon homogenates (30 ml) were centrifuged (15 000 rev/min) for 15 min and aliquots (20 ml) were added to gas-tight bottles. The extracts were stored in the dark at a temperature of 4°C although they were at a higher temperature (35°C; 2 days) during transit from Ghana to the UK. Duplicate extracts of the cocoa bean homogenates were filtered through a 0·45 μm Millex filter (Millipore UK Ltd) and *c.* 1 ml collected.

Preparation of extracts from the dried bean (nib and shell)

The cocoa beans were carefully shelled using a scalpel. To freshly ground shell (2·5 g) or nib (5·0 g), freshly boiled water (30 ml) was added. The liquor was left to cool to room temperature with occasional stirring and then filtered through Whatman No. 1 filter paper. An aliquot (10 ml) of the filtrate was passed through two activated C18 Sep-Pak cartridges (Millipore UK, Cat No. 20805) arranged in tandem to which a 0·45 μm Millex filter (Millipore UK) was attached. The extracts were prepared in duplicate.

HPLC determination of organic acids, sugars and ethanol

Analyses were carried out using an HPLC system which comprised a Waters 520 pump (Waters UK Ltd), a Rheodyne sample injection valve (20 μl), a circulating-water bath (25°C) and a dual detection system: UV detector (215 μm, ACS, Macclesfield, UK) and a Waters R-400 RI detector. An Aminex HPX-87H column fitted with a Cation H^+ Micro-Guard cartridge (Bio-rad Laboratories, Richmond, USA), operated at a temperature of 25°C, was used to achieve the chromatographic separations.

Water-soluble acids (acetic, citric and lactic), sugars (fructose, glucose and sucrose) and ethanol were eluted with 0·013 N sulphuric acid at a flow rate of 0·8 ml/min. The acids were detected both by UV absorbance (215 μm) and RI, while the sugars and ethanol were measured by RI only. Peak heights were measured using Hewlett Packard 3390A integrators. Individual acids, sugars and ethanol were identified by comparison of retention times with authentic standards and their concentrations determined by the external standard method (Tomlins *et al.*, 1990).

Cut-test of the dried cocoa beans

A total of 300 beans were cut using a MAGRA guillotine (Teserba Technischer Service, Herrliberg, Switzerland) and then viewed under standardised daylight illumination (VeriVide, Leslie Hubbe Ltd, Leicester, UK). The beans were classified as fully fermented, partly brown/purple, fully purple, slaty, insect damaged, flat/shrivelled and mouldy. An index of fermentation (IF) was used being (10 × % fully fermented) + (5 × % partly brown/purple) + (0 × % fully purple).

Statistical analysis of the results

Statistical analysis was carried out using M-Stat statistical software on an IBM compatible computer.

RESULTS AND DISCUSSION

Statistical analysis

Cocoa fermentations

Chemical (sucrose, glucose, fructose, ethanol, lactic, citric and acetic acids, and pH) and physical (moisture) variables for both the pulp and cotyledon, and temperature and pulp/cotyledon ratio for the whole bean only, were analysed separately by analysis of variance (ANOVA), doing a factorial analysis with the factors cultivar, fermentation method, pod-storage time, time and sampling level. The design was treated as a split plot for sampling level; this approach was also considered for fermentation time, but as there was no significant difference in the residual variance, time was treated as a main plot factor. Except for the analysis of temperature, no significant second and higher order interactions were found, and all such interactions were incorporated in the residual. In the case of temperature, all third and higher order interactions were incorporated in the residual.

Dried cocoa beans

Chemical (lactic, citric and acetic acids) variables, for both the shell and nib, were analysed separately by analysis of variance (ANOVA), doing a factorial analysis with the factors cultivar, fermentation method, pod-storage time, and drying method. Cut-test (Index of fermentation [IF] and mouldy) variables were analysed as for the chemical variables, but for the whole bean only. Slaty, insect damaged, and flat/shrivelled beans were not included in the analysis as the data comprised mainly zero values with only the occasional positive value.

Discussion of the results

Tables 1 and 2 show the *F* ratios obtained using ANOVA for each of the variables and factors for the fermenting bean and dried bean respectively. Although a large number (104) of significant differences were noted during the fermentation, only one significant difference (mouldy) was noted in the dried bean analysis.

Cocoa fermentations

Cultivar

A comparison of Amazon and Amelonado cultivars in Ghana (Dougan, 1979; Adomako, 1986), found that the Amazon variety tends to have higher concentrations of pulp sugars, produce more lactic acid, and have a higher pulp to cotyledon ratio. However, other studies comparing Amazon, Amelonado and Hybrid cultivars in Ghana (Anon., 1981) concluded that differences in the chemical profiles were not significant, owing to limitations in the sampling procedure, the small sample size and the natural variability of the fermentation process. Two of the above investigations (Dougan, 1979; Anon., 1981) reported that the finished cocoa was inferior to standard commercial Ghana cocoa which was used as a control. Amelonado has been found to have a lower pulp/cotyledon ratio than the other cultivars, and it is suggested that it was the reduction in the ratio that improved the aeration of the ferments leading to the lower acidity (Dougan, 1979; Biehl *et al.*, 1989).

Whereas higher concentrations of pulp sugars have been noted for the Amazon cultivar (Dougan, 1979), the current investigation has found a significantly lower concentration, but this was only apparent during the first 48 h of the fermentation. Furthermore, a cultivar/fermentation method interaction revealed higher concentrations for combinations of Amazon and Amelonado with the box method, and Hybrid with the heap. While the cultivar did not affect the overall concentrations of lactic acid, the concentration differed within the ferments, higher concentrations occurring at

Table 1. Fermenting cocoa beans: analysis of variance (ANOVA; *F* ratios)

Main effects	DF	Sucrose		Glucose		Fructose		Ethanol	
		P	COT	P	COT	P	COT	P	COT
F	1	2·90	< 1	4·07*	1·20	4·50*	< 1	11·3*	8·59*
C	2	< 1	< 1	1·60	1·00	1·27	< 1	2·44	1·68
S	1	< 1	6·66*	18·4*	3·51	41·3*	4·59*	63·8*	32·3*
T	6	4·13*	12·7*	96·9*	3·31*	85·4*	7·59*	7·76*	6·30*
L	2	3·78*	< 1	4·42*	6·71*	7·98*	7·27*	3·45*	6·40*
Interactions									
F * C	2	2·49	3·14	10·6*	3·90*	5·56*	7·16*	4·02*	3·63*
F * S	1	3·87	< 1	1·66	< 1	2·19	1·53	3·32	< 1
F * T	6	< 1	< 1	1·61	< 1	1·52	1·94	1·84	1·70
C * S	2	1·38	< 1	< 1	1·00	1·33	< 1	3·00	1·72
C * T	12	< 1	< 1	2·37*	< 1	2·25*	< 1	< 1	< 1
S * T	6	< 1	·94	11·4*	1·79	9·53*	1·81	3·42*	4·21*
L * F	2	5·37*	1·78	< 1	< 1	< 1	< 1	1·51	2·92
L * C	4	< 1	2·00	< 1	1·00	< 1	1·27	1·63	< 1
L * S	2	2·93	< 1	< 1	2·00	1·91	2·18	1·15	6·69*
L * T	12	1·07	< 1	< 1	2·57*	1·00	2·82*	1·38	< 1

Main effects	DF	Citric		Acetic		Lactic		pH	
		P	COT	P	COT	P	COT	P	COT
F	1	< 1	< 1	< 1	9·15*	3·43	< 1	22·2*	12·6*
C	2	< 1	3·93*	4·79*	16·0*	2·21	2·96	7·29*	< 1
S	1	17·6*	2·71	13·1*	2·40	4·36*	2·67	18·9*	< 1
T	6	< 1	4·38*	17·9*	40·6*	5·77*	3·64*	18·9*	34·5*
L	2	2·90	3·00	2·91	11·7*	11·1*	4·82*	2·37	10·6*
Interactions									
F * C	2	2·14	2·49	4·31*	1·65	< 1	1·50	9·80*	12·1*
F * S	1	< 1	< 1	1·76	< 1	3·37	< 1	1·99	< 1
F * T	6	< 1	1·13	3·81*	2·67*	< 1	< 1	8·00*	1·53
C * S	2	< 1	< 1	1·93	< 1	< 1	< 1	1·51	3·80*
C * T	12	1·44	< 1	1·22	1·35	< 1	< 1	2·22*	1·90
S * T	6	1·58	1·00	2·38*	7·48*	1·50	< 1	1·79	7·16*
L * F	2	1·98	3·92*	4·80*	1·03	1·05	2·15	1·60	< 1
L * C	4	< 1	2·69*	2·83*	3·25*	1·95	3·33*	1·03	1·65
L * S	2	5·34	< 1	2·65	2·78	3·21*	3·48*	2·19	8·08*
L * T	12	< 1	< 1	1·53	1·81	< 1	1·36	1·85*	1·82

Main effects	DF	Moisture		Temp. WB	pulp/cot
		P	COT		
F	1	3·39	< 1	1·84	5·43
C	2	< 1	< 1	< 1	50·2*
S	1	< 1	14·0*	18·1*	19·3*
T	6	2·45*	14·2*	76·6*	36·1*
L	2	< 1	1·12	91·1*	3·92*
Interactions					
F * C	2	< 1	< 1	1·23	5·78*
F * S	1	< 1	5·76*	< 1	13·3*
F * T	6	< 1	1·59	2·42*	1·82
C * S	2	1·66	6·10*	< 1	1·71
C * T	12	< 1	1·68	1·35	< 1
S * T	6	< 1	1·28	4·15*	3·29*
L * F	2	1·81	1·05	7·52*	9·51*
L * C	4	1·92	< 1	< 1	< 1
L * S	2	1·94	1·70	5·39*	< 1
L * T	12	< 1	< 1	3·85*	1·31
L * F * T	12			2·87*	

F, fermentation method; C, cultivar; S, storage time; T, time; L, level; P, pulp; COT, cotyledon; DF, degrees of freedom; WB, whole bean.
* Significantly different (p = 0·05).

Table 2. Dried cocoa beans: analysis of variance (ANOVA; *F* ratios)

	DF	Nib			Shell		
		Citric	Lactic	Acetic	Citric	Lactic	Acetic
Organic acid analysis							
Cultivar	2	< 1	2·1	1·5	< 1	< 1	2·8
Storage time	1	< 1	3·4	1·0	< 1	< 1	< 1
Fermentation	1	< 1	< 1	< 1	< 1	< 1	< 1
Drying method	1	< 1	< 1	< 1	< 1	< 1	< 1
Cut test	DF	IF	M				
Cultivar	2	1·4	< 1				
Storage time	1	< 1	7·5*				
Fermentation	1	< 1	1				
Drying method	1	< 1	2·6				

*, Significantly different ($P = 0{\cdot}05$); cultivar, Amelonado, Amazon, hybrid; storage time, 1, 7 days; fermentation, box, heap; drying method, sun, mechanical; IF, index of fermentation: M, mouldy beans; DF, degrees of freedom.

the bottom of the Amazon and Amelonado fermentations, and at the top of the Hybrid. Analysis of the pulp/cotyledon ratio and acidity (as measured by pH), produced findings similar to those of Dougan (1979) and Biehl *et al.* (1989), that of a lower pulp volume and reduced acidity, particularly for the Amelonado cultivar, except that this was only true when the Amelonado cultivar was fermented in the box. The Hybrid cultivar tended to have a higher pulp volume and acidity. The moisture in the cotyledon was increased when the Amelonado and Amazon cultivars were used in combination with the longer pod-storage time of 7 days.

With level, cultivar has a significant effect on the concentrations of citric, lactic and acetic acid. For cotyledon citric acid, the cultivars differed in that the concentration was more uniform within the Amelonado fermentations. With regard to lactic acid (cotyledon) and acetic acid (pulp and cotyledon), the highest concentrations occurred at the top of the Hybrid fermentations. However, the importance of level is not clear since the beans at the different levels are mixed when the fermentations are turned, thus complicating the interpretation.

Fermentation method

A variety of fermentation methods have been applied to cocoa fermentation (Wood & Lass, 1985). The methods principally used are the heap method in West Africa and the box method in South East Asia and South America. Whereas the heap method is inexpensive, the use of the box method enables the fermentations to be more closely defined and controlled (Howat *et al.*, 1957). Differences in the chemistry of the heap and box methods have been noted (Carr *et al.*, 1979; Anon., 1981). Carr *et al.* (1979) found that the bottom of the box was more aerobic than the bottom of the heap such that, in the former, the sugars were consumed faster with a higher maximum ethanol concentration, a greater increase in acetic acid but a lower concentration of lactic acid. It has been noted (Anon., 1981) that physical (temperature) and chemical (lactic and acetic acids, pH and oxygen) profiles of the heap were more uniform than the box, the profiles for both methods only being similar at the centre location. In both investigations, the sensory characteristics of the finished cocoa from either heap or box fermentations were similar. Moreover, the chocolate was inferior and more acidic than standard commercial Ghana cocoa.

In previous studies (Carr *et al.*, 1979; Anon., 1981), both the heap and box fermentations were carried out in a fermentary at CRIG. However, Ghanaian farmers traditionally ferment the cocoa using the heap method in the field among the trees close to where the pods were harvested and split (Baker, D. M., unpublished). Therefore, in this study, the heaps were fermented in the field. Statistical analysis of the data obtained from this investigation shows that the influence of the fermentation method on the chemistry of cocoa fermentations is dependent, not only on general differences, but on interactions with time, cultivar and pod-storage time. In general, lower concentrations of the sugars, ethanol and acetic acid, and a higher pH are the product of using the box method. Of the interactions, the most important is that of the fermentation method (box) with cultivars (Amelonado and Amazon), the pattern being higher concentrations of sugars, lower concentrations of ethanol, lower acidity as measured by pH, and a lower pulp/cotyledon ratio (box/Amelonado). The concentration of acetic acid varied with time, the box giving a lower concentration towards the end of the fermentation. The temperature in the box also increased at a slower rate than in the heap. Since higher concentrations of sugars, but lower concentrations of ethanol and acetic acid were found for the box method, this implies incomplete fermentation or non-utilisation of the sugars by the yeasts.

Although Carr *et al.* (1979) reported differences between the fermentation methods with respect to level, the results obtained by statistical analysis do not follow the same pattern. With level, there were no differences between the fermentation methods with respect to the concentration of sugars, ethanol or lactic acid. Further-

more, whereas Carr *et al.* (1979) found higher concentrations of acetic acid at the bottom of the box, the present study found the reverse.

Post-harvest pod storage

Storing the cocoa pods prior to splitting resulted in a reduction in the concentration of pulp sugars and a reduced pulp/cotyledon ratio of the fresh beans (Carr *et al.*, 1979; Anon., 1981; Lewis, 1986; Biehl *et al.*, 1989; Meyer *et al.*, 1989). Meyer *et al.* (1989) reported that the post-harvest storage of cocoa pods leads to improved aeration of the ferments resulting in a rapid increase in temperature to greater than 45°C within 20 h, the suppression of lactic acid bacteria, and increased sugar respiration by yeasts over alcoholic fermentation. Significantly lower concentrations of acetic acid and a higher pH occur in the product. Storing pods prior to splitting has also been recommended for cocoa beans which are difficult to ferment (Howard, 1984).

The findings of this investigation are generally in agreement with the previous studies, the longer 7-day pod storage leading to significantly reduced concentrations of sucrose, glucose, fructose and ethanol (pulp and cotyledon), acetic acid (pulp), reductions in the pulp/cotyledon ratio, an increase in pH, and a rapid early rise in temperature. There was also an increase in pulp lactic acid.

Moreover, the application of statistical analysis has highlighted additional effects concerning how other variables such as fermentation method and cultivar interact with storage time. The pulp/cotyledon ratio is dependent on both the fermentation method and storage time, a 7-day storage time and box method giving the lowest pulp volume. Acidity, as measured by the pH analysis, is dependent on both the storage time and cultivar, a longer storage time and the Amelonado cultivar giving lower acidity.

Sampling position

Within the heaps (top, centre and bottom levels) there were significant differences in the concentration of the sugars, ethanol, lactic and acetic acid, pH, pulp/cotyledon ratio and the temperature.

During the fermentations, all the beans were mixed together when the heaps and boxes were turned after 2 and 4 days, Hence the statistical analysis of the cocoa fermentations with respect to level are difficult to interpret and may not be meaningful. However, some trends appear to be important. For example, the pattern for the concentrations of acetic and lactic acid are reversed; that is the concentration of lactic acid is highest at the bottom level, decreasing towards the top, while for acetic acid, the highest concentration occurs at the top level and decreases towards the bottom level. The acidity, as measured by pH, was highest at the top level, decreasing towards the bottom, and follows a similar pattern to acetic acid.

As regards temperature, a second-order interaction between time, level and fermentation method revealed that, for either fermentation method, the temperature increased more rapidly and reached a higher level at the top of the fermentations. Moreover, the fermentation methods differed in that the temperature was more uniform within the box. However, although analysis of the level could help to further the understanding of how cocoa fermentations differ, it is not of direct practical significance for optimising cocoa quality since all the beans would be processed.

Dried beans

Considering the dried cocoa beans, there were no significant differences in the acid content of either the cotyledon or shell part of the bean regardless of the cultivar, post-harvest pod-storage time, fermentation method or method of drying. However, there were differences in the concentrations of the acids between the shell (acetic = 0·53% v/w, lactic = 0·42% w/w, citric = 0·40% w/w) and nib (acetic = 0·29% v/w, lactic = 0·11% w/w, citric = 0·33% w/w). The shell contained higher concentrations of acetic and lactic acids. However, this difference is negligible since the mass of the shell was small in relation to the nib.

There was a significant increase in the proportion of mouldy beans among those subjected to a longer pod-storage time of 7 days prior to fermentation, although the proportion of mouldy beans in either was low (1 day = 0%, 7 day = 0·7%). The reason for the increase in the proportion of mouldy beans is not clear but may be due to the inoculation by moulds during post-harvest pod storage.

CONCLUSIONS

The main conclusions to be drawn from the results of this particular study are that, whereas the cultivar, duration of post-harvest pod storage and fermentation method significantly affect the chemical and physical changes during the course of the fermentation, after either sun or mechanical drying, the quality of the beans, as measured by the cut-test and organic acid concentration, is similar.

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