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Production of Microcrystalline Cellulose from Fibrous Maize Stem: Re-Engineering Food By-Products for Pharmaceutical Use

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1. Abstract

Microcrystalline cellulose (MCC) has become one of the major pharmaceutical excipients in high demand in the world today. The constant use of wood to produce microcrystalline cellulose may lead to the danger of deforestation. Agricultural wastes and byproducts are an alternative source of microcrystalline cellulose. In this study, microcrystalline cellulose was produced from the fibrous stem of Zea mays plant by acid hydrolysis and characterized using Avicel PH 101 as a standard. The bulk, tapped, and true densities of the microcrystalline cellulose produced were comparable with those of Avicel PH 101. However, the angle of repose, Hausner's ratio and compressibility index revealed poor flow properties of the produced microcrystalline cellulose. Tablets formulated from the produced microcrystalline cellulose as well as Avicel PH 101 showed good tablet strengths, uniformity of thickness, binding capacity, elastic recovery, and signs of instability as indicated by increase in thickness after 24 hours of storage. The microcrystalline cellulose produced was used to formulate metronidazole tables in percentage ratios of 50:50, 65:35, 75:25, and 85:15. The reworking potential of these tablet blends were calculated, and it was observed that blend with 85 % of the produced microcrystalline cellulose had the highest reworking potential of 75.28 %. In conclusion, the Microcrystalline cellulose was successfully produced from the fibrous stem of Zea mays with comparable bulk and tapped densities to Avicel PH 101. In addition, it had an excellent reworking potential. However, it exhibited poor flow in comparison with Avicel PH 101.

2. Introduction

Cellulose is the major composition of plant and serves as the primary structural element in their cell walls. It is a linear polymer of β-glucopyranose bound via β-1,4-glucosidic bonds and has gradually become a very preferred polymer in drug delivery system technology because of its rather unique properties which include low cost, excellent biodegradability, and biocompatibility [3]. Cellulose is usually gotten from wood and other plants by-products such as hemp, husk, rice and kenaf. It can also be gotten from other sources apart from plants which include bacteria, algae and fungi [6]. The molecular arrangement of cellulose is greatly determined by its source and method of treatment [1, 6, 15]. Cellulose possesses a complicated dissolution property as it is insoluble in water as well as other non-polar organic solvents. This over the years has been a source of concern to researchers and manufacturers as dissolution of cellulose is a vital step in its use for manufacturing purposes and other areas of application. Its insolubility in non-polar solvents is quite expected as cellulose is a polar molecule with several hydroxyl groups thus possessing good hydrogen bonding

capacity. However, its insolubility in aqueous solvents a bit incomprehensible. This aqueous insolubility has been blamed on the hydrophobic interactions present in the molecules because of the amphiphilic nature of cellulose [17].

Microcrystalline cellulose (MCC) is the most popularly used binder for pharmaceutical manufacturing purposes. Its preferred use is due to its versatility in manufacturing as it can serve as a binder, disintegrant, lubricant, bulking agent and has chemical resistant ability. Different methods exist to produce MCC which include the use of enzymes, acid hydrolysis, and steam explosion [2, 9, 18]. These methods basically involve hydrolysis of cellulose from refined wood pulp at high temperature (170 °C) and pressure. The degree of crystallinity of MCC is determined by the source and method of preparation [7].

Wood pulp is gotten from hardwood or softwood, with the former coming from deciduous plants while the latter originate from conifers. These woods are readily available, however, due to the high demand for MCC in the industrial market, there is a danger of deforestation that will arise with the constant cutting down of wood as raw material thus creating a need to explore other non-wood plant sources of MCC.

Plant agro-waste or by-products have been an area of emphasis as alternative sources of MCC. Studies have been done on jute plant fibers and guinea corn stalk to demonstrate the possibility of producing MCC from non-wood sources. It was reported that MCC was successfully produced from the stalk of guinea corn and that it conformed to the official standards specified in the British Pharmacopoeia for microcrystalline [21].

Another plant with agro-waste material that should be considered to produce MCC is Zea mays. The stem of Zea mays popularly known as maize plant contains a lot of fibrous material and should be studied as a potential source of MCC. Maize plant is an annual plant belonging to the Gramineae family. It is wildly planted as a source of food in almost every part of the world. This work aims at determining and demonstrating the possibility of the production of MCC from the fibrous stem of Zea mays.

3. Materials and Methods

3.1. Materials

Pharmaceutical and laboratory grade chemicals were used as obtained from manufacturers. The chemicals and excipients include sodium hydroxide and hydrochloric acid (May and Baker, England), sodium hypochlorite, JIK (Reckitts and Colman, Nigeria); Distilled and Deionized water (Pharm Tech, UNN), Avicel PH 101 (FMC Corporation, Philadelphia), and Fibrous straw of maize stem (Nanka, Nigeria).

3.2. Methods

3.2.1. Pulping of Cellulose: The soda process was adopted (Whistler and smart 1953; Britt 1970). Sprouts of fibrous straw of maize stem were peeled and cut into chips. The chips were dried at 70 °C for 5 h after which a total weight of 470 g was cooked for 8 h in 6 L of 1 % NaOH at 150 °C. An antifoaming agent (polypropylene glycol 2025) was added to reduce frothing at the initial 2 h of cooking. The cooked chips were washed several times with tap water until neutral to litmus. The lignin was tested in supernatant from above and in the pulped cellulose using phloroglucinol and conc HCl. The supernatant gave a red colour, which is indicative of the presence of lignin whereas the pulped cellulose gave no colour change with the reagent. This shows that the lignin content of the cellulose material has been removed by treatment with 1 % NaOH.

The pulped cellulose was placed in 4 L of 0.4 % sodium hypochlorite. This was maintained at 50 °C for 30 min. The partially bleached pulp was washed several times until neutral to litmus paper.

3.2.2. Dissolution of Beta and Gamma Celluloses: The pulped material was boiled in a 17.5 % NaOH solution for 1 h. This solubilized the beta and gamma cellulose but not the alpha fraction of the cellulose content. The alpha cellulose, which remained, was washed several times with tap water until neutral to litmus and further treated with 0.4 % sodium hypochlorite at 50 °C for 30 min to bleach the pulp material. The cotton-like cellulose was washed several times, until neutral to litmus, to remove traces of the bleaching agent. The cellulose was dried using a fluidized bed drier (FBD/L72, Mostyn Flintshire, U.K). It was then weighed dry.

3.2.3. Partial Acid Hydrolysis of Alpha Cellulose: A 450 g quantity of the dry cellulose was boiled in 1 L of 2N HCl at 95 °C to 100 °C for 1 h with vigorous stirring. The suspension obtained was cooled, washed till the wash water became neutral to litmus paper. It was then filtered under negative pressure. The cake was dried in a fluidized bed drier (FBD/L72, Mostyn Flintshire, UK) for 15 min. The dried mass was milled using an attrition mill (RetschSK1, Germany). The dried powder obtained was passed through a 200 mm aperture sieve.

The yield of the product was calculated from the formula:

$$\frac{\text{Original weight of material}}{\text{weight of product}} * 100 \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (1)$$

4. Evaluation of Powder Properties of Produced Microcrystalline Cellulose

4.1. Scanning Eye-piece Microscopy

Minute samples of the new MCC and Avicel PH 101 were mounted on the Eye-piece Microscope and observed for crystallinity. These samples were observed with Eye-piece microscope of magnification x 400.

4.2. Measurement of Angle of Repose

A thick cylindrical paper of height 17.5 cm and internal diameter of 17.0 cm, open at both ends was placed on a flat solid rubber base of diameter 20 cm. The cylinder was filled with powder and

gradually lifted vertically upwards. The height of the powder heap, thus formed on the rubber base as measured with a graph paper. The experiment was repeated thrice with the new MCC and Avicel PH 101.

The Angle of repose (H) was calculated as:

$\Theta = \tan^{-1} h/r$ …… (2)

Where:

h = height of the powder heap

r = radius of heap base.

4.3. Determination of Bulk and Tapped Density

A 20 g quantity of the new MCC and Avicel PH 101 were carefully poured into a 100 ml measuring cylinder. The bulk volume of the powder was noted. The cylinder was gently tapped on the bench until no change was observed. The final volume (tapped volume) was noted.

Bulk density = $\frac{weight\ of\ sample}{bulk\ volume}$…… (3)

Tapped density = $\frac{weight\ of\ sample}{tapped\ volume}$…… (4)

4.4. Determination of Hausner Quotient

This was calculated using the formula:

$\frac{tapped\ density}{bulk\ density}$ …… (5)

4.5. Compressibility Index

This was calculated using the formula:

$\frac{tapped\ density - bulk\ density}{tapped\ density}$ x 100 ……..……(6)

4.6. Determination of True Density

A 50 mL pycnometer, with weight W1, was used for this experiment. New MCC was weighed, and its weight was noted as W3. The empty pycnometer was filled with acetone (in which both the new MCC and Avicel PH 101 are insoluble) with the excess wiped off. The pycnometer and solvent were weighed together (W2). About 6 mL of acetone contained in the pycnometer was transferred to a clean test tube. A 2.30 g mass of the new MCC was then transferred into the filled pycnometer and made up to volume with the acetone in test tube. The pycnometer, the new MCC and acetone were weighted as W4. W3 of the powder occupied W3/P, where P is unknown true density. The remainder of the 50 mL is solvent so that the total weight is given by:

W4 = W3 + (W2/50) (50-W3/P)

Therefore, P = $\frac{W2 * W3}{50(W2 - W4 + W3)}$ g/ml …… (7)

This procedure was repeated for Avicel PH 101.

5. Evaluation of Tablet Properties

5.1. Compressibility

A 300 mg quantity of Avicel PH 101 and the new MCC were directly compressed at a compression force of 60 kgf at a dwell time of 5 seconds.

5.2. Uniformity of Thickness

Ten tablets were selected at random from each batch of Avicel PH 101 and the new MCC respectively and their thicknesses were measured using a Venier caliper.

5.3. Hardness Test

Ten tablets each of Avicel PH 101 and the new MCC were used. The hardness of each tablet was measured using a hardness tester (TBH 28, Erweka, Germany).

5.4. Determination of Elastic Recovery (ER)

This was calculated using the formula:

$ER = [(H_0 - H_p)/H_p] * 100\%$…… (8)

where H_o is the thickness of the tablets immediately after ejection and Hp is the thickness of the tablets after 24 hours storage. Tablet thicknesses were measured using a Venier caliper.

5.5. Reworking Potential

A 300 mg quantity of new MCC and Avicel PH 101 were compressed using the F3 electric tableting machine at a compression force of 60 kgf and a dwell time of 5 seconds before ejection. The hardness of these tablets was measured. Ten tablets were used for each determination. They were crushed and passed through a 200 µm sieve apparture. The powders were re-compressed at the same compression force and dwell time. The hardness of the re-compressed tablets was measured using the hardness tester. The same was repeated for new MCC and Metronidazole binary mixtures combined at different ratios.

The reworking potential was calculated with the formula:

R.P. = $\frac{Af}{Ai}$ * 100 ……..……(9)

where, Af = final strength (hardness) of tablets

Ai = initial strength of tablets.

5.6. Binding Capacity

This was calculated using the formula:

$\frac{Breaking\ strength\ (N)}{Thickness\ of\ tablet\ (CM)}$…….…… (10)

5.7. Determination of Excipient swelling capacity

A 300 mg tablets compressed at 60 kgf and dwell time of 5 seconds with the new MCC and Avicel PH 101 were used for this test.

Heights of each of the tablet in a graduated cylindrical tube were noted. The tablets were dropped into a petri-dish containing water and the tubes were immediately inverted over them. The tablets were allowed to swell to maximum size. Their vertical swelling capacities were noted on the tubes. The swelling capacity was then calculated as the percentage increase in tablet thickness after maximum swelling.

5.8. Determination of capacity of Excipient

The capacity of the new MCC was studied using Metronidazole. Various percentage ratios of the new MCC and metronidazole

(50:50, 65:35, 75:25, and 85:15) were compressed at 60kgf without lubricant, at a dwell time of 5 seconds. The mechanical properties of the formed compacts were determined. The capacity of the new MCC is given as the lowest concentration of the excipients which forms a stable compact.

6. Results

6.1. Percentage yield

A yield of 49.6 % alpha cellulose was obtained as the intermediate product. A final yield of 37.23 % of the new MCC was also obtained. This is a very high yield value and confirms the material as an abundant source of cellulose.

6.2. Crystallinity of the New Microcrystalline Cellulose

Particles with sponge-like structures, and irregular shapes were observed under the eye-piece microscope. Some fibrils and bundles were also seen. The particle size of both the new MCC and Avicel PH 101 contain particles ranging in size from 2 to 200 microns.

6.3. Flow Properties of Microcrystalline Cellulose

Results of density measurements are present in Table 1. The true, tapped, and bulk densities of the new MCC and Avicel PH 101 are similar.

6.4. Angle of Repose

The values of angles of repose for the new MCC and Avicel PH 101 are recorded in Table 1.

6.5. Tablet Hardness (Breaking Strength)

The Average values of the hardness of ten tablets selected at random from batches of the new MCC and Avicel PH 101 are recorded in Table 2 below.

6.6. Uniformity of Thickness

Thickness of 10 tablets produced from the new MCC and Avicel PH 101 at a compression force of 60 kgf and dwell time of 5 seconds immediately after ejection after 24 hours storage are indicated in Table 3. A 5 % variation in thickness is allowed depending on the type of tablet.

6.7. Elastic Recovery (ER)

The elastic recovery of the new MCC and Avicel PH 101 were found to be -3.06 and -1.81 respectively.

6.8. Binding Capacity

The binding capacity of the produced MCC and Avicel PH 101 were calculated as 24.75 Ncm^{-1} and 34.69 Ncm^{-1}**.**

6.9. Swelling time

The time for maximum swelling and disintegration of both the new MCC produced and Avicel PH 101 were both 8 minutes. The percentage vertical swelling capacity within 8 minutes were 160 % and 150 % for the new MCC and Avicel PH 101 respectively.

6.10. Reworking Potential

The Reworking potential of the new MCC, Avicel PH 101 and those of the produced MCC: metronidazole binary mixtures are recorded in table ii and iv respectively (Table 4).

Table 1: Some Powder properties of the new MCC and Avicel PH 101.

Powder property	New MCC	Avicel PH 101
Bulk density (gcm^{-3})	0.5	0.5
Tapped density (gcm^{-3})	0.68	0.6
True density (gcm^{-3})	0.91	0.91
Compressibility index (%)	26	17
Hausner's quotient	1.36	1.2
Angle of Repose (degrees)	54.4	47.8

Table 2: Average breaking strengths of Avicel PH 101 and the new MCC.

Breaking strengths	New MCC	Avicel PH 101
Initial Breaking Strength (N)	11.67	11.83
Final Breaking Strength (N)	7.93	7.42
Reworking Potential (%)	67.95	62.72

Table 3: Thickness of Tablets (CM).

	New MCC				Avicel PH 101			
	0 hour	% deviation	24 hours	% deviation	0 hours	% deviation	24 hours	% deviation
	0.456	1.32	0.462	3.16	0.324	3.58	0.33	3.5
	0.456	1.32	0.462	3.16	0.326	2.94	0.33	3.5
	0.474	2.53	0.48	0.71	0.326	2.94	0.332	2.95
	0.452	2.21	0.458	4.06	0.334	0.48	0.338	1.12
	0.462	0	0.466	2.27	0.326	2.94	0.338	1.12
	0.486	4.94	0.49	2.73	0.346	3.01	0.352	2.9
	0.462	0	0.468	1.83	0.388	13.51	0.344	0.64
	0.458	0.87	0.462	3.16	0.344	2.44	0.348	1.78
	0.562	17.8	0.468	1.83	0.344	2.44	0.35	2.34
	0.452	2.21	0.55	13.3	0.348	3.56	0.356	3.99
Mean	0.462		0.4766		0.3356		0.3418	

Table 4: The reworking potential of the New MCC: Metronidazole mixtures

	50:50%	65:35%	75:25%	85:15%
Initial Breaking Strength (N)	4.07	6.11	9.99	16.2
	4.89	9.78	10.29	17.22
	4.58	10.04	8.97	16.2
	4.17	8.35	9.27	15.39
	5.7	8.35	10.39	15.49
	5.6	7.54	6.93	16.41
Mean (N)	4.14	7.04	7.98	13.84
Final breaking Strengthening (N)	2.75	4.07	5.3	10.09
	2.24	3.77	5.5	10.39
	3.05	4.17	5.19	10.8
	3.16	4.17	5.81	10.7
	3.26	4.28	5.4	10.29
	3.36	4.48	6.32	10.7
Mean (N)	2.97	4.16	5.58	10.49
Reworking Potential (%)	72.7	59.23	71.8	75.28

7. Discussions

7.1. Percentage Yield

A final yield of 37.23 % of MCC was obtained which can be considered as high when compared to 18% yield obtained from coconut fibres [16] and a maximum yield of 31.33 % obtained from empty fruit bunch [5]. This confirms that the fibrous stem of *Zea mays* is an abundant source of MCC. Lower values have been reported from bagasse and rice husk with 7.4 % and 10.9 % percentage yields respectively [24].

7.2. Flow properties of MCC

Bulk density and true density of the MCC produced was 0.5 and 0.91gcm^{-3} respectively which is the same with that of Avicel PH 101. The tapped density on the other hand is 0.68 gcm^{-3} which is greater than that of Avicel PH 101. This shows that Avicel PH 101 contained a lot more voids and air spaces when left untapped and suggests a freer flowing powder compared to the MCC produced. It may also suggest that the MCC produced had higher individual particle densities which may be directly proportional to their particle sizes. The true density which indicates the crystalline nature of the powders (Stamm, 1964) show that both powders possess the same degrees of crystallization since the value is the same for both.

The Hausner's quotients of the produced MCC and Avicel PH 101 were 1.36 and 1.2 respectively. Hausner observed that the ratio of tapped to bulk density is a function of interparticulate friction and hence affects powder flow (Hausner, 1967). Generally, a Hausner's quotient or ratio of less than 1.25 is considered good, between 1.26 and 1.34 is passable and above 1.34 ranges from poor to very poor

flow as the number increases (USP, 2004). Thus, given a Hausner's ratio of 1.36 it shows that the MCC produced has a poor flow property while Avicel PH 101 has a good flow property. This, however, correlates with values obtained from two batches of *Dendrocalamusasper* derived MCC which bothshowed a Hausner's ratio of 1.45 [25]. MCC derived from *Musa sapientum, Musa paradisiaca* and *Tithonia diversifolia* have also showed high Hausner's ratio of 1.48, 1.42 and 1.46 respectively all indicating poor flow properties of the MCC [22]. This may indirectly infer that MCC obtained from agricultural byproducts might not have good flow properties for manufacturing purposes.

The compressibility index of the MCC produced and Avicel PH 101 are 26 and 17 % respectively. Compressibility index below 20 % is regarded as good, 21- 25 % is regarded as passable and above 26 % ranging from poor to a very poor flow with increase in values (USP, 2004). This indicates that the MCC produced has a poor flow while Avicel PH 101 has a good flow. This correlates with the Hausner's number derived for both powders thus confirming the poor flow properties of the MCC produced. [22] obtained similar results with high compressibility indices of 32.47, 29.61 and 31.35 % respectively for the MCC produced from the three pulp samples (*Musa sapientum, Musa paradisiaca* and *Tithonia diversifolia*) used.

7.3. Angle of Repose

The angle of repose of the produced MCC and Avicel PH 101 is 54.4 and 47.8º respectively. Angle of repose values greater than 40º is characteristic of cohesive powders. Although some powdered formulations with an angle of repose between 40 and 50º have been successfully manufactured, powders with angles of repose greater than 50º cannot be accepted for manufacturing (USP, 2004). In the case of the MCC produced, the angle of repose is far above 40º and 50º as well. This indicates that the MCC is highly cohesive with poor flow characteristics.

7.4. Tablet Hardness (Breaking Strength)

The tablet hardness of Avicel PH 101 (11.83 N) was more than that of the produced MCC (11.67 N) as seen in table 2. This may be the reason why the produced MCC has a higher reworking potential than Avicel PH 101.

7.5. Uniformity of thickness

Among the tablets formulated from the MCC, only one of the ten tablets used for the assay, deviated by more than 5 % immediately after formulation and 24 hours after compression respectively. There was a general increase in tablet thickness for most of the tablets within the first 24 hours of consideration which may be due to the absorption of moisture from the atmosphere. This clearly shows that the tablets are not stable on storage. Appropriate packaging may be used to mitigate this effect.

One tablet formulated with Avicel PH 101 deviated by more than 5 % just immediately after formulation. However, a marked increase in tablet thickness were also observed in most of the tablets which may also suggest absorption of moisture from the atmosphere as seen in the tablets formulated from the MCC produced. This can be related to the research by Nyqvist and Nicklasson (1983) where it was observed that tablets formulated from dry Avicel powder and stored in high relative humidity conditions produced significant decrease in tablet strength because of absorption of moisture from the storage environment. Thus, tablets produced from both MCC and Avicel PH 101 showed signs of instability. This instability may also be due to the elastic recovery of the MCC produced as well as that of Avicel PH 101. Changes in the tablet height or thickness is a function of structural changes inside the tablet (Picker, 2001).

7.6. Elastic Recovery

The elastic recovery of the new MCC and Avicel PH 101 were found to be -3.06 and -1.81 respectively. Elastic recovery may account for some tablet defects especially capping and this feature mostly depends upon the value of the material's elasticity. Negative elastic recovery values are usually acceptable for tablet formulation thus placing both produced MCC and Avicel PH 101 in good elastic recovery positions. However, the MCC produced showed more elastic recovery than Avicel PH 101. Microcrystalline cellulose generally consists of microfibrillar structure which does not easily collapse [4]. However, it still forms hard compacts due to the formation of hydrogen bonds between individual cellulose molecules during compression [14].

The Elastic Recovery (ER) obtained for the new MCC was higher than that of Avicel PH 101. This is indicative of a poorer compactibility of the MCC produced when compared to Avicel PH 101. This may also account for the increase in tablet thickness observed in the tablets as those formulated from the MCC produced showed more increase in tablet thickness than those formulated with Avicel PH 101.

7.7. Binding capacity

It was observed that the binding capacity of Avicel PH 101 was greater than that of the produced MCC. This means that the tablets formulated from Avicel PH 101 will be more compact than that formulated from the produced MCC. This correlates with the hardness of the tablets which shows that Avicel PH 101 tablets are harder than that of the produced MCC. It also correlates with uniformity of thickness of the tablets with the former showing less changes in thickness than the latter hence more stability. Finally, because the Avicel PH 101 tablets were more compact they were more difficult to rework hence the lower reworking potential obtained compared to that of the produced MCC.

7.8. Swelling time

The time for maximum swelling of both the MCC produced and Avicel PH 101 were 8 minutes respectively. The percentage vertical swelling capacity within 8 minutes were 160 % and 150 % for the new MCC and Avicel PH 101 respectively. This is indicative

that the new MCC may have higher disintegrant property than Avicel PH 101 since some disintegrants act by swelling significantly upon contact with water. This correlates with results observed from the uniformity of thickness of tablets which showed a marked increase in tablet thickness within 24 hours of storage.

7.9. Reworking Potential

The reworking potentials of the new MCC was greater than that of Avicel PH 101. This is indicative of the fact that the new MCC could be reworked more easily than Avicel PH 101.

8. Conclusion

Microcrystalline cellulose was successfully produced from the fibrous part of maize stem (*Zea mays*) which is an entirely wasted part of this plant having not been found of any use in our traditional community.

The material was easily pulped to a cotton-like cellulose using the soda process. This was further converted to microcrystalline cellulose by vigorous acid hydrolysis using 2N HCl. The entire process was both cost and time effective.

The yield of the new MCC was 32.23 %. This is a high yield value and it confirms the material as an abundant source of cellulose. The new MCC prepared was evaluated and compared to Avicel PH 101, a commercially available microcrystalline cellulose from FMC Corporation Philadelphia, USA.

The MCC produced exhibited poor flow properties evidenced by their compressibility indices, angles of repose and Hausner's quotients.

The compressibility of the new MCC is comparable to that of Avicel PH 101. The reworking potential of the produced MCC in metronidazole tablet formulation was very high. The swelling test showed that the produced MCC swells significantly when in contact with water. This suggests that it possess some disintegrant properties; a property also possessed by Avicel PH 101.

Finally, the MCC produced was used to satisfactorily formulate tablets with metronidazole; a poorly compressible drug, when mixed at different ratios.

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