

# MATURITY CLASSIFICATIONS INFLUENCE PHENOLOGICAL INTERVALS IN MAIZE

## ABSTRACT

This field trial compared the growth stages of early, intermediate and late varieties of maize and evaluated the effect of maturity group on their phenological intervals. Fifteen maize varieties belonging to three different maturity groups were evaluated in a randomized complete block design with four replicates. Data were collected on the days to leaf expansion from the early seedling stages to flowering. The results showed that maturity group effect was significant for all phenological intervals starting from the third leaf stage. Maturity group had the highest influence on the phenological intervals of the varieties during the period of seedling establishment at the fifth leaf stage. The contribution of maturity group to the total observed variability increased from seedling establishment to the late vegetative stages and peaked at the flowering stages. Varietal differences have a minimal contribution in ranking maturity in maize. In summary, maturity differences in maize varieties are initiated early, but their time-course effects on phenological intervals are more pronounced in the late vegetative and flowering stages.

**KEYWORDS:** Maize, maturity group, phenology, seedling establishment

## INTRODUCTION

Maize (*Zea mays* L.) is a popular staple in sub-Saharan Africa [1,2,3]. Breeders have developed various hybrid seeds for biotic and abiotic stresses and, more recently, with a focus on climate change [4]. With the adoption of hybrid seeds, maturity is an indispensable trait considered by farmers in seed selection [5]. Varieties belonging to different maturity groups were developed as a crop risk aversion strategy to suit diverse agro-ecological conditions. For instance, an early maturing variety provides food early in the season and can be planted to escape drought stress. [6,7]. Grading maize varieties and other crops by maturity period are based on several systems, including growing degree days (GDD) or **FAO- what does it mean?** maturity classes [8,9,10]. The length of the growing cycle is one of the essential traits determining hybrid adaptability to the environment [8].

All maize plants follow a general development pattern, but the specific time interval between phenological stages and canopy size may vary among different hybrids, seasons, treatments, planting dates and locations [11, 28, 29]. Phenological stages describe the time lapse necessary for different organs to come into view or become fully developed [12]. The phenological stages in maize include the days to emergence, the number of days to leaf production, the flowering date, which includes the days to tasselling and anthesis, days to silking, grain filling and physiological maturity. Plant phenology can also be quantitatively assessed by calculating the rate of plant development in terms of dry matter accumulation. In this case, plant growth rate is expressed as the algebraic product of a series of factors which include the plant weight and the size of the assimilatory system, which is usually the leaf area [13]. Two growth indices can be used: Crop growth rate (CGR) and Relative growth rate. RGR is more influenced by environmental conditions than GR [14].

Late and intermediate maturity groups have a higher dry matter accumulation and higher yield than extra-early and early maturity groups. However, growth rate (GR) and relative growth rate (RGR) do not significantly contribute to maturity classification in maize [15]. An early maturing hybrid may have lesser stature and develop fewer leaves or progress faster through its different growth stages. In contrast, a late-maturing hybrid may develop more leaves or progress through the growth stages more slowly [16,17]. However, little is known about the interaction between varietal effects and maturity in defining phenological intervals. Thus, time-course data on maize phenology may be useful for crop management decisions [18]. This study contributes towards understanding the phenology of maize specifically by:

1. Comparing the growth stages of three different maturity groups of maize using days to leaf expansion.
2. Investigating the effect of maturity group on the seedling establishment of early, intermediate and late maize varieties.
3. Quantifying the contribution of maturity group in predicting maize phenological development

## MATERIALS AND METHODS

**Research Location:** This study was conducted at the Obafemi Awolowo University Teaching and Research Farm, Ile-Ife, Nigeria. Ile-Ife is found on Longitude 7, 28'N and Latitude 4, 33'E, at 244 m elevation.

**Plant Material:** The 15 inbred lines used for this experiment were obtained from the International Institute for Tropical Agriculture (IITA), Ibadan, Nigeria. They consist of 5 early, 5 intermediate and 5 late maturing maize varieties.

**Experimental design:** The experiment was laid out in a randomized complete block design in four replicates. The plot used was disc-ploughed and harrowed prior to planting. Each plot consisted of seven rows 6 m long, and planting was done in June 2011 at a rate of three seeds per hole. The plant spacing used was 75 by 50cm. Thinning was done two weeks after planting to obtain two plants per stand with a density of 53,333 plants/ha.

**Fertilizer application and Weed control:** (NPK 15:15:15) was applied at two weeks after planting and four weeks after planting at the rate of 50 Kg/ha. Atrazine and gramoxone were used to control weeds at a concentration of 4.5 L/ha and was applied at pre-emergence and post-planting.

**Phenological data:** Data were also collected on the different growth stages of each variety from planting to flowering using the BBCH codes as outlined in [19](refer to the supplemental Table for more details). The data taken on phenological intervals were days to leaf expansion, tasseling, silking and anthesis. Every phenological interval was taken as days after planting (DAP) when about 70% of the population attained these growth stages.

**Statistical analysis:** All data were subjected to Analysis of Variance (ANOVA) using the generalized linear model of the SAS statistical package. Separation of means was also performed using the Duncan multiple range test.

## RESULTS

### **Maturity influences phenology more than varietal differences in maize**

More than any other factor investigated, maturity mean square values significantly contributed to the total variation recorded for all the phenological intervals of the 15 varieties (Table 1). The effect of maturity group was pronounced in all the phenological intervals, excluding stage 12 when two leaves were fully expanded. During the seedling stage, maturity group had the highest significance ( $p < 0.0001$ ) at phenological interval 15, when 5 leaves were fully expanded. The coefficient of variability (CV) was very high for all the varieties at their seedling stages, indicating that the data was relatively not stable (Table 5). Similarly, the high  $R^2$  values recorded for all the varieties signify the effects of unaccountable sources of variation in the data.

The effect of replication on the number of days to leaf expansion was very high during the stages preceding seedling establishment. As all the varieties advanced in their growth stages, the effect of replication became lower. The influence of maturity group became higher and significant as all the varieties approached their reproductive phase (Table 1). Variety (maturity group) accounts for the variations in growth behavior of all the varieties within a maturity group, and was only significant when the plants were at the advanced vegetative stages. These results suggest that the total variation observed in the growth pattern of these varieties after stand establishment is more attributed to their maturity group than varietal differences.

**Table 1: Mean square values of the maize varieties with respect to their growth stages**

<b>BBCH Phenological Intervals</b>	<b>Rep. (df= 3)</b>	<b>Mat.group (df=2)</b>	<b>Variety(mat. group) (df=12)</b>	<b>Error (df=42)</b>	<b>CV</b>	<b>R<sup>2</sup></b>
12	10.911***	1.117	0.383	0.399	7.153	70.237***
13	13.244***	5.000*	1.033	1.054	8.148	58.396***
14	15.178***	4.200*	1.133	0.987	6.022	61.957***
15	6.417	24.950***	2.425	2.405	7.318	43.310*
16	7.756**	11.816**	0.966	1.434	4.745	49.270*
17	5.083	27.950**	4.392	4.548	7.241	39.336
18	1.800	8.467**	1.417	1.157	3.016	44.731*
19	0.244	21.667***	1.033	0.768	2.273	63.636***
20	2.222	25.800***	4.067**	1.127	2.552	69.344***
21	2.244	71.717***	6.242**	1.934	3.080	73.471***
22	4.644	61.117***	6.542***	1.192	2.248	81.088***
23	4.111	75.800***	5.025***	1.171	2.126	82.017***
24	4.044	108.200***	3.267**	1.140	2.007	84.833***
25	3.311	138.317***	3.192**	0.930	1.741	89.265***
26	2.417	136.267***	2.867**	0.905	1.661	89.210***
27	1.839*	147.150***	2.391**	0.827	1.534	90.438***
28	0.986	65.383***	0.951	0.711	1.357	87.570
29	0.379	46.180	0.300	0.452	1.032	89.179***
30	0.152	-	0.251	0.827	1.342	21.95
59	2.469	598.752***	1.55	1.220	1.784	96.04***
63	4.328*	675.45***	2.017	1.494	1.927	95.674***
65	3.217	845.517***	2.641	2.443	2.423	94.409***
ASI	0.578	12.867***	0.833	1.149	114.86	43.701*

\*, \*\* and \*\*\* significant at 0.05, 0.001, and 0.0001 levels of probability respectively

df: degree of freedom

CV: Coefficient of variability

R<sup>2</sup>: Coefficient of determination

ASI-Anthesis-silking interval

Rep: Replication

-: Most individuals have produced the flag leaf before this stage, hence no sufficient data for analysis

### **Seedling establishment marks the initiation of the stages determining maturity**

The early and the late maturity groups were significantly different right from phenological interval 12 when 2 leaves were fully expanded, and this difference was maintained till when they got to the later growth stages (Table 2). Between phenological intervals 12 and 14, the intermediate maturity group was not significant from the early and late maturity group as it exhibited both early and late maturity traits. The intermediate and late maturity group took longer days (more than 4 days) between phenological interval 14 and 15 while the early maturity group took only 4 days (Table 2). This indicates that the transition from phenological intervals 14 and 15 was the longest period in maize seedling establishment of the intermediate and late varieties.

As the varieties advanced in age, the differences in days to attaining their phenological intervals became more apparent. The three maturity groups were significantly different at phenological interval 23 when 13 leaves were fully expanded. At this phenological interval, the early maturity group took about 48 days to produce fully expanded thirteenth leaf while the intermediate and late maturity groups took about 51 and 52 days respectively. The differences among the maturity groups increased and widened till the later vegetative and flowering stages. At about 58 days after planting, the early maturity group produced the fully expanded last leaf (flag leaf). In comparison, the intermediate and the late maturity groups produced their flag leaf at 62 days after planting and 67 days after planting respectively. From Table 2, it was evident that the early maturity group had the lowest number of leaves produced (18 leaves) while the late maturity group had the highest number of leaves produced (20 leaves).

The three maturity groups were phenotypically different at all the flowering phenological intervals. The early maturity group was the first to silk at around 57 DAP. In comparison, the intermediate and the late maturity group attained silking at approximately 65 DAP and 70 DAP, respectively, where DAP refers to days after planting.

**Table 2: Mean values for the different phenological intervals for the early, late and intermediate maturity groups.**

<b>BBCH PHENOLOGICAL INTERVALS</b>	<b>EARLY</b>	<b>INTERMEDIATE</b>	<b>LATE</b>
12	8.650 <sup>b</sup>	8.750 <sup>ab</sup>	9.100 <sup>a</sup>
13	12.100 <sup>b</sup>	12.600 <sup>ab</sup>	13.100 <sup>a</sup>
14	16.000 <sup>b</sup>	16.600 <sup>ab</sup>	16.900 <sup>a</sup>
15	20.000 <sup>b</sup>	22.150 <sup>a</sup>	21.600 <sup>a</sup>
16	24.350 <sup>b</sup>	25.750 <sup>a</sup>	25.750 <sup>a</sup>
17	28.950 <sup>b</sup>	30.800 <sup>a</sup>	28.600 <sup>b</sup>
18	34.000 <sup>b</sup>	35.300 <sup>a</sup>	34.600 <sup>b</sup>
19	37.400 <sup>b</sup>	39.400 <sup>a</sup>	38.900 <sup>a</sup>
20	40.300 <sup>b</sup>	42.400 <sup>a</sup>	42.100 <sup>a</sup>
21	43.050 <sup>c</sup>	46.700 <sup>a</sup>	45.750 <sup>b</sup>
22	46.550 <sup>b</sup>	49.650 <sup>a</sup>	49.500 <sup>a</sup>
23	48.700 <sup>c</sup>	51.600 <sup>b</sup>	52.400 <sup>a</sup>
24	50.700 <sup>c</sup>	53.600 <sup>b</sup>	55.300 <sup>a</sup>
25	52.650 <sup>c</sup>	55.550 <sup>b</sup>	57.900 <sup>a</sup>
26	54.550 <sup>c</sup>	57.550 <sup>b</sup>	59.750 <sup>a</sup>
18	34.000 <sup>b</sup>	35.300 <sup>a</sup>	34.600 <sup>b</sup>
19	37.400 <sup>b</sup>	39.400 <sup>a</sup>	38.900 <sup>a</sup>
20	40.300 <sup>b</sup>	42.400 <sup>a</sup>	42.100 <sup>a</sup>
21	43.050 <sup>c</sup>	46.700 <sup>a</sup>	45.750 <sup>b</sup>
22	46.550 <sup>b</sup>	49.650 <sup>a</sup>	49.500 <sup>a</sup>
23	48.700 <sup>c</sup>	51.600 <sup>b</sup>	52.400 <sup>a</sup>
24	50.700 <sup>c</sup>	53.600 <sup>b</sup>	55.300 <sup>a</sup>
25	52.650 <sup>c</sup>	55.550 <sup>b</sup>	57.900 <sup>a</sup>
26	54.550 <sup>c</sup>	57.550 <sup>b</sup>	59.750 <sup>a</sup>
27	56.400 <sup>c</sup>	59.550 <sup>b</sup>	61.800 <sup>a</sup>
28	58.600 <sup>c</sup>	61.316 <sup>b</sup>	63.850 <sup>a</sup>
29	-	62.200 <sup>b</sup>	65.850 <sup>a</sup>
30	-	-	67.786 <sup>a</sup>
59	56.158 <sup>c</sup>	62.100 <sup>b</sup>	67.200 <sup>a</sup>
63	57.300 <sup>c</sup>	64.200 <sup>b</sup>	68.850 <sup>a</sup>
65	57.700 <sup>c</sup>	65.200 <sup>b</sup>	70.650 <sup>a</sup>
ASI	0.100 <sup>c</sup>	1.000 <sup>b</sup>	1.700 <sup>a</sup>

Means with the same letter along the same row are not significantly different  
 -: Most individuals have produced the flag leaf before this stage, hence no sufficient data for analysis  
 ASI-Anthesis-silking interval

### **Maturity differences are more evident at the late vegetative and flowering stages**

From Table 3, it can be deduced that maturity group contributed less than 10% to the variations in the growth pattern of the varieties planted at phenological interval 3 when three leaves were fully expanded while replication had the highest contribution (37.34%) to the variations in the growth pattern of the varieties. The contribution of maturity group to seedling establishment got to the peak at phenological interval 15 when 5 leaves were fully expanded. At this growth stage, maturity group effect influenced about one-quarter of the total observed variability to the days each maturity group studied took in attaining this growth stage. The coefficient of variability (CV) for all phenological intervals, excluding the anthesis-silking interval was below 10.

The contribution of maturity group effect increased up till when the last leaf (flag leaf) was produced in each maturity group. The effect of replication and error on the phenological intervals of the varieties also reduced as the varieties approached the late vegetative stages and flowering. At the flowering stage, the contribution of maturity group to the total variations in days to 70% tasseling, silking and anthesis was above 90%. Replication contributed below 1% to the total variations in days to flowering, while error contributed below 6%. The growth behavior of the three maturity groups as to reaching phenological intervals at this stage is in line with the criteria used to classify maize into maturity groups (the days from sowing to silking).

**Table 3: Percentage of the total sum of squares from analysis of variance for the various phenological intervals.**

<b>BBCH PHENOLOGICAL INTERVALS</b>	<b>REPLICATION (%)</b>	<b>MAT.GRP (%)</b>	<b>VAR.(MAT. GRP) (%)</b>	<b>ERROR (%)</b>
12	58.104	3.95	8.166	29.764
13	37.343	9.399	11.654	41.603
14	41.773	7.706	12.477	38.037
15	9.661	25.044	14.605	50.690
16	19.595	19.902	9.710	50.730
17	4.844	17.754	17.691	60.664
18	6.141	19.390	19.333	55.269
19	0.836	48.832	13.975	36.363
20	4.313	33.420	31.606	30.656
21	2.197	46.823	24.451	26.529
22	5.262	46.171	29.653	18.912
23	4.510	55.450	22.056	17.983
24	3.844	68.568	12.421	15.167
25	2.729	76.011	10.524	10.735
26	2.059	77.384	9.768	10.790
27	1.517	81.019	7.900	9.561
28	1.784	78.829	5.161	12.432
29	1.817	73.721	2.398	10.822
59	0.586	94.696	1.478	3.955
63	0.526	92.155	1.728	5.591
65	0.895	93.111	1.668	4.326
ASI	2.021	30.015	11.664	56.298

ASI-Anthesis silking interval

## DISCUSSION

The high contribution of replication and error by the statistical model to the phenological intervals of the maturity groups during seedling establishment and the early vegetative stages (Table 2) indicate the substantial interplay of many factors in maize early growth stages. According to [20], the relative growth rate of maize seedlings is strongly dependent on the environment. [21] showed that maize seedlings do not obtain sufficient photosynthetic capacity to supply energy equal to that supplied by the endosperm until the seedlings were 10 days old with 2 leaves fully emerged. It has also been observed by [22] that leaf growth stages below the third leaf stage are early periods of seedling establishment when the root hairs on the nodal roots of the maize seedlings are not well developed and seminal root growth is still in progress. At this time, the leaves of the varieties across the maturity groups have not reached their full photosynthetic capacity with seedling growth depending on stored seed reserves. Thus, maturity effects were not evident in the phenological intervals preceding seedling establishment as a result of the joint effect of environmental factors and low photosynthetic apparatus.

The observations in stages 14 and 15 signify that these phenological intervals are the most important phases in seedling establishment. There is a transition from partial dependence on food reserves in the endosperm to full dependence on photosynthesis, which is the autotrophic phase. Earlier reports showed that the most critical and longest stage of maize seedling establishment was the transition from the fourth to the fifth leaf stage and hybrid differences influenced the interval more than seed maturity and mechanical damage [23]. Our data showed a sharp drop in replicational effect and rapid increase in the contribution of maturity group at the fifth leaf stage (stage 15). The contribution of maturity group at this stage is stronger than that of varietal differences within each maturity group; notwithstanding, unaccountable sources of variation have the largest effect in triggering the days in attaining the fifth leaf stage. At this stage, the early maturing varieties were quite distinguishable from the late and intermediate maturing varieties (Table 2). This shows that the days in attaining the fifth leaf stage may indicate seedling maturity group observations on the field. However, this period has a short window and may be unreliable as several factors also control it.

Plant growth is a function of the increase in dry matter accumulation. The quantity of dry matter accumulated by the varieties depends on the efficiency of the assimilatory system, which is the leaf. At the early vegetative stages, the days to phenological intervals of the late and intermediate varieties were not in the order of their maturity classification due to the significant interference of variety (maturity group). The high effect of variety (maturity group) resulted from external conditions largely influencing their relative growth rate. These external conditions not investigated in the experiment also culminated in the higher contribution of error at this growth stage. The unaccountable sources of variation (error) are the other factors contributing to the days in reaching this phenological interval apart from maturity group, variety (maturity group) and replication. Such sources include day length, temperature, irradiation, relative humidity and soil nutrient supply [24]. All these factors are important determinants of maize performance in terms of leaf initiation and stem elongation. It has been reported that seedling traits or seedling growth indices are not reliable indices for maize maturity classification [15]. Our data further show that the trend of dry matter accumulation in the early vegetative phenological intervals in maize is not reliable for maturity classification.

The three maturity groups were quite distinguishable at stage 23 when 13 leaves were fully expanded, with maturity group accounting for more than half of the observed variability at this stage. According to [25], the late vegetative stage shortly after the 12th leaf is the first peak in dry matter accumulation rate as the other peak occurs during grain filling. This implies that the timing of the peaks in dry matter accumulation patterns is positively related to maturity. This stage also specifies the advanced stages of leaf development, triggering the initiation of floral primordia. The heavy effect of maturity group on the majority of the observed variations in the late vegetative and reproductive stages indicates the usefulness of these phenological intervals in ranking maturity in maize. The low influence of maturity group on the anthesis-silking interval of the varieties resulted, as this stage is seriously affected by many environmental factors, including drought and limiting resources [26, 27].

## CONCLUSION

Maturity group had significant effects on the days to phenological intervals of the varieties planted. The timing of growth stages in maize is controlled by maturity with genotypic differences having little impact. During the period of seedling establishment, maturity group had the highest influence on phenological interval 15 when 5 leaves were fully expanded. Across the maturity groups investigated, maturity group had more influence on their growth stages at the later vegetative and flowering stages than at seedling establishment. Though this experiment was carried out in a rainforest location, analysing the phenology of different maize maturity under multiple agro-ecological conditions and complex environments will increase the precision in using phenological data for modelling crop maturity.

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**Supplemental Table: The BBCH identification codes for the different maize phenological stages.**

<b>Principal growth stage</b>	<b>BBCH identification code</b>	<b>Meaning</b>
<b>Leaf development</b>	10	First leaf through the coleoptile
	11	First leaf unfolded
	12	2 leaves unfolded
	13	3 leaves unfolded
	14	4 leaves unfolded
	15	5 leaves unfolded
	16	6 leaves unfolded
	17	7 leaves unfolded
	18	8 leaves unfolded
	19	9 leaves unfolded
	20	10 leaves unfolded
	21 continuous till 30	11 leaves unfolded, stages continuous till when 20 leaves are unfolded
<b>Inflorescence emergence</b>	59	Tassel fully emerged and separated
<b>Flowering and anthesis</b>	63	Beginning of pollen shedding (anthesis)
	65	Female stigmata fully emerged (silk)

Adapted from Lancashire [19].

#### COMPETING INTERESTS

#### DISCLAIMER:

Authors have declared that no competing interests exist. The products used for this research are commonly and predominantly used products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.