IMAGE – RICE GRAIN SCANNER: A THREE-DIMENSIONALLY FULL AUTOMATED NEW ASSESSMENT OF GRAIN SIZE AND QUALITY TRAITS FOR RICE BREEDING

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INTRODUCTION

Majority of rice breeding programs primarily focus on yield as the most important trait. Nevertheless, grain quality is a trait also evaluated and used for selection in breeding programs. Rice quality however is a set of traits which are not easy to measure, and quite often are evaluated as qualitative instead of quantitative one. Even traits as grain size, directly related to grain quality, typically a quantitative trait, are too much depended on sample size and can lead to basic sampling errors, which, on the other hand, will lead to a wrong estimation of average grain size. If chalkiness is the trait to be evaluated it became even more difficult to get good evaluations, since usually it is treated as a qualitative trait. Another problem is that chalkiness is irregularly distributed in the grain. Sometimes chalkiness affects a small part of many grains, other times lead to complete chalked grains, but only a few ones are affected. So, it is not only important to know how many grains have chalked areas, but how large is this area affected throughout the grains sample.

The solutions available for rice scientists to handle the selection of such complex traits are softwares that handle images took usually by an ordinary scanner, which measure and analyze grains in two dimensions. Examples are 'SmartGrain' (TANABATA et al., 2012), or the Rice/Grain Analyzer Software 6980 (Osaw Ind. Products Pvt. Ltd., Indian). There are solutions that bring together device and software, like the Classifier S21 – Rice Statistical Analiser (Máquinas Suzuki S.A., Brazil), the RN300 Rice Quality Analyzer (Kett, US), the Satake RSQI10A Grain Scanner (Satake, Australia), the SeedCount SC5000 Rice Analyser (Next Instr. Pty Ltd, Australia), and the QSorter Explorer (QualySense AG, Switzerland). There are even other approaches given by softwares (WHAN et al., 2014; MRUTYUNJAYA et al., 2014). Such devices are the update possibilities of breeding for cereal quality worldwide, but not always are specific for rice.

The 'Image Classifier' was developed by Selgron (Brazil) to support the rice industry by quickly analyzing samples of about 3300 milled grains (100g) in twelve minutes. The device verifies how each sample match to the different rice type classifications of the Agriculture Ministry of Brazil at its norms (MAPA, 'Instrução Normativa nº 06', Feb 16th 2009).

However, for the research use of this scanner, changes, adaptations and the way to organize the rough data were necessary. The software platform to manage samples at the Image Rice Grain Scanner was so, developed by Selgron under Epagri's scientists demand, advisement and supervision, so that ensures a full use applied to the breeding/research needs. This includes some routines, like sample identification through a bar code reader, step by step protocol for analysis, data layout at the spreadsheet, and even new approaches, like the introduction of standard deviation at size measurements, total sample grain chalked area (besides percentage of chalked grains), and milling quality estimates. The final version of the Image Research Software Platform was upgraded at May 2016 and the final product was called "Image – Rice Grain Scanner" (MARSCHALEK et al., 2017).

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MATERIAL AND METHODS

HARDWARE & SOFTWARE: The equipment (100W; 220V, 50-60Hz, weight 43 kg) is operated by means of a microcomputer integrated with the image scanner. The device consists of the scanner itself (Figure 1), computer, barcode reader and screen.

SCANNING: rice samples can be identified by a field tag with a bare code, according field plots at the field trials. To prepare the samples for the analysis at the Image, each sample of rough (paddy) rice must be dehulled and milled to produce white milled rice. At this point, the advantage of the software designed by Epagri/Selgron over the traditional evaluations of the milled yield quality will start to become evident. The Image Rice Grain Scanner is set up to an initial sample of rough rice, of 100g, 50g or 25g. Only this three weights of rough rice should be used for a good estimation of milling quality (milling yield). The sample is finally placed at Image's helical plate at once, and in a few minutes all the grains of the whole sample are analyzed. The device orders the dehulled grains ensuring that each would pass a sensor individually, in free fall, while the grain is three-dimensionally analyzed, which seems to be a unique feature for rice scanners at the market, allowing the complete coverage of the whole grain surface (Figure 2). The software allows a precise measurement of grain geometry, shape, size, grain classes, chalkiness, other grain defects and an estimation of milling quality, resulting in the output of 40 different information.

THE SOFTWARE PLATFORM: Once finished the sample analysis (run)at the scanner, the data will appear at the spreadsheet on the screen at a clear six kind of columns division. The first one is composed by columns with traits coming from field data. The second division is related to sample identification, like complete code of the sample, followed by the trial code and the plot (sample) number itself. The third division of data output gives information about the size of the whole sample (length, width and thickness), including all grains (even broken ones), standard deviations (length, width and thickness), length/width relation and total number of grains analyzed. Also the fifth and sixth division of columns will contain the different classes of the analyzed grains and the last columns will contain milled quality data.

There is possible to apply a 'filter' to the already obtained grain size data just before saving the data of each sample. The filter is useful to determine the real size of the unbroken grains of a genotype. If a filter of 6.5 mm is applied to the length the software will recalculate de average of the length, width and thickness taking in account only the grains over 6.5 mm of length. This will give the breeder a real idea of new line/cultivar grain's size.

RESULTS AND DISCUSSION

PERFORMANCE: The Image Scanner takes about 3 minutes to analyze 35g milled rice (starting with 50g rough rice). Since 35g milled rice usually represent about 1,000 grains, it is a good representative sample of a line or variety.

Next can be find the 40 output information given by the Image at the consolidate spreadsheet. Sample identification columns: 'Complete Field trial code'; Field experiment/trial code; Plot (Sample) number (inside each trial). Sample grain size columns (green header at Figure 3): grain length (mm), standard deviation of grain length, grain width (mm), standard deviation of grain width, grain Thickness (mm), standard deviation of grain thickness, length/width Ratio, number of grains analyzed.

Applied settings for the filter will fill up the next group of columns, called 'Amostral com filtro' (Sample with filter) (red header at Figure 3) composed by: grain length (mm), standard deviation of grain length, grain width (mm), standard deviation of grain width, grain thickness (mm), standard deviation of grain thickness, length/width Ratio and number of grains analyzed.

The 'Totalizador' (totalizer) group of columns (orange header at Figure 3) refers to the different classes and/or defects according the Brazilian Ministry of Agriculture, Livestock and Supply (MAPA, 'Instrução Normativa nº 06', Feb 16th 2009). Adjustments at the class building parameters can easily be done by Selgron regarding to the needs or specific

regulations of each country. The totalizer columns are composed by: estimated sample weight, good grains (%), white bally grains (%), moldy and burnt grains (%), chopped/stug and stained grains (by insects or fungi) (%), total chalky area (%), no chalked grains (%), chalked grains (%), striped, yellow and color outsiders (%), long-thin grains (%), medium grains (%), short grains (%), whole grains (%), broken grains (%), very small grain pieces (%), impurities (%).

The last group of columns at the output spreadsheet file are the Milling Quality columns (black header, not shown at Figure 3), given by the percentage of whole and broken grains , and the total milling quality percentage.

Regarding the chalkiness trait, as commented at the introduction, sometimes this defect affects a small part of many grains, or depreciate many grains which show large chalked areas. Other times lead to totally chalked grains, but only a few grains are really affected, remaining many no chalked grains. This are different aspects of chalkiness variations, and the Image Rice Grain Scanner is able to handle it by given out two different measurements, once, by the total chalked area of the sample's surface (all grains), and on the other hand, counting the number of grains that had some chalked area, even if the grain has a very small chalked area. This two parameters of the same trait are really useful for breeders to check the chalkiness patterns of new lines.

CONCLUSION

The Image Rice Grain Scanner has been successful used at Epagri's Rice Breeding Laboratory (LAMGEN) up to January 2016. The scanner and its software proved to be easy to operate, stable and reliable. The data output from samples are currently used for selection, proving to be very reliable and useful to Epagri's rice breeding team. All this give breeders the real possibility of analyzing a big set of samples in a very fast and accurate way. Therefore, an efficient selection for quality can be done at very early breeding stages. Probably this equipment could also be successfully used in phytopathology or entomology studies, and initial assesstments have been done on this way. The device can certainly be applied to studies on abiotic stresses like cold or heat on grain quality, which first assesstments are in progress, since many defects can be measured, and so, a decrease in quality traits can be accurately measured.

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Figure 1: Front and back views of the Image Rice Grain Scanner

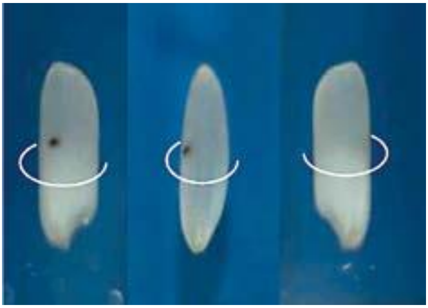

Figure 2: Three-dimensionall images during the free fall covering the whole grain surface

INFORMAÇÕES	AMOSTRAL								AMOSTRAL COM FILTRO									
Código	Comprimento do grão	Desvio padrão Comprimento	Largura Grão	Desvio padrão Largura	Espessura grão	Desvio padrão espessura	Relação compr./largura	QUANTIDADE	Valor do filtro Aplicado	Condição Aplicada	Comprimento do grão	Desvio padrão Comprimento	Largura Grão	Desvio padrão Largura	Espessura grão	Relação compr./largura	QUANTIDADE
Epagri 104	6,59	0,56	2,07	0,14	1,72	0,11	3,18	2735	6,50	>=	6,92	0,27	2,08	0,12	1,73	3,32	1702
Epagri 105	6,43	0,54	2,26	0,17	1,72	0,13	2,85	2698	6,50	>=	6,81	0,23	2,27	0,14	1,73	2,99	1311
Epagri 106	6,23	0,54	2,16	0,15	1,70	0,11	2,88	2796	6,50	>=	6,83	0,28	2,18	0,14	1,71	3,13	866
Epagri 107	6,39	0,66	1,97	0,13	1,61	0,10	3,24	3146	6,50	>=	6,87	0,33	1,98	0,12	1,63	3,47	1457
Epagri 108	6,54	0,79	2,18	0,17	1,69	0,13	3,00	1820	6,50	>=	7,03	0,37	2,18	0,13	1,71	3,23	1049
Epagri 109	6,45	0,74	2,17	0,15	1,68	0,13	2,98	2258	6,50	>=	6,95	0,32	2,17	0,12	1,70	3,21	1216
SCS112	6,62	0,92	2,10	0,14	1,69	0,11	3,15	2848	6,50	>=	7,20	0,41	2,11	0,12	1,69	3,42	1752
SCSBRS Tio Taka	6,57	0,72	2,16	0,14	1,68	0,11	3,04	2200	6,50	>=	7,02	0,35	2,16	0,12	1,69	3,25	1309
SCS114 Andosan	6,53	0,62	2,18	0,16	1,68	0,12	3,00	2361	6,50	>=	6,95	0,33	2,19	0,13	1,70	3,18	1276
SCS116 Satoru	6,68	0,72	2,20	0,16	1,70	0,12	3,04	1480	6,50	>=	7,04	0,35	2,21	0,13	1,71	3,18	1011
SCS117 CL	6,81	0,69	2,26	0,19	1,72	0,14	3,02	2109	6,50	>=	7,07	0,35	2,27	0,14	1,73	3,12	1583
SCS118 Marques	6,50	0,72	2,17	0,14	1,67	0,12	2,99	1898	6,50	>=	7,00	0,35	2,18	0,12	1,68	3,22	1017
SCS121 CL	6,54	0,73	2,17	0,15	1,71	0,11	3,02	1888	6,50	>=	7,01	0,35	2,18	0,13	1,72	3,22	1099
SCS122 Miura	6,54	0,78	2,10	0,16	1,69	0,12	3,11	1980	6,50	>=	7,00	0,33	2,11	0,12	1,69	3,31	1191

INFORMAÇÕES	TOTALIZADOR																			
Código	PESO TOTAL DA AMOSTRA	Bom %	Barriga Branca %	Bom + Barriga Branca %	Mofado e Ardidos %	Picados ou Manchados %	Área Gessada % (Centro Branco Oficial Brasil)	Grãos não gessados %	Grãos gessados %	Rajados %	Amarelos e Destoante %	Longo Fino %	Longo %	Médio %	Curto %	Inteiro %	Quebrados %	Quirera %	Impureza (Semente) %
Epagri 104	56,29	92,31	0,00	92,31	0,00	0,73	5,67	91,85	8,15	0,00	0,69	6,16	83,34	8,75	0,47	99,31	0,17	0,00	0
Epagri 105	59,03	95,29	0,00	95,29	0,00	0,90	2,76	94,85	5,15	0,00	0,49	0,44	83,33	8,93	0,15	99,41	0,09	0,00	0
Epagri 106	56,10	95,82	0,00	95,82	0,00	0,48	2,77	95,14	4,86	0,06	0,33	0,88	68,99	23,92	0,30	99,41	0,10	0,00	0
Epagri 107	56,09	98,86	0,00	98,86	0,00	0,32	0,12	98,97	1,03	0,00	0,40	22,15	60,67	13,81	1,86	98,77	0,92	0,00	0
Epagri 108	38,45	97,94	0,00	97,94	0,00	0,42	0,43	97,21	2,79	0,00	0,45	0,99	82,21	9,26	0,86	98,65	0,59	0,00	0
Epagri 109	46,61	94,10	0,00	94,10	0,00	0,21	4,72	92,46	7,54	0,06	0,25	0,69	79,58	11,37	0,79	98,86	0,49	0,00	0
SCS112	58,56	98,29	0,00	98,29	0,00	0,54	0,12	98,50	1,50	0,00	0,56	3,53	76,38	12,83	1,82	98,95	0,57	0,00	0
SCSBRS Tio Taka	45,94	91,53	0,00	91,53	0,00	0,28	7,01	90,74	9,26	0,00	0,52	1,73	82,19	9,94	0,66	98,95	0,39	0,00	0
SCS114 Andosan	49,54	98,30	0,00	98,30	0,00	0,35	0,31	97,93	2,07	0,00	0,37	1,92	83,92	9,35	0,30	99,26	0,12	0,00	0
SCS116 Satoru	32,49	98,59	0,00	98,59	0,00	0,04	0,24	96,93	3,07	0,00	0,26	0,83	88,70	4,73	0,45	98,71	0,43	0,00	0
SCS117 CL	49,04	98,18	0,00	98,18	0,00	0,29	0,65	97,58	2,42	0,00	0,20	0,63	91,78	3,06	0,36	99,09	0,23	0,00	0
SCS118 Marques	39,28	95,98	0,00	95,98	0,00	0,25	2,85	94,08	5,92	0,00	0,31	0,90	82,14	8,87	0,81	99,11	0,28	0,00	0
SCS121 CL	40,03	98,29	0,00	98,29	0,00	0,40	0,27	97,86	2,14	0,00	0,34	1,73	82,82	7,87	0,77	98,95	0,35	0,00	0
SCS122 Miura	40,30	95,80	0,00	95,80	0,00	0,57	2,39	94,63	5,37	0,00	0,51	4,16	79,78	9,81	1,49	98,76	0,52	0,00	0

Figure 3: Partial spreadsheet of an output data file.