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UTICAJ TIPA I SABIJENOSTI ZEMLJIŠTA NA PRINOS KUKURUZA

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Sažetak: Cilj ovoga rada bio je da se utvrdi prinos različitih hibrida kukuruza (*Zea mays* L.) u zavisnosti od tipa i sabijenosti zemljišta. Ogled je postavljen tokom 2016. i 2017. godine na teritoriji opštine Leskovac, na tri različita tipa zemljišta (aluvijum, smonica i parapodzol). U ogledu je bilo uključeno 9 hibrida kukuruza različitih FAO grupa zrenja (NS 4051, AGR DIAN, ZP 427, NS 5211, ZP 555, AS 534, NS 6030, ZP 666 i Bećar). Najveći prosečni prinosi dobijeni su na aluvijumu, potom parapodzolu, a najmanji na smonici. Najveći prosečan prinos za sve tipove zemljišta dobijen je gajenjem hibrida iz FAO grupe zrenja 500, a najmanji gajenjem hibrida iz FAO grupe zrenja 400. Na zemljištu tipa smonice, najveći prosečni prinos dobijen je gajenjem hibrida iz FAO grupa zrenja 500, na zemljištu tipa parapodzol gajenjem hibrida iz FAO grupe zrenja 400, a na zemljištu tipa aluvijum, gajenjem hibrida iz FAO grupe zrenja 600. Najveći prosečni prinos (11,90 t ha^{-1}) dobijen je gajenjem hibrida ZP 666 na zemljištu tipa aluvijum, a najmanji (4,60 t ha^{-1}) gajenjem hibrida NS 6030, na zemljištu tipa smonice. Najveća sabijenost zemljišta u orničnom sloju utvrđena je na smonici, potom na parapodzolu, a najmanja na aluvijumu. Ovi podaci o prosečnim prinosima govore o pogodnostima pojedinih tipova zemljišta za proizvodnju kukuruza i daju smernice proizvođačima za odabir najrodnijih hibrida koje žele gajiti na svojim njivama.

Ključne reči: hibridi, FAO grupa zrenja, smonica, parapodzol, aluvijum.

Uvod

Zemljište predstavlja izuzetno dinamičnu sredinu, jedan je od najvažnijih elemenata biljne proizvodnje i osnovni je preduslov visokih i stabilnih prinosa. Sastav zemljišta i njegove osobine su osnove plodnosti zemljišta, pa su prinosi biljaka na različitim tipovima zemljišta promenjivi (Sekulić et al., 2004). Zemljište

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je osnovni supstrat ukorenjavanja biljaka, a rast korena zavisi od dubine sloja, biljne vrste, karakteristika korena, sabijenosti zemljišta, vlage i sl. (Navaz et al., 2013). Živanović (2012) proučavajući uticaj tipa zemljišta (černozem i gajnjača), đubrenja i hibrida na prinos kukuruza ističe da je tip zemljišta imao vrlo značajan uticaj na prinos kukuruza, veći od hibrida i đubrenja. Jevtić (1986) ističe da prinos zrna kukuruza zavisi od hibrida (46–51%), agroekoloških uslova (9–23%) i nivoa primenjene tehnologije gajenja (31–40%).

Williamson i Neilsen (2000) ističu da se u modernoj poljoprivredi većina operacija od setve do žetve obavlja mehanizovano, pomoću teških traktora koji sabijaju zemljište u svakom prohodu. U takvim zemljištima vladaju nepovoljni uslovi za razvoj korenovog sistema, slabija je mikrobiološka aktivnost, što za posledicu ima smanjenje prinosa i povećanje troškova proizvodnje za 20–40%. Od svih oblika degradacije zemljišta, sabijanje učestvuje sa 11% i predstavlja jedan od glavnih oblika degradacije zemljišta (Lynden, 2000).

Radojević et al. (2006) i Mueller et al. (2010) ističu da teške mašine utiču na kvarenje strukture i degradaciju zemljišta u gornjem (površinskom) i donjem (dubokom) sloju čime se smanjuje njegova produktivnost i povećava potrošnja energije i opasnost od erozije. Nikolić et al. (2002) navode da veliki pritisci mehanizacije prouzrokuju prekomerno sabijanje zemljišta, što za posledicu ima smanjenje prinosa za 10–25%, dok Kuht and Reintam (2004) ističu i značajnije smanjenje prinosa, preko 40%. Ishaq et al. (2001) i Sakib et al. (2004a) ističu da je ukupni efekat sabijanja zemljišta na prinos biljaka negativan. Koliki je uticaj sabijenosti zemljišta na prinos kukuruza govore podaci do kojih su u svojim istraživanjima došli Barać et al. (2016) koji ističu da je prinos kukuruza na uvratinama, gde je veći broj prohoda mehanizacije, bio manji za 38,74% nego prinos u unutrašnjosti parcele. Zemljišta se razlikuju po sadržaju organske materije, vode, strukture i teksture i na njihovu sabijenost utiče broj prohoda mehanizacije, broj i dimenzija pneumatika, brzina kretanja mašina i sl. (Sakai et al., 2008). Smanjenjem broja operacija u obradi zemljišta čuvaju se fizičke osobine zemljišta čime se čuva sadržaj organske materije u zemljištu, poboljšava se biodiverzitet zemljišta i smanjuju troškovi proizvodnje (Morris et al., 2010; Soane et al., 2012). Sadržaj vode u zemljištu veoma je bitan u procesu sabijanja zemljišta i direktno utiče na opterećenje pri pritisku na zemljište (Hamza i Anderson, 2005). Posebno je bitan sadržaj vode u gornjem sloju. Na zemljištima koja se obrađuju sa manjiim brojem prohoda smanjena je evapotranspiracija a povećan sadržaj vode, posebno u gornjim slojevima (Bescansa et al., 2006).

Sabijenost zemljišta zavisi od gajenja pojedinih biljaka. Tako Milošev et al. (2007) navode da proizvodnja šećerne repe i kukuruza najviše uzrokuje sabijanje zemljišta, dok to nije slučaj sa pšenicom.

Opšte posmatrano, sabijanje zemljišta dovodi do usporavanja apsorpcije vode i hranjivih materija, slabijeg razvoja korenovog sistema, njegove dužine i prodiranja u dublje slojeve, usporen rast biljaka, što sve skupa ima za posledicu slabiji razvoj biljaka i umanjenje prinosa (Chen i Veil, 2011; Lipiec et al., 2012; Nosalevicz i Lipiec, 2014; Prakash et al., 2014; Dimitri i Destain, 2016).

Cilj ovoga rada bio je da se utvrdi prinos različitih hibrida kukuruza (*Zea mays* L.) u zavisnosti od tipa i sabijenosti zemljišta.

Materijal i metode

Ogled je postavljen tokom 2016. i 2017. godine na teritoriji opštine Leskovac (Južna Srbija), na tri različita tipa zemljišta (aluvijum, smonica i parapodzol). U ogledu je bilo uključeno 9 hibrida kukuruza različitih FAO grupa zrenja (NS 4051, AGR DIAN, ZP 427, NS 5211, ZP 555, AS 534, NS 6030, ZP 666 i Bećar). Pre postavljanja ogleda sa parcela su uzeti uzorci zemljišta za hemijsku analizu. Ogled je postavljen po planu blok sistema u tri ponavljanja. Veličina elementarne parcele iznosila je 28 m^2. Sejana su po 4 reda svakog hibrida. Međuredno i rastojanje u redu je iznosilo: 70 cm x 25 cm (FAO grupa 400), 70 cm x 30 cm (FAO grupa 500) i 70 cm x 35 cm (FAO grupa 600), tako da bi hibridi imali optimalnu gustinu. Predusev je bila ozima pšenica. Priprema zemljišta podrazumevala je jesenje oranje na dubinu od 30 cm gde je unešeno i 250 kg ha^{-1} NPK (16:16:16) đubriva. Predsetvena priprema je obavljena neposredno pred setvu setvospremačem. Setva je obavljena u drugoj polovini aprila meseca. Tretiranje protiv semenskih korova (*Echinochloa crus – galli, Solanum nigrum, Sorghum halepense, Chenopodium spp.*) obavljeno je dan nakon setve, preparatima *Basar* i *Rezon*. Tretiranje protiv širokolisnih i uskolisnih korova obavljeno je preparatima *Siran* i *Maton*. Prihrana KAN-om je obavljena u fazi 3–5 listova, nakon prve međuredne kultivacije, u količini od 350 kg ha^{-1}. Tokom vegetacije nije bilo prisutnih bolesti i štetočina. Berba kukuruza je obavljena u tehnološkoj zrelosti. Prinos je obračunat na svakoj parceli i sveden na 14% vlage u zrnu. Prinos različitih hibrida kukuruza u zavisnosti od tipa i sabijenosti zemljišta obrađen je statistički, analizom varijanse i Pearson-ovim koeficijentom korelacije, uz upotrebu softvera WASP 1.0.

Pošto se radi o različitim tipovima zemljišta određivali smo i njihovu sabijenost. Sabijenost je merena nakon setve i nakon berbe kukuruza, penetrologerom Eijkelkamp hardver verzije 6,0, softver verzije 6,03. Merenje sabijenosti je obavljeno u skladu sa standardom NEN 5140, brzinom penetracije od 2 cm sec^{-1}, pri čemu odstupanje nije bilo veće od 0,5 cm s^{-1} a sve prema standardu (ASAE S313.1). Pre početka merenja postavljena je referentna ploča, određen položaj parcele (GPS) i vlažnost zemljišta. Vlažnost zemljišta u momentu merenja sabijenosti određivana je Theta sondom, a izražena je u % vol. Merenja su obavljena na unutrašnjem delu parcele na dubini od 0 do 40 cm u 5 ponavljanja. Rezultati sabijenosti su predstavljeni kao prosečni i prikazani su tabelarno.

Klimatske i zemljišne karakteristike

Leskovačka kotlina, mesto izvođenja ogleda, dugačka je oko 50 km, a široka oko 45 km i ima nadmorsku visinu od oko 230 m. Odlikuje se plodnim zemljištima i povoljnim klimatskim uslovima za biljnu proizvodnju. Šušić (2000) ističe da su na ovom području najrasprostranjeniji tipovi zemljišta aluvijum (duž reke Veternice), smonice, deluvijumi i parapodzoli (u podnožju pobrđa).

U tabeli 1 prikazane su ukupne mesečne padavine i prosečne mesečne temperature u toku vegetacionog perioda kukuruza. U toku 2016. godine, koja je bila povoljnija za proizvodnju kukuruza, ukupna suma padavina u toku vegetacije iznosila je 438,8 mm. Posebno treba naglasati količinu padavina u junu, julu i avgustu (207 mm) koja je veoma bitna za formiranje prinosa kukuruza. Sa aspekta ukupne količine padavina (308 mm), 2017. godina je bila lošija, posebno padavina u junu, julu i avgustu mesecu (73 mm) i može se smatrati nepovoljnom za biljnu proizvodnju.

Tabela 1. Padavine (mm) i prosečne temperature (^{0}C) za Leskovac.
Table 1. Precipitation (mm) and mean temperatures (^{0}C) in Leskovac.

	Apr.	Maj	Jun	Jul	Avg.	Sep.	Okt.	Apr./Okt.
Sezona 2016/*The 2016 growing season*								
mm	24,2	69,6	63	114	30	56	82	438,8
^{0}C	13,7	14,7	21,9	22,8	21,4	17,9	11,9	17,7
Sezona 2017/*The 2017 growing season*								
mm	69	82	19	34	20	20	64	308,0
^{0}C	11,3	16,7	21,9	23,5	23,3	18,7	12,5	18,2
Višegodišnji prosek/*Multi-year average 1985-2014*								
mm	48	46	37	25	24	30	36	246
^{0}C	12,5	16,5	19,5	22,0	22,5	18,0	14,0	17,8

U poređenju sa višegodišnjim prosekom (246 mm), obe godine su imale veću količinu padavina. Prosečne temperature u toku vegetacije kukuruza u 2017. godini bile su nešto veće nego u 2016. godini, posebno one u julu, avgustu i septembru. Prosečne temperature, u obe godine, mogu se smatrati povoljnim za proizvodnju kukuruza. U poređenju sa višegodišnjim prosekom, prosečne mesečne temperature u junu i julu bile su veće u obe godine, dok su prosečne mesečne temperature u oktobru bile niže. Ukupne padavine u toku vegetacije, u obe godine, bile su veće za 62, odnosno 192,8 mm u odnosu na višegodišnji prosek.

U tabeli 2 prikazane su hemijske osobine zemljišta. Kiselost zemljišta je određena metodom po Kapenu, humus je određen metodom po Kotzmanu, ukupni azot metodom po Kjeldahlu, a dostupni fosfor i kalijum Engner-Riehm Al metodom.

Tabela 2. Hemijske osobine zemljišta.
Table 2. Chemical properties of the soil.

Tip zemljišta *Type of soil*	pH – *pH*		Humus *Humus*	Azot *Nitrogen*	Dostupno *Available (mg/100g of soil)*	
	H_2O	KCl	(%)	(%)	P_2O_5	K_2O
Smonica – *Vertisol*	5,94	4,74	2,12	0,12	9,78	30,00
Parapodzol – *Parapodzol*	6,14	5,04	2,31	0,12	9,42	20,00
Aluvijum – *Alluvium*	7,74	5,78	2,65	0,13	17,56	34,00

Prema vrednostima pH u KCl-u smonica (5,78) i parapodzol (5,04) pripadaju grupi kiselih, a aluvijum (5,78) grupi umereno kiselih zemljišta. Po sadržaju humusa u orničnom sloju, sva tri tipa zemljišta pripadaju grupi slabo humoznih zemljišta (Škorić, 1991). Sadržaj ukupnog azota, kod sva tri tipa zemljišta, bio je približno isti i zemljišta su srednje obezbeđena ovim elementom. Sadržaj fosfora kod smonice (9,78) i parapodzola (9,42) bio je nizak, dok je aluvijum (17,56) sa ovim elementom optimalno obezbeđen. Zemljišta smonica (30,00) i aluvijum (34,00) su visoko obezbeđena kalijumom, dok je parapodzol (20,00) optimalno obezbeđen. Iako ova zemljišta pripadaju grupi potencijalno plodnih zemljišta, njihovo intenzivno korišćenje uglavnom zahteva primenu meliorativnih mera.

Rezultati i diskusija

Odavno je poznato da su hibrid, agrotehnika i klima osnovni faktori za visoke prinose kukuruza. Zbog sigurnije proizvodnje, neophodno je gajiti nekoliko hibrida različite dužine vegetacije. Tako su u ovim istraživanjima odabrana po tri hibrida koja pripadaju FAO grupama zrenja 400, 500 i 600. U tabeli 3 dat je prikaz prinosa hibrida različitih FAO grupa zrenja u zavisnosti od tipa zemljišta.

Tip zemljišta je imao vrlo značajan uticaj na prosečni prinos kukuruza. Tako je prosečan prinos kukuruza na zemljištu tipa aluvijum iznosio 10,34 t ha^{-1} i bio je vrlo značajno veći nego prosečan prinos kukuruza na zemljištima tipa smonica (5,68 t ha^{-1}) i parapodzol (7,61 t ha^{-1}). Takođe, vrlo značajno veći prinos ostvaren je na zemljištu tipa parapodzol u odnosu na smonicu. Sličnu tendenciju razlika u prinosu u zavisnosti od tipa zemljišta imali su i pojedinačni hibridi. Bekavac (2012) ističe da su zahtevi kukuruza prema zemljištu izraženi, s obzirom na to da je to visoko produktivna biljna vrsta. Ovako izražene razlike u prinosu mogu biti rezultat hemijskih osobina datih zemljišta, s obzirom na to da su smonica i parapodzol zemljišta kisele reakcije i da imaju nizak sadržaj fosfora za razliku od aluvijuma. Uzrok mogu biti i drugi agroklimatski faktori bitni za formiranje prinosa. Tako su lokacije na kojima su se ispitivani tipovi zemljišta nalazili, na različitim nadmorskim visinama, međusobno udaljene od 20 do 25 km. Imajući u vidu lokalni karakter padavina, može se pretpostaviti da su neke lokacije imale više, a neke manje padavina tokom vegetacije. Tako Bekavac et al. (2010) ističu da u poslednjih 25 godina visina prinosa zrna u sve većoj meri zavisi od meteoroloških uslova tokom vegetacionog perioda kukuruza, a koji se vrlo često karakterišu pojavom „ekstremnih klimatskih događaja".

Tabela 3. Uticaj tipa zemljišta i hibrida na prinos kukuruza (t ha $^{-1}$) (2016/2017).
Table 3. The impact of soil type and hybrids on maize yield (t ha $^{-1}$) (2016/2017).

B. Hibridi *Hybrids*	A. Tipovi zemljišta/*Types of soil*					
	Smonica *Vertisol*		Parapodzol *Parapodzol*		Aluvijum *Alluvium*	Prosek B *Average*
NS 4051	5,95		8,60		9,18	7,91
AGR DIAN	5,45		8,55		10,49	8,16
ZP 427	5,25		7,05		8,95	7,08
Prosek za FAO 400 *Average for FAO 400*	5,55		8,07		9,54	7,72
NS 5211	6,10		8,85		11,20	8,72
ZP 555	6,65		8,55		10,10	8,43
AS 534	5,70		5,60		10,15	7,15
Prosek za FAO 500 *Average for FAO 500*	6,15		7,66		10,48	8,10
NS 6030	4,60		6,30		10,05	6,98
ZP 666	5,55		8,20		11,90	8,55
Bećar	5,90		6,75		11,05	7,90
Prosek za FAO 600 *Average for FAO 600*	5,35		7,08		11,00	7,81
Prosek A *Average*	5,68		7,61		10,34	7,87
	A		B		A x B	
LSD	5%	1%	5%	1%	5%	1%
	0,184	0,245	0,319	0,422	0,552	0,736

Najveći prosečni prinos za sve tipove zemljišta (8,10 t ha^{-1}) dobijen je gajenjem hibrida iz FAO grupe zrenja 500, a najmanji (7,72 t ha^{-1}) gajenjem hibrida iz FAO grupe zrenja 400. Razlike u prosečnom prinosu između FAO grupa zrenja nisu bile statistički visoko značajne. Međutim, prosečni prinosi po FAO grupama zrenja razlikovali su se u zavisnosti od tipa zemljišta. Tako je na zemljištu tipa smonice, najveći prosečni prinos (6,15 t ha^{-1}) dobijen gajenjem hibrida iz FAO grupe zrenja 500 i on je bio statistički značajno veći nego prosečni prinosi hibrida iz FAO grupa 400 i 600. Najveći prosečni prinos (8,07 t ha^{-1}) na zemljištu tipa parapodzol, dobijen je gajenjem hibrida iz FAO grupe zrenja 400 i on je bio statistički značajno veći nego prosečni prinosi hibrida iz FAO grupa 500 i 600. Ovo zemljište se nalazilo na nešto većoj nadmorskoj visini od ostalih tipova. Na zemljištu tipa aluvijum, najveći prosečni prinos (11,00 t ha^{-1}) dobijen je gajenjem hibrida iz FAO grupe zrenja 600 i on je bio statistički značajno veći nego prosečni prinosi hibrida iz FAO grupa zrenja 400 i 500.

Najveći prosečni prinos za sve tipove zemljišta u FAO grupi zrenja 400 imao je hibrid AGR DIAN (8,16 t ha^{-1}), u FAO grupi zrenja 500 hibrid NS 5211 (8,72 t ha^{-1}), a u FAO grupi zrenja 600 hibrid ZP 666 (8,55 t ha^{-1}). Ako se analiziraju svi tipovi zemljišta i svi hibridi, vidljivo je da je najveći prosečni prinos (11,90 t ha^{-1}) dobijen gajenjem hibrida ZP 666 na zemljištu tipa aluvijum, a najmanji (4,60 t ha^{-1}) gajenjem hibrida NS 6030, na zemljištu tipa smonice. Prema ostvarenim prinosima, za preporuku je da se na smonici gaje hibridi iz FAO grupe zrenja 500, na parapodzolu hibridi iz FAO grupe zrenja 400, a na aluvijumu hibridi iz FAO grupe zrenja 600. Naši rezultati su u saglasnosti sa rezultatima Živanovića (2012) koji ističe da je tip zemljišta imao vrlo značajan uticaj na prinos kukuruza. Ovi podaci o prosečnim prinosima na različitim tipovima zemljišta upućuju proizvođače na odabir najrodnijih hibrida koje će gajiti na svojim njivama.

Sabijenost zemljišta

Gajenje pojedinih biljaka utiče na sabijenost zemljišta. Sabijanje zemljišta dovodi do usporavanja apsorpcije vode i hranjivih materija, slabijeg razvoja korenovog sistema, njegove dužine i prodiranja u dublje slojeve i usporen rast biljaka, što sve skupa ima za posledicu slabiji razvoj biljaka i umanjenje prinosa. U tabeli 4 dat je prikaz sabijenosti pojedinih tipova zemljišta, merene posle setve i posle berbe kukuruza.

Zemljišta su se međusobno razlikovala, kako po sabijenosti, tako i po vremenu merenja. Tako je najveća prosečna sabijenost (do 40 cm) izmerena na smonici, potom na parapodzolu a najmanja na aluvijumu. Naši rezultati su slični rezultatima koje navode Alakuku i Pavo (1994), gde ističu da sabijanje zemljišta nije isto po profilu, i najizraženije je do dubine od 50 cm.

Kod svih tipova zemljišta izmerena je veća sabijenost posle berbe nego posle setve kukuruza. Naši rezultati su u saglasnosti sa rezultatima koje navode Nikolić et al. (2006) i Simikić et al. (2005), ističući da je otpornost merena na proleće manja od one merene u jesen, što je rezultat višestrukog prohoda mehanizacije tokom sezone.

Sa porastom dubine dolazi i do porasta sabijenosti zemljišta. Najveći porast sabijenosti zabeležen je na dubini od 30 do 40 cm. Kod smonice i parapodzola, na 40 cm dubine, posle setve, izmerena je sabijenost od 2,94 i 2,37 MPa, a kod aluvijuma 2,12 MPa, što su vrednosti u kojima je otežan razvoj korenovog sistema. Kod svih zemljišta, u periodu posle berbe, izmerene su visoke vrednosti sabijenosti zemljišta (smonica 5,00 MPa; parapodzol 3,82 MPa i aluvijum 3,18 MPa), što pokazuje da je kukuruz u drugom delu vegetacije imao lošije zemljišne uslove. Da sabijenost zemljišta direktno utiče na smanjenje prinosa govore podaci koje navode Marinković et al. (1999), naglašavajući da to smanjenje kod kukuruza iznosi od 4,7 do 21,3% na našim zemljištima.

Sadržaj vlage u orničnom sloju, kod svih tipova zemljišta, bio je veći posle setve nego posle berbe. Tako je najveći sadržaj vlage (32%) bio kod parapodzola, dok je kod smonice (23%) i aluvijuma (24%) bio približno isti. Posle berbe, najveći sadržaj vlage (19%) bio je kod parapodzola, a najmanji (13%) kod aluvijuma. Tamo gde je sadržaj vlage bio veći, sabijenost je bila manja, što potvrđuju i rezultati do kojih su došli Savin et al. (2011a) i koji ističu da vlaga u zemljištu utiče na smanjenje sabijenosti.

Tabela 4. Sabijenost zemljišta (MPa).
Table 4. Soil compaction (MPa).

Period *Period*	Vlaga % *Moisture*	Dubina (cm)/*Depth (cm)*					Prosek *Average*
		1	10	20	30	40	
		Smonica/*Vertisol*					
Posle setve *After sowing*	23	0,35	0,79	1,22	1,61	2,94	1,38
Posle berbe *After harvesting*	17	0,39	1,52	3,32	4,01	5,00	2,69
		Parapodzol/*Parapodzol*					
Posle setve *After sowing*	32	0,14	0,73	1,33	1,55	2,37	1,22
Posle berbe *After harvesting*	19	0,42	1,14	2,41	3,37	3,82	2,23
		Aluvijum/*Alluvium*					
Posle setve *After sowing*	24	0,15	0,51	1,08	1,30	2,12	1,03
Posle berbe *After harvesting*	13	0,25	1,81	1,82	2,54	3,18	1,72
Pearson-ov koeficijent korelacije između prinosa i sabijenosti zemljišta *Pearson's coefficient of correlation between yield and soil compaction*							r = -0,99

Na osnovu koeficijenta korelacije (r=-0,99) utvrđeno je da postoji jaka negativna korelacija između prinosa kukuruza i sabijenosti zemljišta. Riedell et al. (2005) su utvrdili postojanje negativne korelacije između sabijenosti zemljišta i prinosa kukuruza, sa čime su saglasni i naši rezultati.

Ovi podaci o vezi prinosa i sabijenosti zemljišta upućuju proizvođače da na teškim i sabijenim zemljištima unose više organske materije i stajnjaka kako bi se popravili uslovi za bolji rast korenovog sistema čime bi i prinosi bili veći.

Zaključak

Na osnovu dvogodišnjih rezultata o uticaju hibrida, tipa i sabijenosti zemljišta na prinos kukuruza može se zaključiti sledeće: tip zemljišta je vrlo značajano uticao na prosečni prinos kukuruza. Najveći prosečni prinosi dobijeni su na aluvijumu, potom parapodzolu, a najmanji na smonici. Na zemljištu tipa smonice, najveći prosečni prinos dobijen je gajenjem hibrida iz FAO grupe zrenja 500, na zemljištu tipa parapodzol hibrida iz FAO grupe zrenja 400, a na zemljištu tipa aluvijum hibrida iz FAO grupe zrenja 600. Najveći prosečni prinos za sve tipove zemljišta u FAO grupi zrenja 400 imao je hibrid AGR DIAN, u FAO grupi zrenja 500 hibrid NS 5211, a u FAO grupi zrenja 600 hibrid ZP 666. Najveća sabijenost zemljišta u orničnom sloju bila je na smonici, potom na parapodzolu a najmanja na aluvijumu. Između sabijenosti zemljišta i prinosa kukuruza utvrđena je jaka negativna korelacija. Ovi podaci o prosečnim prinosima govore o značaju odabira hibrida i pogodnostima pojedinih tipova zemljišta za proizvodnju kukuruza.

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INFLUENCE OF SOIL TYPE AND COMPACTION ON MAIZE YIELD

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A b s t r a c t

The goal of this paper was to determine the yield of different maize hybrids depending on soil type and compaction. The experiment was carried out in 2016 and 2017 on the territory of the Leskovac municipality, including three different soil types (alluvium, vertisol, and parapodzol). The experiment comprised nine hybrids of different FAO maturity groups (NS 4051, AGR DIAN, ZP 427, NS 5211, ZP 555, AS 534, NS 6030, ZP 666 and Bećar). The highest average yields were achieved on alluvium, then on parapodzol, and the lowest yields were obtained on vertisol. The highest average yields on all examined soil types were achieved in the FAO 500 maturity group, and the lowest in the FAO 400 maturity group. The highest average yield on vertisol was achieved in the FAO 500 maturity group, on parapodzol in the FAO 400 maturity group, and on alluvium in the FAO 600 maturity group. The highest average yield (11.90 t ha^{-1}) was achieved with the ZP 666 hybrid on alluvium, and the lowest (4.60 t ha^{-1}) with the NS 6030 hybrid on vertisol. The largest soil compaction in the ploughing layer was determined in vertisol, then in parapodzol, and the smallest in alluvium. These data on average yields indicate the convenience of certain soil types for maize production, and provide guidelines for growers to select the most yielding hybrids for their fields.

Key words: hybrids, FAO maturity group, vertisol, parapodzol, alluvium.

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EFFECT OF SOYBEAN SEED PRIMING ON GERMINATION ENERGY

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Abstract: Six varieties were chosen from three different maturity groups: Galina and NS Princeza (0), Sava and NS Apolo (I), and Rubin and NS Zita (II) in order to examine the effect of priming on germination energy depending on soybean seed age. The seeds were produced in the period from 2012 to 2014, i.e., one- to three-year-old seeds were used. The following treatments were used for priming: distilled water – hydropriming (H_2O), ascorbic acid 250 mg/l (ASA), folic acid 15 mg/l (Fol.), hydrogen peroxide 1% (H_2O_2), and control – non-treated seeds (C). Results showed that the effects of priming depended on the variety, seed age, as well as on the applied treatment. Some varieties reacted to priming very well, while others had an inhibited reaction, so germination energy was significantly reduced. Rubin variety had the most favourable reaction, whereby all treatments of this variety led to an increase of quality, with an average increase of 2–8%. The application of primers on one-year-old soybean seeds had a weaker effect than on two- and three-year-old seeds. However, one-year-old seeds had minor oscillations in quality due to priming. The application of priming on one-year-old seeds can lead to an increase or decrease of germination energy by 3%, while two- and three-year-old seeds reached 7%. The results showed that seed priming in ascorbic acid had a positive effect, while the application of other primers did not affect the value of germination energy, nor did it lead to a decrease of its value. Ascorbic acid activity led to the largest increase in the quality of three-year-old soybean seeds by 2.83%, while the increase in the quality of two- and three-year-old seeds was 1.87%. Therefore, we can conclude that there is no universal use of one single primer, as it might not be suitable for each particular variety and can ultimately result in the seed quality decrease.

Key words: germination energy, priming, seed aging, soybean.

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Introduction

The maximum potential of germination and vigour of soybean seed is accomplished in the physiological maturity phase, while during aging, this value decreases (Balešević-Tubić and Miladinović, 2014). The aging process is accelerated due to unfavourable conditions of storage and under the effect of high temperature and relative air humidity (Dolatabadiand et al., 2008). During aging, unsaturated fatty acids, components of the lipid, turn into free radicals under the effect of enzymes and atmospheric oxygen. The changes which occur affect permeability of the cell membranes, which leads to loss of seed vitality (Vimala and Pratap, 2014). Aging has a harmful effect on enzymes which are essential for nutrition of the embryo and obtaining normal seedlings (Iqbal et al., 2002). The application of a particular solution for seed priming can encourage biological activity and hence increase germination capacity (Schopfer et al., 2001). Priming leads to the partial hydration of seed up to the moment when germination processes are initiated, but are not finished (Basra et al., 2005). This treatment increases seed performances, and enables quicker and more uniform germination and emergence (Berhanu and Gebremedhn, 2013). Furthermore, it ensures an optimal flow of molecular and biological processes during germination, stimulates activation of different enzymes, mobilises protein reserves and prepares cells for division (Soleimanzadeh, 2013). Useful effects of priming are related to nucleic acid repair and development, increased protein synthesis, and repair of cell membranes and mitochondria (McDonald, 2000). Likewise, seed antioxidant mechanism is recovered (Siri et al., 2013).

Priming has a practical agronomic significance, primarily in unfavourable environmental conditions, such as a high content of salt in the soil (Foti et al., 2008), low and high temperature (Wahid and Shabbir, 2005), etc. The application of this measure successfully improves seed germination of diverse plants, especially vegetables and grasses (Arif et al., 2007). Useful effects of immersion have also been determined in other field crops, such as sugar beet (Sadeghian and Yavari, 2004), barley (Abdulrahmani et al., 2007), and rape seed (Ghassemi-Golezani et al., 2010).

There is little information in the literature about the effect of priming on germination energy of naturally aged seeds. Therefore, the aim of this research was to examine how soybean seeds of a particular variety and age react to priming, as well as to determine which primer gives the best results.

Materials and Methods

The experiment was carried out in 2015 at the Institute of Field and Vegetable Crops in Novi Sad. Testing was conducted on six soybean varieties from three maturity groups: Galina and NS Princeza (0), Sava and NS Apolo (I), and Rubin and NS Zita (II). One variety from each group was chosen, which had already been widely produced, while the other variety was new in the assortment. Three-, two- and one-year-old seeds were produced in the experimental fields of PSS Ruma in 2012, 2013, and 2014, with variations in annual agro-meteorological conditions. The seeds were stored in a commercial seed storing facility, in which storage conditions depended on environmental factors.

The following treatments were used for priming:

1. Distilled water – hydropriming (H_2O);
2. Ascorbic acid 250 mg/l (ASA);
3. Folic acid 15 mg/l (Fol);
4. Hydrogen peroxide 1% (H_2O_2);
5. Control – non-treated seeds (C).

After six hours of priming, seeds were dried until they reached 11% of moisture content. After that, 4x50 seeds were taken from each variant and tested using standard laboratory methods at 25 °C in the period of five days (ISTA, 2009).

The obtained results were statistically processed by the analysis of variance of a trifactorial split-split-plot experiment (A – variety, B – seed age, C – priming). Data were processed using the analytics software package Statistica 8, while results were sorted on the basis of the Duncan's test for the significance threshold of 5% (Hadživuković, 1991).

Results and Discussion

The obtained results showed that the effect of seed priming, as a pre-sowing treatment, depended on variety, seed age, and the applied treatment. Some varieties reacted very well to this pre-sowing practice, while others had an inhibited reaction, thus significantly reducing germination energy. The variety Rubin had the most favourable reaction, whereby all treatments led to the increase of quality, by an average of 2–8%. The application of this pre-sowing practice led to significant differences in germination energy, even within the same maturity group. The smallest difference was determined within the 0 maturity group. Varieties Galina and NS Princeza reacted well to the use of all primers, except to distilled water, i.e., hydropriming. On average, the largest effect was accomplished with ascorbic acid which increased germination energy by 6%, i.e., by 4%. The varieties of the I maturity group, Sava and NS Apolo, mostly reacted to germination energy. A decrease of germination energy by 4–8% was observed in the variety Sava, while NS Apolo reacted only to the application of ascorbic acid (Table 1).

The greatest difference in reaction was determined within the II maturity group. While Rubin had the most favourable reaction to priming, particularly to the application of ascorbic acid, and both hydrogen peroxide and folic acid as well, this practice led to a decrease in quality of NS Zita by 5–7%. Examining a wheat sample, Dezfuli (2008) determined that the effect of priming on seed germination mostly depended on the genotype. Miladinov et al. (2014) determined different responses to priming by the seed of the same genotype but from different soybean lots.

Table 1. The priming effect on soybean seed germination energy (%).

Variety (A)	Seed age (B)	Treatment (C)					Mean (A*B)
		H_2O	ASA	Fol	H_2O_2	C	
Galina (0)	Three-year-old	21	28	28	29	22	27*
	Two-year-old	65	77	75	70	70	72
	One-year-old	95	98	96	96	94	96
Mean (A*C)		60	68*	66*	65*	62	
NS Princeza (0)	Three-year-old	15	21	20	21	16	19*
	Two-year-old	64	72	69	66	66	68
	One-year-old	92	95	93	92	92	93
Mean (A*C)		57	62*	61*	60	58	
Sava (I)	Three-year-old	23	27	22	21	30	23*
	Two-year-old	78	72	68	70	78	72*
	One-year-old	88	90	88	89	92	89
Mean (A*C)		63*	63*	59*	60*	67	
NS Apolo (I)	Three-year-old	23	37	34	24	32	30
	Two-year-old	71	81	81	69	78	76
	One-year-old	91	94	93	92	91	93
Mean (A*C)		62*	71*	69	62*	67	
Rubin (II)	Three-year-old	25	31	28	30	24	29*
	Two-year-old	77	86	82	81	76	82*
	One-year-old	93	95	92	93	90	93*
Mean (A*C)		65	71*	67*	68*	63	
NS Zita (II)	Three-year-old	29	35	30	32	38	32*
	Two-year-old	74	70	70	74	79	72*
	One-year-old	86	88	87	87	90	87
Mean (A*C)		63*	64*	62*	64*	69	
Mean(C)		62	67*	64	63	64	

*significant difference.

$LSD_{0.05}$	Treatment	Variety x Seed age	Variety x Treatment
	2.54	2.87	2.83

The largest effect was accomplished with the use of ascorbic acid by 3%. Ascorbic acid is also an important metabolite included in many cell processes. First of all, it participates in cell division (De Gara et al., 2003). The exogenous application of ascorbic acid may affect a set of different processes in plants including seed germination, since it increases its absorption in different tissues, and participates in biosynthesis of other hormones like gibberellic acid and ethylene, which are necessary in the germination process (Arrigoni and Detullio, 2000). Stasolla and Yeung (2001) determined the use of large quantities of ascorbic acid in the early stages of seed germination.

The reaction of a variety to the application of this pre-sowing treatment also depends on seed age. The use of primers on one-year-old soybean seeds had a weaker effect than on the two-or three-year-old seeds. On the other hand, fewer oscillations in the quality of one-year-old seeds were caused by priming. The application of primers can increase or decrease germination energy in one-year-old seeds by 3%, while this value reaches up to the 7% increase as seen in two- and three-year-old seeds, depending on the variety.

Three-year-old seed varieties Galina and NS Princeza achieved the 5% and 3% increase of germination energy, while there was no significant increase in the quality of two- and one-year-old seeds. The varieties from the I maturity group had an inhibited reaction to this practice in terms of seed quality, regardless of its age. The largest 7% decrease in the quality was noted in the three-year-old seeds of the variety Sava. The variety Rubin reacted positively in all cases, but germination energy in one-year-old seeds had a lower increase than in two- and three-year-old seeds. Unlike the variety Rubin, the application of this practice on the variety NS Zita resulted in a decrease of the quality by 3 to 7%. Improvement of seed quality depends on the level of deterioration, and the increase of germination energy can be achieved by priming seed into a particular solution, but only to a very limited extent (Wattanakulpakin et al., 2012).

Results showed that immersing seeds into ascorbic acid had a positive effect, while the application of other primers did not have any effects on the germination energy value, nor did it lead to the decrease of its value (Table 2).

Table 2. The effect of treatment and interaction with seed age on soybean seed germination energy (%).

Seed age (B)	Treatment (C)					Mean (B)
	H_2O	ASA	Fol	H_2O_2	C	
Three-year-old	22.67*	29.83*	27.00	26.17	27.00	26.53*
Two-year-old	71.50*	76.33*	74.17	71.67*	74.50	73.63*
One-year-old	90.83	93.33*	91.50	91.50	91.50	91.73*

*significant difference.

	Seed age	Seed age x Treatment
$LSD_{0.05}$	10.37	2.33

Ascorbic acid had the largest effect on the increase of the quality of three-year-old soybean seeds by 2.83%, while the increase of two- and three-year-old seed quality was the same – 1.87%. The application of this practice on onion showed a higher increase of germination in older seeds (Patil and Manjare, 2013), while the best results were achieved in treatments of fresh seeds of vetch and sunflower (Karta et al., 2011). It was determined that the application of ascorbic acid and hydrogen peroxide on aged sunflower seeds can increase germination even by 12.23% (Dolatabadian et al., 2008). The same authors determined that the application of ascorbic acid on safflower seeds did not lead to positive results, while the application of hydrogen peroxide resulted in the increase of seed quality, since hydrogen peroxide acidifies the inhibitor which is found in seed coat and thus increases germination energy. The application of hydrogen peroxide on wheat had a stimulative effect on seed germination, since it increases the activity of peroxidase enzyme (Liheng et al., 2009).

Conclusion

The results showed that the effect of this pre-sowing practice depended on variety, seed age, and the applied treatment. Some varieties reacted very well to priming, while others had an inhibited reaction, so the seed quality was significantly reduced. Hence, it was observed that the application of one solution on one variety affected the increase in the quality of some parameters, while it had an inhibitory effect on the quality of others. Therefore, we can conclude that there is no universal use of one single primer, as it might not be suitable for each particular variety and can ultimately result in the seed quality decrease.

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UTICAJ POTAPANJA SEMENA SOJE NA ENERGIJU KLIJANJA

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R e z i m e

Da bi se ispitao efekat potapanja na energiju klijanja starog semena soje, za analizu je odabrano šest sorti iz tri različite grupe zrenja: Galina i NS Princeza (0 grupa), Sava i NS Apolo (I grupa), Rubin i NS Zita (II grupa zrenja). Seme je proizvedeno u periodu od 2012. do 2014. godine, odnosno seme starosti tri, dve i jednu godinu. Za potapanje su korišćeni sledeći tretmani: destilovana voda (H_2O), askorbinska kiselina 250 mg/l (ASA), folna kiselina 15 mg/l (Fol.), vodonik-peroksid 1% (H_2O_2) i kontrola – netretirano seme (C). Rezultati su pokazali da efekat potapanja zavisi od sorte, starosti semena, ali i tretmana koji se koristi. Neke sorte veoma dobro reaguju na potapanje, dok kod drugih dolazi do inhibitornog delovanja, pa se energija klijanja semena značajno smanjuje. Sorta Rubin je najbolje reagovala i jedino su kod ove sorte svi tretmani doveli do povećanja kvaliteta, u proseku od 2% do 8%. Primena prajmera kod jednogodišnjeg semena soje ostvarila je slabiji efekat nego kod dvogodišnjeg i trogodišnjeg semena. Međutim, kod jednogodišnjeg semena manje su oscilacije u kvalitetu usled potapanja. Kod jednogodišnjeg semena dejstvom prajmera energija klijanja može da se poveća odnosno smanji za 3%, dok se kod dvogodišnjeg i trogodišnjeg ta vrednost kreće do 7%. Rezultati pokazuju da je potapanje semena u askorbinsku kiselinu ostvarilo pozitivan efekat, dok primena ostalih prajmera nije uticala na vrednost energije klijanja ili je dovela do pada njene vrednosti. U proseku, dejstvom askorbinske kiseline najviše je došlo do povećanja kvaliteta trogodišnjeg semena soje – 2,83%, dok je povećanje dvogodišnjeg i trogodišnjeg bilo isto i iznosilo je 1.87%. Zbog toga se ne može govoriti o univerzalnoj primeni samo jednog prajmera, jer se može dogoditi da on ne odgovara određenoj sorti, ali i starosti semena, pa može doći do pada kvaliteta semena.

Ključne reči: energija klijanja, potapanje, starenje semena, soja.

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EFFECTS OF THE ENVIRONMENTAL CHARACTERS ON GERMINATION PROPERTIES OF SEEDS OF *THYMUS DAENENSIS* AND *T.VULGARIS*

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Abstract: This research was conducted to investigate the effects of salinity, temperature, pH and sowing depth on germination characters of two species of thyme in 2017. Two separate experiments with four replications using a randomized complete design were conducted in laboratory conditions. Treatments contained: a) fluctuating temperatures in a germinator 5/15, 10/20, 15/25, 15/30 and 20/35^{0}C (day/night), b) concentrations of salinity by using solutions of 0, 10, 20, 40, 80, 160 and 320 mM NaCl, c) sowing depths of 0, 1.5, 3 and 6 cm, pH values of 5, 6, 7, 8 and 9. Results showed that the effects of different treatments were significant on germination percentage, germination rate and seed vigour. In most cases, upper concentrations of treatments had negative effects on germination indices. The emergence decreased with an increased concentration of salinity, planting depth and pH. In both species, the highest germination percentage (94%) was obtained in the combination treatment of 15/25^{0}C (day/night), 0 mM of NaCl, 1 cm of planting depth and pH=7. Germination was stopped in the combination of treatments of 35/20 (day/night), 320mM of NaCl, 6 cm of planting depth and pH=9. The combination of treatments in the upper level had a more inhibitory and destructive effect than single treatments.

Key words: pH, salinity, sowing depth, temperature, thyme.

Introduction

Thyme, one of the most important spices, is used all over the world, and includes many species. Two of the most important commercially grown species are *Thymus vulgaris* L. and *T. daenensis* Celak, members of the family Lamiaceae. *Thymus* species are commonly used as herbal teas, flavouring agents (condiments and spices) and medicinal plants (Hudaib and Aburjai, 2007). The major components are phenols (mainly thymol and carvacrol), monoterpen hydrocarbon and alcohol that have insecticidal activity. Among these, thymol and carvacrol are the main compounds (Yadegari, 2015a). Thyme species are commonly used as

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flavouring agents and medicinal plants (Chauhan and Johnson, 2008; Yadegari, 2017a, b). Seeds of this plant have no dormancy (Yadegari, 2015b). Thyme morphological diversity can affect ecological factors of plant competition, time of germination, flowering time and genetic effects (Corticchiato et al., 1998). Germination is a key to the success of plants in agro-ecosystems (Chauhan and Johnson, 2008; Keller and Kollmann, 1999; Hubbard et al., 2010). The affecting factors on germination and emergence are temperature, osmotic pressure of the solution, the position of the seed in the soil seed bank and soil texture. Temperature is an important environmental factor regulating germination (Ren et al., 2002). Seed germination and emergence belong to the depth in the soil. Emergence of seedling reduces more than optimum depth. The effect of pH on germination potential varied in different plants. Seed germination, in particular, appears to be extremely sensitive to soil salinity (Amiri et al., 2012). Field salinization is a growing problem worldwide and it is a major abiotic stress reducing the yield of a wide variety of crops all over the world (Bourgou et al., 2012). Some species require more acidic conditions, and some prefer alkaline or neutral pH, while some others do not show any reactions (Susko et al., 1999). Germination is a critical stage in the life cycle of plants, and often controls population dynamics, with major practical implications (Keller and Kollmann, 1999). Overall germination events are regulated by several environmental factors such as temperature, salinity, pH and moisture (Hubbard et al., 2010). There are few studies examining environmental characters on germination properties of seeds of thyme. Objectives of this study were to determine the effects of environmental characters (salinity, temperature, pH and sowing depth) on germination properties of seed germination of *T. daenensis* and *T.vulgaris* for better establishment in rangelands.

Materials and Methods

Laboratory experiment and conditions

To study the effects of salinity, temperature, pH and sowing depth on germination characters of two species of thyme, two separate experiments with four replications using a randomized complete design in a factorial layout were carried out. This study was conducted in 2017 in the Center of Medicinal and Aromatic Plants of Islamic Azad University Branch of Shahrekord in laboratory conditions. Seeds of thyme species were collected naturally from mountains of Chaharmahal and Bakhtiari province (Tables 1and 2).

Treatments contained: a) fluctuating temperatures in a germinator 5/15, 10/20, 15/25, 15/30 and 20/35^{0}C night/day; b) concentrations of salinity by using solutions of 0, 10, 20, 40, 80, 160 and 320 mM NaCl; c) sowing depths of 0, 1.5, 3 and 6 cm, and d) pH of 5, 6, 7, 8 and 9 (Chauhan and Johnson, 2008). Seeds were placed on two moistened paper towels. After covering the seeds with a third sheet of paper, the three towels were loosely rolled to form a tube and placed in plastic bags (23×33 cm) to prevent evaporation. Seeds were observed twice daily and considered germinated when the radicle was approximately 2 mm in length (Soltani et al., 2001).

Table 1. Geographic and climatic properties in collection sites.

Average minimum temperature	Average maximum temperature	Average annual temperature	Average annual rainfall	Elevation	Geographic properties	Mountain
	^0C		Mm	meter		
-1.4	24.5	12.1	516.9	3670	32010/N-50030/E	Saldaran
-4.5	17.5	9.5	875.5	3904	320 5/N-16051/E	Mili
4	27.8	18.4	500	3640	31, 82 N-50, 96 E	Kallar

Table 2. Soil properties in collection sites.

Elevation	Mountain	Texture	pH	O.C	EC	B	Mn	Fe	Cu	Zn	N	K	P
				%	ds/m	mg/lit							
2500	Saldaran	Clay loam	7.76	1.65	0.361	1.12	8.37	3.62	0.94	0.81	0.1	254	12.7
2500	Mili	Clay	7.12	2.2	0.38	1.3	11.1	3.8	0.55	0.52	0.3	355	19.1
2500	Kallar	Clay loam	7.93	2.85	0.481	1.23	9.38	5.32	1.02	0.77	0.311	291	34.6

Analysis of morphological traits

To evaluate the potential salinity on seed germination reduction, the three parameters of x, x50 and b were used (Chauhan and Johnson, 2008):

$$Y=a/ [1+(x/x50)^{b}] \quad \text{Eq. (1)}$$

Y: germination at the salinity level of X (%), a: maximum germination (%), X50: the salinity level required for 50% inhibition of maximum germination and b: a slope represents reduced germination by increasing salinity. Seed percentage, germination percentage and seed vigour were measured by the following equations (Chachalis and Ready, 2000):

S.P= X1 /Y1 + (X2-X1)/Y2 + …. + (Xn-Xn-1)/Yn Eq. (2)

G.P = Number of germinated seeds/ Number of total seeds Eq. (3)

S.V = Mean of length of seedling × G.P (measured with a calliper) Eq. (4)

Quality of treatments

A 2-mM potassium hydrogen phthalate buffer solution was adjusted to pH 4 with 1 N HCl. A 2-mM solution of MES [2-(N-morpholino) ethane sulfonic acid] was adjusted to pH 5 or 6 with 1 N HCl or NaOH. A 2-mM solution of HEPES [N-(2-hydroxymethyl) Piperazine-N ’-(2-ethanesulfonic acid)] was adjusted to pH 7 or 8 with 1 N NaOH. pH 9 buffer was prepared with 2-mM tricine [N-Tris (hydroxymethyl) methyl glycine] and adjusted with 1 N NaOH. Non-buffered deionized water (pH 6.3) was used as control. In glass bottles with height of 15 cm, the effects of seed sowing depths on plant seedling emergence were studied.

Statistical analysis

After the normality and homogeny test of variance, the logarithmic transformation of data was done and then all data were subjected to ANOVA using the statistical computer package SAS ver.8 and treatment means were separated using the L.S.D multiple range test at the $P<0.05$ level.

Results and Discussion

The effects of treatments on germination rate, germination percentage and seed vigour in two species were significant (Tables 3 and 4). In single treatments, the highest percentage of germination temperature was in the treatment of 15/25 (day/night) with 91% (Figure 1). Salinity was significant in measured parameters. The highest percentage of germination in seeds of *T. vulgaris* and *T. daenensis* was in 0 mM treatment amounting to 91% and 94% respectively. There was no germination in both of species regarding the salinity of 320 mM sodium chloride. Decreased 50% germination occurred at the salinity of 160 mM sodium chloride (Figure 2). The percentage and germination rates of both species of thyme seeds were influenced by acidity. The highest and lowest germination was achieved at pH 7 (94%) and pH 9 (19%) respectively (Figure 3). Seedling emergence was affected by planting depth so that significant differences were observed at various depths. The statistic groups of emergence at the soil surface (86%) and at the depth of 1.5 cm (84%) were the same (Figure 4). Temperature had a significant effect on seed germination. This represents the tolerance of the plant in the different temperature ranges. This is because the mentioned plant in the Zagros Mountain, able to adapt to different temperatures and with germination in a relatively wide temperature range, guarantees its survival. Light and temperature are two environmental factors that impact on multiple levels of germination. The highest germination rate was observed in the treatment combination of 15/25 (day/night), 0mM of NaCl, 1cm of planting depth and pH=7. Alternating temperatures increase germination of seeds (Martinez-Ghersa et al., 2003).

Table 3. Analysis of mean of variances for effects of salinity, temperature, pH and sowing depth on germination characters of *Thymus daenensis* and *T. vulgaris* in the first step.

Source	d.f	G.R	S.V	G.P	G.R	S.V	G.P
		Thymus daenensis			*T. vulgaris*		
Salinity (A)	6	12.2^{**}	35.5^{**}	64.4^{**}	14.5^{**}	48.98^{**}	55.32^{**}
Temperature (B)	4	8.8^{**}	24.3^{**}	42.2^{**}	9.14^{**}	35.78^{**}	27.1^{**}
pH (C)	4	7.2^{**}	23.3^{**}	63.3^{**}	14.5^{**}	37.98^{**}	29.89^{**}
Sowing depth (D)	3	15.6^{**}	47.7^{**}	77.8^{**}	14.9^{**}	55.98^{**}	114.2^{**}
A× B	24	1.5^{*}	3.95^{*}	18.76^{**}	1.37^{*}	5.03^{*}	24.5^{**}
A× C	24	1.54^{*}	4.01^{*}	6.62^{*}	1.45^{*}	4.99^{*}	45.76^{**}
A× D	18	1.52^{*}	4.19^{*}	7.02^{*}	1.41^{*}	5.63^{*}	117.8^{**}
B× C	16	2.7^{**}	4.25^{*}	7.12^{*}	4.51^{**}	5.71^{*}	8.44^{*}
B× D	12	3.5^{**}	4.56^{*}	7.63^{*}	6.97^{**}	6.15^{*}	9.2^{*}
C × D	12	4.4^{**}	5.01^{*}	7.55^{*}	12.7^{**}	6.18^{*}	9.43^{*}
A× B ×C	96	0.94^{*}	2.58^{*}	4.32^{*}	0.89^{*}	3.48^{*}	5.14^{*}
A× B× D	72	1.14^{*}	3.16^{*}	5.29^{*}	1.06^{*}	4.26^{*}	6.31^{*}
A× C× D	72	1.16^{*}	3.2^{*}	5.15^{*}	1.02^{*}	4.29^{*}	6.37^{*}
B × C× D	48	1.265^{*}	3.49^{*}	5.85^{*}	1.14^{*}	4.71^{*}	6.99^{*}
A× B ×C× D	288	0.0045^{ns}	0.0005^{ns}	0.024^{*}	0.00056^{ns}	0.021^{*}	0.03^{*}
Error	210	1.1	3.04	5.09	0.99	4.08	6.03
C.V		11.1	14.5	8.7	7.4	6.5	5.9

Maximum germination in seeds was performed at red light and alternating temperatures (Tang et al., 2008). Annual seeds sprout in late spring or early summer to complete their life cycle. Seeds of plants must be exposed to high temperatures in the summer months (Baskin and Baskin, 1998). In *Phalaris arundinacea* better germination was observed in 16 hours of lighting (Lindig-Cisneros and Zedler, 2001), but in another study it has been reported that the best temperature was 20° C as well as light regime of 12 h in dark conditions (Kon et al., 2007). The germination and maturity responses to latitude, elevation, soil moisture, soil nutrients, temperature and vegetation density of habitat destruction are different (Baskin and Baskin, 1998). Germination and seedling growth can be reduced by some non-living factors such as salinity and drought, which are the most important abiotic stresses to limit the number of seedlings and seedling growth (Atak et al., 2006; Kaya et al., 2006). Salinity is an important factor in the peripheral condition that threatens the sustainability of arid and semiarid regions, especially in areas where evapotranspiration is greater than precipitation (Szabolcs, 1994). High salinity usually decreases the rate and extent of germination. Salinity inhibits germination of seeds by reduced water availability or interferes with some aspects of metabolism like changing the balance of growth regulators. In canary grass, the highest percentage of germination (96.5 %) in the control treatment was observed and in salinity of 320 mM sodium chloride, germination was stopped (Ahmadi et al., 2013). In this research, fresh weight and dry mass yield of plants slightly decreased as the salinity increased. The highest tolerance to salinity in the seeds of the medicinal herbs of *Langematia iberica, Plantago major, Anethum graveolens, Cuminum cyminum, Trifolium subterraneum, Trachyspermum ammi, Origanum majorana L., Lactuca sativa, Sesamum indicum, Trigonella foenum, Alyssum desertorum,* and *Portulaca oleracea* L. was in *Portulaca oleracea L., Alyssum desertorum*, and *Trigonella foenum*, and it was up to 450 mM/l while the lowest tolerance was reported in *Plantago major, Langematia iberica*, and *Anethum graveolens* (Yadegari, 2015b). In a similar study on *Linum usitatissimum* L., *Echinacea angustifolia, Carthamus tinctorius* L., and *Cynara scolymus* L., the highest salinity stress tolerance was observed in the seeds of *Linum usitatissimum* L. and *Carthamus tinctorius* L. at the germination stage (Gholizadeh et al., 2016). A response to several salinity concentrations depends on many reasons, for example species of plant and osmotic regulation. Yield and biomass reductions are very common under salt stress conditions, especially for salt-sensitive crops, due to osmotic effects and ionic imbalances (Bannayan et al., 2008; Lattanzio et al., 2009; Myung et al., 2009). Ahmadi et al. (2013) found similar results in a study on germination of *Phalaris minor*. Percentage and germination rates of seeds of canary grass were influenced by acidity (Ahmadi et al., 2013). The most important effect of pH is the availability of nutrients in soil. The elements such as calcium, phosphorus and potassium are leached or insoluble in low pH, and on the other hand, the elements such as iron, manganese and other micro-nutrients are unavailable in high pH (Yadegari, 2017a, b). Most studies about the effect of depth on seedling emergence of plants showed that by increasing of sowing depth, emergence of seedlings reduced (Benvenuti, 2003; Mohler, 2001). Yield and biomass reductions are very common under salt stress conditions, especially for salt-sensitive crops, due to osmotic effects and ionic imbalances (Attia et al., 2011). Biological reasons for the lack of germination have not been specified yet completely. The seedling emergence in different depths depends on the seed energy reserves (Ren et al., 2002).

Table 4. Analysis of mean of variances for effects of salinity, temperature, pH and sowing depth on germination characters of *Thymus daenensis* and *T. vulgaris* in the second step.

Source	d.f	G.R	S.V	G.P	G.R	S.V	G.P
		Thymus daenensis			*T. vulgaris*		
Salinity (A)	6	42.2**	57.7**	89.9**	55.87**	77.93**	77.19**
Temperature (B)	4	12.8**	44.3**	66.8**	89.54**	65.98**	34.65**
pH (C)	4	14.2**	88.5**	89.86**	44.35**	87.65**	44.73**
Sowing depth (D)	3	19.6**	66.7**	82.76**	33.65**	63.78**	69.77**
A× B	24	15.5**	14.78**	45.65**	5.44*	6.78*	64.78**
A× C	24	14.78**	6.6*	9.43*	5.51*	6.81*	77.94**
A× D	18	1.62*	7.05*	9.95*	5.82*	7.22*	93.75**
B× C	16	5.5**	7.15*	10.22*	5.92**	7.33*	11.84*
B× D	12	7.7**	7.62*	10.85*	6.35*	7.89*	12.95*
C × D	12	12.4**	7.65*	11.16*	44.66**	7.85*	12.94*
A× B ×C	96	0.99*	4.38*	6.17*	3.63*	4.45*	6.98*
A× B× D	72	1.23*	5.36*	7.55*	4.41*	5.51*	8.56*
A× C× D	72	5.8**	5.42*	8.05*	4.47*	5.58*	8.51*
B × C× D	48	1.33*	5.92*	8.31*	4.77*	6.021*	9.34*
A× B ×C× D	288	0.0009ns	0.000075ns	0.00024ns	0.0000081ns	0.026*	0.042*
Error	2100	1.21	5.08	7.17	4.156	5.19	8.16
C.V		9.93	8.18	7.76	9.14	5.8	4.42

ns: Not significant, * and ** : Significant at P ≤ 0.05 and P ≤ 0.01 levels respectively. (G.P: germination percentage, G.R: germination rate, S.V: seed vigour).

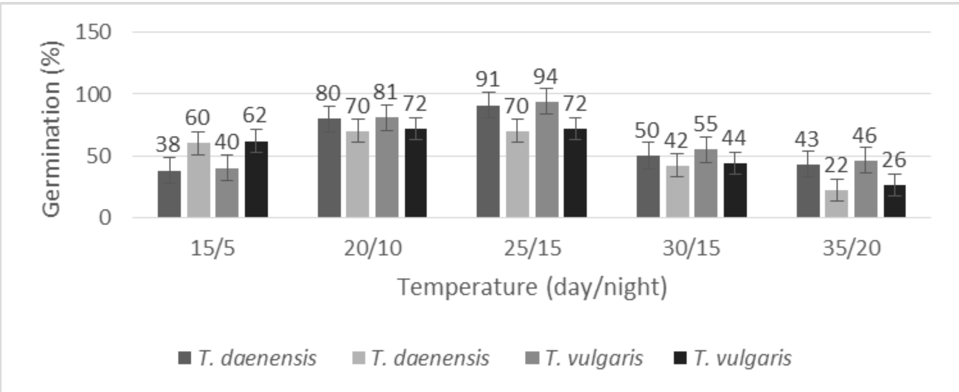

Figure 1. The effect of alternating temperatures (day/night) on *T. daenensis* and *T. vulgaris* seed germination.

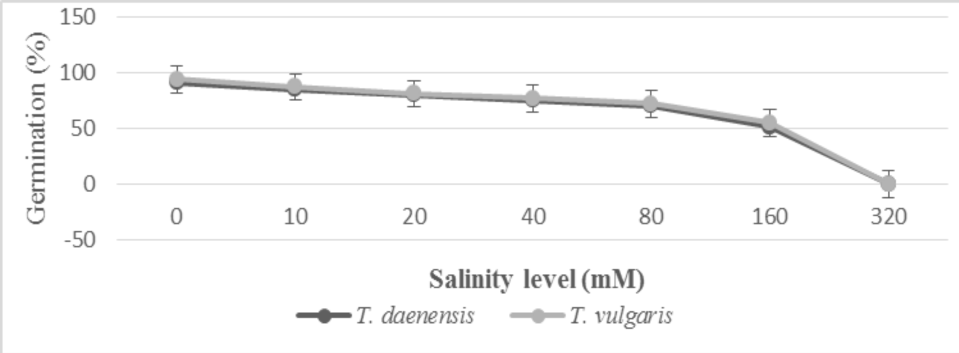

Figure 2. Final germination percentage of *T. daenensis* and *T. vulgaris* under different salt stresses.

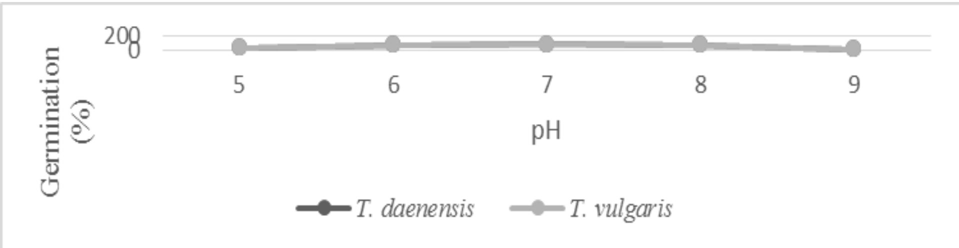

Figure 3. Germination percentage of *T. daenensis* and *T. vulgaris* under various pHs and temperatures 15°/25° C (day/night).

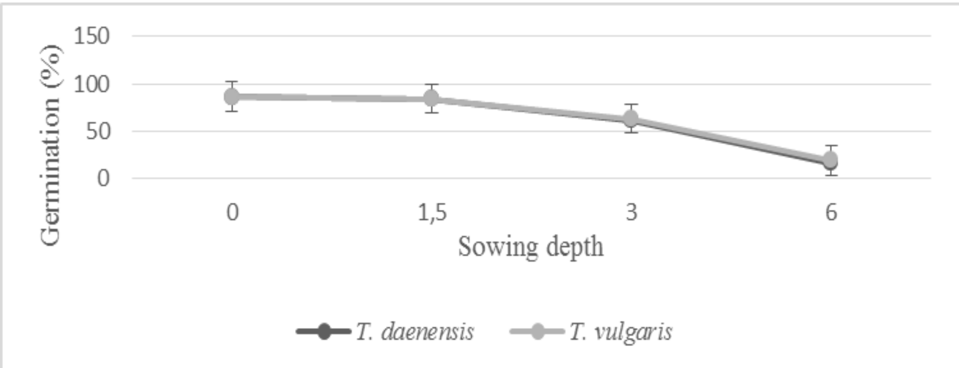

Figure 4. Germination percentage of *T. daenensis* and *T. vulgaris* under various sowing depths and temperatures 15°/25°C (day/night).

A high concentration of NaCl in lettuce in nutrient solution strongly affected the germination rate and root elongation, seedling and mature vegetative growth of both sesame and lettuce (Myung et al., 2009). By increasing of salinity levels, seedling emergence of medicinal plants decreased. Responses to several salinity concentrations depended on many reasons. In addition, seed germination and seedling emergence are influenced by moisture availability, temperature and light levels (Chauhan and Johnson, 2008). Enhanced radiation decreased plant height, dry weight of individual stem and yield per plant, plant growth and development, photosynthesis and biomass production (Liu et al., 2013; Choudhary and Agrawal, 2014). With the increase in levels of drought stress by increasing of sowing depth, seed accessing to water was reduced. In this way, it is possible that the germination percentage will be reduced (Ansari et al., 2012). Generally, with the increase of drought stress, the ability of suction of water by seeds will be decreased and the necessary duration for water sucking will be increased and consequently the start of germination processes will be postponed (Ghaderi et al., 2010). Destructive effects of salinity levels and a decrease of growth parameters in *Thymus broussonetii* Boiss (Belaqziz and Romane, 2014), *Nigella sativa* (Bourgou et al., 2012), *Suaeda maritime* (Gazala et al., 2013), *Artemisia annua* L. (Irfan Qureshi et al., 2013), *Schinopsis quebracho* (Meloni et al., 2008), *Carthamus tinctorius* L. (Salem et al., 2014) and *Capsicum* (Patade et al., 2011) were previously reported.

Conclusion

Treatments with superior levels of salinity, pH, sowing depth and temperature had more negative effects on germination characters than other treatments. Emergence decreased with an increased concentration of salinity, planting depth and pH. In two species, the greatest germination percentage was made by combination of treatment of 15/25 (day/night), 0mM of NaCl, 1cm of planting depth and pH=7. The least germination percentage was produced by the combination of treatments of 35/20 (day/night), 320mM of NaCl, 6cm of planting depth and pH=9. This study provides some useful information about the efficacy of environmental effects (salinity, temperature, pH and sowing depth) on germination characters of two species of thyme. These methods are relatively new and need further improvement with regard to rates, timing, and techniques.

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UTICAJ FAKTORA ŽIVOTNE SREDINE NA KARAKTERISTIKE KLIJAVOSTI SEMENA *THYMUS DAENENSIS* I *T.VULGARIS*

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R e z i m e

Ovo istraživanje je sprovedeno radi ispitivanja uticaja saliniteta, temperature, pH i dubine setve na karakteristike klijavosti dve vrste majčine dušice 2017. godine. Tretmani su sadržavali: a) promenljive temperature u klijalištu 5/15, 10/20, 15/25, 15/30 i 20/35 0C (dan/noć), b) različite koncentracije rastvora soli 0, 10, 20 , 40, 80, 160 i 320 mM NaCl, c) dubinu setve od 0, 1,5, 3 i 6 cm, pH vrednosti od 5, 6, 7, 8 i 9. Rezultati su pokazali da su efekti različitih tretmana bili značajni na procenat klijavosti, stopu klijavosti i vigor semena. U većini slučajeva, najveće koncentracije svih tretmana su imale negativan efekat na karakteristike klijavosti. Klijavost se smanjivala sa povećanom koncentracijom soli, dubinom setve i pH vrednosti. Kod obe vrste, najveći procenat klijavosti (94%) postignut je kombinovanim tretmanom 15/25^{0}C (dan/noć), 0 mM NaCl, 1 cm dubine setve i pH=7. Klijanje je zaustavljeno u kombinaciji tretmana 35/20 (dan/noć), 320 mM NaCl, 6 cm dubine setve i pH=9. Pomenuta kombinacija tretmana imala je jači inhibitorni i destruktivni efekat od pojedinačnih tretmana.

Ključne reči: pH, salinitet, dubina setve, temperatura, majčina dušica.

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PHYSICO-CHEMICAL CHARACTERISTICS OF POMEGRANATE ACCESSIONS FROM THE KURDISTAN REGION, IRAQ

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Abstract: Some physico-chemical properties of ten pomegranate accessions collected from different districts in the Kurdistan region of Iraq were investigated in this paper. Considerable correlations between the characteristics studied were found and valuable pomological traits were observed. Cluster analysis showed the homonyms between some pomegranate accessions. Principle component analysis reported that the component describing the greatest variability also positively correlated with fruit weight, total aril weight, total peel weight, volume of juice, total soluble solids (TSS), fruit length, fruit diameter, pH, aril length, and 100-seed fresh weight, but negatively correlated with titratable acidity (TA). Fruit weight was firmly correlated with total aril weight, total peel weight, volume of juice, TSS, aril length, 100-seed fresh weight, fruit length and fruit diameter. The volume of juice was correlated with TSS, aril length, 100-seed fresh weight, fruit length, fruit diameter and it was observed that with an increase in the fruit size, the volume of juice increased as well. The correlation between total phenolic compounds and antioxidant capacity was not observed. The associations found among physical and chemical traits suggest that consumers should use large fruits with large arils so that they have more juice. Thus, 'Choman', 'Raniyeh' and 'Halabja' were juicier than other accessions.

Key words: physical properties, accessions, fruits, antioxidants, phenolic compounds.

Introduction

Pomegranate (*Punica granatum* L.) has a highly distinctive fruit and it is the individual from two species belonging to the *Punicaceae* family (LaRue, 1980). *Punica granatum* L. *(Punicaceae)* is one of the oldest domesticated fruit trees that people have been consuming for a long time. With respect to numerous organic product species, pomegranate fruit is a significant natural product for human

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wellbeing as a result of its rich cancer prevention activity and high polyphenol content (Hernandez et al., 1999). The most plentiful phytochemicals in pomegranate juice are polyphenols, including the hydrolyzable tannins called ellagitannins framed when gallic acid binds with a sugar to shape pomegranate ellagitannins, otherwise called punicalagins (Singh et al., 2002). The red color of juice can be ascribed to anthocyanins, for example, delphinidin, cyanidin, and pelargonidin glycosides (Hernandez et al., 1999). For the most part, an expansion in juice pigmentation happens in natural product aging (Hernandez et al., 1999). Contrasted with the pulp, the unpalatable pomegranate peel contains as much as three times of the aggregate sum of polyphenols (Singh et al., 2002), including condensed tannins (Ben-Nasr et al., 1996), catechins, gallocatechins and prodelphinidins (Morton, 1987).

The pomegranate tree normally develops up to 4–6 m, and has numerous spiked branches, as proven by trees at Versailles, France, known to be more than 200 years of age. The leaves are polished and spear molded, and the bark of the tree turns dark as the tree ages. The blooms are large, red, white, or variegated and have a tubular calyx that turns into fruits (LaRue, 1980). Pomegranate originated in the area of Iran, and has been developed since old circumstances all through the Mediterranean district and northern India. Today, it is broadly spread all through the Middle East and Caucasus district, north and tropical Africa, the Indian subcontinent, Central Asia, the drier parts of Southeast Asia, and parts of the Mediterranean Basin (Chidambara-Murthy et al., 2002).

Kurdistan is a local point of source for pomegranate and has more suitable conditions than any place where pomegranate is cultivated. Neighborhood pomegranates are various and adjusted to different natural states of Kurdistan climate. The pomegranates have been developed using traditional techniques for many years and can be used for various purposes; namely, protection from infections, irritations, cold, and dry season. Furthermore, it can be used for modern procedures like natural juice production. Accordingly, the losses in genetically arranged assortment in crop species because of commercialization have driven the need to protect the current genetic sources. The principle objectives of this study were: a) to portray and analyze pomegranate that developed in 10 locations of Kurdistan, and b) to decide the fluctuation inside characters utilized as a part of morpho-pomological and chemical compound examinations.

Materials and Methods

Plant preparation

Pomegranate fruits of accessions were harvested from pomegranate orchards located in the ‘Choman’, ‘Soran’, ‘Hewler’, ‘Sidakan, ‘Barzan, ‘Raniyeh’, ‘Halabja’, ‘Kerkuk’, ‘Harir’, ‘Balakayati’, in the Kurdistan region of Iraq. They were brought to the laboratory of the General Science Department of the Faculty of Education of the Soran University, in 2016. Ripe fresh fruits were picked from different mature trees randomly (by a completely randomized design of four trees per sample of ten fruits per replications) selected to represent the population of the plantation.

Physical properties: Harvested fruits were sorted according to their size, uniformity, shape and weight. All fruits were first flushed with tap water before the peel, pulp and seed fractions were carefully separated. The peel and the pulp were separated manually after fruit fresh weight and fruit density determination.

Fruit fresh and aril weights were determined by weighting the fruits in the air on a precision digital balance (Mettler AJ50) with an accuracy of 0.0001 g. Then peel thickness, aril and fruit lengths and diameter were measured by a digital caliper with 0.01 mm accuracy. Aril, juice and seed weights were measured as above. Fruit juice extraction content was measured manually. Then the fruit juice was analyzed for total phenols and antioxidant activity.

Chemical analysis: Total soluble solids (Brix) in the juice were determined with a digital refractometer (ATAGO RX-5000) at 20°C, calibrated using distilled water. Titrable acidity was estimated by juice titration with 0.1 N NaOH to the titration end point of pH 8.3, monitored with a pH meter (Labtron) and expressed as citric acid content (mg/100ml). For pH determinations, the samples were homogenized and measured with a pH meter (Labtron).

Total phenolic content (TPC)

TPC was determined by the Folin-Ciocalteau technique as described by Singleton et al. (1999) with minor adjustments, as indicated by colorimetric oxidation/decrease response of phenols. The polyphenol extraction was completed using 10 ml of 85% methanol added to 1g of fine powder of pomegranate. To prepare 250 µl of concentrate, 250 µl of sterile refined water was used, and after that 2.5 ml of the Folin-Cicalteau reagent and 2 ml of sodium carbonate 7.5% were added. The samples were shaken for 1.5 to 2 hours. The absorbance of tests was estimated at 765 nm by a PG Instruments T80+ UV/VIS spectrophotometer. Gallic acid was utilized for a regulation curve. Results were expressed as mg GAE/100 g FW.

Total antioxidant capacity

The antioxidant properties were estimated by the scavenging of 2, 2-diphenyl-2-picrylhydrazyl hydrate (DPPH) radicals as per Brand-Williams et al. (1995) with minor adjustments. The purple color intensity of DPPH solution decays and the change of absorbance were followed spectrophotometrically (PG Instruments ltd – T80 + UV/VIS) at 517 nm. Quickly, a 0.15 mM solution of DPPH in methanol was prepared. Then, 2 ml of this solution was added to 1 ml of methanol concentrates of pomegranate natural products. The substance of the tubes was blended and taken after to remain for 30 min and absorbances were determined at 517 nm. The antioxidant properties were communicated as the level of free radical scavenging. The inhibition rate for each sample was figured as follows:

% inhibition = 100 (A0 – Ax)/A0, where A0 is the absorbance of a DPPH blank and Ax is the absorbance of juice solution.

Statistical analysis

Investigation of variance, correlation, principle component analysis (PCA) and cluster dendograph were performed using the SAS (variant 9.2) program. LSD values were computed at $p \leq 0.01$.

Results and Discussion

According to data, differences among accessions for all the traits were significant at the 1% level ($p \leq 0.01$). Mean comparison properties (Table 1) showed that ‘Choman’ accessions reached the highest values of fruit weight, total aril weight, and total peel weight, seed fresh weight, fruit length and fruit diameter. ‘Harir’ accessions reached the highest values of peel thickness and aril length. Peel thickness varied from 1.3 mm to 3.6 mm, and the average values of peel thickness share in pomegranate fruits between 4.2% and 5.9% were reported (Martinez et al., 2012). The vast majority favor pomegranate fruits with thin peel since they have less waste and are simple to separate (Radunic et al., 2015). Also, 100-seed fresh weight, pH and TSS/TA reached the highest values in ‘Halabja’ in comparison to other accessions. The TSS/TA proportion is a quality factor that is critical for assurance of taste and harvest time. According to the results, TSS/TA values varied from 4 (‘Balakayati’) to 6.34 (‘Halabja’). The TSS/TA ratio for Italian pomegranate was in the range of 5.4 to 37.7 (Cristofer et al., 2010) and for Spanish pomegranate between 37.4 and 56.9 (Martinez et al., 2006). According to Table 1, the lowest and highest TA contents were observed in ‘Halabja’ (1.8%) and ‘Sidakan’ (2.2%) with ‘Balakayati’ (2.2%) accessions respectively. TSS varied between 8.7 and 11.3%. The volume of juice differed significantly from 34 ml (‘Soran’) to 135 ml (‘Raniyeh’). Juice quality is incredibly impacted by cultivar and type of extraction method as the extraction of juice using a blender is more different than when using mechanical methods (Rajaseker et al., 2012).

Table 1. Mean comparison of different fruit characteristics in ten pomegranate accessions.

Accessions	Fruit weight (g)	Total aril weight (g)	Total peel weight (g)	Peel thickness (mm)	Volume of juice (ml)	pH	TA (total acidity) (%)	TSS (total soluble solids) (%)
Choman	418.6a	230.67a	148.10a	3b	127.3a	3.63b	1.8de	11.1ab
Raniyeh	376.0b	198.33b	96.87bc	2.6bc	135a	3.47c	1.8cd	11bc
Halabja	361b	220.33a	97.23bc	1.3e	134a	3.79a	1.8e	11.3a
Harir	355.4bc	179.00c	110.43b	3.6a	126.6a	3.1d	2b	10.3e
Kerkuk	333.7c	110.67e	97.67bc	2.3bc	126.6b	3.6b	1.9c	10.8cd
Sidakan	254.5d	136.33d	67.67d	1.3e	91b	2.83e	2.2a	8.7g
Barzan	253.1d	121.17e	84.43c	1.8d	73.3c	3.48c	1.9c	10.9bcd
Balakayati	224.1e	78.67f	55.30d	1.3e	56.6d	2.83e	2.2a	8.9g
Hewler	191.1f	75.00f	53.93d	2.8b	57d	3.52bc	1.8e	10.7d
Soran	169.5f	59.67g	60.98d	3.6a	34e	3.6b	2b	10f
Accessions	TSS (total soluble solids) (%)	Taste index (TSS/TA)	Aril length (mm)	Aril diameter (mm)	Seed fresh weight (g)	100-seed fresh weight (g)	Fruit length (mm)	Fruit diameter (mm)
Choman	11.1ab	6.12ab	9b	4b	3.9a	6.4e	88.3a	100a
Raniyeh	11bc	6.07b	9.3ab	4.3ab	2.1bcd	7.3c	87.6ab	87.6b
Halabja	11.3a	6.34a	8.3c	4.6a	2.5b	8.8a	85.3ab	87b
Harir	10.3e	5.1d	9.6a	4.6a	1.4e	7.2c	86ab	85.3b
Kerkuk	10.8cd	5.7c	7.3e	4.3ab	1.8cde	6.7d	84.3b	85.3b
Sidakan	8.7g	3.9f	7.6de	4b	1.6de	5.9f	75.3d	76.3c
Barzan	10.9bcd	5.8c	8.3c	4.6a	1.8cde	8b	79c	75.6c
Balakayati	8.9g	4f	6f	2.66c	1.32e	4.5f	66.3f	65.6d
Hewler	10.7d	6.1ab	8cd	4.3ab	1.23f	5.2g	68.6ef	68d
Soran	10f	5d	7.6de	4.3ab	2.4bc	6.1f	70e	65.3d

Means of three replicates followed by the same letters were not statistically significant ($P \leq 0.01$).

Antioxidant capacity of the studied accessions is shown in Figure 1. According to antioxidant content, ‘Raniyeh’ and ‘Hewler’ accessions showed the highest antioxidant content (85.1% and 87.3% respectively) and ‘Choman’, ‘Harir’ and ‘Halabja’ showed the lowest (75.3%, 74.7% and 77.7%). ‘Kerkuk’, ‘Sidakan’, 'Barzan' and 'Balakayati' accessions did not show any differences in antioxidant capacity between fruits. According to total phenolic compounds (Figure 2), 'Soran', 'Sidakan' and 'Raniyeh' accessions showed the highest content of phenolic compounds (94, 95 and 90.4 mg/100g respectively) while 'Harir', 'Barzan', 'Balakayati' and 'Hewler' accessions showed the lowest content (75.1, 73.2, 74.5 and 71.5 mg/100g). It is interesting that commercial juices had higher antioxidant activity and, on the other hand, experimental juices produced by pressing the arils had a lower activity (Gil et al., 2000). Therefore, it can be presumed that the maintenance of antioxidant potential might be exceedingly affected by preparing techniques. Phenolic compounds are essential for their contribution to sensory attributes, and in addition, they have an extraordinary medical advantage in organic products (Gil et al., 2000).

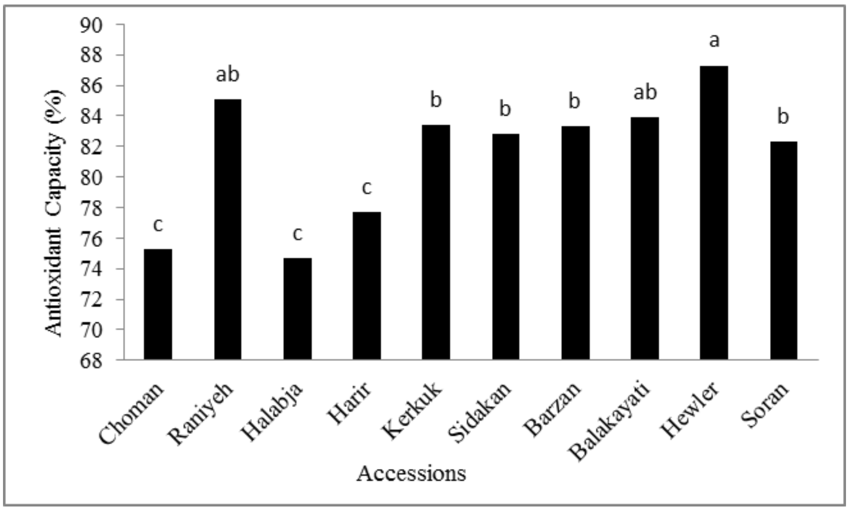

Figure 1. Antioxidant capacity in eleven pomegranate accessions. Bars with the same letters were not statistically significant (P≤0.01).

PCA (Principal component analysis) was applied to study the characteristics for distinguishing the most important factors of changeability and to portray the connection among the variables (Tables 3 and 4). PCA produced two parts representing a total of 95.3% of variability. The most critical variables incorporated by the first component (81.9% of variability) were TSS/TA, TSS, pH, 100-seed fresh weight, seed fresh weight, aril diameter and peel thickness; while negative correlations were observed in TA, total phenolic compounds and antioxidant capacity. The second part (13.3% of variability) was predominantly related to fruit weight, fruit diameter, fruit length, volume of juice, total aril weight, total peel weight and aril length, while a negative correlation was observed in antioxidant capacity.

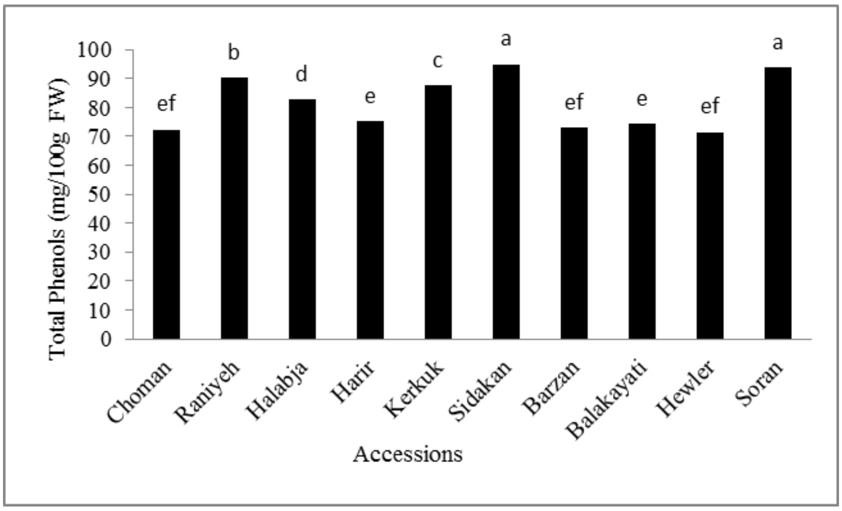

Figure 2. Total phenols in fruits of ten pomegranate accessions. Bars with the same letters were not statistically significant (P≤0.01).

The reliance of the variable was determined by analysis of correlation (Table 2). Although large pomegranates are more delicious to the customer, these will probably be sweetened. A positive correlation was observed between fruit weight and TSS (r =0.85). In addition, there was a positive correlation between fruit weight and other traits: total aril weight (r = 0.79), total peel weight (r = 0.76), volume of juice (r = 0.88), fruit length (r = 0.85), fruit diameter (r = 0.92). TSS was adversely correlated with TA (r = -0.9). Similarly, Melgarejo et al. (2000) found that sour pomegranate contained more TA and less TSS than sweet pomegranate. Total aril weight correlated with total peel weight (r = 0.59), volume of juice (r = 0.84), 100-seed fresh weight (r = 0.56), fruit length (r = 0.72) and fruit diameter (r = 0.79) suggesting that aril weight had a positive relationship with fruit size and juice content. A positive correlation was observed between total peel weight and volume of juice (r = 0.53), aril length (r = 0.57), fruit length (r = 0.68) and fruit diameter (r = 0.88) suggesting that large fruits had more peel than small fruits. Also, there was a positive correlation between volume of juice and fruit length and fruit width (r = 0.81 for both) demonstrating that large fruits might be better for making juice. On the contrary, Jalikop and Kumar (1998) detailed that small fruits are juicer than large fruits. No significant relationships were found between total phenolic compounds and antioxidants with different characteristics that can be of importance to the values for different fruits (Wang et al., 1996).

Table 2. Results of simple correlation analysis for different fruit characteristics of ten pomegranate accessions.

Total phenolics	Antioxidant capacity	Fruit diameter	Fruit length	100-seed fresh weight	Seed fresh weight	Aril diameter	Aril length	TSS/TA	TSS	TA	pH	Volume of juice	Peel thickness	Total peel weight	Total aril weight	Fruit weight	
-0.04	-0.45	**0.92****	**0.85****	**0.51****	0.29	0.16	**0.51****	0.37*	**0.85****	-0.30	0.22	**0.88****	0.08	**0.76****	**0.79****	1	Fruit weight
-0.02	-0.37	**0.79****	**0.72****	**0.56****	0.31	0.16	0.23	0.40	0.45	0.34	0.24	**0.84****	0.08	**0.59****	1		Total aril weight
-0.1	-0.34	**0.88****	**0.68****	0.38	0.09	0.41	**0.57****	0.26	0.35	-0.23	0.19	**0.53****	0.33	1			Total peel weight
-0.09	-0.06	0.18	0.09	-0.06	0.03	0.3	**0.46****	0.14	0.14	-0.14	0.21	-0.04	1				Peel thickness
-0.01	-0.37	**0.81****	**0.81****	**0.6****	0.33	0.17	**0.53****	0.40	**0.47****	-0.33	0.17	1					Volume of juice
-0.06	-0.18	0.27	0.34	**0.47****	0.37	0.41	0.25	**0.68****	**0.67****	**-0.6****	1						pH
0.21	0.16	-0.34	-0.33	**-0.4****	-0.27	-0.28	-0.33	**-0.9****	**-0.9****	1							TA (total acidity)
-0.15	-0.23	0.45	0.45	**0.62****	0.32	0.34	0.41	0.69	1								TSS (total soluble solids)
-0.19	0.16	0.38	0.37	**0.52****	0.33	0.29	0.36	1									Taste index(TSS/TA)
-0.12	-0.27	**0.58****	**0.54****	**0.51****	0.2	**0.61****	1										Aril length
-0.19	-0.13	0.32	0.30	0.52	-0.14	1											Aril diameter
-0.10	-0.27	0.30	0.24	0.25	1												Seed fresh weight
0.06	-0.31	**0.49****	**0.62****	1													100-seed fresh weight
0.03	-0.2	**0.85****	1														Fruit length
-0.06	-0.38	1															Fruit diameter
-0.04	1																Antioxidant capacity
1																	Total phenolics

******Different significant values ($P \leq 0.01$). Absolute linear correlations ≥ 0.01 are marked in bold.

Table 3. Specific values, variance and cumulative percent of variances for seven main factors.

Factors	Specific value	Variance (%)	Cumulative (%)
1	493.3	**81.9**	81.9
2	80.5	**13.3**	95.3
3	10.9	1.83	97.1
4	6.9	1.1	98.3
5	3.8	0.6	98.9
6	2.5	0.43	99.4
7	2.4	0.4	99.8

Table 4. The variable selected by the factor and explained cumulated proportion of variation for the two eigenvectors.

	Factor 1 (81.9%)	Factor 2 (13.3%)
TSS/TA	**1**	0.00
TSS	**0.97**	0.10
pH	**0.68**	-0.004
100-seed fresh weight	**0.52**	0.40
Seed fresh weight	**0.33**	0.19
Aril diameter	**0.29**	0.19
Peel thickness	**0.14**	0.06
Total phenolics	**-0.19**	0.04
TA	**-0.98**	0.05
Fruit weight	0.37	**0.88**
Fruit diameter	0.38	**0.87**
Fruit length	0.37	**0.80**
Volume of juice	0.40	**0.78**
Total peel weight	0.26	**0.77**
Total aril weight	0.40	**0.73**
Aril length	0.36	**0.49**
Antioxidant capacity	-0.16	**-0.39**

Cluster analyses (Figure 3) produced five clusters showing relatedness between accessions ‘Choman’, ‘Raniyeh’ and ‘Halabja’ and ‘Harir’, ‘Kerkuk’, ‘Sidakan’ and ‘Barzan’, ‘Balakayati’ and ‘Hewler’ and ‘Soran’. A high disparity level was found in accessions ‘Choman’ and ‘Kerkuk’, being very heterogeneous among the considered accessions. Groups one and three were highly heterogeneous in comparison with others. In addition, group two had highly similar fruit weight, total peel weight, volume of juice, fruit length and fruit diameter. In group four, fruit weight, seed fresh weight and fruit diameter were also similar. In group five, total peel weight, pH, aril length, aril diameter, 100-seed fresh weight, fruit diameter and fruit weight were similar too. Fruit weight and size were main factors in grouping of accessions.

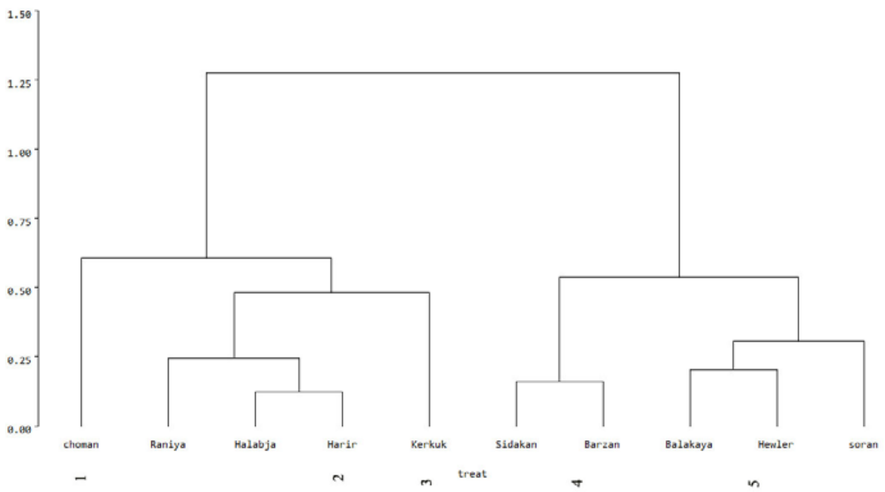

Figure 3. Grouping of ten pomegranate accessions based on fruit characteristics by the Ward method.

Conclusion

We have estimated for the first time the physical and chemical properties of pomegranate accessions from the Kurdistan region of Iraq. All of accessions varied significantly in studied traits. In this study, no significant correlation was found between total phenolic compounds and antioxidant capacity. To obtain more juice from fruits in food industries, fruits and aril size should be considered, and according to this, ‘Choman’, ‘Raniyeh’ and ‘Halabja’ accessions, due to large fruit size, had more juice. Principle component analysis (PCA) indicated that physico-chemical properties of pomegranate fruit were significantly affected by genotype. Also, physical properties had the importance factor in close relationship between accessions. Finally, this study gives us more information about physical and chemical properties in pomegranate and can be useful to producers and breeders for increasing fruit quality.

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FIZIČKO-HEMIJSKE KARAKTERISTIKE GENOTIPOVA NARA IZ REGIONA KURDISTANA U IRAKU

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R e z i m e

U ovom radu istraživane su neke fizičko-hemijske osobine deset genotipova nara prikupljenih iz različitih okruga u regionu Kurdistana u Iraku. Utvrđene su značajne korelacije između ispitivanih karakteristika i uočene su korisne pomološke karakteristike. Klaster analizom su ustanovljeni homonimi između nekih genotipova nara. Analiza glavnih komponenti je pokazala da je komponenta kojom se opisuje najveća varijabilnost u pozitivnoj korelaciji sa masom ploda, ukupnom masom zrna nara, ukupnom masom kore, zapreminom soka, ukupnim rastvorljivim suvim materijama (TSS), dužinom ploda, prečnikom ploda, pH, dužinom zrna, i masom 100 semena u svežem stanju, ali je u negativnoj korelaciji sa titracionom kiselošću (TA). Masa ploda je bila u jakoj korelaciji sa ukupnom masom zrna, ukupnom masom kore, zapreminom soka, TSS, dužinom zrna, masom 100 semena, dužinom ploda i prečnikom ploda. Zapremina soka je bila u korelaciji sa TSS, dužinom zrna, masom 100 semena, dužinom ploda, prečnikom ploda i uočeno je da se sa povećanjem veličine ploda, zapremina soka takođe povećavala. Korelacija između ukupnih fenolnih jedinjenja i antioksidativnog kapaciteta nije uočena. Veze među fizičkim i hemijskim osobinama sugerišu da bi potrošači trebalo da koriste krupne plodove sa velikim zrnima kako bi dobili više soka. Stoga su genotipovi ‘Choman’, ‘Raniyeh’ i ‘Halabja’ bili sočniji od drugih genotipova.

Ključne reči: fizičke osobine, genotipovi, plodovi, antioksidanti, fenolna jedinjenja.

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ASSESSMENT OF *STRIGA* RESISTANCE IN WILD RELATIVES OF SORGHUM UNDER FIELD CONDITIONS

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Abstract: The witch-weeds (*Striga spp.*) are destructive root parasites of many crops. They result in considerable crop damage, especially in the semiarid tropic parts of the world. The parasite control is difficult due to the complexity of the parasite life cycle, and the large number of seeds produced by the parasite with prolonged viability. The most promising way of controlling the parasite is through the development of the resistant crop varieties. Identification of different sources of resistance will enhance breeding for resistant varieties. Wild relatives of sorghum are rich in genetic diversity and have a broad genetic base including novel and valuable traits like *Striga* and disease resistance. In this context, 55 wild sorghum lines were collected from three regions of Sudan including eastern Sudan (Gadaref), central Sudan (Gezira), and western Sudan (North Kordofan). The collected germplasm was assessed for *Striga* resistance using artificial infestation. The results showed a significant difference in the number of *Striga* emerged plants compared to the checks. Hence, the significant difference was observed in the number of days to 50% plants to reach flowering, plant height, and grain yield per hectare. The wild relatives were also morphologically characterized and the result showed 55 lines structured in six groups independently from their geographical regions.

Key words: *Striga* resistance, morphological characterization, wild relatives, sorghum.

Introduction

Wild relatives of sorghum have two subspecies of *S. bicolor*: *S. bicolor Verticilliflorum* and *S. bicolor* subsp. *Drummondii* (Harlan and De Wet, 1972; Dogget, 1988). Wild relatives of sorghum are rich in genetic diversity and have a broad genetic base including novel traits like disease resistance (Gurney et al., 2002; Kamala et al., 2002; Reed et al., 2002; Rao Kameswara et al., 2003; Rich et al., 2004).

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Identification and utilization of conserved genetic material of wild species help to sustain crop improvement, particularly in breeding for biotic and abiotic stresses. Information from genetic diversity studies permits the classification of genotypes into heterotic groups which are important for hybrid development and estimating the relative strengths of the factors affecting the genetic makeup as mutation, natural selection, migration, and genetic drift. Understanding of genetic variability is useful to create segregating populations with maximum genetic variability for further selection (Barrett and Kidwell, 1998). This information is also useful for better understanding of evolutionary trends and will help in gene bank management and strategies for collection and conservation of the germplasm.

Sorghum is originated in the area bordering between Ethiopia and Sudan (Dogget, 1988). In Sudan, wild relatives of sorghum have been little investigated, and few results have been published. In this context, we collected wild relatives of sorghum from *Striga* (*Striga hermonthica*) infested fields of eastern (Gadaref), central (Gezira) and western (North Kordofan) parts of Sudan.

The objectives of this study were to assess *Striga* resistance in wild relatives of sorghum as well as to characterize and determine the genetic diversity of Sudanese wild sorghum accessions.

Materials and Methods

Plant material

Fifty-five wild relatives of sorghum entities were collected from three areas in Sudan: central (Gezira), eastern (Gadaref area), and western (North Kordofan), which can be described as irrigated, high and low rainfall areas, respectively (Figure 1). Four cultivated sorghum cultivars were used as out-groups.

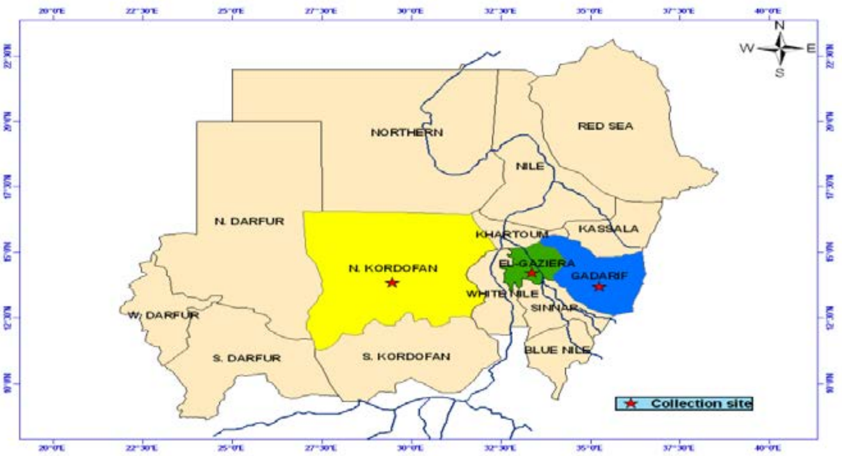

Figure 1. Sampling areas of Sudan.

Field testing

A selected wild relative of sorghum (55 lines) collected germplasm was tested in the *Striga* infested plot at the Elobied Agricultural Research Station farm. Data was collected for the number of emerged *Striga* plants, 45 and 60 days after crop emergence. The progenies were also evaluated for days to 50% flowering, plant height, and yield. Four lines SRN39-40, Tex623B, Tex623A and Carper-R were used as control. A randomized complete block design (RCBD) with three replicates was used.

The selected genotypes were also morphologically characterized using ten parameters including: awn, midrib color, senescence, waxy layer, plant height, exertion, plant color, 50% flowering, seed shattering, and head shape.

The statistical analysis was carried out using Statistix8.1 and GenSTAT for mean separation and clusters respectively.

Results and Discussion

Striga count

The number of emerged *Striga* plants was collected at 45, and 60 days after planting. Analysis of variance of the first and second *Striga* emerged plants revealed significant differences (P = 0.05) between the wild relatives and checks in the *Striga* infested field (Tables 1, 2 and 3). No significant difference was observed between the 20 wild sorghum accessions including: WS-1, WS-2, WS-8, WS-9, WS-16, WS-17, WS-18, WS-21, WS-22, WS-23, WS-31, WS-32, WS-33, WS-37, WS-42, WS-43, WS-47, WS-51, WS-52 and WS-56 (Table 1). These accessions expressed better *Striga* resistance comparing to the resistant line SRN-39.

50% flowering

The results showed significant differences in all tested lines, comparing with the resistant check SRN-39. Differences appeared with the use of LSD at the 0.05 level of probability (Tables 1 and 4).

Plant height

Field screening results showed a significant difference (Table 1) for all wild relatives and the resistant cultivar SRN-39. The other three lines as Tex623A, Tex623B and Carber-R were wiped out. The examined wild sorghum accessions and the resistant control SRN-39 showed a normal plant height. Differences appeared using the least significant differences (LSD) at the 0.05 level of probability (Table 5).

Grain yield

Analysis of variance of the grain yield revealed significant differences (P= 0.05) between the wild relatives and the resistant check SRN-39 in *Striga* infested plots (Tables 1 and 6). Mean grain yield for each genotype in the experiment is presented in Table 1. The highest grain yield was produced by the resistant cultivar SRN-39, while the wild relatives showed lower grain yield (Table 1).

Table 1. Results of the wild relatives of sorghum in the *Striga* infested field.

No.	Genotype	Region	1^{st} count of *Striga*	2^{nd} count of *Striga*	50% flowering	P. height	Yield kg/ha
1	WS-1	G	0^{i}	0^{h}	58^{bc}	188.7^{abcdef}	$164.7^{fghijklmn}$
2	WS-2	G	0^{i}	0^{h}	56.0^{bcdefg}	201.0^{a}	174.7^{cdefg}
3	WS-3	G	1^{h}	1^{g}	$54.7^{bcdefghi}$	200.0^{ab}	150^{opq}
4	WS-4	C	2^{f}	2^{f}	$53.7^{cdefghij}$	202.7^{a}	119.3^{u}
5	WS-5	C	3^{e}	3^{e}	$53.7^{cdefghij}$	$182.3^{cdefghij}$	149.7^{opqr}
6	WS-6	G	1^{h}	1^{g}	$54.0^{cdefghij}$	$174.7^{fghijklmnop}$	134.7^{t}
7	WS-7	G	2^{f}	2^{f}	$55.0^{bcdefghi}$	$175.0^{fghijklmnop}$	$166.7^{efghijklm}$
8	WS-8	G	0^{i}	0^{h}	51.3^{ijkl}	192.7^{abcde}	150^{opq}
9	WS-9	G	0^{i}	0^{h}	52.0^{ghijkl}	194.3^{abcd}	178.7^{bcde}
10	WS-10	W	0^{i}	0^{h}	51.3^{ijkl}	$186.3^{abcdefgh}$	136^{st}
11	WS-11	C	1^{h}	1^{g}	$54.0^{cdefghij}$	$181.0^{cdefghik}$	156.3^{klmnop}
12	WS-12	W	1.7^{g}	2^{f}	51.7^{hijkl}	$177.0^{efghijklmnop}$	156.7^{klmnop}
13	WS-13	C	3^{e}	3^{e}	49.3^{kl}	$168.7^{ijklmnop}$	138^{qrst}
14	WS-14	W	1^{h}	1^{g}	$54.7^{bcdefghi}$	189.7^{abcdef}	153.3^{nop}
15	WS-15	G	1.7^{g}	2^{f}	51.3^{ijkl}	190.3^{abcdef}	154.3^{mnop}
16	WS-16	W	0^{i}	0^{h}	48.7^{l}	200.7^{a}	136^{st}
17	WS-17	W	0^{i}	0^{h}	50.3^{jkl}	163.3^{nop}	148.3^{pqrs}
18	WS-18	C	0^{i}	0^{h}	58.3^{b}	$168.7^{ijklmnop}$	155.7^{lmnop}
19	WS-19	G	2^{f}	2^{f}	$52.3^{fghijkl}$	$186.3^{abcdefgh}$	$166.3^{efghijklmn}$
20	WS-20	G	1^{h}	1^{g}	51.7^{hijkl}	$183.7^{bcdefghi}$	176.7^{bcdef}
21	WS-21	G	0^{i}	0^{h}	51.7^{hijkl}	133.3^{q}	136.7^{rst}
22	WS-22	G	0^{i}	0^{h}	$52.3^{fghijkl}$	161.0^{p}	127.7^{tu}
23	WS-23	G	0^{i}	0^{h}	$53.3^{defghijk}$	$181.3^{cdefghijk}$	188^{b}
24	WS-24	G	2^{f}	2^{f}	$54.0^{cdefghij}$	$180.0^{cdefghijklm}$	185^{bc}
25	WS-25	C	1^{h}	1^{g}	$52.3^{fghijkl}$	188.7^{abcdef}	$167^{efghijklm}$
26	WS-26	G	3^{e}	3^{e}	$54.7^{bcdefghi}$	$177.7^{efghijklmno}$	$159^{hijklmnop}$
27	WS-27	C	2^{f}	2^{f}	$53.7^{cdefghij}$	$175.3^{fghijklmnop}$	131.3^{tu}
28	WS-28	C	2^{f}	2^{f}	$53.0^{efghijk}$	$171.3^{ghijklmnop}$	$169^{efghijk}$
29	WS-29	W	3^{e}	3^{e}	58^{bc}	201.0^{a}	$165^{fghijklmn}$
30	WS-30	W	1^{h}	1^{g}	56.0^{bcdefg}	$166.7^{jklmnop}$	178.3^{bcde}
31	WS-31	G	0^{i}	0^{h}	56.0^{bcdefg}	$177.3^{efghijklmnop}$	189^{b}
32	WS-32	W	0^{i}	0^{h}	$53.0^{efghijk}$	190.0^{abcdef}	$168.7^{efghijk}$

Table 1. Continued**.**

No.	Genotype	Region	1st count of *Striga*	2nd count of *Striga*	50% flowering	P. height	Yield kg/ha
33	WS-33	G	0^{i}	0^{h}	55.7^{bcdefgh}	196.3^{abc}	171.3^{defghi}
34	WS-34	G	2^{f}	2^{f}	51.7^{hijkl}	188.0^{abcdef}	182.3^{bcd}
35	WS-35	W	1^{h}	1^{g}	54.0^{cdefghij}	178.7^{cdefghijklm}	162.7^{ghijklmno}
36	WS-36	W	3e	3e	56.3^{bcdef}	166.7^{jklmnop}	167.7^{efghijkl}
37	WS-37	G	0^{i}	0^{h}	56.7^{bcde}	167.0^{jklmnop}	158.0^{jklmnop}
38	WS-38	G	0^{i}	0^{h}	56.0^{bcdefg}	164.3^{lmnop}	149.3^{pqr}
39	WS-39	W	2^{f}	2^{f}	54.0^{cdefghij}	166.0^{jklmnop}	149.0^{pqrs}
40	SRN39	Out-group	4^{d}	4^{d}	62.7^{a}	119.0^{q}	320^{a}
41	WS-41	W	1^{h}	1^{g}	54.0^{cdefghij}	177.0^{efghijklmnop}	166.7^{efghijklm}
42	WS-42	G	0^{i}	0^{h}	54.7^{bcdefghi}	186.3^{abcdefgh}	165.7^{efghijklmn}
43	WS-43	G	0^{i}	0^{h}	55.7^{bcdefgh}	177.3^{efghijklmnop}	176.3^{bcdef}
44	WS-44	W	3^{e}	3^{e}	53.3^{defghijk}	168.7^{ijklmnop}	185.7^{bc}
45	WS-45	W	2^{f}	2^{f}	53.0^{efghijk}	164.3^{lmnop}	170.0^{defghij}
46	WS-46	W	1^{h}	1^{g}	52.3^{fghijkl}	165.0^{klmnop}	171.7^{defgh}
47	WS-47	W	0^{i}	0^{h}	51.3^{ijkl}	169.3^{ijklmnop}	178.7^{bcde}
48	WS-48	W	3^{e}	3^{e}	53.7^{cdefghij}	178.3^{cdefghijklmn}	158.3^{ijklmnop}
49	WS-49	W	2^{f}	2^{f}	52.0^{ghijkl}	187.7^{abcdefg}	167^{efghijklm}
50	WS-50	W	1^{h}	1^{g}	50.3^{jkl}	180.7^{cdefghijkl}	158^{jklmnop}
51	WS-51	W	0^{i}	0^{h}	55.7^{bcdefgh}	170.3^{hijklmnop}	148.3^{pqrs}
52	WS-52	W	0^{i}	0^{h}	54.0^{cdefghij}	165.7^{klmnop}	150.3^{opq}
53	WS-53	W	2^{f}	2^{f}	57.3^{bcd}	167.0^{jklmnop}	169.3^{efghijk}
54	WS-54	W	1^{h}	1^{g}	56.3^{bcdef}	164.0^{mnop}	167.7^{efghijkl}
55	WS-55	W	3^{e}	3^{e}	58^{bc}	161.7^{op}	177^{bcdef}
56	WS-56	W	0^{i}	0^{h}	56.0^{bcdefg}	170.0^{hijklmnop}	188.3^{b}
57	Tex623B	out-group	28^{c}	34^{a}			
58	Tex623A	out-group	30^{a}	32^{b}			
59	Carper-R	out-group	29^{b}	30^{c}			
Mean			2.58	2.75	54	176.96	164.11
SE±			0.09	0.06	2.18	8.2467	6.63
C.V			4.12	2.73	4.94	5.71	4.95

Legend: C: Central, G: Gezira, W: Western, O: Out-group.
**Means with different superscript letters in the same column are significantly different at P = 0.05.

Table 2. ANOVA table of the *Striga* 1st count.

Source of variation	d. f.	s. s.	m. s.	v. r.	F. pr.
Blocks	2	0.04520	0.02260	2.04	
Genotypes	58	6843.72881	117.99532	10625.79	<.001
Residuals	116	1.28814	0.01110		
Total	176	6845.06215			

Table 3. ANOVA table of the *Striga* 2nd count.

Source of variation	d. f.	s. s.	m. s.	v. r.	F. pr.
Blocks	2	0.31638	0.15819	2.89	
Genotypes	58	8356.39548	144.07578	2631.82	<.001
Residuals	116	6.35028	0.05474		
Total	176	8363.06215			

Table 4. ANOVA table of days to 50% flowering.

Source of variation	d. f.	s. s.	m. s.	v. r.	F. pr.
Blocks	2	572.576	286.288	147.32	
Genotypes	58	25979.559	447.923	230.50	<.001
Residuals	116	225.424	1.943		
Total	176	26777.559			

Table 5. ANOVA table of plant height.

Source of variation	d. f.	s. s.	m. s.	v. r.	F. pr.
Blocks	2	3927.49	1963.75	30.38	
Genotypes	58	306158.46	5278.59	81.67	<.001
Residuals	116	7497.84	64.64		
Total	176	317583.80			

Table 6. ANOVA table of plant height.

Source of variation	d. f.	s. s.	m. s.	v. r.	F. pr.
Blocks	2	6196.37	3098.19	298.42	
Genotypes	58	348159.37	6002.75	578.20	<.001
Residuals	116	1204.29	10.38		
Total	176	355560.03			

Cluster analysis and the effect of cultivated region

The cluster analysis developed based on ten morphological characters included: awn, midrib color, senescence, waxiness, plant height, exertion, plant color, 50% flowering, seed shattering, and head shape. The results showed the accessions structured in 4 major groups (Figure 2). The wild relatives were structured in different groups and subgroups independently from their geographical regions.

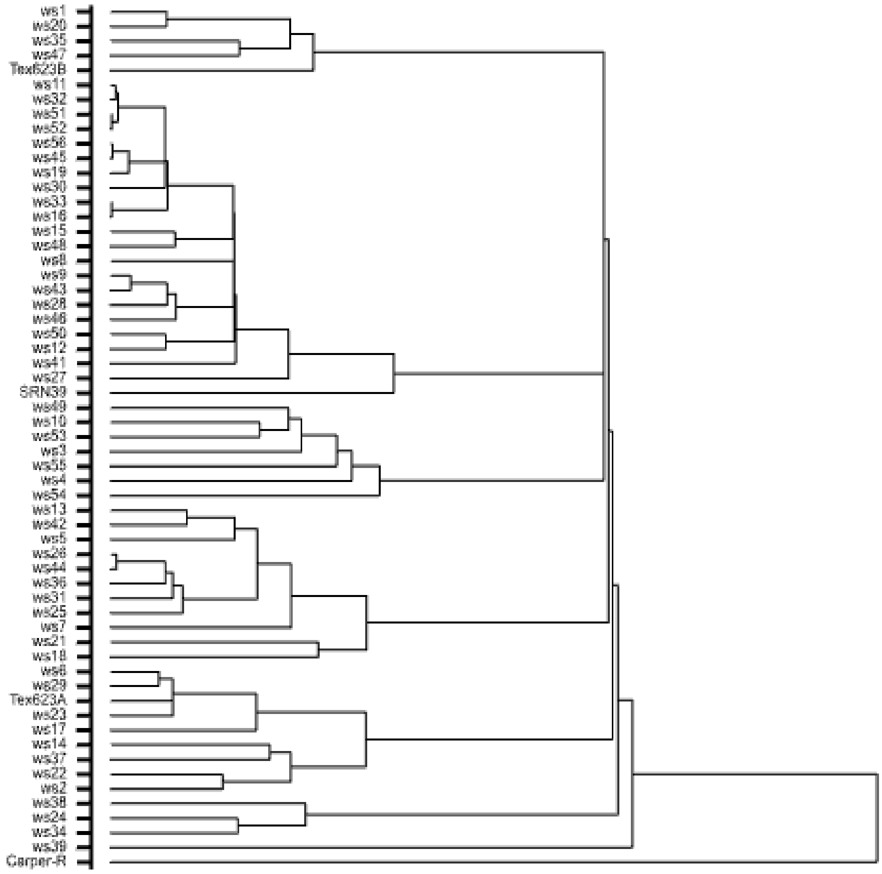

Figure 2. Cluster analysis of 59 sorghum genotypes.

This work is part of the project that aimed to introgress *Striga* resistance into Hageen Dur1 (F1 hybrid generation), which is one of the hybrids released for commercial use, showing good performance in irrigated and high rainfall areas of Sudan. To achieve this goal, 55 wild relatives of sorghum were collected, assessed for *Striga* resistance and morphologically characterized. *Striga* resistance in wild and related species has not been fully exploited, and a few surveys of wild sorghums for *Striga* resistance have been reported (Deodikar, 1951; Lane et al., 1994; Mohamed et al., 2003).

Through this work, 55 wild relatives of sorghum were tested in *Striga* infested plots, with the resistant cultivar, SRN-39, as a resistant control. However, the parental lines of Hageen Dura-1, Tex623A, Tex623B and Carber-R, were used as the susceptible control.

Counting *Striga* plants is considered as one of the field measurements that can determine host plant resistance. Our data indicated that the susceptible checks, Tex623A, Tex623B and Carber-R, had the highest number of the emerged *Striga* plants among the tested genotypes. This may be due to production or exudation of the stimulant from the three checks. However, wild sorghum showed a lower number of *Striga* emerged plants, some of which showed immunity in which no *Striga* plants were observed surrounding the host plant. Wild sorghums may be sources of unique resistance traits lacking in cultivars since they have evolved under selective pressures imposed by *Striga* spp. (Rich et al., 2004). It is noteworthy that *Striga* resistant sorghum varieties may produce the low number of *Striga* plants and the number of viable seeds in the soil, but they are often not locally adapted and morphologically inferior (Haussmann et al., 2004). The wild relatives were surveyed and tested in the field, but they were not examined for the resistance mechanisms. In the previous studies, several mechanisms of *Striga* resistance were mentioned in sorghum and its wild relatives (Ejeta et al., 2000; Gurney et al., 2002; Heller and Wegmann, 2000).

The susceptible control showed no flowering even one hundred and twenty days after germination; it was wiped out by the end of the season. The observed results correspond with the previously published reports stating that sorghum yield losses may reach 100% on heavily infested soils (Parker and Riches, 1993; Khidir, 1983; Obilana, 1983). The results confirmed the high susceptibility of the cultivars, Tex623A, Tex623B and Carber-R, and a high level of infestation in the naturally infested plot.

A significant difference was observed in plant height among these genotypes. The susceptible ones were poorly developed and stunted and genotypes with resistance had normal height. This result is coherent and corresponding with our previous field evaluation of 19 sorghum lines developed by using marker assisted selection in our local breeding program (Gamar and Abdalla, 2013).

The study also measured the grain yield. The three susceptible parents showed no seeds (highly affected by *Striga* damage), while the resistant control SRN-39 achieved the highest grain yield. The wild relatives of sorghum showed a normal feature of growing, which means the tested lines were not affected by *Striga* infection. Empirical breeding for *Striga* resistance in field crops has relied on selection of host plants that allow the emergence of few parasitic plants and show little or no loss in productivity of the crop (Mohamed et al., 2003).

The cluster analysis of 55 wild relatives revealed that Tex623A and Tex623B were structured in one group, while Carper-R, the restorer line, was structured in a different group. This showed the ability of morphological markers to discriminate between the tested lines. The wild relatives of sorghum were structured in different groups independently from their geographical regions and *Striga* resistance.

Conclusion

The study revealed the *Striga* resistance in wild relatives of sorghum under field conditions. The wild relatives of sorghum showed immunity against *Striga* invasion. The lines showed that immunity was expected to have an impact on breeding for *Striga* resistance in sorghum.

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PROCENA OTPORNOSTI PREMA PARAZITIMA RODA *STRIGA* KOD DIVLJIH SRODNIKA SIRKA U POLJSKIM USLOVIMA

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R e z i m e

Vrste roda *Striga* su destruktivni paraziti korena mnogih useva. Uzrokuju znatne štete na usevima, posebno u semiaridnim tropskim delovima sveta. Kontrola parazita je teška zbog složenosti životnog ciklusa, i velikog broja semena koje parazit proizvodi sa produženom životnom sposobnošću. Način kontrolisanja ovog parazita, koji najviše obećava, je putem razvoja otpornih sorti useva. Prepoznavanje različitih izvora otpornosti poboljšaće oplemenjivanje za otporne varijetete. Divlji srodnici sirka su bogati genetskom raznovrsnošću i imaju široku genetsku osnovu, koja uključuje nove i vredne osobine poput otpornosti prema parazitu roda *Striga* i bolestima. U ovom kontekstu, 55 linija divljeg sirka sakupljene su iz tri regiona Sudana uključujući istočni Sudan (Gadaref), centralni Sudan (Gezira) i zapadni Sudan (severni Kordofan). Prikupljena germplazma procenjena je na otpornost prema parazitima roda *Striga* korišćenjem veštačke infestacije. Rezultati su ukazali na značajnu razliku u broju niklih biljaka zaraženih parazitima roda *Striga* u poređenju sa kontrolom. Značajna razlika je zabeležena u broju dana do kada 50% biljaka procveta, visini biljke i prinosu zrna po hektaru. Divlji srodnici su takođe bili morfološki okarakterisani i rezultat je pokazao da je 55 linija strukturirano u šest grupa nezavisno od njihovih geografskih regiona.

Ključne reči: otpornost prema parazitima roda *Striga*, morfološka karakterizacija, divlji srodnici, sirak.

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WEED BIOMASS AND PRODUCTIVITY OF OKRA (*ABELMOSCHUS ESCULENTUS* (L) MOENCH) AS INFLUENCED BY SPACING AND PENDIMETHALIN-BASED WEED MANAGEMENT

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Abstract: Field trials were conducted at the Teaching and Research Farm of the Kwara State University, Malete, Nigeria, to determine the effect of plant spacing and weed control methods on weed infestation, growth and yield of okra during the 2016 and 2017 cropping seasons. The experiment consisted of twelve treatments comprising six weed control methods and two plant spacings. The method of weed control consisted of pre-emergence application of pendimethalin at 1.0 kg a.i. ha^{-1}, pendimethalin at 2.0 kg a.i. ha^{-1}, pendimathalin at 1.0 kg a.i. ha^{-1} + one supplementary hoe weeding (SHW) at 6 weeks after sowing (WAS), two hoe weedings at 3 and 6 weeks after sowing (WAS), weeding at 4 and 8 weeks after sowing (WAS) and a weedy check. The plant spacings were 60cm x 30cm and 60cm x 50cm. These treatments were laid out in a randomized complete block design (RCBD) with split plot arrangements and three replications. Weed control and plant spacing treatments were allocated to the subplots and the main plot respectively. Results showed that a plant spacing of 60cm x 30cm minimized weed infestation and resulted in a higher total number of pods/plot and okra fresh weight, while pendimethalin at 1.0 kg a.i. ha^{-1} + one supplementary hoe weeding at 6 WAS minimized weed infestation in the plots, and led to the highest total number of pods and yield of okra. This combination also promoted higher economic returns.

Key words: weed control method, okra productivity, southern Guinea savanna, Nigeria.

Introduction

Okra (*Abelmoschus esculentus* (L.) Moench) is an important vegetable crop grown in the wet season and to a little extent in the dry season (Alegbajo and Ogunlana, 2006).

The world's greatest producer of okra is India, producing 70% of the total world's production estimated to be 6 million tons per year (FAOSTAT, 2012),

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followed by Nigeria and Sudan (Varmudy, 2011). In Nigeria, okra ranks third in terms of consumption and production area following tomato and pepper (Odeleye et al., 2005).

Adeyemi et al. (2008) have reported that in Nigeria, the limiting factors in okra production, among others, are sub-optimal planting density, weed management, tillage practices and low yielding varieties. Yield loss as a result of uncontrolled weeds in okra fields was reported to be up to 91% in the northern Guinea savanna of Nigeria (Adejonwo et al., 1989). Similarly, Olabode et al. (2006) reported 85% fruit yield loss in okra when comparing the performance of okra in uncontrolled weed plots and mulched plots in the southern Guinea savanna of Nigeria.

Hoe weeding is the most common weed control practice in okra cultivation in Africa, but this method has been reported to be cumbersome, time-consuming and expensive. Apart from high cost of hoe weeding, scarcity of labor is a common problem usually experienced during the peak period of farming operations and can sometimes result in abandonment of field crops (Adigun et al., 2005; Osupitan, 2017).

Although efforts are being intensified to promote the use of chemical weed control among farmers in developed agriculture, the sustained use of herbicides has caused shifts in weed flora of arable fields or in increasing environmental concerns over their use (Racjan et al., 2001; Hyvonen et al., 2002). Furthermore, some workers have reported that the use of herbicides alone for weed control in okra can hardly give season-long weed control (Olabode et al., 2006), especially in the southern Guinea savanna where higher rainfall and relative humidity favor rapid and prolonged weed growth. Consequently, integrated approach to weed management remains very important (Gianessi and Williams, 2001).

A lot of research has been done on the effect of spacing on the productivity of okra. However, different spacings have been recommended for higher okra yield in different agro-ecological conditions (Vikash et al., 2016; Paththinige et al., 2008). Therefore, the need to determine the correct spacing for higher production of okra variety NHAe47-4 in Malete, Kwara State, Nigeria is imperative.

Work done on the use of integrated weed management for effective weed control and for increasing the pod yield of okra includes that of Smith et al. (2009), who have reported that the use of pendimethalin-based integrated weed management system (IWMS) will enhance production of okra and farmers' livelihood in polyculture-based small farms. Similarly, pendimethalin either alone or in mixtures with broadleaf herbicides and supplemented with other control methods, especially hand weeding, has given effective control of weeds (Dhann Appal and Gowda, 1996; Imoloame, 2017).

Pendimethalin is one of the herbicides commonly used for weed control in okra cultivation in Malete, Kwara State, Nigeria. This could be a result of its low price and availability in agro-chemical stores all the time. However, because most farmers in the area are illiterate and lack the knowledge of using herbicides, these agrochemicals are indiscriminately applied with attendant consequences of low and high costs of weed control, environmental pollution, high crop mortality, poor crop yields, low incomes and standard of living. There is, therefore, the need to determine the correct dose of pendimethalin herbicide and better weed control options that will provide season-long weed control, higher okra pod yield, and increase cash returns to farmers.

The hypothesis of this study is that a single method of weed control will not provide season-long weed control, maximum okra yield and economic returns in Malete, Kwara State. Therefore, IWM involving a lower dose of pendimethalin integrated with a supplementary hoe weeding at 6WAS and narrower spacing of okra will not only give season-long weed control in okra, but it will also increase okra yield and cash returns to the farmers. The objectives of the study are to determine the correct spacing and better pendimethalin weed management option that will be more effective in controlling weeds and increasing yield and cash income in the production of okra.

Materials and Methods

Site description

The experiment was conducted during the 2016 and 2017 cropping seasons at the Kwara State University Teaching and Research (T&R) Farm, Malete (Lat.08^{o} 71^{l}N; Long.04^{o} 44^{o}E), Kwara State, in a southern Guinea savanna ecological zone of Nigeria. The experimental site was characterized by two peaks of rainfall in June and September and the soil was sandy loam with a low water retaining capacity.

Treatment and experimental design

The experiment consisted of twelve treatments comprising six weed control methods and two plant spacings. The methods of weed control were: application of pendimethalin at 1.0 kg a.i. ha^{-1}, pendimethalin at 2.0 kg a.i. ha^{-1}, pendimathalin at 1.0 kg a.i. ha^{-1} + 1 supplementary hoe weeding (SHW) at 6 WAS, weeding at 3 and 6 weeks after sowing (WAS), weeding at 4 and 8 weeks after sowing (WAS) and a weedy check. Weeding at 3 and 6 WAS served as the control plot. The weed-free treatment was not, therefore, necessary since the critical period of weed competition in okra occurred between 3 and 7 WAS (William and Warren, 1975). In addition, okra plots weeded twice have been reported to give maximum pod yield which was comparable to the weed-free plots (Olabode et al., 2010). The plant spacings were 60cm x 30cm and 60cm x 50cm giving plant populations of 55,556 and 33,333 plants ha^{-1} respectively. These treatments were laid out in a randomized complete block design (RCBD) fitted into split plot arrangements and replicated three times.

The land used for the experiment was first mechanically plowed and harrowed, then it was leveled and marked out into plots measuring 3m x 3m each. A space of 0.5m was left between plots, while 1m was left between replicates. There were a total of 36 plots.

In order to provide nutrient supply, the NPK 15:15:15 was applied to each plot at the recommended rate of 300 kg ha^{-1}. The mineral fertilizer was applied in two split doses – the first dose applied to each plot before planting and the remaining dose applied at 6 WAS.

Sowing was done on the 18^{th} and 14^{th} of July, in 2016 and 2017 respectively, using treated seeds of okra variety NHAe47-4 obtained from the National Horticultural Research Institute (NIHORT), Ibadan, Nigeria. Three seeds were planted per hole and the resultant seedlings were thinned to one plant per strand at 3 WAS at spacings of 60cm x 30cm and 60cm x 50cm.

Herbicide (pendimethalin) was applied as pre-emergence a day after sowing using a CP 15 knapsack sprayer and a green nozzle calibrated to deliver 208l ha^{-1} of the herbicide spray volume. Karate insecticide (lambdacyhalothrin) at the rate of 30ml /10 liters of water was used to control insects, especially leaf eater beetles.

Harvesting of okra seed was done periodically in both September and October in the two years of the experiment.

The following parameters were measured:

Weed dry matter (kg ha^{-1})

Weed dry matter was determined by harvested weeds from one square meter quadrat, randomly placed in three locations within each plot. The weeds were put in well-labeled envelopes which were later oven-dried at a temperature of 80^{o}C for 2 days to a constant weight before the final weights were taken. The weed dry matter was taken at 6 and 9 WAS.

Weed cover score

Weed cover score was determined at 3 and 6 WAS by visual observation using a scale of 0–9, where 0 means weed-free plots and 9 means a complete weed cover of plots.

Weed density (kg ha^{-1})

Weed density was determined at 6 and 9 WAS by counting the number of weed species within a 50cm x 50cm quadrat, randomly placed in three locations within each plot and the total number of weed species per unit area was recorded.

Number of leaves/plant

The number of leaves per plant was determined at 6 and 9 WAS. Five plants from each plot were selected at random and the number of leaves on them was counted. The average of the total number of leaves was recorded as the number of leaves per plant.

Leaf area (cm)

Leaf area of okra was determined at 6 and 9 WAS by using the expression: Leaf area (LA) = Length (L) x breadth (B) x 0.62. The leaf area was obtained by measuring the length and breadth of leaves from five randomly selected plants from each plot and the average of these measurements was multiplied by a factor of 0.62 to give the leaf area per plant.

Number of pods/plot

The number of pods per plot was determined by counting the total number of harvested pods from each net plot.

Fresh weight (kg ha^{-1})

Fresh weight was determined by weighing the pods harvested from each net plot and the weight was converted to kilogram per hectare using the equation below:

$$\text{Fresh weight} = \frac{\text{Pod yield per net plot x } 10000m^2}{\text{Net plot size } (m^2)} \qquad \text{Eq 1}$$

Effect of plant spacing and different methods of weed control on the economics of producing okra

The effect of plant spacing and methods of weed control on the economics of producing okra was determined by calculating the total cost of production, gross revenue, net revenue (Eni et al., 2013) for each treatment as follows:

Production cost (PC): This was computed by adding the cost of inputs and those of all farm operations. These included cost of okra seeds, pendimethalin herbicide at different rates, insecticide, fertilizers, land preparation, planting, herbicide application, one and two hoe weeding(s), fertilizer application, harvesting and bagging. This is represented by the following equation: PC = (PC1+PC2+PC3+……..PCn) (Eni et al., 2013);

Gross revenue (R): This was obtained by multiplying the okra fresh weight in kg/ha by the farm gate price as follows: Gross revenue = Crop yield (Y) x Farm gate price (P) (Eni et al., 2013);

Net revenue (NR): This was calculated by subtracting the total production cost from the gross revenue as follows: NR= GR-PC;

Cost: benefit ratio = Total cost of production/Total revenue (Joshua and Gworgwor, 2001).

Data analysis

All the data collected were subjected to analysis of variance (ANOVA) using SAS 9.0 package and the means were separated using the least significant difference (LSD) at the 5% level of probability. The yield data in the economic analysis was separated using Duncan's Multiple Range Test (DMRT) at the 5% level of probability.

Results and Discussion

Rainfall

The total rainfall recorded in 2016 and 2017 amounted to 1414mm and 1015.7mm respectively. The highest rainfall was recorded in the months of September and August in both years, respectively (Figures 1 and 2).

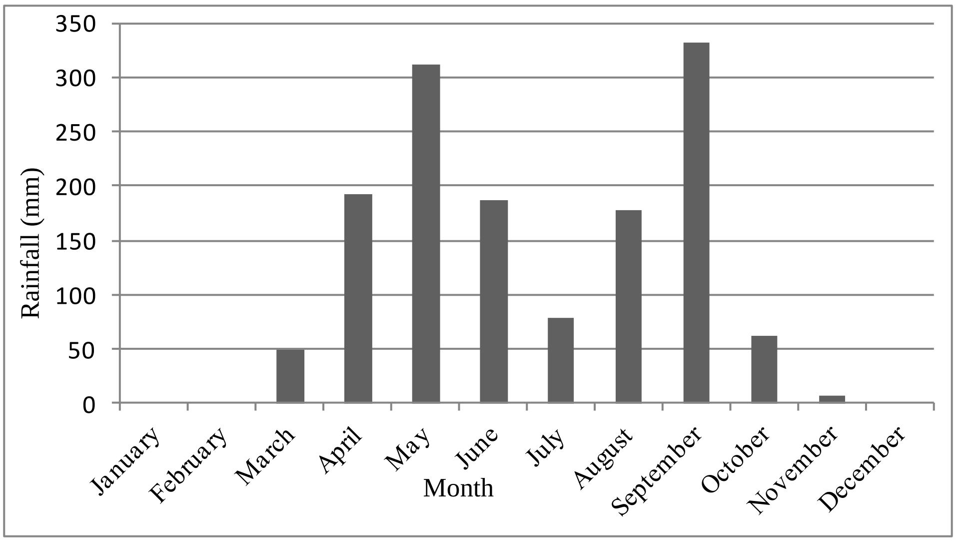

Figure 1. Rainfalls in 2016; Total – 1, 398.5.
Source: Lower Niger River Basin and Rural Development Authority, Hydrology Station.

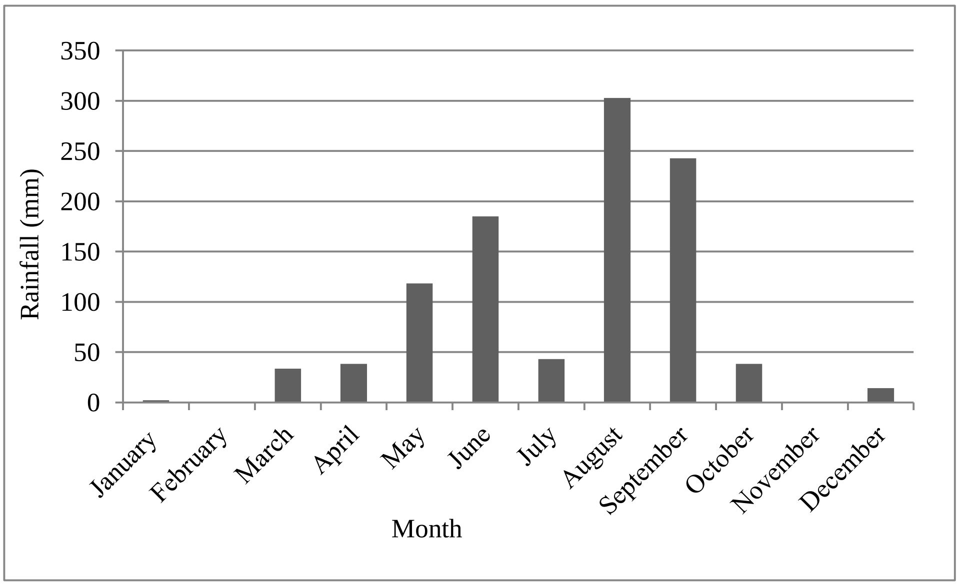

Figure 2. Rainfalls in 2017; Total = 1017mm.
Source: Lower Niger River Basin and Rural Development Authority Hydrology Station.

Effect of spacing and pendimethalin-based weed management options on weed biomass and weed density

Wider spacing promoted a higher amount of weed biomass and weed density in both years and average of the two years compared to narrower spacing. The difference was statistically significant only in 2016 (Figures 3, 4, 5, 9, 10 and 11). Pendimethalin at 1.0 kg a.i. ha^{-1} plus one SHW and two hoe weedings at 3 and 6, and 4 and 8 WAS significantly reduced weed biomass and weed density and provided season-long effective weed control throughout the life of the crop. Generally, higher values of weed biomass and weed density were recorded in 2016 than in 2017 (Figures 6, 7, 8, 12, 13 and 14). The significant reduction in weed biomass and weed density by the narrower spacing in 2016 could have been a result of the higher rainfall recorded in that year which could have promoted better growth and early canopy closure of okra required for smoother weeds. The broadcast method of producing sesame was found to suppress weeds better than the drilling method as a result of quick and early canopy closure and reduction in light penetration (Dalley et al., 2004; Imoloame, 2007). Similarly, one SHW at 6 WAS integrated with pendimethalin at 1.0 kg a.i. ha^{-1} increased the efficacy of the herbicide to provide season-long weed control. This method of weed control can serve as an alternative to two hoe weedings at 3 and 6 or 4 and 8 WAS, which is considered to be laborious and associated with drudgery. The higher rainfall recorded could have promoted higher weed biomass in 2016 compared to 2017. There was no significant interaction between spacing and the weed control method.

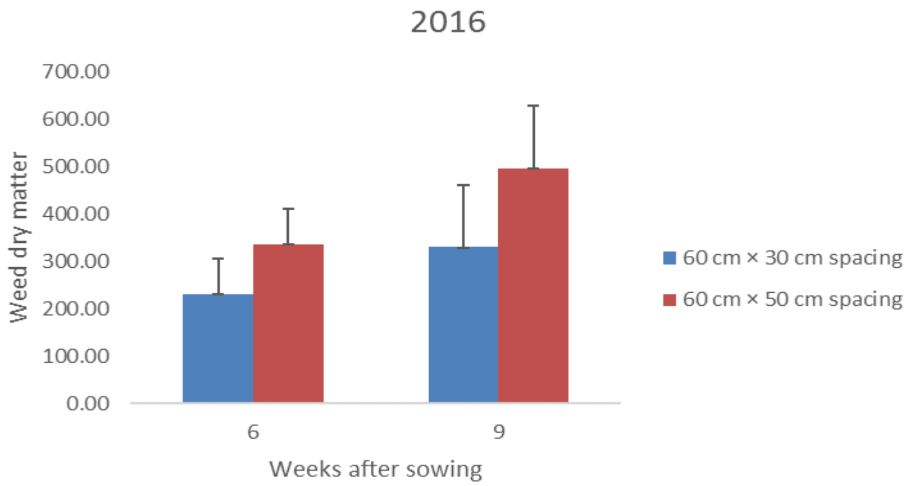

Figure 3. The effect of spacing on weed dry matter (2016).
LSD (0.05) at 6 WAS = 73.59; LSD (0.05) at 9 WAS = 131.76.

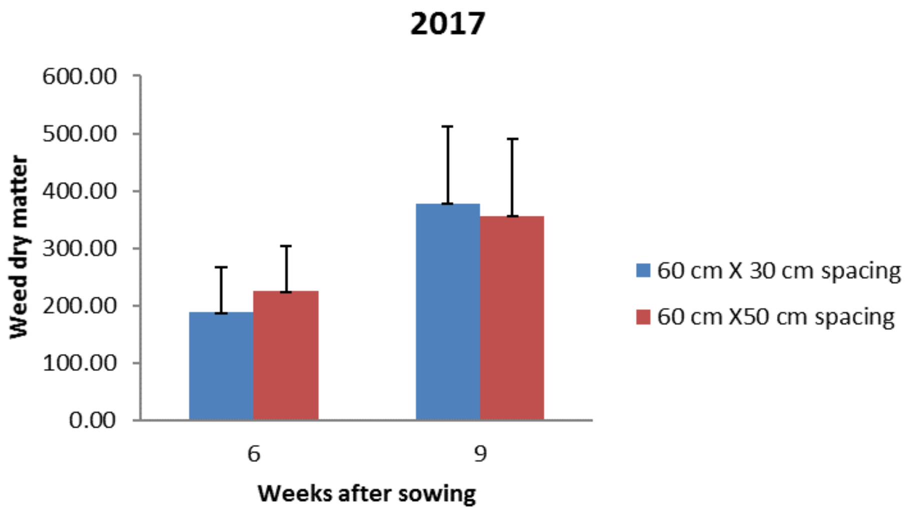

Figure 4. The effect of spacing on weed dry matter (2017).
LSD (0.05) at 6 WAS = 79.70; LSD (0.05) at 9 WAS = 134.00.

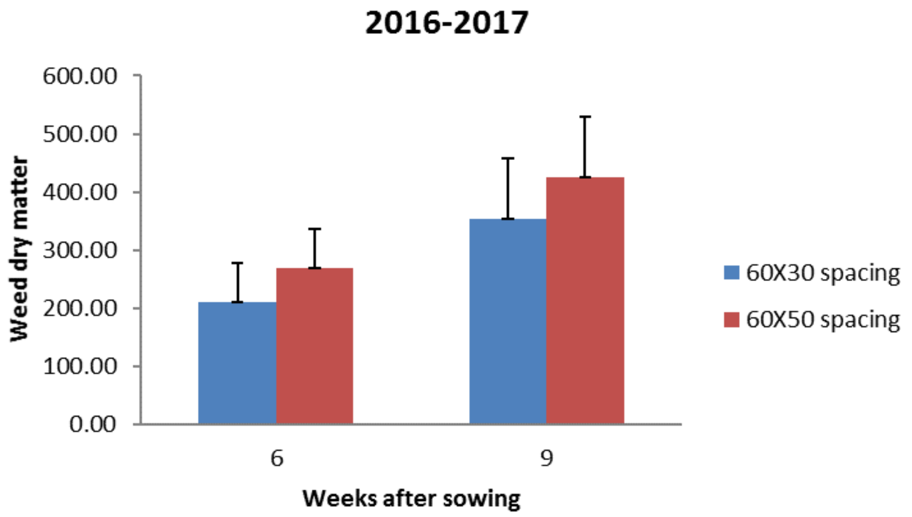

Figure 5. The Effect of spacing on averaged weed dry matter.
LSD (0.05) at 6WAS = 66.76; LSD (0.05) at 9WAS = 103.02.

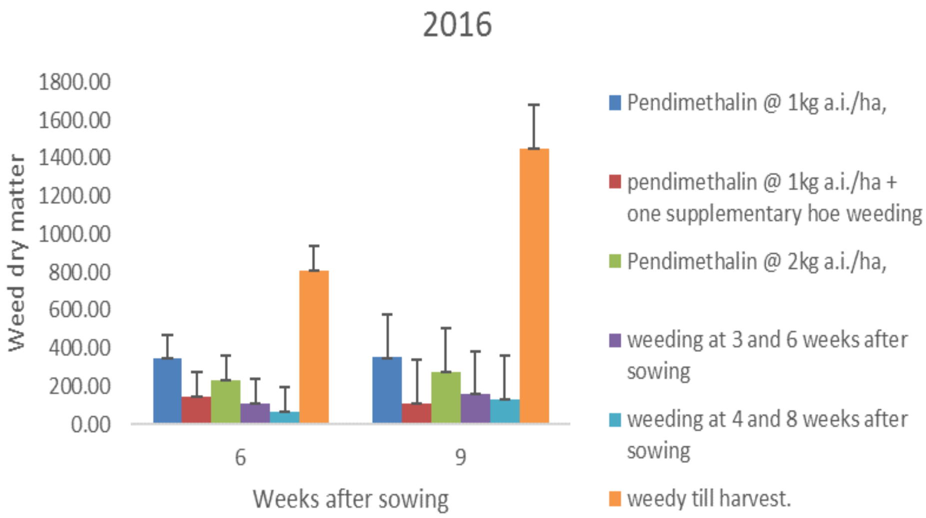

Figure 6. The effect of weed control on weed dry matter (2016).
LSD (0.05) at 6 WAS = 127.45; LSD (0.05) at 9 WAS = 228.07.

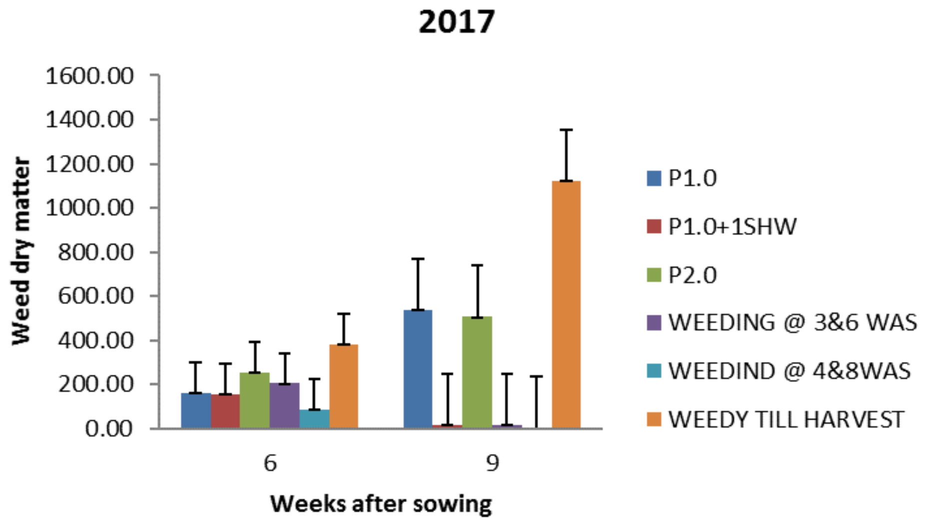

Figure 7. The effect of weed control on weed dry matter (2017).
LSD (0.05) at 6 WAS = 138.04; LSD (0.05) at 9 WAS = 232.09.

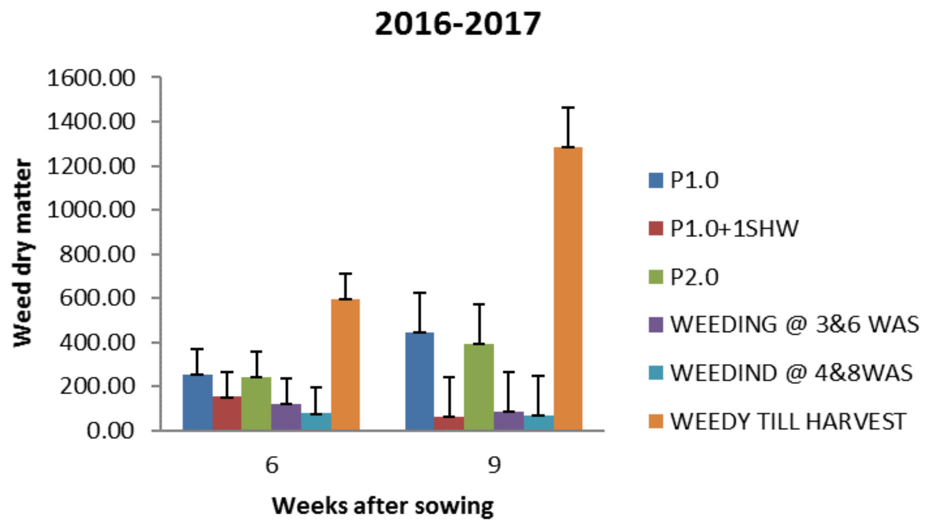

Figure 8. The effect of weed control on weed dry matter at the mean.
LSD (0.05) at 6 WAS = 175.62; LSD (0.05) at 9 WAS = 178.44.

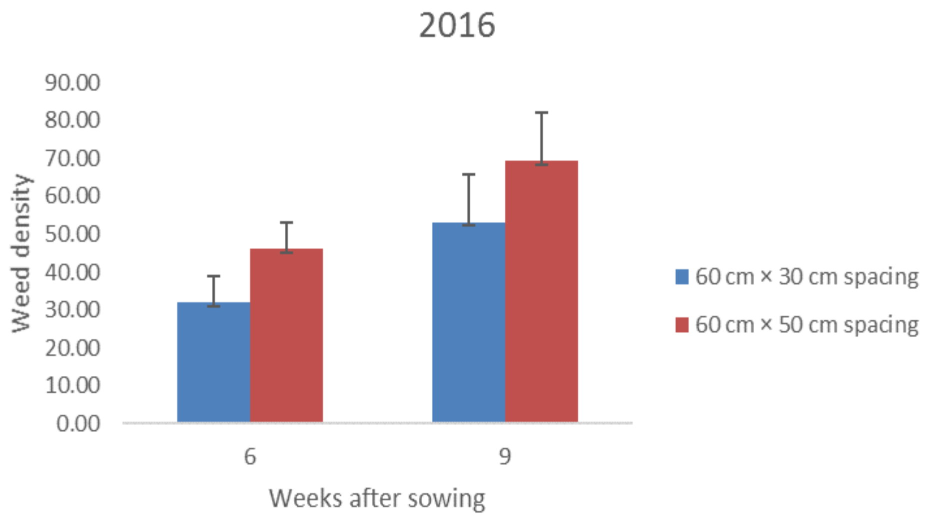

Figure 9. The effect of spacing on weed density (2016).
LSD (0.05) at 6 WAS = 0.85; LSD (0.05) at 9 WAS = 12.74.

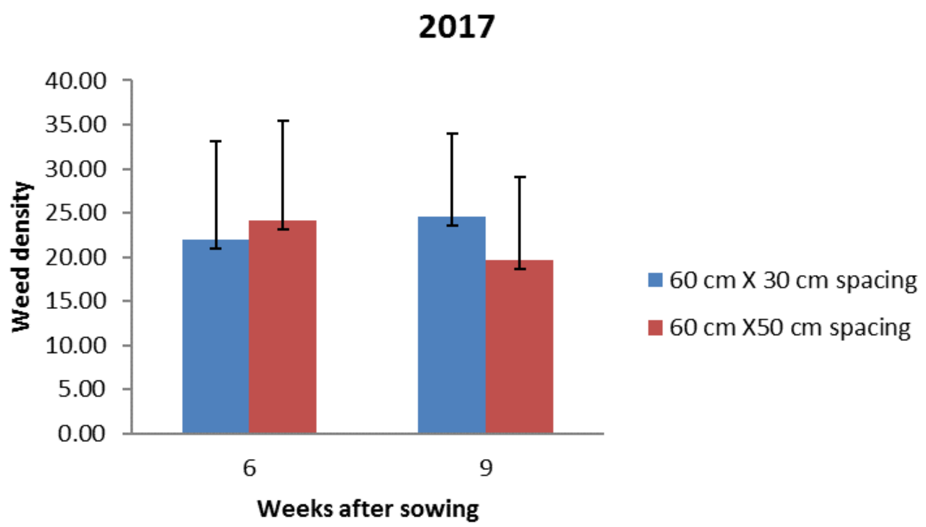

Figure 10. The effect of spacing on weed density (2017).
LSD (0.05) at 6 WAS = 11.22, LSD (0.05) at 9 WAS = 9.42

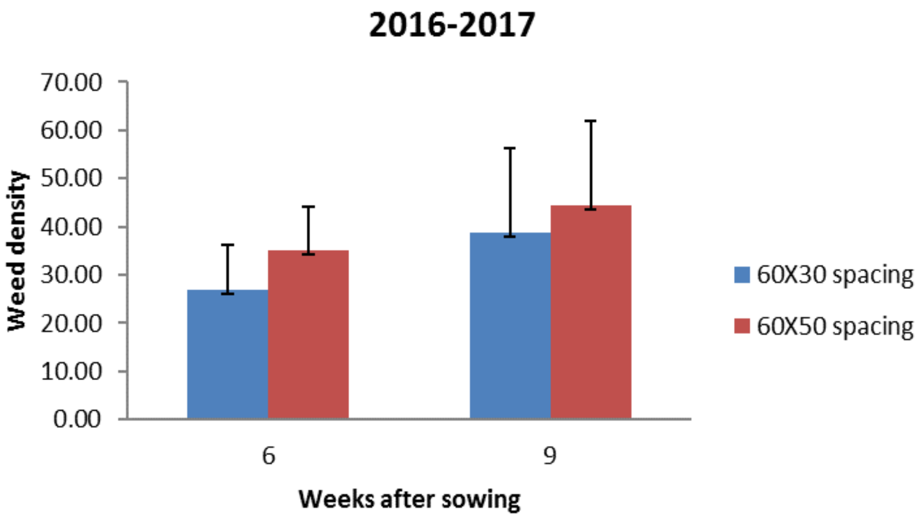

Figure 11. The effect of spacing on weed density at the mean.
LSD (0.05) at 6 WAS = 9.05; LSD (0.05) at 9 WAS = 17.41.

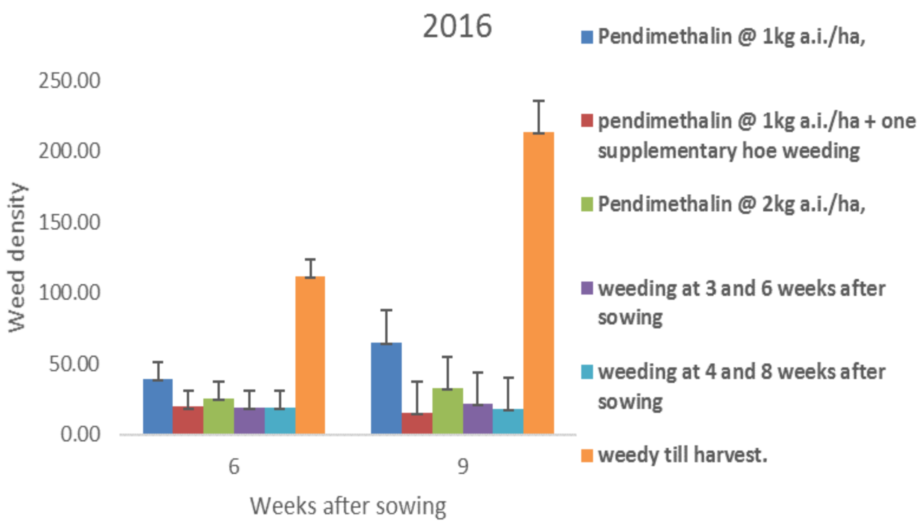

Figure 12. The effect of weed control on weed density (2016).
LSD (0.05) at 6 WAS = 11.86; LSD (0.05) at 9 WAS = 22.06.

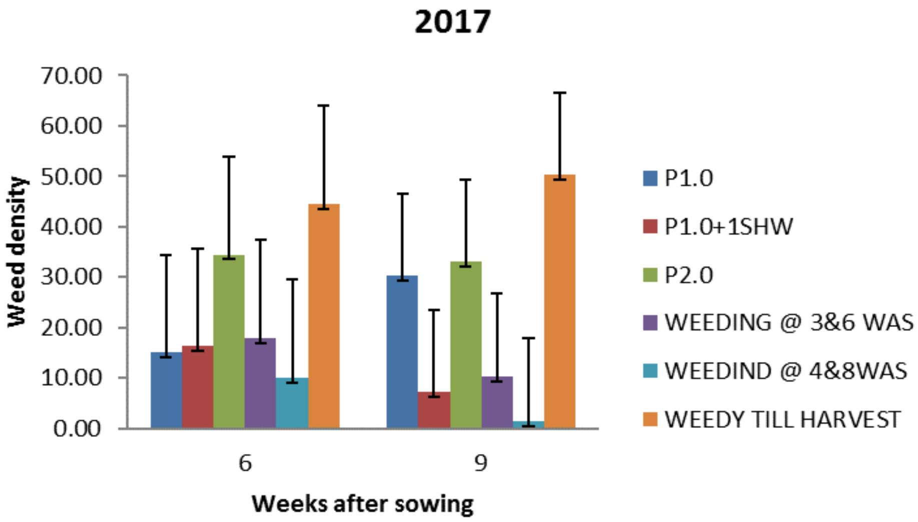

Figure 13. The effect of weed control on weed density (2017).
LSD (0.05) at 6 WAS = 19.44; LSD (0.05) at 9 WAS = 16.32.

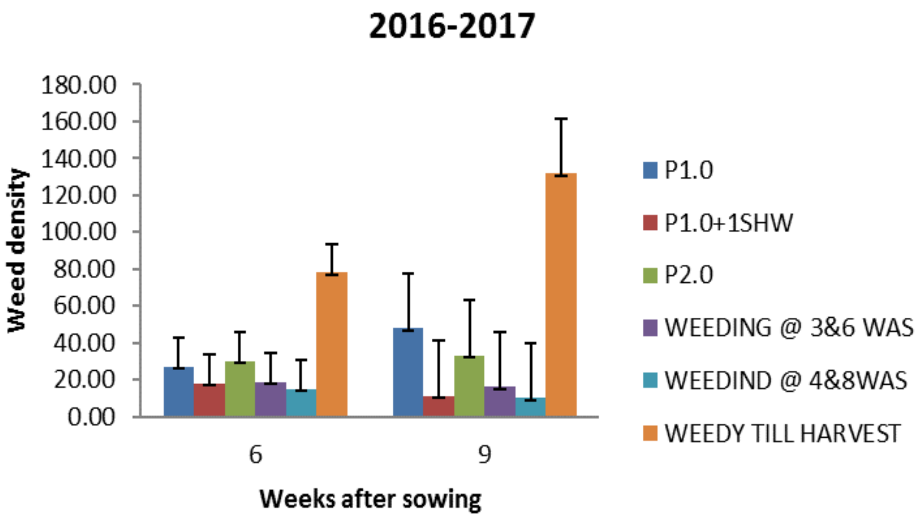

Figure 14. The effect of weed control on weed density at the mean.
LSD (0.05) at 6 WAS = 15.68, LSD (0.05) at 9 WAS = 29.98.

Effect of spacing and pendimethalin-based weed management options on leaf area and number of leaves/plant

There was no significant difference in the leaf area of okra spaced at 60cm x 30cm and 60cm x 50 cm in the two years of the study and the mean (Table 1). However, pendimethalin at 1.0 kg a.i. ha^{-1} plus one SHW gave rise to crops with leaf areas significantly larger than those from other treatments, but was comparable with two hoe weedings at 3 and 6 and 4 and 8 WAS (Table 2).

Table 1. The effect of spacing and weed control method on leaf area of okra (cm).

	Leaf area/plant					
Treatment	6 WAS			9 WAS		
	2016	2017	Mean	2016	2017	Mean
Plant spacing (S)						
60cm x 30cm	69.7	54.3	62.0	87.1	76.9	81.9
60cm x 50cm	74.2	56.3	65.2	88.4	72.3	80.4
LSD (0.05)	11.99	7.57	9.07	14.12	9.86	10.10
Method of weed control (WC)						
Pendimethalin at 1.0kg a.i.ha^{-1}	72.2	50.9	61.8	86.4	70.9	78.6
Pendimethalin at 1.0kg a.i./ha^{-1} + one SHW at 6 WAS	109.7	49.5	79.6	129.4	79.6	104.5
Pendimethalin at 2.0kg a.i./ha^{-1}	65.7	55.5	60.6	85.6	73.3	79.5
Hoe weeding at 3 and 6 WAS	81.6	64.9	73.3	108.9	80.4	94.7
Hoe weeding at 4 and 8 WAS	78.9	65.2	72.1	90.8	89.2	90.0
Weedy check	22.9	45.9	34.4	25.3	54.3	39.8
LSD (0.05)	20.90	13.10	15.72	24.45	17.08	17.49
Interaction S X WC	NS	NS	NS	NS	NS	NS

SHW = Supplementary hoe weeding; WAS = Weeks after sowing.

Similarly, okra crops spaced at 60cm x 50cm produced a significantly higher number of leaves than narrower spaced crops at 9 WAS in 2016 and in the average for two years (Table 2). In addition, a pre-emergence application of pendimethalin at 1.0 kg a.i.ha^{-1} plus one SHW resulted in crops with a higher number of okra leaves, which was significantly different from crops in other treatments except for two hoe weedings at 3 and 6 and 4 and 8 WAS at 9 WAS in 2017. There was no significant interactive effect between spacing and weed control methods on the number of leaves/plant (Table 2). The higher rainfall coupled with wider spacing of okra enhanced the uptake of growth factors which could have been responsible for the production of a significantly higher number of leaves in 2016. The effectiveness of weed control methods, namely, pendimethalin at 1.0 kg a.i./ha^{-1} plus one SHW at 6 WAS and two hoe weedings minimized weed infestation significantly. This freed enough growth resources of moisture, light and soil nutrients for better performance of the crop. This agrees with the report of Jalendhar et al. (2012) that integrated weed management is more effective in controlling weeds.

Table 2. The effect of spacing and method of weed control on the number of leaves/plant.

	Number of leaves/plant					
Treatment	6 WAS			9 WAS		
	2016	2017	Mean	2016	2017	Mean
Plant spacing (S)						
60cm x 30cm	6.08	6.5	6.3	7.5	8.9	8.2
60cm x 50cm	5.9	6.9	6.4	9.3	8.9	9.1
LSD (0.05)	0.23	0.48	0.31	1.02	1.06	0.82
Method of weed control (WC)						
Pendimethalin at 1.0g a.i.ha^{-1}	5.8	6.7	6.2	7.4	9.1	8.2
Pendimethalin at 1.0kg a.i./ha^{-1} + one SHW at 6 WAS	7.2	6.8	7.0	11.8	10.2	11.0
Pendimethalin at 2.0kg a.i./ha^{-1}	6.3	6.3	6.3	8.6	8.2	8.4
Hoe weeding at 3 and 6 WAS	6.1	7.6	6.9	10.7	10.5	10.6
Hoe weeding at 4 and 8 WAS	5.9	6.8	6.3	8.1	10.2	9.1
Weedy check	4.8	5.9	5.4	3.9	5.5c	4.7
LSD (0.05)	0.39	0.84	0.53	1.77	1.84	1.43
Interaction S X WC	NS	NS	NS	NS	NS	NS

SHW = Supplementary hoe weeding; WAS = Weeks after sowing.

Effect of spacing and weed control methods on yield components and yield of okra

Spacing had a significant influence on the total number of pods and fresh pod yield of okra in 2016 and the mean of two years (Table 3). Okra crops spaced at 60cm x 30cm recorded a significantly higher total number of pods and fresh pod weight than those spaced at wider spacing. The utilization of adequate growth resources resulting from better weed control and the higher population of okra plants in plots treated with narrower spacing could have accounted for a higher number of okra pods and better performances. This agrees with the findings of Paththinige (2008) and Okunowo (2012) that narrow plant spacing produced higher yield and fresh weight of okra. The higher rainfall recorded in 2016 compared to 2017 could have caused the significant difference in yield of okra between the two spacings of 60cm x 30cm and 60cm x 50cm. Plots treated with pendimethalin at 1.0 kg a.i.ha^{-1} plus one SHW produced significantly higher fresh okra pod weight than those from the other treatments in both years and their means. This has further proved the efficacy of this method of weed control which was able to minimize weed infestation and enhance the utilization of growth resources and assimilate for the production of a higher number of pods and fresh pod weight of okra. This is in line with the report of Jalendar et al. (2012), that integrated weed management produced the highest total number of pods and fresh weight of okra.

Table 3. The effect of spacing and the method of weed control on the total number of pods and fresh okra pod yield.

Treatment	Total number of pods/plot			Fresh pod weight		
	2016	2017	Mean	2016	2017	Mean
Plant spacing (S)						
60cm x 30cm	91.8	95.0	93.4	904.5	1104.7	1004.6
60cm x 50cm	68.2	86.1	77.1	665.4	967.5	816.5
LSD (0.05)	18.76	16.95	15.07	200.34	256.97	181.99
Method of weed control (WC)						
Pendimethalin at 1.0kg a.i.ha^{-1}	74.0	72.0	73.0	705.5	776.1	740.8
Pendimethalin at 1.0kg a.i. ha^{-1} + one SHW at 6 WAS	149.3	122.0	135.7	1519.4	1445.6	1482.5
Pendimethalin at 2.0kg a.i. ha^{-1}	61.5	54.0	57.9	574.2	605.2	589.7
Hoe weeding at 3 and 6 WAS	112.3	128.8	120.6	1109.3	1608.35	1358.8
Hoe weeding at 4 and 8 WAS	66.5	144.0	105.3	648.1	1618.9	1133.5
Weedy check	16.3	22.0	19.2	153.3	162.42	157.9
LSD (0.05)	32.50	29.36	26.10	347.01	445.08	315.21
Interaction S X WC	NS	NS	NS	NS	NS	NS

SHW = Supplementary hoe weeding; WAS = Weeks after sowing.

Economic performance of the interaction of spacing and methods of weed control

In both years and their mean, plots treated with pendimethalin at 1.0 kg a.i. ha^{1} plus one SHW with the crop spacing of 30cm x 60cm had yields significantly greater than the other treatments. However, this was comparable to two hoe weedings at 3 and 6 WAS in 2017 and the mean (Table 4). The highest cost of production (N116, 000.00) was incurred in plots treated with two hoe weedings at 3 and 6 and 4 and 8 WAS followed by those treated with a combination of pendimethalin at 1.0 kg a.i.ha^{-1} plus one SHW at 6 WAS. On the other hand, the cost of weed control in plots treated with only pendimethalin and a weedy check were lower. Manual weed control has been reported to be an expensive weed control method (Imoloame, 2013, 2014, 2017). The additional cost of one SHW at 6 WAS increased the cost of weed control in plots applied with pendimethalin at 1.0 kg a.i. ha^{-1} plus one SHW (Table 4). The pre-emergence application of pendimethalin at 1.0 kg a.i. ha^{-1} plus one SHW together with the crop spacing of 60cm x 30cm generated the highest revenue (N239, 708.00 ha^{-1}) followed by two hoe weedings at 3 and 6 WAS with the crop spacing of 60 cm x 30cm. Revenues from plots that received only herbicide treatments and a weedy check generated the lowest revenue. A similar trend was recorded with regards to gross margin as pendimethalin at 1.0 kg a.i. ha[1] plus one SHW and two hoe weedings with the spacing of 60cm x 30cm resulted in the highest gross margins, N124, 542.00 and N76, 374.00 ha^{-1} respectively. This is a demonstration of the ability of integrated weed management to increase okra yield and enhance revenue and profitability. This result is similar to the findings of Imoloame (2013) in which the application of herbicide mixture plus one SHW at 6 WAS resulted in higher yield and profitability from the production of soybean.

The lowest cost: benefit ratio was recorded under a treatment combination of the pre-emergence application of pendimethalin at 1.0 kg $a.i.ha^{-1}$ plus one SHW and with the spacing of 60cm x 30cm. The significant increase in okra yield caused by this method of weed control could have been responsible for this.

Table 4. The effect of spacing and the method of weed control on the economics of production of okra.

Treatment	Yield kg/ha		Mean	Average cost (₦)	Average revenue(₦)	Gross margin (₦)	Cost: benefit ratio
	2016	2017					
P1.0kga.i,/ha^{-1} x 60x30cm	780.2a[1]	1001.2bc	890.7bcd	106,500.00	124,698.00	18,198.00	1:0.854
P1.0kga.i. ha^{-1} x60x50cm	630.8def	551.3cd	590.9cdef	106,500.00	82,600.00	-23,900.00	1:1.289
P1.0 kg a.i..+One SHWx60x30cm	1837.9a	1586.5ab	1712.2a	115,160.00	239,708.00	124.542.00	1:0.480
P1.0kg+One SHWx60x50cm	1200.9bc	1304.7ab	1252.9ab	115,166.00	175,406.00	60,240.00	1:0.657
P 2.0 kg $a.i.ha^{-1}$ x 60x30cm	685.2cde	614.3cd	649.6cde	111,555.00	90,944.00	-20,611.00	1:1.227
P2.0 kg $a.i.ha^{-1}$ x 60x50cm	463.2def	596.3cd	529.8def	111,555.00	74,172.00	-37,383.00	1:1.504
Weedy at 3&6 WAS x60x30cm	1320.4b	1427.9ab	1374.1ab	116,000.00	192,374.00	76,371.00	1:0.603
Weedy at 3&6 WAS x60x50cm	898.2bcd	1788.8a	1343.6ab	116,000.00	188,104.00	72,104.00	1:0.617
Weedy at 4&8 WAS (60x30cm)	619.3def	1812.5a	1215.9b	116,000.00	170.226.00	54,226.00	1:0.681
Weedy at 4&8 WAS x60x50cm	677.3cde	1425.4ab	1051.9bc	116,000.00	147.266.00	31,266.00	1:0.788
Weedy check x 60x30cm	184.0ef	185.9d	184.9ef	98,000.00	25,886.00	-72,114.00	1:3.786

SHW = Supplementary hoe weeds; Market price of okra in 2016/2017 = N140.00/kg; P = Pendimethalin.

Conclusion

The treatment combination of pendimethalin at 1.0 kg a.i.ha^{-1} integrated with one SHW at 6 WAS together with the spacing of 60cm x 30cm caused a significant reduction in weed infestation and enhanced the pod yield of okra. This combination was also found to be more cost-effective and increased cash returns compared to other treatments. Therefore, this method of weed control together with the above-mentioned spacing can be recommended to farmers in the southern Guinea savanna of Nigeria.

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UTICAJ GUSTINE SETVE I HERBICIDA PENDIMENTALINA NA BIOMASU KOROVA I PRODUKTIVNOST BAMIJE (*ABELMOSCHUS ESCULENTUS* (L) MOENCH)

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R e z i m e

Poljski ogledi su sprovedeni na oglednoj stanici Državnog univerziteta u Kvari, Malete, Nigerija, kako bi se odredilo uticaj gustine useva i načina kontrole korova na zakorovljenost, rast i prinos bamije tokom vegetacionog perioda 2016. i 2017. godine. Ogled se sastojao od dvanaest tretmana koji su obuhvatali šest načina kontrole korova i dve gustine useva. Kontrola korova sastojala se od primene pendimetalina pre nicanja korišćenjem doze 1,0 kg a.m. ha^{-1}, pendimetalina doze 2,0 kg a.m. ha^{-1}, pendimatalina doze 1,0 kg a.m. ha^{-1} + jednog dodatnog ručnog okopavanja 6 nedelja posle setve, dva ručna okopavanja 3 i 6 nedelja posle setve, plevljenja 4 i 8 nedelja posle setve i apsolutne kontrole (bez kontrole korova). Oba međuredna rastojanja u usevu bamije bila su ista 60 cm, s tim da je u jednoj varijanti rastojanje između biljaka u redu bilo 30 cm, a u drugoj 50 cm. Ovi tretmani su postavljeni po potpuno slučajnom blok sistemu sa podeljenim parcelama u tri ponavljanja. Rezultati su pokazali da je tretman sa većom gustinom biljaka (60 x 30 cm) smanjio zakorovljenost i doveo do povećanja ukupnog broja mahuna i sveže mase bamije, dok se u tretmanu sa pendimetalinom u dozi od 1,0 kg a.m. ha^{-1} + jedno ručno okopavanje 6 nedelja posle setve smanjila zakorovljenost i u njoj je dobijen najveći ukupan broj mahuna i prinos bamije. Ova kombinacija, takođe je imala i veći ekonomski efekat.

Ključne reči: načini kontrole korova, produktivnost bamije, južnogvinejska savana, Nigerija.

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A CORRELATION BETWEEN THE INCREASED TEMPERATURES AND THE PRODUCTIVITY OF LADANG IN KUTAI BARAT REGENCY, THE PROVINCE OF EAST KALIMANTAN, INDONESIA

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Abstract: The studies on the impact of global warming, particularly in the field of food production, could provide more accurate analysis to support a policy of food security. This research was conducted to find out the trends and the impact of the increase in the air temperature on the production of ladang (swidden field) in the Kutai Barat Regency. The results of the research carried out in some of the traditional rice fields indicate a technology that has barely changed, and organic based cultivation. During the period 1990–2015, air temperature increased leading to daily average of 21.2ºC–25.2ºC or an average increased by 0.16ºC/year. In the same period, the productivity of ladang increased by 1.872–3.195 kg ha^{-1} or an average increased by 261 kg/hectare/year. Linear regression analysis results at the 5% level of significance showed the existence of the real correlation (r=0.7) between the average air temperature change and the productivity of the paddy fields.

Key words: global warming, swidden fields, productivity, correlation, organic cultivation, temperature.

Introduction

The impact of changes in the dynamics of the weather at increased temperatures has been actually identified since early 1990s. Indonesia seems to have not prepared a comprehensive policy and operational strategies to adapt to changes in the dynamics of global weather. Some of the recommendations of the World Development Report (2008) are as follows: plant varieties that are highly adaptive, change the planting period adjusting to weather, and practice agriculture with a shorter growing period. In the context of Indonesia, farmers

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have a high level of vulnerability. In addition, due to the very small land holdings as well as weak access to various agricultural inputs as well as limitations on market access and agricultural processing, farmers' knowledge and 'know how' are very minimal about strategy on the adaptation of agricultural production to changes in global weather dynamics. Therefore, it is urgent to determine the impacts of climate change on crop production. This issue is worthwhile of concern as Eitzinger et al. (2010) have noted that such variability has undesirable effects on food production. This agronomic impact of climate variability on crop yields could trigger economic impulses reflected through its effects on agricultural prices, production, demand, trade, regional comparative advantage and producers' as well as consumers' welfare (Li et al., 2011). A considerable amount of literature has been published on paddy production. These studies show that its cultivation is constrained by factors such as solar radiation, temperature and water.

In East Borneo (Indonesia), shifting cultivation is a common practice for farmers to meet their own personal needs and the needs of fellow villagers (Ave and King, 1986). Swidden or shifting cultivation is an agricultural system practiced mainly in the tropics and is very prevalent in Indonesia (Angelsen, 1995). A so-called lading (swidden field) is planted with rice for one to several years, and rice is often intercropped with other useful food crops such as chilli peppers, cassava or bananas. When sufficient time has passed to restore fertility and reduce the weed population and agricultural pests, the field will be cleared and then reused for cultivation. The lading and fallow are traditionally controlled by the individual (usually male) by whom they were originally cultivated, or by his descendants. This swidden model has historically been practiced by upriver indigenous Dayaks (Inoue and Lahjie, 1990).

Materials and Methods

Kutai Barat Regency, the province of East Kalimantan, is geographically located at 113°45'05"–116°31'19" L and between 1°31'35"N and 1°10'16"S. The total area of the County reaches 31.629 km^2. The topography is dominated by the mountains comprising 1,586,552.08 hectares (approximately 50%), while the flat topography of the region amounts to 10.35% or 327,400.84 hectares. Most of its territory is covered by forests of dipterocarpaceae lowlands (Anonymous, 2012).

The research was carried out in Kutai Barat Regency in several villages where there are still the actively managed traditional fields of rice cultivation system. We have used descriptive research methods, i.e., methods that discuss several possibilities to solve the problem with the actual path of collecting data, compiling, analysing, and interpreting them. The descriptive method of analysis was applied by describing the facts, followed by analysis, data analysis, research conducted using linear equations with two variables in the form of the following equation:

Y = a + bX, where,

Y = the change in productivity of rice fields ($kgha^{-1}$) and

X = annual air temperature changes averaging (^{o}C).

A simple correlation analysis (bivariate correlation) is used to find out the significance of the relationship between the two variables (productivity of paddy fields and temperature changes) as well as the direction of the relationship going on with the Pearson method:

$$r_{xy} = \frac{n\sum XY - \sum X \sum Y}{\sqrt{(n\sum X^2 - (\sum x)^2)(n\sum Y^2 - (\sum Y)^2)}} \qquad \text{Eq. 1}$$

Results and Discussion

The research results of diversity of local rice Kutai Barat are the capital base that is very valuable for the development of the agricultural sector of food crops in support of the national food self-sufficiency program. Using microsatellite markers 30, Thomson et al. (2009) obtained 183 local rice cultivars collected from 15 villages along the Bahau and Kayan. From the fields of rice, 183 cultivars were analysed using DNA markers through the kinship, 80% of cultivars were identified and grouped into tropical Japonica and 20% were identified as Indica. In addition, Soedjito (1999) identified at least 44 local rice cultivars in the area of Kantu. In addition, Nurhasanah and Sunaryo (2015) collected 44 local rice cultivars in Kutai Barat Regency, consisting of 39 rice and 5 glutinous rice cultivars. Figure 1 showed that land area of paddy fields continued to decline in the last 10 years with the lowest extents in the year of 2015 (6,403 hectares) in comparison to the year of 1990 when the the productivity of paddy fields showed the highest value of 3.195 kg ha^{-1} (Central Bureau of Statistics[a-e]). The vast acreage of paddy fields decreased due to the more active awareness of traditional belief not to open fields with a 'slash and burn methods'. Therefore, more investors engaged in the plantation sector managed the traditional fields of the community as the concession (working area).

Air temperature data obtained from various institutions (Figure 2) shows an increase in the annual average temperature which is linier over a period of 25 years, 21.2–25.2ºC (Central Bureau of Statistics[a-e]). Therefore, in Kutai Barat Regency, air temperature changed by 4.0ºC or increased by 0.16ºC$year^{-1}$. The relationship between changes in the production of paddy fields (Y) and the change (increase) in the air temperature (X) is shown through the equation Y = 1.66 + 0.85X, significant and strongly correlated (r^2 = 0.48).

Changes in air quality, microclimate, and soil moisture will produce bio-climates for new production systems of agriculture, particularly rice production systems. The main characteristic of the bio-climate among others is the CO_2 concentration, the higher the temperature the more heat, and extreme climate (El-Nino/La-Nina) will occur more frequently. International Rice Research Institute (IRRI) synthesises the influence of climate parameters on changing climate conditions, against the result and the production of rice. The increase in CO_2 gives rise to a positive impact on the biomass of rice, but depends on decreased results due to the rise in air temperature. For every 75 ppm CO_2 concentration increases, the yield of rice will rise by 0.5 t ha^{-1}. However, the result of rice will drop by 500 kg ha^{-1} for every $10^{\circ}C$ rise in temperature. The results of research using the FACE (free-air CO_2's) showed that the increase in yield due to CO_2 concentration is not as big as the one of a study using a closed system (enclosure chambers). The studies about wheat production affected by climate change are mainly concerned with future CO_2 concentrations (Lal, 2005; Yinhong et al., 2009).

Temperature rise and extreme weather events are one of the indicators of the changes in the dynamics of the weather (Mirza, 2003). Temperature is an indication of the amount of heat energy that is contained in a system or the mass. Temperature affects plants through its influence on the rate of metabolic processes. In addition, the influence of temperature is also seen on the development, the establishment of leaves, the initiation of productive organ, the maturation of the fruit and the age of the plant. Rising temperatures will accelerate the process of photosynthesis and biochemistry of plant development and accelerate the process of respiration. Respiration is limited to the oxidation of carbohydrates into CO_2 and H_2O (Las et al., 2007; Amthor, 2002). Temperature increases crop development to some extent. The relationship of temperature with plant growth shows a linear relationship to a certain extent, having reached the point of maximum (peak) of the relationship of the two variables that indicates the relationship of the parabola.

Temperatures increase the rate of growth to form a straight line (linear) where the curve is the exponential function together with temperature. At this stage, the heat energy can turn the entire system (device) growth. Hence, the efficiency of the use of thermal energy by plants is large. Thermal energy is being wasted on a small amount of heat energy, or captured molecules can increase the movement of molecules in the tissues of plants (Allen, 2000; Anwar et al., 2007; Koesmaryono et al., 2002). Research shows that the productivity of rice in China will decrease by 5–12% in a temperature increase of 3.6ºC.The same case on wheat production in Bangladesh is decreasing as its neighbors in 2050 compared to current production if a temperature increase occurs. Possible effects of global warming on the rice cultivation in Indonesia are not much different than in

China or Bangladesh, or perhaps they are much worse during the long dry season when the rainy season fails to come (Challinor, 2008; Jerry and Prueger, 2015).

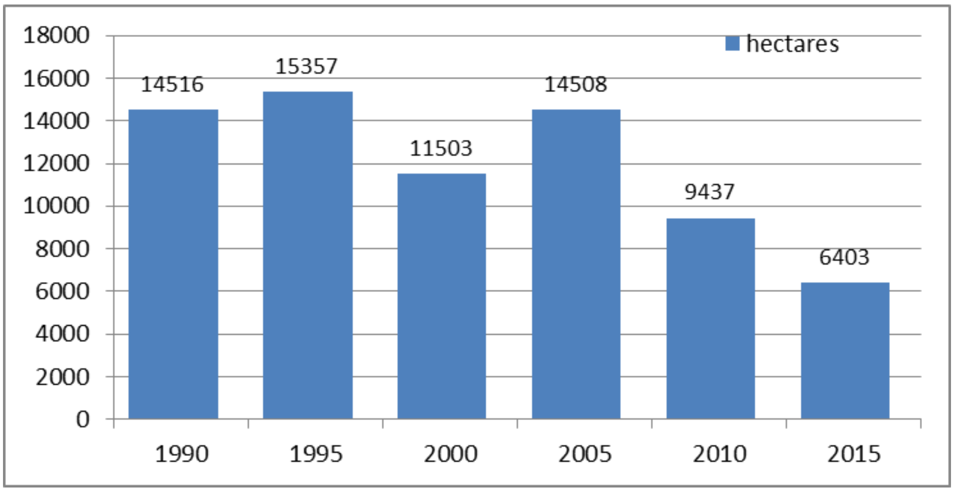

Figure 1. Land area of paddy fields.
(Central Bureau of Statistics[a-e])

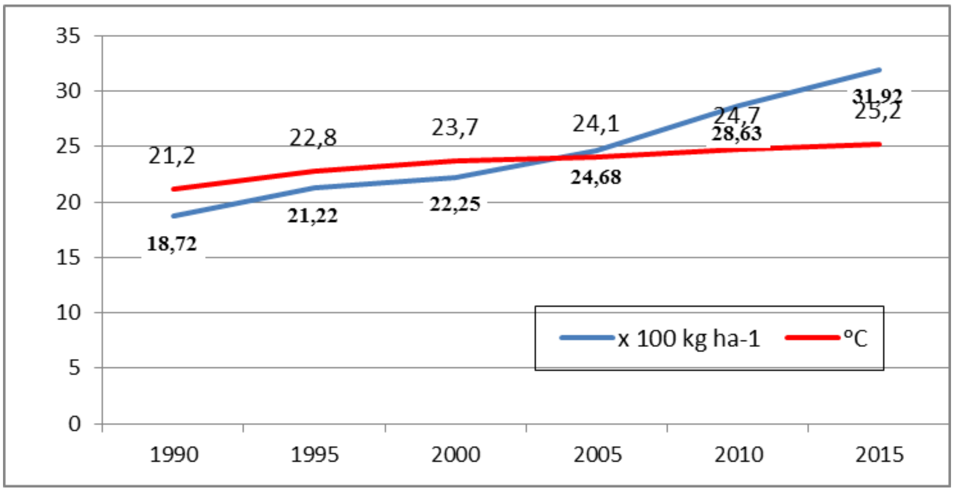

Figure 2. Temperature average (°C).
(Productivity kgha^{-1})

The contribution of Kutai Barat Regency in sufficient food needs (rice) nationally is still low, although it has the potential of vast paddy fields. This is due to the level of a relatively low soil fertility causing the productivity of rice (the rice and paddy fields) in Kutai Barat to be low, i.e. 2.5 to 3.5 tha^{-1}. Regarding the rice plant, a floret, anthers and pollen are more sensitive to high temperature than ovules, and floret sterility at temperatures ≥30°C has been correlated with diminished anther dehiscence, production of fewer pollen grains, pollen sterility, and reduced *in vivo* pollen germination. Grain sterility temperature quickly rises, more than 35°C (Mitsui et al., 2000; Jagadish et al., 2010), and the increase in CO_2 in conjunction with high temperatures can make things worse, because of the fall in the cooling of the plant through transpiration (Amthor, 2001; Yinhong et al., 2009).

The average air temperature in the House plant belongs to the optimum temperature for rice crops, namely within the range of 20–28ºC (Yamaguchi, 1983). Salisbury and Ross (1995) describe the thermo-periodisme that is a phenomenon that shows that the growth and development of crops are enhanced by the day and night temperature alternately. The establishment of a rice grain enhanced by a low night temperature rise in the temperature of the night has been the main cause of the rise in global temperatures since the mid-20th century and the results showed negative correlations of rice with nighttime temperatures. The reason for this negative correlation is variation of solar radiation, the loss due to respiration or the effect of differential influence of night temperature. A significant difference of grain yield between low night temperature and high night temperature treatment was observed in all experiments. Grain yield at high night temperature was decreased by 16.7%, 9.1%, 9.6% and 8.0% in the four consecutive seasons, respectively (Sheehy et al., 2005)

Warming will accelerate a lot of processes in the system soil-microbiology puddles, which is consequently in a cycle of N and C. Soil temperature rise can also raise the CO_2 auto-trop from land loss due to roots, root exudate and turnover of fine roots. Rice plant that grows at high ground temperature can change the partition of C and N compared with this growing at low soil temperatures (Lynch and St. Clair, 2004). The temperature of the heat is the appropriate temperature for rice crops. A rice plant can grow well at a temperature of 24–30ºC. The optimum temperature is the right temperature for rice growth and development circumstances. The relationship of temperature with plant growth is explained in a ‘remainder index’ or heat unit, i.e. a method of approach between agronomy and climatology. Temperature of a raw plant was measured in a controlled experiment in the growth chamber. Cardinal temperature is a temperature point that shows the non-occurrence of the physiological processes of the plant. Raw temperature varies for each plant and in every process of development. Examples of raw temperatures are as follows: for potato 7.2ºC, corn 10ºC, rice 10ºC, soybean 7.8ºC and cotton 16.6ºC (Hatfield et al., 2011). Figure 2 shows the temperature conditions based on an annual average in the center of rice fields are in the optimum temperature range for rice cultivation including the paddy fields.

Conclusion

The area of paddy fields at Kutai Barat Regency shows a declining trend, with increasing air temperature positively and strongly (r^2 = 0.48) with increased production of rice fields, because the temperatures of the center of paddy production are in the optimum temperature range.

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KORELACIJA IZMEĐU POVEĆANIH TEMPERATURA I PRODUKTIVNOSTI NA GAZDINSTVIMA U ADMINISTRATIVNOJ OBLASTI KUTAI BARAT, PROVINCIJA ISTOČNI KALIMANTAN, INDONEZIJA

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R e z i m e

Istraživanja o uticaju globalnog zagrevanja, posebno u oblasti proizvodnje hrane, mogla bi pružiti tačniju analizu, koja bi podržala politiku prehrambene sigurnosti. Ovo istraživanje je sprovedeno kako bi se utvrdili trendovi i uticaj povećanja temperature vazduha na proizvodnju na gazdinstvu (raskrčeno zemljište) u administrativnoj oblasti Kutai Barat. Rezultati istraživanja sprovedenog na tradicionalnim pirinčanim poljima obuhvataju tradicionalnu tehnologiju i organski sistem proizvodnje. Tokom perioda 1990–2015, temperatura vazduha se povećala vodeći ka dnevnom proseku od 21,2ºC–25,2ºC i prosečno se povećala za 0,16ºC godišnje. U istom periodu, produktivnost proizvodnje na gazdinstvu povećala se za 1,872–3,195 kg ha^{-1}, što je u proseku bilo povećanje od 261 kg po hektaru godišnje. Rezultati linearne regresione analize na nivou značajnosti od 5% pokazali su da postoji realna korelacija (r=0,7) između promene prosečne temperature vazduha i produktivnosti pirinčanih polja.

Ključne reči: globalno zagrevanje, raskrčeno zemljište, produktivnost, korelacija, organska poljoprivreda, temperatura.

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IMPROVING THE METABOLISABLE ENERGY VALUE OF BREWERS' DRIED GRAINS WITH ENZYME COCKTAILS IN POULTRY NUTRITION

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Abstract: The determination of the positive effects of exogenous enzymes is essential to ensure their inclusion in poultry feed formulation. This study was conducted to determine the effect of enzymes on the apparent metabolisable energy (AME) value of brewers' dried grain (BDG). Xylanase, phytase and multipurpose enzymes were used in a completely randomised design to determine the effects of individual exogenous enzymes and their cocktails on poultry metabolisable energy using adult cockerels. There were eight treatments comprising a control and seven experimental treatments with BDG and one, two or three enzymes. The AME values were determined using the intubation method. Data collected were analysed using the statistical analysis system. Enzymes individually and as a cocktail improved the AME value of BDG compared to the control. An increase in the AME value was 3.48%, 5.39%, 5.92%, 14.29%, 18.13%, 23.21% and 29.58% respectively for phytase, xylanase, cocktail of xylanase and phytase, multipurpose enzyme, cocktail of multipurpose enzyme and phytase, cocktail of xylanase and multipurpose enzyme and cocktail of xylanase, phytase and multipurpose enzyme. Cocktails of enzymes were significantly better ($P<0.05$) than individual enzymes in their effects on apparent metabolisable energy of BDG. Phytase gave a marginal increase in AME of the studied feedstuff. It has been concluded that the cocktail of enzymes is better than individual enzymes in their effects on AME of BDG. If different enzymes are available, it is recommended that the enzyme with higher units should be used.

Key words: cockerel, enzyme, cocktail, metabolisable, energy, intubation.

Introduction

Brewers' dried grain (BDG) is a solid waste from the brewery industries. It is available and cheap, but difficult to dry to low moisture content for easy storage and use, especially during the raining/wet season. Breweries in Nigeria use cereals

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such as wheat, maize, millet and sorghum in different combinations, which vary from one brewery to another. This, therefore, has resulted in the production of BDG with variable physical and chemical composition (Oluponna et al., 2002). Brewers' dried grain is a good source of quality protein and dietary fibre for the poultry, with a good amino acid profile, high mineral and B-vitamin content (Ironkwe and Bamgbose, 2011). It does not form food for humans nor does it have any other industrial use for now. It is usually dried and sold as feedstuff for livestock. Brewers' dried grain is reported to have better available protein, energy and ash composition than maize and wheat offal (Aletor, 1986; Babatunde, 1989).

The nutritional value of BDG in poultry feeding is limited by its fibrous nature though it has a relatively high protein value of about 19–25% (Kwari et al., 1999). Onifade and Babatunde (1998) reported that broilers fed BDG-based diet had poor efficiency of feed utilisation and this was attributed to its high crude fibre. The presence of this high crude fibre with non-starch polysaccharide (NSP) limits its desirability in poultry diets (Kwari et al., 1999; Oluponna et al., 2002). Brewers' dried grains have 33.02% of hemicellulose and 34.20% of cellulose and these are polymers of xylans and arabinans. Xylans play an important role in the integrity of the plant cell wall and increase cell wall recalcitrance to enzyme digestion (Faik, 2013). Comwell et al. (1993) reported that dietary inclusion of BDG was normally limited to 5–10% in swine and poultry diets. Brewers' dried grain has 10–22% crude fibre, metabolisable energy value of 7379.82 Kj/kg (Dogari, 1985) and gross energy value of 19339.54 Kj/kg (Amaefule et al., 2009). High fibrous feedstuff like BDG has energy locked up in the fibre content and this cannot be digested by the monogastric animal. Therefore, to enhance its nutritional value as a poultry feeding, exogenous enzymes have been added. Olajide et al. (2013) reported that substitution of 20% of maize with BDG supplemented with *Grindazyme*® enzyme resulted in better performance and gave a higher net profit compared with other treatments. It was concluded that BDG with enzyme supplementation could be adopted to alleviate the problem of high costs of maize. In addition, because of the difference in profile of enzymes and the complex nature of crude fibre, the need for the cocktail of enzymes has been suggested.

Jimoh and Atteh (2017) reported that cocktails of enzymes were significantly better than the respective individual enzymes in their effects on *in vitro* digestibility of fibre fractions of BDG. It was reported that the enzyme cocktails significantly improved the measured parameters, namely crude fibre, fibre fractions and crude protein compared to individual enzymes and the control treatment. It was, therefore, the objective of this study to determine the effect of exogenous enzymes individually and as a cocktail on the apparent metabolisable energy of brewers' dried grains in adult cockerels.

Materials and Methods

Experimental design

Twenty-four (24) Harco black adult cockerels of twenty-six weeks of age and about 2.2 kg each were randomly allotted to the experimental treatments. There were eight treatments comprising one control and seven experimental treatments in a completely randomised design as shown in Table 1. Each treatment was replicated three times with one bird per replicate. Three enzymes, namely xylanase, multipurpose enzyme and phytase were used individually and as cocktails following the manufacturers' recommended inclusion levels. The choice of xylanase and multipurpose enzymes was based on the cellulose and hemicellulose content of brewers' dried grains. BDG has 33.02% hemicellulose and 34.20% cellulose which are mainly composed of xylans and arabinans. Phytase was included to investigate its ability to release nutrients bound to phytate, especially carbohydrates (Ravindran et al., 1995). For cocktails, the enzymes were included at ratio of 100 ppm: 150 ppm: 150 ppm (xylanase: multipurpose enzyme: phytase).

The xylanase is a bacterial enzyme preparation obtained from *Bacillus substilis* and contains 9000 units of endo-1, 4-β-xylanase enzymes per gram. It is recommended for rations with high content of arabinoxylans which are present in grains and their by-products. As an endoxylanase, it specialises in splitting the glycosidic bonds within the polysaccharide chain. This causes a dramatic decrease in digesta viscosity and increases the liberation of entrapped nutrients. It also produces a large number of smaller fragments of oligosaccharides, each with a reducing terminal monosaccharide.

The multipurpose enzyme is derived from *Trichoderma viride*. Each gram of the enzyme complex has 8000 units of cellulase, 18000 units of β-glucanase and 26000 units of xylanase.

The phytase used is a 3-phytase enzyme obtained from *Aspergillus niger*. It is granular in nature and it has activity of 5000 FTU/gram as stated by the manufacturer. One FTU (phytase unit) is the amount of enzyme which liberates 1 micromole (1 µmol) of inorganic phosphate per minute from sodium phytate at pH 5.5 and 37^{o}C. Phytate usually chelates with cations, proteins and carbohydrates (Ravindran et al., 1995). Thus, a breakdown of the phytate will release other nutrients like protein, carbohydrate and minerals, in addition to phosphorus. This explains why phytase could lead to improvement in the digestibility of other nutrients apart from phosphorus. The use of microbial phytase in poultry diet has increased in response to concerns over phosphorus pollution of effluents from intensive animal farming operations and the skyrocketing price of inorganic phosphates. Phytic acid, also known as 1, 2, 3, 4, 5, 6, hexakisdihydrogen phosphate is an organic phosphate and is abundant in plant seeds. Its salt is known as phytate.

Table 1. Composition of experimental treatments.

Test material	Treatments							
	NE	Xy	Mp	Ph	Xy+Mp	Xy+Ph	Mp+Ph	Xy+Mp+Ph
BDG (%)	100	100	100	100	100	100	100	100
Xy[1] (ppm)	--	100	--	--	100	100	--	100
Mp[2](ppm)	--	--	150	--	150	--	150	150
Ph[3](ppm)	--	--	--	150	--	150	150	150

1: Xylanase enzyme 2: Multipurpose enzyme 3: Phytase enzyme; NE = No enzyme, Xy = Xylanase enzyme alone, Mp = Multipurpose enzyme alone, Ph = Phytase enzyme alone, Xy+Mp = Cocktail of xylanase and multipurpose enzyme, Xy+Ph = Cocktail of xylanase and phytase, Mp+Ph = Cocktail of multipurpose enzyme and phytase, Xy+Mp+Ph = Cocktail of xylanase, multipurpose enzyme and phytase.

Feeding trials

The cockerels were randomly allocated to the battery cage system with one bird in a cell representing a replicate. The birds were provided with *ad libitum* feed and water before the experiment. Brewers' dried grain was obtained from a commercial feed mill in Ilorin, north central Nigeria. It was ground into mash form. The exogenous enzymes were obtained from appointed distributors of the feed additives in Lagos, Nigeria. The feeding trial was done using the intubation method as described by Sibbald (1976) with some modifications. Feed was withdrawn from all the birds for 21 hours prior to the administration of the treatment so as to empty the digestive system. At exactly 21 hours, a cockerel was removed from its cell and a tube of about 8mm of internal diameter was inserted into the crop of the cockerel via the oesophagus. A plastic funnel was placed on top of the tube. Sixty grams of the treatment (BDG plus a respective individual enzyme or a cocktail) in the form of mash was placed in the funnel and gently pushed down with the aid of a glass rod. Water was then added to rinse the feedstuff off the funnel and the tube. After this procedure, the fed bird was returned to the cell and this procedure was repeated for each of the birds. The time for the intubation for each bird was recorded. Immediately after the feeding for each bird, an excreta collection tray was placed under the individual cell and excreta samples were collected over a period of 24 hours after the intubation of all the cockerels. Adequate water was provided for the birds prior to and after the intubation. At exactly 24 hours post intubation, the excreta collection tray was removed from each of the cells. The sample was collected, weighed, oven dried at 72^0C for 24 hours and weighed again.

Gross energy determination

Gross energy determination of the excreta sample was done using a bomb calorimeter (Gallenkamp Ballistic Bomb calorimeter).

Calculations

Apparent metabolisable energy for each treatment was calculated using the formula below:

$$AME\ (kj/g) = \frac{(GE_f \times X) - (y_{ef} - y),}{X} \qquad \text{Eq.1}$$

where:
AME = Apparent metabolisable energy of experimental diet;
GE_f = Gross energy of experimental feedstuff in Kj/g;
X = Weight of feed given to the cockerel (60g);
y_{ef} = Gross energy of feaces of birds fed with experimental treatment in Kj/g;
y = Weight of feaces voided by fed birds in g;
Percentage increase in AME (%**)** =

$$\frac{\text{AME value for treatment - AME value for control}}{\text{AME Value for control}} \text{ X 100.}$$

Statistical analyses

Values obtained for apparent metabolisable energy and percentage increase in apparent metabolisable energy values were subjected to analysis of variance suitable for a completely randomized design using a general linear procedure of the statistical analysis system (SAS, 2002). Significant differences between treatments' means were determined using Duncan`s multiple range test (Duncan, 1955).

Results and Discussion

All the enzymes individually and as a cocktail improved the AME of BDG compared to the control (Table 2). There were significant differences ($P<0.05$) between the individual enzymes and cocktails in their effects on AME of BDG. Among the individual enzymes, multipurpose enzyme gave the highest AME while phytase gave the lowest AME value of 10236.03 Kj/kg, meanwhile, a cocktail of the three enzymes gave the highest AME (12817.64 Kj/kg) and this was significantly different ($P<0.05$) from other cocktails. However, there was no significant difference ($P>0.05$) between the effects of xylanase enzyme and those of the cocktail of xylanase and phytase enzymes.

Table 2. Effects of enzymes on apparent metabolisable energy of brewers' dried grains.

Parameters	Treatments								
	NE	Xy	Mp	Ph	Xy+Mp	Xy+Ph	Mp+Ph	Xy+Mp+Ph	SEM
AME, Kj/Kg	9891.68[g]	10424.43[e]	11304.95[d]	10236.03[f]	12187.57[b]	10477.61[e]	11684.86[c]	12817.64[a]	49.62

[a, b, c, d, e, f] Means in the same row with the same superscript are not significantly different (P>0.05); AME = Apparent metabolisable energy, NE = No enzyme, Xy = Xylanase enzyme alone, Mp = Multipurpose enzyme alone, Ph = Phytase enzyme alone, Xy+Mp = Cocktail of xylanase and multipurpose enzyme, Xy+Ph = Cocktail of xylanase and phytase, Mp+Ph = Cocktail of multipurpose enzyme and phytase, Xy+Mp+Ph = Cocktail of xylanase, multipurpose enzyme and phytase.

There was no significant difference between xylanase and phytase in their effects on AME increment (5.39% vs. 3.48%) as shown in Figure 1. There was also no significant difference (P>0.05) between xylanase and the cocktail of xylanase and phytase in their effects on AME increment of BDG (5.39% vs. 5.92%). The cocktail of the three enzymes gave the highest increment on AME of BDG (29.58%) and this was significantly different from other treatments.

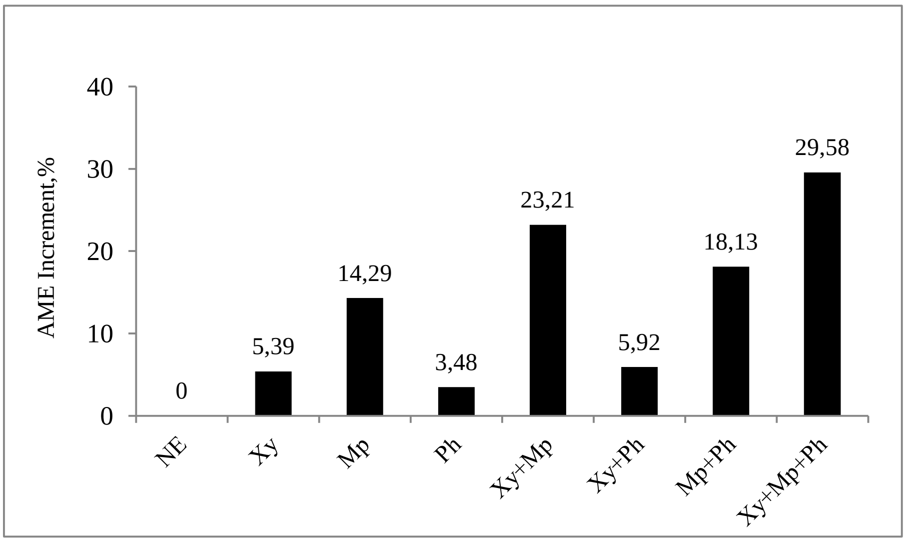

Figure 1. Percentage increase in apparent metabolisable energy value of brewers' dried grains following addition of enzymes.

AME = Apparent metabolisable energy, NE = No enzyme, Xy = Xylanase enzyme alone, Mp = Multipurpose enzyme alone, Ph = Phytase enzyme alone, Xy+Mp = Cocktail of xylanase and multipurpose enzyme, Xy+Ph = Cocktail of xylanase and phytase, Mp+Ph = Cocktail of multipurpose enzyme and phytase, Xy+Mp+Ph = Cocktail of xylanase, multipurpose enzyme and phytase.

Onifade and Babatunde (1998) have reported that the increased inclusion level of brewers' dried grains in broiler diets is known to increase the feed intake but without a corresponding increase in weight gain. This has been attributed to the high fibre level of the feedstuff prompting the birds to eat more so as to satisfy their energy requirement. The excessive use of high fibre sources like brewers' dried grains in the diet may increase the viscosity of the intestinal content with a resulting decrease in the digestion and bioavailability of nutrients which adversely affect body weight gain. Results of the present study showed that enzyme supplementation resulted in the improved AME of brewers' dried grains. Similar results have been reported by Iyayi and Tewe (1998) in layers as well as Alabi et al. (2014) in broilers. Alabi et al. (2014) reported that an increase in the dietary level of BDG without commercial enzyme supplementation significantly decreased weight gain, and increased feed conversion ratio and nutrient digestibility of broilers. Hypertrophy of the digestive organs was also observed in the birds fed brewers' dried grain without enzyme supplementation, but this effect was ameliorated with the inclusion of commercial enzymes.

For energy to be derived from nonstarch polysaccharides, the polysaccharides like cellulose, xylans, arabinans, glycans must be broken down first to oligosaccharides and then to monosaccharides that are absorbable by the animal. Polysaccharides are sugar polymers containing twenty or more monosaccharide units and some have hundreds or thousands of units. Polysaccharides, also called glycans, differ from each other in the identity of their recurring monosaccharide units, in the length of their chains, in the types of bonds linking the units, and in the degree of branching (David and Michael, 2004). Endo-xylanase enzymes specialise in breaking down polysaccharides from within the polymer, and this results in the production of many oligosaccharides each with terminal reducing sugars. However, oligosaccharides cannot be absorbed except if they are broken down further. Exo-xylanase enzymes are then involved in the breakdown of the oligosaccharides into monosaccharides that are absorbable by the villus of the animal. Values obtained for AME in this study are indicative of the efficacy of the individual enzymes and their combinations in performing this role. This implies that the non-starch polysaccharides in the brewers' dried grains have been broken down to oligosaccharides and eventually to absorbable monosaccharides. Furthermore, this ability of the enzymes varies from enzyme to enzyme depending on the profile and activity and this explains the variations obtained in the performance of the individual enzymes. Jimoh and Atteh (2017) reported that the cocktail of xylanase and multipurpose enzymes performed significantly better than the individual enzymes in their effects on *in vitro* digestibility of proximate components and fibre fractions of brewers' dried grains. The efficacy of these enzymes in this regard will depend on the profile and activity of the respective enzymes (McDonald et al., 2010). Thus, among the three enzymes used in this study, the multipurpose enzyme had the highest activity and units of the carbohydrase enzymes. The values obtained in this study are indicative of this attribute of the enzyme complex.

The marginal effect observed for the phytase enzyme in this study was probably due to the matrix effect of enzymes. The enzyme is primarily designed for digestion of phytate. Phytic acid is known to bind with minerals such as calcium, magnesium and other nutrients like protein, carbohydrate and ether extract (Lesson, 1993), and it is, therefore, obvious that phytate-bound nutrients will also be released along with the free phosphorus. This effect known as an ‘extra phosphoric effect’ resulted in improved digestibility of crude fibre, protein and ether extract attributed to phytase compared to the control (Shelton et al., 2004). Thus, it could be inferred that even if the phytase is used for the primary aim (to release phosphorus), its positive effect on fibre, crude protein and ether extract must also be taken into consideration. Therefore, the 3.48% increase in AME of BDG in this study may be ascribed to the ‘extra phosphoric effect’ of phytase. This value is significant to any poultry business and practical application can lead to substantial reduction in feed cost.

Furthermore, results of this study show that the fungal enzyme (the multipurpose enzyme) was better than the bacterial enzyme (the xylanase), both of which are carbohydrases. Fungal enzymes have more active sites than bacterial enzymes (Krisana et al., 2005; Kar et al., 2006) and this effect may have manifested in the better digestibility of the nonstarch polysaccharides in addition to the higher activity of the fungal enzyme (18,000,000 units of xylanase for the multipurpose enzyme compared to 9,000 units for the xylanase). Adeniji and Jimoh (2007) also reported that the multipurpose enzyme was better than the single purpose xylanase enzyme in their effects on the digestibility of the bovine rumen content used as a replacement for maize in the diets of pullets.

Conclusion

Generally, in this study, exogenous enzymes improved energy digestibility and this effect was more pronounced with NSP-degrading enzymes than with the phytase enzyme. An expanded enzyme matrix containing xylanase, cellulase, hemicellulase and β-glucanase combinations gave better results than the individual enzymes. The values of AME obtained in this study are practically useful in the presence of the respective enzymes and their cocktails. The practical application of these values in feed formulation will ensure that the anticipated effects of exogenous enzymes in poultry feed are taken into consideration even during formulation. In conclusion, the cocktails of enzymes in degrading NSP have a future to improve nutrient utilisation of agricultural by-products in poultry.

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POBOLJŠANJE SADRŽAJA METABOLIČKE ENERGIJE U SUVOM PIVSKOM TROPU KORIŠĆENJEM ENZIMSKIH KOKTELA U ISHRANI ŽIVINE

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R e z i m e

Utvrđivanje pozitivnog efekta dodatih enzima je od primarnog značaja za njihovo korišćenje pri formulisanju obroka za živinu. Ovo istraživanje je sprovedeno kako bi se odredio uticaj enzima na vrednost prividne metaboličke energije (engl. *apparent metabolisable energy* – AME) suvog pivskog tropa (engl. *brewers' dried grain* – BDG). Ksilanaza, fitaza i višenamenski enzimi korišćeni su u potpuno slučajnom dizajnu kako bi se odredili uticaji pojedinačnih egzogenih enzima i njihovih koktela na metaboličku energiju za živinu korišćenjem odraslih petlića. Bilo je osam tretmana uključujući kontrolu i sedam eksperimetalnih tretmana sa BDG i sa jednim, dva ili tri enzima. Vrednosti AME određene su korišćenjem metode intubacije. Prikupljeni podaci su analizirani pomoću sistema statističke analize. Enzimi pojedinačno ili kao koktel poboljšali su vrednost AME suvog pivskog tropa u poređenju sa kontrolom. Povećanje vrednosti AME bilo je 3,48%, 5,39%, 5,92%, 14,29%, 18,13%, 23,21% odnosno 29,58% za fitazu, ksilanazu, koktel ksilanaze i fitaze, višenamenski enzim, koktel višenamenskog enzima i fitaze, koktel ksilanaze i višenamenskog enzima i koktel ksinalaze, fitaze i višenamenskog enzima. Kokteli enzima su bili značajno bolji ($P<0,05$) nego pojedinačni enzimi u svojim uticajima na prividnu metaboličku energiju BDG. Fitaza je dovela do marginalnog povećanja AME proučavanih hraniva. Zaključeno je da je koktel enzima bolji nego pojedinačni enzimi u pogledu efekta na vrednost AME suvog pivskog tropa. Ukoliko su različiti enzimi dostupni, preporučuje se da se koriste enzimi sa većom aktivnošću.

Ključne reči: petlić, enzim, koktel, metabolička, energija, intubacija.

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The examples of listing references are the following:

Periodicals

Gvozdenović, S., Saftić Panković, D., Jocić, S., & Radić, V. (2009). Correlation between heterosis and genetic distance based on SSR markers in sunflower (Helianthus annuus L.). *Journal of Agricultural Sciences*, *54*, 1-10.

Books

Steel, R.G.D., & Torrie, J.H. (1980). *Principles and procedures of statistics*. New York: McGraw-Hill Book Company.

Book chapter

Bell, R.L., Quamme, H.A., Layne, R.E.C., & Skirvin, R. M. (1996). Pears. In J. Janick & J.N. Moore (Eds.), *Fruit breeding, Volume I: Tree and tropical fruits.* (pp. 441-514). New York: John Wiley and Sons, Inc.

Proceedings
Behera, T.K., Staub, J.E., Behera, S., Rao, A.R., & Mason, S. (2008). One cycle of phenotypic selection combined with marker assisted selection for improving yield and quality in cucumber. In M. Pitrat (Ed.), *Proceedings of the IXth EUCARPIA meeting on genetics and breeding of Cucurbitaceae* (pp. 115-121). Avignon, France.

Thesis
Singh, N.K. (1985). *The structure and genetic control of endosperm proteins in wheat and rye*. University of Adelaide.

Report
Ballard, J. (1998). *Some significant apple breeding stations around the world*. Selah, Washington.

Web site
Platnick, N.I. (2010). The world spider catalog, version 10.5. *American Museum of Natural History*. Retrieved February 12, 2016, from http://research.amnh.org/entomology/spiders/catalog/index.html

Summary
The summary in Serbian is given at the end of the paper and should comprise 100 to 250 words. Before the main text of the summary, as well as in English, the title of the paper, first name, middle initial(s) and last (family) name of all authors and the names and addresses of affiliations should be given. The title of the summary is centred and written separately. Below the title, the text of the summary should follow, without any indentation, and immediately after the text of the summary, the key words are given with the full stop at the end. The e-mail address of the corresponding author should be given at the bottom of the page.

Tables
Tables numbered with Arabic numerals (1, 2, etc.), followed by the title should be placed in the text using 9 font size and a maximum width of 13 cm. They should be clear, simple and unambiguous. The vertical sections should be avoided, and the number of columns should be limited so that the table is not too wide. Also, an unnecessary usage of horizontal sections should be avoided. The title of the table, single spaced above the table, justified, and with the full stop at the end should be given. The detailed explanation of abbreviations, symbols and signs used in the table should be provided below the table. Each table must be mentioned in the text.

Illustrations
All graphs, diagrams and photographs should be titled "Figure" (1, 2, etc.). They should be placed in the text. Graphs and diagrams should be computer drawn, using 9 font size and a maximum width of 13 cm, so that they can be legible and distinct after the size reduction. The overuse of colours and hues should be avoided for aesthetic reasons. The detailed legend without abbreviations for each graph and

diagram should be given. The photographs must be of high quality so that they can technically be well reproduced. They should be submitted in "TIF" or "JPG" format, and they will be printed in black and white. The title of the illustration should be justified, with a full stop at the end, single spaced from the illustration and given below it. Each illustration should be mentioned in the text.

Abbreviations and units

Only standardised abbreviations should be used in the paper. Measure units should be expressed using International System of Units (SI). The abbreviations can be used for other expressions provided these expressions are stated in the full form when appear for the first time with the abbreviated form in the brackets. Values from 1 to 9 can be written in letters, but others numerically.

Nomenclature

The complete nomenclature (chemical and biochemical, taxonomical, genetic etc.) must be adjusted to international codes and commissions, such as *International Union of Pure and Applied Chemistry, IUPAC-IUB Combined Commission on Biochemical Nomenclature, Enzyme Nomenclature, International Code of Botanical Nomenclature, International Code of Nomenclature of Bacteria* etc.

Formulae

All formulae and equations in the paper should be worked out by means of the programme "WORD Equation". An ample space should be left around the formulae for the sake of visibility. Subscripts and superscripts should be clear. Greek letters and other non-Latin symbols should be explained when they are first used. The meaning of all symbols should be given immediately after the equation where these symbols are first used. Equations should be numbered by Arabic numerals, serially in brackets, at the right-hand side. Each equation must be mentioned in the text as Eq. (1), Eq. (2), etc.

The corresponding author will be sent a free copy of the journal after it has been published.

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Editorial Board of the Journal
Journal of Agricultural Sciences

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Za obradu teksta treba koristiti program MS-Word. Rukopise treba slati u jednom od sledećih formata .doc, .docx, koristiti font Times New Roman, veličina 12, jednostruki prored, margine 2,5 cm. Strane ne treba numerisati.
Originalan naučni rad – Rad koji sadrži prethodno neobjavljivane rezultate sopstvenih istraživanja. Obim ovog rada treba da iznosi od 6 do 12 strana.
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Obavezna poglavlja svakog originalnog naučnog rada i prethodnog saopštenja su sledeća: naslov rada, imena autora, naziv ustanove autora, sažetak, ključne reči, uvod, materijal i metode, rezultati i diskusija, zaključak, zahvalnica, literatura i rezime na srpskom jeziku (ako je rad na engleskom i obrnuto). Pregledni rad mora da sadrži: naslov rada, imena autora, naziv ustanove autora, sažetak, ključne reči, uvod, analizu-diskusiju određene teme, zaključak, literaturu i rezime na srpskom jeziku (ako je rad na engleskom i obrnuto). Ako su radovi na engleskom jeziku, prednost se daje britanskoj varijanti ovog jezika.

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Sažetak
Sažetak je kratak informativni prikaz sadržaja članka koji čitaocu omogućava da brzo i tačno odredi njegovu relevantnost. U interesu je autora da sažetak sadrži termine koji se koriste za indeksiranje i pretraživanje. Sažetak ne sme da sadrži reference. Sastavni delovi sažetka su cilj istraživanja, metode, rezultati i zaključak. Sažetak treba da ima od 100 do 250 reči. Reč „Sažetak“ piše se boldovano i uvlači jednim tabulatorom, nakon čega slede dve tačke, a zatim tekst sažetka.

Ključne reči
Ključne reči su termini ili fraze koje najbolje opisuju sadržaj članka za potrebe indeksiranja i pretraživanja. Broj ključnih reči može biti od 3 do 10. Navode se ispod sažetka. Naslov „Ključne reči“ piše se boldovano i uvlači jednim tabulatorom. Nakon toga slede dve tačke, a zatim nabrajanje ključnih reči malim slovima, sa tačkom na kraju. Treba izbegavati korišćenje ključnih reči koje se nalaze u naslovu rada. Ključne reči se dostavljaju na srpskom i engleskom jeziku posle sažetaka na oba jezika.

Uvod

Uvod treba da sadrži informacije o dosadašnjim istraživanjima po navedenom pitanju i šta se datim istraživanjem želi postići. Prilikom osvrta na literaturu, navesti autora i godinu, a autora citirati u spisku literature. Naslov „Uvod" piše se sa prvim velikim slovom, centrirano i boldovano, nakon čega sa jednim razmakom ispod naslova sledi tekst uvoda poravnat po levoj i desnoj margini. Svaki novi pasus uvlači se jednim tabulatorom. Ova pravila važe i za sva ostala poglavlja.

Materijal i metode

Materijal i metode treba izložiti jasno uz objašnjenje svih primenjenih postupaka u radu. Opšte poznate metode izložiti kratko, a detaljnije ih objasniti ukoliko se odstupa od ranije objavljenih postupaka. Za radove eksperimentalnog karaktera obavezno navesti način statističke obrade podataka. U ovom poglavlju, kao i u poglavlju „Rezultati i diskusija", po potrebi se mogu dati i određena podpoglavlja.

Rezultati i diskusija

U poglavlju „Rezultati i diskusija" interpretiraju se podaci dobijeni na osnovu zapažanja i izvršenih eksperimenata. U komentaru rezultata treba se pozivati na literaturu koja se navodi na kraju rada, čime se obezbeđuje poređenje dobijenih rezultata sa dosadašnjim saznanjima u toj oblasti.

Zaključak

U zaključku treba ukratko navesti najznačajnije rezultate dobijene u radu. Izbegavati nabrajanje svih rezultata istraživanja sa ponavljanjem brojčanih vrednosti koje su prethodno već navedene u poglavlju „Rezultati i diskusija". Zaključak ne sme da sadrži reference.

Zahvalnica

Zahvalnica treba da sadrži naziv i broj projekta, odnosno naziv programa u okviru koga je rad nastao, kao i naziv institucije koja je finansirala projekat ili program.

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Literatura koja je citirana u tekstu navodi se u spisku referenci u originalnom obliku, po abecednom redu, bez numeracije. Ako se citira veći broj radova istog autora najpre se navode radovi kada je autor sam, a zatim kada su prisutna dva i više autora. Ako se u nekoj od ovih kategorija javlja veći broj radova, treba ih hronološki srediti po godinama (1997, 2002, 2006, itd.), a ako se u istoj godini javlja veći broj radova dodaju se slova (2005a, 2005b, 2005c, itd.). Literaturni podatak treba da sadrži: prezime autora, početno slovo imena, godinu izdanja u zagradi, naslov rada, naziv časopisa, volumen i broj stranica (prva-poslednja). Prilikom citiranja knjiga navodi se izdavač i mesto izdavanja. Redovi svake reference posle prvog reda moraju biti uvučeni. U časopisu se koristi APA - Publication Manual of the American Psychological Association citatni stil.

Primeri navođenja referenci su sledeći:

Periodičan časopis

Gvozdenović, S., Saftić Panković, D., Jocić, S., & Radić, V. (2009). Correlation between heterosis and genetic distance based on SSR markers in sunflower (Helianthus annuus L.). *Journal of Agricultural Sciences*, *54*, 1-10.

Knjiga

Steel, R.G.D., & Torrie, J.H. (1980). *Principles and procedures of statistics*. New York: McGraw-Hill Book Company.

Poglavlje u knjizi

Bell, R.L., Quamme, H.A., Layne, R.E.C., & Skirvin, R.M. (1996). Pears. In J. Janick & J.N. Moore (Eds.), *Fruit breeding, Volume I: Tree and tropical fruits.* (pp. 441-514). New York: John Wiley and Sons, Inc.

Zbornik

Behera, T.K., Staub, J.E., Behera, S., Rao, A.R., & Mason, S. (2008). One cycle of phenotypic selection combined with marker assisted selection for improving yield and quality in cucumber. In M. Pitrat (Ed.), *Proceedings of the IXth EUCARPIA meeting on genetics and breeding of Cucurbitaceae* (pp. 115-121). Avignon.

Teza

Singh, N.K. (1985). *The structure and genetic control of endosperm proteins in wheat and rye*. University of Adelaide.

Izveštaj

Ballard, J. (1998). *Some significant apple breeding stations around the world*. Selah, Washington.

Veb sajt

Platnick, N.I. (2010). The world spider catalog, version 10.5. *American Museum of Natural History*. Retrieved February 12, 2016, from http://research.amnh.org/entomology/spiders/catalog/index.html

Rezime

Rezime na srpskom jeziku (za radove napisane na engleskom jeziku) ili na engleskom jeziku (za radove napisane na srpskom jeziku) navodi se na kraju rada i treba da ima od 100 do 250 reči. Ispred osnovnog teksta rezimea, navodi se naslov rada, puno ime, srednje slovo i prezime svih autora i naziv i adresa ustanove autora. Naslov „Rezime" piše se razmaknuto i centrirano. Nakon naslova sledi jedan razmak, a zatim tekst rezimea, uvučen jednim tabulatorom. Neposredno nakon teksta rezimea, navode se ključne reči, sa tačkom na kraju. E-mail adresa autora za kontakt navodi se ispod crte, pri dnu stranice.

Tabele

Tabele obeležene arapskim brojevima (1, 2, itd.) praćene naslovom treba da se nalaze na odgovarajućem mestu u tekstu, u fontu 9. Maksimalna širina tabela treba da bude 13 cm. One treba da budu jasne, što jednostavnije i pregledne. Treba izbegavati vertikalne crte, a broj kolona ograničiti tako da tabela ne bi bila preširoka. Takođe, treba izbegavati nepotrebnu upotrebu horizontalnih crta. Naslov tabele, poravnat po levoj i desnoj margini, sa tačkom na kraju, navodi se sa jednim razmakom iznad tabele. Ispod tabele treba dati detaljno objašnjenje skraćenica, simbola i znakova korišćenih u samoj tabeli. Svaka tabela mora biti pomenuta u tekstu.

Ilustracije

Svi grafikoni, dijagrami i fotografije treba da se nazovu „Slika" (1, 2, itd.). Prilažu se na odgovarajućem mestu u tekstu. Grafikone i dijagrame treba uraditi fontom 9, u crno-beloj tehnici i sa maksimalnom širinom od 13 cm. Voditi računa da oni budu čitki i jasni i nakon redukcije veličine. Za svaki grafikon i dijagram treba obezbediti detaljnu legendu bez skraćenica. Fotografije moraju biti visokog kvaliteta da bi se tehnički mogle dobro reprodukovati. Prilažu se u „TIF" ili „JPG" formatu, u crno-beloj tehnici. Naslov ilustracije, poravnat po levoj i desnoj margini, sa tačkom na kraju, navodi se sa jednim razmakom ispod ilustracije. Svaka ilustracija mora biti pomenuta u tekstu.

Skraćenice i jedinice

U radu treba koristiti samo standardne skraćenice. Merne jedinice treba izražavati u internacionalnom sistemu jedinica (SI). Kod navođenja jedinica posle broja treba da stoji razmak (osim za % i °C). Skraćenice se mogu koristiti i za druge izraze pod uslovom da se ti izrazi navedu u punom obliku prilikom prvog pominjanja, sa skraćenim oblikom u zagradi. Vrednosti od 1 do 9 mogu se izražavati slovima, a ostali brojevi isključivo numerički.

Nomenklatura

Celokupna nomenklatura (hemijska i biohemijska, taksonomska, genetička itd.) mora biti usklađena sa međunarodnim kodeksima i komisijama, kao što su *International Union of Pure and Applied Chemistry, IUPAC-IUB Combined Commission on Biochemical Nomenclature, Enzyme Nomenclature, International Code of Botanical Nomenclature, International Code of Nomenclature of Bacteria* itd.

Formule
Sve formule i jednačine u radu moraju biti urađene pomoću programa „Word Equation“. Pri pisanju formula, radi preglednosti, ostaviti dovoljno praznog prostora oko same formule. Subskripti i superskripti treba da budu jasni. Prilikom pisanja jednačina treba dati smisao svih simbola odmah posle jednačine u kojoj se simbol prvi put koristi. Jednačine treba da budu numerisane arapskim brojevima, serijski u zagradama, na desnoj strani linije. Svaka jednačina mora biti pomenuta u tekstu kao Eq. (1), Eq. (2), itd.

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