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ASSESSMENT OF FERMENTED DAIRY PRODUCTS ADEQUACY IN DIET OF LACTOSE INTOLERANT PERSONS

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Original scientific paper

Summary

Introduction and objective: Milk and dairy products are an important source of many nutrients that are known to have many beneficial effects on human health. On the other hand, milk and dairy products can cause allergies and intolerances. Allergy is caused by milk proteins that lead to immune reactions, while intolerance is caused by the milk sugar, lactose, due to reduced activity of enzyme lactase which digests it. To avoid the unpleasant symptoms, lactose intolerant persons can consume fermented milk products that are known to have reduced lactose content. The aim of this study was to determine the content of lactose in commercially available fermented dairy beverages by HPLC method and to assess their adequacy in diet of lactose intolerant persons.

Methods: Altogether 21 commercially available type of fermented milk products was analysed of which 13 were plain yogurts, while remaining 8 belonged to the group of functional products. Lactose content was determined by HPLC method. Lactose detection was achieved by refraction index detector and its quantification by external standard method.

Results: Lactose content of analysed products ranged from 2.65 g/100 g up to 4.05 g/100 g in plain yogurts, and from 2.61 g/100 g up to 4.63 g/100 g in functional products.

Conclusions: Based on determined lactose content and a presumption that most of lactose intolerant persons can digest up to 6 g of lactose on a daily basis without obvious symptoms it is assessed that daily acceptable amount of the analysed products ranges from 130 to 230 g.

Keywords: lactose, lactose intolerance, fermented milk products, diet, HPLC method

Introduction

Milk is one of the most complete foods. For infants it represents the exclusive food which enables their normal growth and development, while in an adult diet plays an important role as a source of highly valued proteins (Mandić, 2007). Milk and dairy products are also an important source of vitamins, essential fatty acids and minerals, and the main source of calcium in human nutrition.

Milk and dairy products can cause health problems, and among them allergies and intolerances are the most common. Allergies on milk and dairy products are commonly caused by milk proteins, while sugar lactose causes intolerance. Namely, one third of the milk caloric value is contributed by the lactose, main dairy carbohydrate, commonly known as the milk sugar (HAH, 2009). Lactose intolerance implies incapability of lactose digestion caused by decreased activity of the enzyme lactase (Brown-Esters et al., 2012) which causes various gastrointestinal problems (Lomer et al., 2008). If lactase activity is decreased by 50% or more maldigestion of lactose occurs. Unabsorbed lactose furthermore raises the amount of water and electrolytes in the large bowel lumen accelerating bowel content movement and resulting in diarrhoea. Bacterial β -galaktosidase unchains glucose

and galactose from the lactose and makes them accessible for the bacterial fermentation which occurs in the bowel and results in abdominal pain, bloating, flatulence and diarrhoea. On the other hand, some bacteria reduce carbon dioxide to methane and that may cause constipation (Lomer et al., 2008).

Hypolactasia, or lactase deficiency, exists in three distinct forms: congenital, primary and secondary (Lomer et al., 2008).

Congenital lactase deficiency is characterised by the lowest lactase activity. It is believed to be an autosomal recessive trait and the main symptom is infantile diarrhoea from the first exposure to breast milk (Madry i sur., 2010). Congenital lactase deficiency is extremely rare and a lifelong disorder (Lomer et al., 2008).

The most common type found in humans is primary lactose malabsorption or lactase non-persistence (adult type hypolactasia). It can be described as a genetically predetermined physiological condition inherited through an autosomal recessive mode. In this type, lactase activity decreases after weaning. Namely, healthy infants normally display an adequate expression of lactase which decreases during the weaning period. This might be an evolutionary adaptation since milk, in most adults, is not a main dietary product. The prevalence of this type of adult

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hypolactasia in Europe increases toward the South and East and reaches 70% in southern Italy and Turkey (Madry et al., 2010).

Secondary lactase deficiency is acquired as a result of gastrointestinal disease which damages the brush border of the small intestine (giardiasis, coeliac disease, viral gastroenteritis, Cronhn's disease, radiotherapy) (Madry et al., 2010; Lomer et al., 2008). The most common management of lactose intolerance relies on reducing lactose exposure by avoiding milk, dairy products and all milk-containing products from the diet (Brown – Esters et al., 2012). Many persons who avoid milk and dairy products which constitute main source of calcium, vitamin D and other nutrients are not ingesting adequate amounts of these essential nutrients (Suchy et al., 2010). This rises their risk of health problems such as rickets, osteoporosis and osteomalacia (Rong et al., 2011), and therefore many studies suggest alternative approaches. These alternative approaches imply consumption of products developed for lactose-intolerant persons (lactose-reduced milk, probiotic yoghurts) and steadily increase of the dietary lactose load which allows the colon to adapt over a period of time (Shaukat et al., 2010). One of the recently studied strategies is a consumption of fermented dairy products (Shaukat et al., 2010), in which, due to the fermentation reactions, lactose content is reduced, which makes them more tolerable for consumption in lactose-intolerant persons (HAH, 2009; Tudor, 2009). Lately, lactase enzyme is commercially available in the form of tablets, what enables lactose digestion. Yet it should be considered that this is an option for cases of unintended lactose consumption like trips or out of home lunches and not preparation for daily use.

There are four different factors that may play a role in lactose digestion. One of them is dose of lactose consumed, where direct relationship between lactose consumed and the severity of maldigestion symptoms exists. In most lactose intolerant persons, lactose doses below 12 g causes no symptoms while higher doses (20-50 g) cause considerable symptoms (Brown-Esters et al., 2012).

The aim of this study was to determine the content of lactose in commercially available fermented dairy beverages by high-performance liquid chromatography (HPLC) method and assess their adequacy in diet of lactose intolerant persons.

Materials and methods

Samples

To conduct this study altogether 21 commercially available types of fermented milk products were

purchased from the supermarkets in the area of city Osijek. Samples were divided into two groups. First group encompassed plain yogurts (13 samples), while second group (8 samples) included products declared as functional foods by the producer. Purchased samples were stored as indicated by the producer (refrigerated) and analysed within the “use by” date.

Lactose extraction from the samples

Carbohydrates were extracted from samples using the method described by Indyk et al. (1996).

Sample (5 g) was dissolved in warm demineralised water and transferred into a graduated 50 mL flask. Carrez reagents 1 and 2 were added (0.5 mL of each) sequentially with mixing and content was left to stand for about 20 minutes. Afterwards, extracts were made to volume with water and filtered. First few mL of filtrate were discharged.

Filtered sample solution was passed through a 0.45 µm nylon membrane filter.

High-performance liquid chromatography of the lactose content in samples

Analysis was performed on Varian instrument comprising of ternary Solvent Delivery Module (ProStar 230), Column Valve Module (ProStar 500) and Refractive Index Detector (ProStar 350). Instrument was supported with ProStar Chromatography Workstation 5.

To enable lactose quantification an external calibration method was applied. Lactose solutions for the calibration (D-lactose monohydrate for HPLC analyses, ≥99.5%, Sigma-Aldrich, Buchs, Switzerland) were prepared in range from 0.1 up to 5.0%. Each standard solution was analysed twice.

Samples were analysed using a Zorbax NH₂ (Agilent Technologies) column (4.6 x 250 mm, 5 µm particle size).

Separation of present carbohydrates was achieved applying an isocratic elution with acetonitrile/water (70/30 v/v). Acetonitrile (Merck, Darmstadt, Germany) of HPLC gradient grade purity and ultrapure water were used to prepare mobile phase solution.

Lactose identification was achieved based on the retention time and quantification based on the external calibration curve obtained based on the standard solutions prepared as described above. 10 µL loop was used for all analyses.

All analyses were conducted in two replicates, and each sample solution was injected twice.

Lactose content is expressed as g/100 g of the original sample.

Results and discussion

Lactose content of analysed plain yoghurts ranged from 2.65 g/100 g up to 4.05 g/100 g (Table 1). Vinko et al. (2011) studied changes of lactose content in dairy production and reported lactose

content of 3.12 g/100 g of product after one day refrigerated storage and 2.80 g/100 of product after 28 days of refrigerated storage for plain yoghurt fermented using yoghurt culture of *Streptococcus thermophilus* and *Lactobacillus delbrueckii* subsp. *bulgaricus*.

Table 1 Basic characteristics and lactose content of analysed fermented milk products

Sample code	Yoghurt type	Fat content declared by producer (g/100 g)	Lactose (g/100 g)
Fermented products			
Y-7	Set	3.2	2.81 ± 0.24
Y-8	Set	3.2	3.50 ± 0.15
Y-9	Drinkable	2.8	4.05 ± 0.31
Y-10	Drinkable	0.1	3.35 ± 0.15
Y-11	Drinkable	3.2	3.05 ± 0.04
Y-12	Drinkable	0.9	3.93 ± 0.17
Y-13	Drinkable	0.9	3.14 ± 0.20
Y-14	Drinkable	2.8	2.65 ± 0.09
Y-17	Drinkable	0.5	2.88 ± 0.19
Y-18	Set	3.2	2.89 ± 0.06
Y-19	Drinkable	2.8	3.07 ± 0.33
Y-20	Drinkable	0.05	2.82 ± 0.22
Y-23	Drinkable	2.8	2.96 ± 0.11
Probiotic fermented products			
FY-2	Drinkable	3.2	2.87 ± 0.13
FY-3	Drinkable	1.5	3.33 ± 0.32
FY-4	Drinkable	3.5	3.49 ± 0.11
FY-5	Drinkable	1.0	2.77 ± 0.09
FY-6	Drinkable	0.5	2.61 ± 0.17
FY-21	Drinkable	0.9	4.63 ± 0.10
FY-22	Drinkable	3.5	4.33 ± 0.15
FY-24	Drinkable	3.1	3.17 ± 0.15

Lactose content of 8 functional dairy products varied from 2.61 g/100 g up to 4.63 g/100 g. Functional milk-based products available in the Italian market and provided from large stores were HPLC analysed by Manzi et al. (2007). They reported 2.32 up to 4.50 g of lactose per 100 g of product for probiotic fermented milk products. O'Brien (1999) determined lactose content of cultured dairy products available in the UK market by enzymatic method. He analysed 94 various products and reported average lactose content of 3.67% in live yoghurts (plain) and 2.54% in drinking yoghurts. O'Brien draw attention to the fact that sugar content of milk and dairy products published in food composition tables are lower from those which he obtained, and explained that by the wide range of products which were developed and broaden to the market recently. He concluded that fermented dairy products have significantly lower lactose contents than unfermented milk and as such might be appropriate for lactose-intolerant individuals (O'Brien, 1999).

Vinko et al. (2011) have reported that lactose content is influenced by the age of product, and drop in

lactose content during the 28 days long refrigerated storage (within the „use by“ date) reaches up to 38.73% of initial lactose content. Our study did not encompass the influence of storage time on lactose content since the exact time of inoculation and fermentation is not consumer available information, and durability of products varies from the few days up to few weeks. Proper storage, and the analysis within the „use by“ date was the only criteria used.

Besides product age, lactose content is influenced by the microbial β -galaktosidase activity during the storage period. Vinko et al. (2011) reported that thermophilic cultures converted about 30% of lactose to lactic acid, while mesophilic culture converted 16-20% indicating that choice of culture can also play an important role in estimation of dairy product suitability for lactose intolerant persons.

Significance of a product in a person's diet depends not only on the content of the observed compound per 100 g of product but also on the frequency of consumption and the portion size. As a result, from the desirable compound point of view, important food source are even those food product which

contains small amount of the compound but it is consumed often and in large amounts. At the same time, from the undesirable compound point of view, product does not have to present a risk if it is consumed occasionally and in small amounts. Using that framework in the lactose intolerance perspective, although fermented milk products contain lactose, its content is lower due to fermentation, and they could be suitable for lactose intolerant persons. The amount of product should not exceed tolerable level of

lactose. Namely, literature data purport the fact that most of the lactose-intolerant individuals can digest up to 6 g of lactose on a daily basis(HAH, 2009). Based on that value, acceptable daily amount of product is estimated for all samples analysed in our study. Results presented in Fig. 1 indicate that most of the lactose intolerant persons could ingest from 130 g up to 230 g of yoghurt daily (in dependence of the product type choice) without feeling discomfort caused by lactose digestion problems.

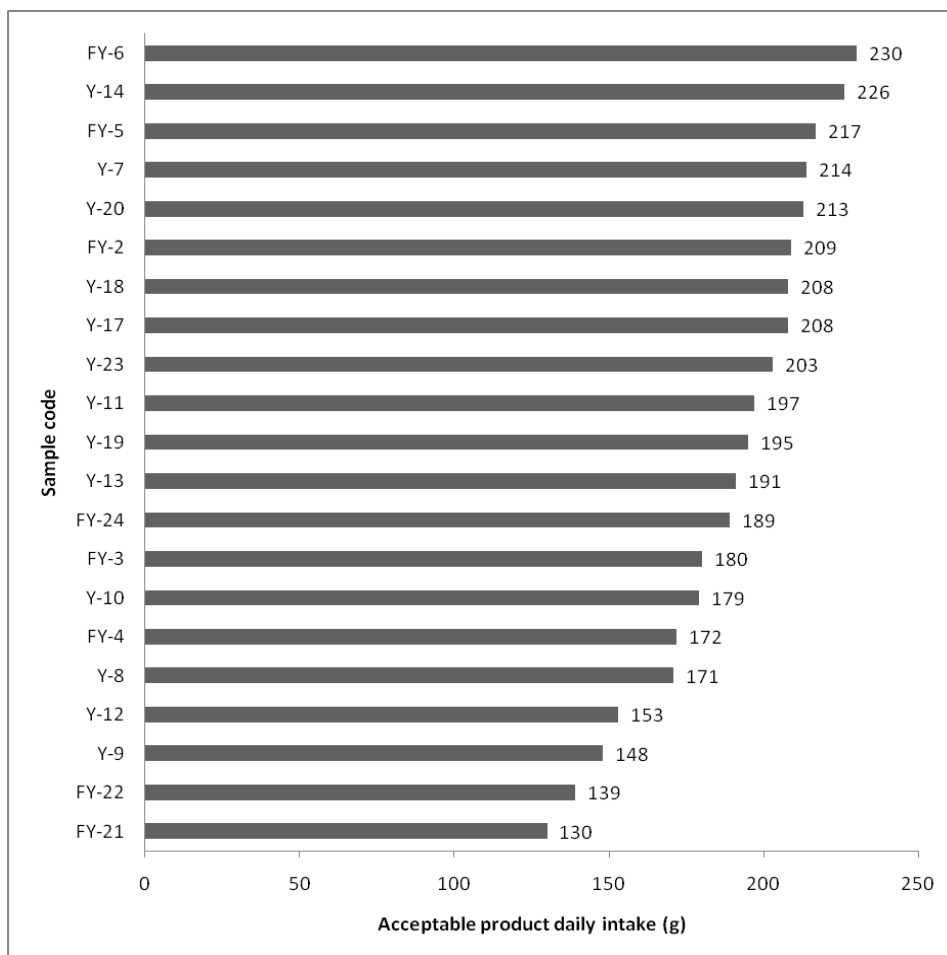


Fig. 1. The individually assessed acceptable amount of daily intake of analysed fermented dairy products in lactose-intolerant persons for plain yoghurts (Y) and functional yoghurts (FY) under the presumption that person can digest 6 g of lactose daily with no or minor symptoms

Suchy et al. (2010) reported that adults and adolescents with diagnosed lactose malabsorption could ingest at least 12 g of lactose when administered in a single dose with no or minor symptoms, and even larger amounts if ingested with meals and distributed throughout the day. The intake of up to 12 g was supported by Brown-Esters et al. (2012) based on the results of studies which indicate how colonic bacteria develop an increased ability to

ferment lactose over prolonged period of time with lactose ingestion. Dairy consumption should therefore not be restricted but managed, and the useful approach is to ingest fermented milk products throughout the day in small portions.

Individual differences in sensitivity are influenced by many factors among which ethnicity and age are studied most intensively. Lomer et al. (2008) reported that incidence is lower in adults from northern

European countries and Australia, while higher in South America, Africa and Asia. Acceptable tolerable daily amount is assumed to vary according to the same pattern. Croatian Food Agency (2009) reports higher lactose intolerance incidence in elderly adult due to diminished lactase production with age. Vonk et al. (2003) concluded that the major difference in intolerance symptoms is caused by differences in the colonic processing of maldigested lactose.

Altogether, determined patterns and mechanisms, as well as the individual differences, show that ingestion of lactose containing products should be personalised.

Conclusions

Lactose content of analysed fermented dairy products ranged from 2.65 g/100 g up to 4.05 g/100 g in plain yogurts, and from 2.61 g/100 g up to 4.63 g/100 g in functional products. Values are in line with those found in literature for the same product type.

Based on determined lactose content and a presumption that most of lactose intolerant persons can digest up to 6 g of lactose on a daily basis without obvious symptoms it is assessed that acceptable amount of analysed products range from 130 to 230 g. Nevertheless, individual differences should be considered prior to ingestion.

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RAZLIKE U PREHRAMBENIM NAVIKAMA KOD UČENIKA UZRASTA 15-18 GODINA U ODNOSU NA SPOL NA PROSTORU OPĆINE TRAVNIK

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Izvorni znanstveni rad

Sažetak

Uvod: Pravilna prehrana je važan faktor za unapređenje i očuvanje zdravlja. Savremen način života koji karakteriše dug radni dan, neredovan unos hrane, unos bar jednog obroka u toku dana van kuće i promjena navika u prehrani u pogledu izbora, vrste, načina pripreme i količine namirnica u svakodnevnoj prehrani, uz smanjenu fizičku aktivnost, doveli su do porasta broja zdravstvenih problema u čijoj osnovi je nepravilna prehrana.

Cilj rada: Cilj ovog istraživanja bio je ispitati općenito prehrambene navike i status uhranjenosti srednjoškolaca na području općine Travnik, te postoje li određene razlike u navedenim karakteristikama s obzirom na spol.

Metodologija: U istraživanje je uključeno 229 učenika uzrasta 15-18 godina, 90 učenika muškog spola i 139 ženskog spola. Kao instrumenti mjerenja korišteni su anketni upitnik koji je izrađen u svrhu ovog istraživanja (22 pitanja o prehrambenim i životnim navikama) i antropometrijska mjerenja (visina i težina). Procjena stepena uhranjenosti provedena je na osnovu percentilnih krivulja, tj. na osnovu izračunatih vrijednosti za visinu i masu u odnosu na godine.

Rezultati: Istraživanjem je utvrđeno da prema percentilnim krivuljama 196 (85,59 %) učenika uzrasta od 15-18 godina ima poželjnu tjelesnu težinu, 29 (12,67 %) ima prekomjernu tjelesnu težinu, dok pretilih učenika nema. Kada je u pitanju konzumiranje kuhanog ručka na osnovu dobivenih rezultata postoji statistički značajna razlika ($X^2=10.400$; $df=3$; $Sig.=0.015$ ($p=0.05$)), veći je postotak djevojaka 62 (44,60 %) koje svakodnevno konzumiraju isti u odnosu na mladiće 23 (25,56 %).

Zaključak: Rezultati dobiveni ovim istraživanjem pokazali su da je najveći postotak ispitanika muškog 74 (82,22 %) i ženskog spola 122 (87,77 %) normalno uhranjeno, prekomjernu tjelesnu težinu ima 15 (16,67 %) mladića i 14 (10,07 %) djevojaka. Pravilna prehrana je veoma značajna za očuvanje zdravlja i za dug, kvalitetan život. Djeci i mladima je potrebna raznovrsna i uravnotežena prehrana, kako bi povoljno utjecala na rast i razvoj, dobru funkciju imunološkog sistema te mentalnu i radnu sposobnost.

Ključne riječi: stanje uhranjenosti, pravilna prehrana, prehrambene navike

Uvod

Pravilna prehrana je važan faktor za unapređenje i očuvanje zdravlja, posebno u razdoblju odrastanja. Osiguravanjem uvjeta za odgovarajući psihofizički razvoj, zdrave prehrambene navike koje djeca steknu u ranom djetinjstvu utječu i na odabir namirnica i način prehrane i u kasnijem životnom razdoblju, a time i na zdravlje u odrasloj dobi (MZRH, 2013).

Savremen način života koji karakteriše dug radni dan, neredovan unos hrane, unos bar jednog obroka u toku dana van kuće i promjena navika u prehrani u pogledu izbora, vrste, načina pripreme i količine namirnica u svakodnevnoj prehrani, uz smanjenu fizičku aktivnost, doveli su do porasta broja zdravstvenih problema u čijoj osnovi je nepravilna prehrana (MZCG, 2009.). Nepravilna prehrana može značajno utjecati na rast i razvoj djece i omladine, te privremeno ili trajno ugroziti njihovo zdravstveno stanje i vremenom dovesti prvo do gojaznosti, a kasnije i do dijabetesa, kardiovaskularnih oboljenja, oboljenja koštano-mišićnog sistema, oboljenja digestivnog sistema, malignih oboljenja debelog

crijeva i dr (Tandir i Karakaš, 2013). Istraživanja pokazuju da raste broj djece koja obolijevaju od dijabetesa tipa 2 upravo zbog pretilosti (Hunter i sur., 2000.; Wang i sur., 2006). Pored nepravilne prehrane, u industrijaliziranim zemljama većina žena i muškaraca danas slijede trend negativnog razvoja razine tjelesne aktivnosti i vode uglavnom sedentarni način života, ili su aktivni povremeno, što rezultira velikim brojem tjelesno neaktivne populacije, značajnim povećanjem osoba s prekomjernom masom i pretilih, posebno među mladima i djecom. Niža tjelesna aktivnost povezuje se sa višim ITM i u drugim istraživanjima (Baruki i sur., 2006.), a što doprinosi različitim zdravstvenim posljedicama i psihičkim poremećajima koji se posebno reflektiraju među djecom i adolescentima (Wang i Lobstein, 2006).

Prehrambene potrebe u adolescenciji pod utjecajem su prvenstveno pubertetskog zamaha rasta i smatra se da su veće nego u bilo kojim drugim životnim razdobljima. U tom periodu kod mladih često se sreću poremećaji prehrane ili nepravilan odnos prema hrani (HZJZ, 2012). Načela pravilne prehrane

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predstavljaju važan dio zdravog stila života, te ih je zbog njihovog pozitivnog utjecaja na cjelokupno zdravlje stanovništva neophodno usvojiti već u najranijoj životnoj dobi (Larson i Neumark-Sztainer, 2009). Zdrava pravilna i raznovrsna prehrana, u kombinaciji sa redovnom tjelesnom aktivnošću može doprinijeti nižoj prevalenciji rizika ili spriječiti pojavu kardiovaskularnih i drugih bolesti (Stamler i sur., 1999.). Praćenje zdravstvenog stanja populacije uključuje i ocjenu stanja uhranjenosti pojedinih populacijskih skupina. Ovakva istraživanja omogućuju utvrđivanje pojavnosti deficitarnih stanja i prehrambenih poremećaja, te procjenu rizika za zdravlje (Cepić, 2007).

Cilj ovog istraživanja bio je ispitati općenito prehrambene navike i status uhranjenosti srednjoškolaca na području općine Travnik, te postoje li određene razlike u navedenim karakteristikama s obzirom na spol.

Materijal i metode

Istraživanje je provedeno u toku 2013/2014. godine. Istraživanjem je obuhvaćeno ukupno 229 učenika uzrasta 15-18 godina (90 učenika muškog spola i 139 ženskog spola) iz šest srednjih škola na prostoru općine Travnik. Ispitanici su bili odabrani metodom slučajnog odabira. Kao instrumenti mjerenja korišteni su: anketni upitnik koji je izrađen u svrhu ovog istraživanja i antropometrijska mjerenja (visina i težina).

Upitnik se sastojao od 22 pitanja koja su se odnosila na opće informacije (naziv škole, mjesto stanovanja, razred, dob i spol) i prehrambene navike (broj glavnih obroka, učestalost konzumiranja kuhanog ručka, doručak prije odlaska u školu, izbor međuobroka, izbor hrane u školi za vrijeme odmora, da li više konzumiraju vodu ili sok, koliko često (nikada, jednom sedmično, 2-4 dana u sedmici, jednom dnevno i više puta dnevno) konzumiraju voće, povrće, slatkiše, grickalice, žitarice, bijeli i crni hljeb i koliko često piju gazirana pića. Tjelesna visina mjerena je visinomjermom s tačnošću od 0,1 cm.

Ispitanik je stajao na ravnoj podlozi, skupljenih peta. Mjerena je udaljenost od podloge do tjemena glave. Masa tijela mjerena je medicinskom vagom sa tačnošću 0,5 kg. Sva mjerenja su bila provedena standardnim postupcima i instrumentima. Procjena stepena uhranjenosti provedena je na osnovu percentilnih krivulja, tj. na osnovu izračunatih vrijednosti za visinu i masu u odnosu na godine. Izmjereni BMI između 15. i 85. percentilne krivulje pokazuje normalnu uhranjenost, BMI između 3. i 15. percentilne krivulje potranjenost, ispod 3. percentilne krivulje neuhranjenost, BMI između 85. i 97. percentilne krivulje pokazuje prekomjernu tjelesnu težinu, a iznad 97. percentilne krivulje pretilost djece (WHO, 2007).

Statistička obrada podataka je urađena uz pomoć statističkog paketa SPSS 20.

Rezultati i rasprava

Prema percentilnim krivuljama rezultati su pokazali da od ukupno 229 učenika uzrasta 15-18 godina 196 (85,59 %) učenika ima normalnu tjelesnu težinu, 29 (12,66 %) učenika ima prekomjernu tjelesnu težinu, pretilih učenika nema, dok su 3 (1,31 %) učenika pothranjena (Tablica 1).

Prema podacima iz Nacionalne zdravstvene ankete stanovništva Crne Gore, koju je 2008. godine provelo Ministarstvo zdravlja, rada i socijalnog staranja Republike Crne Gore, tri četvrtine djece i adolescenata uzrasta od 7 do 19 godina u Crnoj Gori, imalo je zdravu težinu (MZCG, 2009).

U Hrvatskoj je procjena uhranjenosti učenika uzrasta od 7 do 15 godina, za razdoblje od 1997-2002. godine, pokazala da je pravilno uhranjeno, u prosjeku, 69,5 % djece, dok je prekomjernu tjelesnu težinu imalo 11,0% djece, a pretilo je bilo 5,2 % učenika (Antonić-Degač i sur., 2004).

U Srbiji, prema objektivnim mjerenjima visine i težine, procenat pothranjenog stanovništva starosti od 15 do 24 godine, iznosi 10,4 %, dok procenat normalno uhranjenih iznosi 67,8 % (IPSOS, 2014).

Tablica 1. Stanje uhranjenosti prema percentilnim krivuljama ispitanika uzrasta od 15 do 18 godina

Table 1. State of nourishment according to the percentiles of the study participants aged 15 to 18 years

Stanje uhranjenosti	%
Pretilost	0.00
Prekomjerna tjelesna težina	12.66
Normalna tjelesna težina	85.59
Pothranjenost	1.31
Neuhranjenost	0.44
Ukupno	100%

Dobiveni rezultati stanja uhranjenosti prema percentilnim krivuljama ispitanika uzrasta 15-18 godina u odnosu na spol (Tablica 2) su pokazali da je nešto veći postotak mladića koji imaju prekomjernu tjelesnu težinu 15 (16,67 %) u odnosu na djevojke 14

(10,07%). Nešto je veći postotak djevojaka sa normalnom tjelesnom težinom 122 (87,77 %) u odnosu na mladiće 74 (82,22 %), a kada je u pitanju pothranjenost 2 (1,44 %) djevojaka je pothranjeno u odnosu na mladiće 1 (1,11 %).

Tablica 2. Stanje uhranjenosti prema percentilnim krivuljama ispitanika uzrasta od 15 do 18 godina, u odnosu na spol

Table 2. State of nourishment according to the percentiles of the study participants aged 15 to 18 years, depending on gender

Stanje uhranjenosti	Muški	Ženski
Pretilost	0.00 %	0.00 %
Prekomjerna tjelesna težina	16.67 %	10.07 %
Normalna tjelesna težina	82.22 %	87.77 %
Pothranjenost	1.11 %	1.44 %
Neuhranjenost	0.00 %	0.72 %

Doručak je najvažniji obrok u danu, budući da slijedi nakon perioda noćnog gladovanja, kada su energetske zalihe iscrpljene, a trebaju biti nadopunjene kako bi učenik mogao izvršavati sve obaveze koje pred njega stavlja novi dan. Nadalje je redovan doručak pokazan kao efikasna mjera u smanjenju pretilosti. (HZJZ, 2012). Na pitanje o učestalosti konzumiranja doručka (Tablica 3) od ukupnog broja ispitanika 35 (25,18 %) djevojaka redovno konzumira doručak, dok je nešto

niži postotak mladića 19 (21,11 %), veliki je postotak ispitanika oba spola koji ne konzumiraju redovno doručak 34 (37,78 %) mladića i 45 (32,37 %) djevojaka, dok udio djevojaka koje 2-4 dana u sedmici konzumiraju doručak iznosi 59 (42,45 %), a mladića 37 (41,11 %). Na osnovu dobivenih rezultata ($X^2=0.869$; $df=2$; $Sig.=0.648$ ($p=0.05$)) ne postoji statistički značajna razlika u odnosu na spol.

Tablica 3. Konzumiranje doručka ispitanika uzrasta od 15 do 18 godina, u odnosu na spol

Table 3. Breakfast consumption among the study participants aged 15 to 18 years, depending on gender

Spol	Konzumiranje doručka			Ukupno
	Da	Ne	2-4 dana u sedmici	
Muški	19	34	37	90
Ženski	35	45	59	139
Ukupno	54	79	96	229

$X^2=0.869$; $df=2$; $Sig.=0.648$ ($p=0.05$)

Istraživanje u Hrvatskoj je pokazalo zabrinjavajući podatak da od početka praćenja u 2002. godini pa do 2010. godine značajni dio djece uopće ne doručkuje (u 2010. 15 % radnim danom, a 6 % vikendom; u 2006. 15 % radnim danom, a 5 % vikendom; u 2002. 15 % radnim danom a 4 % vikendom). U 2010. godini radnim je danom uvijek doručkovalo 59 % učenika i 54 % učenica, što je vrlo slično podacima za 2006. godinu (60 % dječaka i 54 % djevojčica), a predstavlja pogoršanje u odnosu na 2002. godinu (72 % dječaka i 68 % djevojčica). Kao i prethodnih godina i 2010. godine s porastom dobi sve se manje doručkovalo. Djevojčice su doručkovala manje od dječaka, tako da u dobi od 15 godina tek svaka druga djevojčica (48 %) redovno doručkuje radnim danom. Preskakanje doručka često uzrokuje mučninu tokom jutra i ometa procese učenja i pamćenja. Doručak, pogotovo onaj bogat žitaricama, dobar je početak

dana, a pridonosi i boljem raspoloženju, pažnji i postizanju boljih školskih rezultata. Oni koji preskaču doručak zbog jakog osjećaja gladi koji se javlja tokom ostatka dana češće konzumiraju hranu bogatu mastima i jednostavnim šećerima, a siromašnu vlaknima i hranjivim tvarima (HZJZ, 2012).

U Crnoj Gori 87,2 % djece i adolescenata uzrasta od 7 do 19 godina doručkovalo je svakog dana, a 76,3 % imalo tri obroka dnevno svakog dana (MZCG, 2009), dok je udio djece i mladih uzrasta 7 do 14 godina u Srbiji koji svakodnevno doručkuju (93,8 %) značajno veći u poređenju sa 2006. godinom (90,5 %) (IPSOS, 2013).

Kada je u pitanju konzumiranje kuhanog ručka na osnovu dobivenih rezultata ($X^2=10.400$; $df=3$; $Sig.=0.015$ ($p=0.05$)) postoji statistički značajna razlika (Tablica 4) veći je postotak djevojaka 62 (44,60 %) koje svakodnevno konzumiraju isti u odnosu na mladiće 23 (25,56 %). Veći je postotak

mladića 50 (55,56 %) koji kuhani ručak konzumiraju više puta sedmično u odnosu na djevojke 52 (37,41 %), također nešto je veći postotak mladića njih 16 (17,78 %) koji 2-4 dana u sedmici konzumiraju kuhani ručak u odnosu na djevojke 21 (15,10 %). U

jednom istraživanju u Hrvatskoj (sa područja Grada Zaprešića) dobiveni su podaci da kuhani obrok rijetko ili nikada ima 20,5 % učenika I. razreda srednje škole (Koprivnjak, 2008).

Tablica 4. Učestalost konzumiranja kuhanog ručka ispitanika uzrasta od 15 do 18 godina, u odnosu na spol

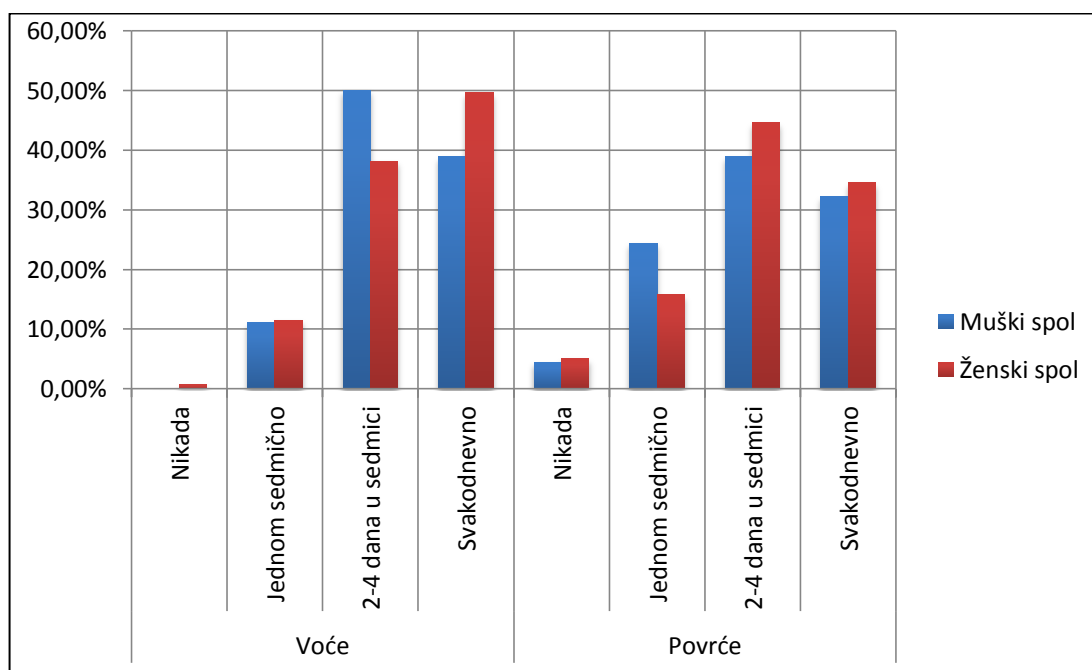
Table 4. Frequency of consumption of cooked lunch among the study participants aged 15 to 18 years, depending on gender

Spol	Učestalost konzumiranja kuhanog ručka				Ukupno
	Svakodnevno	Više puta sedmično	2-4 dana u sedmici	Jednom sedmično	
Muški spol	23	50	16	1	90
Ženski spol	62	52	21	4	139
Ukupno	85	102	37	5	229

$X^2=10.400$; $df=3$; $Sig.=0.015$ ($p=0.05$)

Rezultati ovog istraživanja su pokazali (Slika 1) da je veći postotak djevojaka 69 (49,64 %) koje svakodnevno konzumiraju voće u odnosu na mladiće 35 (38,89 %). Udio mladića koji konzumiraju voće više puta sedmično iznosi 45 (50,00 %), dok je udio djevojaka nešto niži 53 (38,13 %), podjednak je udio oba spola kada je u pitanju konzumiranje voća jednom sedmično $M=10$ (11,11%) i $Ž=16$ (11,51 %). Povrće svakodnevno konzumira 48 (34,53 %)

djevojaka i mladića 29 (32,22 %), udio djevojaka koje više puta sedmično konzumiraju povrće je 62 (44,60 %) u odnosu na mladiće 35 (38,89 %). Kada je u pitanju konzumiranje povrća jednom sedmično veći je postotak mladića 22 (24,44 %) u odnosu na djevojke 22 (15,83 %). Statistički značajna razlika u učestalosti konzumiranja voća ($X^2=4.067$; $df=4$; $Sig.=0.397$ ($p=0.05$)) i povrća ($X^2=2.674$; $df=4$; $Sig.=0.614$ ($p=0.05$)) ne postoji.



voće: $X^2=4.067$; $df=4$; $Sig.=0.397$ ($p=0.05$); povrće: $X^2=2.674$; $df=4$; $Sig.=0.614$ ($p=0.05$)

Slika 1. Učestalost konzumiranja voća i povrća ispitanika uzrasta od 15 do 18 godina, u odnosu na spol
Fig. 1. Frequency of consumption of fruits and vegetables among the study participants aged 15 to 18 years, depending on gender

U Federaciji Bosne i Hercegovine istraživanje je pokazalo da preko trećine ispitanika (36,2 %) konzumira svježe žuto i narandžasto povrće manje od jednom sedmično. Svježe lisnato povrće se rijetko koristi, preko polovine ispitanika ga konzumira 1-2 puta sedmično ili manje (41,5 % odnosno 14,5 %). Također, 16,0 % ispitanika konzumira mahunarke manje od jednom sedmično. Svježe voće, osim cistrusnog, 6-7 puta sedmično konzumira samo 11,6 % ispitanika. Skoro petina ispitanika konzumira svježe citrusno voće manje od jednom sedmično (19,2 %) (FMZ i sur., 2012). Rezultati u Crnoj Gori su pokazali da je dnevni unos svježeg voća u nedjelji dana prijavilo 41,5% djece i adolescenata uzrasta 7-19 godina koji idu u školu, a 33,4 % iste grupe jelo je svježe povrće svakog dana (MZCG, 2009).

U Hrvatskoj konzumiranje voća i povrća kod najvećeg dijela mladih nije zadovoljavalo stručne preporuke. Naime, podaci za 2010. pokazuju da ukupno 66 % učenika ne jede voće, a 76 % ne jede povrće svaki dan (HZJZ, 2012).

U Srbiji više od polovine djece i mladih svakodnevno konzumira voće 51 %, dok je 49 % njih koji voće konzumiraju nedovoljno ili ga nikada ne konzumiraju. Slična je situacija i kada je riječ o povrću – 56,0 % djece i mladih koji imaju od 7 do 14

godina konzumira svakodnevno povrće, dok 43,4 % njih ne konzumira povrće u dovoljnoj količini ili ga ne konzumira uopšte (IPSOS, 2013).

Energijom bogate, a nutritivno siromašne namirnice (Tablica 5), se često konzumiraju i to svakodnevno 62 (44,60 %) djevojaka i 31 (34,44 %) mladića. Kada je u pitanju konzumiranje istih više puta u sedmici udio mladića je 32 (35,56 %) u odnosu na djevojke 30 (21,58 %), dok jednom sedmično slatkiše konzumira 19 (21,11 %) mladića i 29 (20,86 %) djevojaka. Na osnovu vrijednosti hi-kvadrata može se uvidjeti da ne postoji statistički značajna razlika između mladića i djevojaka u konzumiranju slatkiša i grickalica ($X^2=6.203$; $df=4$; $Sig.=0.184$ ($p=0.05$)).

Što se tiče konzumiranja slatkiša i grickalica u Federaciji Bosne i Hercegovine dobiveni su sljedeći podaci: slatkiše svakodnevno konzumira petina ispitanika (21,1 %), a među djecom uzrasta 5-15 godina njih gotovo trećina (31,3 %). I keksi i kolači, slatka bezalkoholna pića, kao i čips i grickalice se također učestalo konzumiraju, 3-5 puta sedmično ih konzumira preko trećina ispitanika, a čak 16,9 % djece uzrasta 5-15 godina konzumira grickalice svakodnevno (FMZ i sur., 2012).

Rezultati istraživanja u Hrvatskoj su pokazali nešto veći postotak učenika koji konzumiraju slatkiše i grickalice i on iznosi 84 % (HZJZ, 2012).

Tablica 5. Učestalost konzumiranja slatkiša i grickalica ispitanika uzrasta od 15 do 18 godina, u odnosu na spol

Table 5. Frequency of consumption of sweets and snacks among the study participants aged 15 to 18 years, depending on gender

Spol	Učestalost konzumiranja slatkiša i grickalica					Ukupno
	Nikada	Jednom sedmično	2-4 dana u sedmici	Jednom dnevno	Više puta dnevno	
Muški spol	8	19	32	17	14	90
Ženski spol	18	29	30	32	30	139
Ukupno	26	48	62	49	44	229

$X^2=6.203$; $df=4$; $Sig.=0.184$ ($p=0.05$)

Mlijeko i mliječni proizvodi su najvažniji prehrambeni izvor kalcija. Iako se kalcij može unijeti i putem ostalih prehrambenih izvora, mlijeko sadržava i druge nutrijente, u prvome redu vitamin D, a potom i fosfor, magnezij, cink te proteine koji su nužni za metabolizam kostiju (Alebić, 2008). Rezultati su pokazali da postoji statistički značajna razlika ($X^2=15.968$; $df=4$; $Sig.=0.003$ ($p=0.05$)) u konzumiranju mlijeka i mliječnih proizvoda, u odnosu na spol. Što se tiče svakodnevnog konzumiranja mlijeka i mliječnih proizvoda (Tablica 6) nešto veći je postotak mladića 33 (36,67 %) u odnosu na djevojke 48

(34,53 %). Također, veći je udio mladića 42 (46,67 %) koji više puta sedmično konzumiraju navedene proizvode u odnosu na djevojke 45 (32,37 %), dok je veći postotak djevojaka 32 (23,02 %) koje jednom sedmično konzumiraju mlijeko i mliječne proizvode u odnosu na mladiće 12 (13,33 %).

Tablica 6. Učestalost konzumiranja mlijeka i mliječnih proizvoda ispitanika uzrasta od 15 do 18 godina, u odnosu na spol
Table 6. Frequency of consumption of milk and dairy products among the study participants aged 15 to 18 years, depending on gender

Spol	Učestalost konzumiranja mlijeka i mliječnih proizvoda					Ukupno
	Nikada	Jednom sedmično	2-4 dana u sedmici	Jednom dnevno	Više puta dnevno	
Muški spol	3	12	42	13	20	90
Ženski spol	14	32	45	33	15	139
Ukupno	17	44	87	46	35	229

$X^2=15.968$; $df=4$; $Sig.=0.003$ ($p=0.05$)

Prema nacionalnoj zdravstvenoj anketi u Crnoj Gori, 49,4 % djece i adolescenata je pilo mlijeko najmanje 6-7 puta sedmično, odnosno konzumiralo mliječne proizvode (jogurt i slično) najmanje 6-7 puta sedmično, ili je pilo mlijeko 3-5 puta sedmično, kao i konzumiralo mliječne proizvode 3-5 sedmično (MZCG, 2009).

Kada je riječ o konzumiranju mliječnih proizvoda u Srbiji kod djece uzrasta 7-14 godina, više od dvije trećine djece i mladih 74,2 % svakodnevno popije bar jednu šolju mlijeka ili mliječnih proizvoda, što je značajno više od nivoa konzumacije mlijeka iz 2006. godine – tada je 60,4 % djece i mladih navelo da dnevno popije barem jednu šolju mlijeka ili mliječnih proizvoda (IPSOS, 2013).

Najbolji izvori ugljikohidrata su cjelovite žitarice kao što su zobene pahuljice, integralni hljeb, smeđa riža i dr (Harvard, 2011.) Predstavljaju niskokalorične namirnice, sa niskim sadržajem masti i umjerenim sadržajem proteina (Rosić i Stanišić-Stojić, 2012). U odnosu na spol ne postoje statistički značajne razlike ($X^2=8.924$; $df=4$; $Sig.=0.063$ ($p=0.05$)). Žitarice koje spadaju u visoko preporučenu vrstu hrane za doručak svakodnevno konzumira 30 (21,58 %) djevojaka u odnosu na mladiće 15 (16,67 %). Više puta sedmično 26 (18,70 %) djevojaka konzumira iste, dok 16 (17,78 %) mladića. Jednom sedmično 33 (36,67 %) mladića konzumira žitarice, a 40 (28,78 %) djevojaka, dok je veliki postotak i onih koji nikada ne konzumiraju žitarice djevojaka 43 (30,93 %) i mladića 26 (28,89 %) (Tablica 7).

Tablica 7. Učestalost konzumiranja žitarica ispitanika uzrasta od 15 do 18 godina, u odnosu na spol
Table 7. Frequency of consumption of cereals among the study participants aged 15 to 18 years, depending on gender

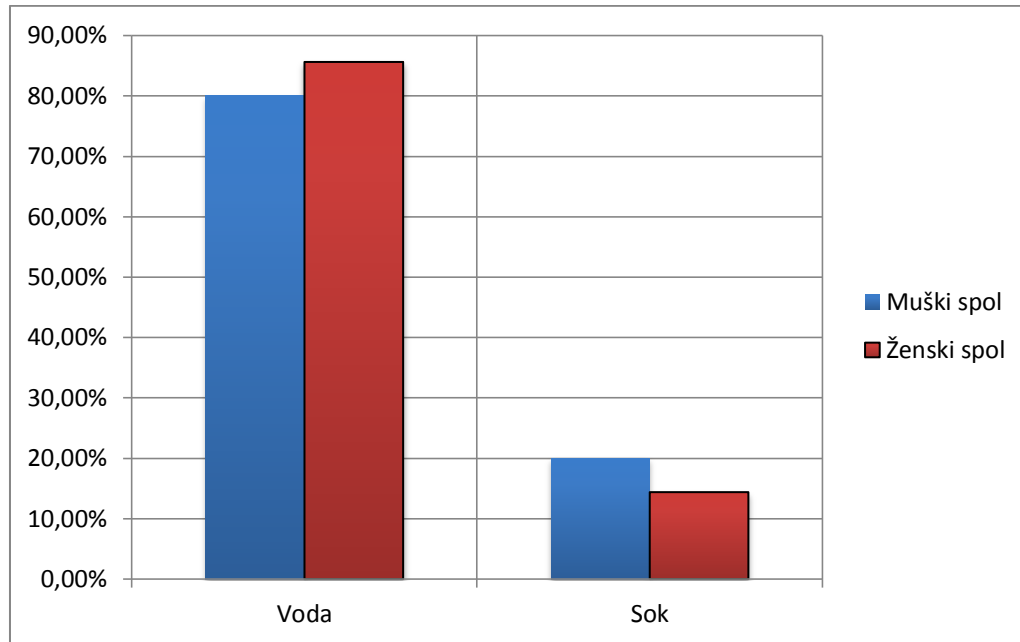
Spol	Učestalost konzumiranja žitarica					Ukupno
	Nikada	Jednom sedmično	2-4 dana u sedmici	Jednom dnevno	Više puta dnevno	
Muški spol	26	33	16	4	11	90
Ženski spol	43	40	26	21	9	139
Ukupno	69	73	42	25	20	229

$X^2=8.924$; $df=4$; $Sig.=0.063$ ($p=0.05$)

Sve veća konzumacija slatkih pića na prijelazu iz dječije u adolescentnu dob posljedica je smanjenog roditeljskog utjecaja na prehranu. Djeca veliki dio vremena svakodnevno provode u školi te je zbog toga dostupnost ukusnih visokovrijednih namirnica upravo u školi važna u pravilnoj prehrani učenika (HZJZ, 2012). Kada je u pitanju konzumiranje tečnosti (Slika 2) udio ispitanika koji konzumiraju vodu iznosi 119 (85,61 %) djevojaka i 72 (80,00 %) mladića, dok je veći udio mladića 18 (20,00 %) koji konzumiraju sok u odnosu na djevojke 20 (14,39 %). Što se tiče konzumiranja tečnosti, u odnosu na spol, ne postoji statistički značajna razlika ($X^2=1.243$; $df=1$; $Sig.=0.265$ ($p=0.05$)).

Različiti izvori potvrđuju da djeca jedu previše ugljikohidrata. Slatka pića, šećer i čokolada vodeći su

izvori nemliječnog izvanjskog šećera u prehrani djece, a previše šećera nepovoljno utječe na tjelesnu težinu i razvoj zubnog karijesa, te ometa unos nutritivno vrijednijih namirnica. U Hrvatskoj u 2010. godini, slatka pića svaki dan pilo je 30 % dječaka i 22 % djevojčica što predstavlja izvjesno poboljšanje u odnosu na prethodne godine (u 2006. 35 % dječaka i 28 % djevojčica; u 2002. 33 % dječaka i 31 % djevojčica). Dječaci piju više slatkih pića nego djevojčice što se nije promijenilo u odnosu na prethodna istraživanja. Najniži udio učenika i učenica koji svakodnevno piju slatka pića zabilježen je u zemljama sjeverne Europe, te u pribaltičkim zemljama dok je Hrvatska kao i u prethodnom istraživanju zauzela poziciju u gornjoj polovici ljestvice (HZJZ, 2012).



$$X^2=1.243; df=1; Sig.=0.265 (p=0.05)$$

Slika 2. Konzumiranje tečnosti ispitanika uzrasta od 15 do 18 godina, u odnosu na spol
Fig. 2. Consumption of fluids among the study participants aged 15 to 18 years, depending on gender

Zaključci

Rezultati dobiveni ovim istraživanjem pokazali su da je najveći postotak ispitanika muškog 74 (82,22 %) i ženskog spola 122 (87,77 %) normalno uhranjeno, prekomjernu tjelesnu težinu ima 15 (16,67 %) mladića i 14 (10,07 %) djevojaka. Što se tiče statistički značajnih razlika u odnosu na spol one ne postoje kada je u pitanju učestalost konzumiranja doručka, voća i povrća, slatkiša i grickalica te žitarica i tečnosti. Statistički značajne razlike postoje kada je u pitanju učestalost konzumiranja kuhanog ručka $X^2=10,400$; $df=3$; $Sig.=0,015$ ($p=0,05$), veći je postotak djevojaka 62 (44,60 %) koje svakodnevno konzumiraju isti u odnosu na mladiće 23 (25,56 %), dok je veći je postotak mladića 50 (55,56 %) u odnosu na djevojke 52 (37,41 %) koji kuhani ručak konzumiraju više puta sedmično. Što se tiče učestalosti konzumiranja mlijeka i mliječnih proizvoda $X^2=15,968$; $df=4$; $Sig.=0,003$ ($p=0,05$), veći je udio mladića 42 (46,67 %) koji više puta sedmično konzumiraju navedene proizvode u odnosu na djevojke 45 (32,37 %), dok je veći postotak djevojaka 32 (23,02 %) koje jednom sedmično konzumiraju mlijeko i mliječne proizvode u odnosu na mladiće 12 (13,33 %). Rezultati dobiveni ovim istraživanjem, vezani za status uhranjenosti, su dosta ohrabrujući u odnosu na druge zemlje gdje je

povećana tjelesna težina kod djece u stalnom porastu. Da bismo održali ovako visok udio normalno uhranjene djece, i vremenom dobili još bolje rezultate, potrebno je osigurati sve neophodne uslove, kako bi se kod djece i mladih razvile pravilne prehrabene i životne navike koje bi povoljno utjecale na rast i razvoj, dobru funkciju imunološkog sistema, te mentalnu i radnu sposobnost. Primarnom edukacijom o načelima pravilne prehrane i redovne fizičke aktivnosti, djecu se od najranije životne dobi može naučiti o pozitivnom utjecaju istih na zdravlje, kao i na zadovoljenje osnovnih tjelesnih i emocionalnih potreba.

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DIFFERENCES IN EATING HABITS AMONG STUDENTS AGED 15-18 YEARS IN RELATION TO SEX IN THE AREA OF MUNICIPALITY TRAVNIK

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Original scientific paper

Summary

Introduction: Proper nutrition is an important factor for promotion and preservation of health. The modern way of life which is characterized by long working hours, irregular food intake, intake of at least one meal during the day out of the house and changes in dietary habits in terms of choice, type, method of preparation and amount of foods in the daily diet, with reduced physical activity, have led to increase the number of health problems that are based on unbalanced diet.

Objective: The aim of this study was to examine overall dietary habits and nutritional status of high school students in the municipality of Travnik, and whether there are differences in these characteristics with regard to gender.

Methodology: The study included 229 students aged 15-18 years, 90 were male students and 139 female. As instruments of measurement were used a questionnaire that was developed for this study (22 questions about dietary and lifestyle) and anthropometric measurements (height and weight). Assessment of nutritional state was carried out on the basis of percentile curves, ie. on the basis of the calculated values for height and weight compared to the year.

Results: It was found that according to the percentile curves 196 (85.59%) students age 15-18 had a desirable body weight, 29 (12.67%) were overweight, and there were not obese students. When they were asked about the consumption of cooked lunch on the basis of the results there is a statistically significant difference ($X^2=10.400$, $df=3$, $Sig.=0.015$ ($p=0.05$)) there was a higher percentage of girls 62 (44.60%) who daily consume the same in relation to boys 23 (25.56%).

Conclusion: The results of this study showed that the highest percentage of male respondents 74 (82.22%) and 122 females (87.77%) were normally nourished, overweight was 15 (16.67%) males and 14 (10.7%) girls. Proper nutrition is very important for health care and for a long, productive life. Children and young people need a varied and balanced diet, in order to favorably influence the growth and development, good function of the immune system and mental and working ability.

Keywords: nutritional status, proper nutrition, eating habits

POLYCYSTIC OVARY SYNDROME (PCOS) – PILOT STUDY ON DIET QUALITY

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Original scientific paper

Summary

Introduction: Polycystic Ovary Syndrome (PCOS) is a complex gynecological and endocrinological disorder affecting 6% to 21% of reproductive age women. The main characteristics of PCOS include hyperandrogenism, irregular menstrual cycles, lack of ovulation, enlarged ovaries with numerous cysts and infertility. Symptoms vary widely between women, but most commonly include obesity, hyperinsulinemia and insulin resistance (approximately 65% to 70% of women with PCOS). Therefore, uniform therapy and dietary recommendations fail.

Objective: The objective of this study was to analyze the quality of diet and dietary patterns of women with PCOS. Actual lifestyle habits and the diet of the women with PCOS have not been extensively researched internationally and never in Croatia.

Methods: The study included women of reproductive age diagnosed with PCOS (N=12), recruited at the Gynecological Office Lončar, Karlovac, Croatia. Study participants completed a general questionnaire on socio-economic characteristics and two questionnaires regarding their gynecological health and PCOS symptoms. Anthropometry was measured (Seca) while biochemical data was provided by the gynecologist. Diet quality was assessed with a 24-hour dietary recall and quality of diet and exercise habits were assessed with a questionnaire developed specifically for this study.

Results: Women with PCOS had an average daily energy intake of 82.3% of the RDA, i.e. 2333 kcal/day. When analyzing the contribution of separate macronutrients to that daily energy intake, discrepancies from the recommendations were found. The main discrepancies are high contribution of fats (40.1%) and increased intake of proteins (17.7%). Total fat intake showed a statistically significant positive correlation with waist circumference and waist to hips ratio. In addition, a statistically significant correlation was found between age and intake of total carbohydrates and plant proteins.

Conclusion: Study findings show the potential for a larger-scale study on Croatian women with PCOS.

Keywords: Polycystic Ovary Syndrome, diet, dietary habits

Introduction

Polycystic Ovary Syndrome (PCOS) is a complex endocrine condition with 6% to 21% prevalence in the population of the reproductive age women (Goss et al., 2014; Moran et al., 2013a; Graff et al., 2013). The main characteristics of PCOS are endocrine (hyperandrogenism) and biochemical abnormalities, irregular menstrual cycles, lack of ovulation, increased ovaries with numerous cysts and infertility (Lucidi, 2014; Moran et al., 2013b; Escott-Stump et al., 2012; Lim et al., 2012; Abercrombie et al. 2010). This disorder is accompanied by obesity, hyperinsulinemia and insulin resistance (Escott-Stump et al., 2012; Teede et al., 2007), but symptoms differ between women (Spritzer, 2014; Cronin et al., 1998). Beside medications, the main treatments for PCOS are weight reduction (Lucidi, 2014; Moran et al., 2013a; Moran et al., 2013b) and lifestyle management practices with equal therapy for obese and non-obese patients (Marsh et al., 2005).

Actual lifestyle habits and the diet of the women with the PCOS have not been extensively researched. Nevertheless, a relatively small number of studies show that these women, compared with their healthy counterparts, consume larger quantities of high glycemic index (GI) foods (Altieri et al., 2013; Graff et al., 2013; Moran et al., 2013b; Douglas et al., 2006). Other researchers have found that PCOS itself, increased energy intake, intake of high GI foods, lower physical activity and some other variables are all independently associated with higher BMI (Moran et al., 2013b).

Until today, only a few studies dealt with the lifestyle and diet interplay involved in PCOS development. None of these studies were performed in Croatia. Therefore, the main aim of this pilot study was to determine the diet quality and dietary patterns of women with PCOS in North West Croatia and subsequently to determine the potential for a larger scale study that could be used as a basis for an intervention study on women with PCOS.

Subjects and methods

Study Participants and Study Protocol

A total of 12 women of reproductive age (aged 18-39) with PCOS diagnosed by the gynecologist (Gynecological Office Lončar, Karlovac, Croatia) were recruited, in a period of 6 months. After recruitment, each woman filled "The Survey on the Basic Data, Dietary Habits, and Physical Activity", and "The Questionnaire on the Polycystic Ovary Syndrome" (adapted according to Cronin et al., 1998). The Survey on the Basic Data, Dietary Habits, and Physical Activity" investigated the following: basic data (age, socio-economic data, anthropometrical data, etc.), dietary habits (number of meals per day; place of consumption; desire to experiment with food; food preferences; eating after feeling satiety; eating breakfast; eating dinner; consumption of dairy products, fruits and vegetables, potatoes, meat, fish, salt, fast food, candy, water, juices, coffee, sugar, alcohol, and dietary supplements) and degree of physical activity (during working time, during sport, and during free time). Research was conducted at Josip Juraj Strossmayer University of Osijek, Faculty of Food Technology Osijek, Department of Food and Nutritional Research laboratory. Medical documentation on the history and course of the disease for the women diagnosed with PCOS was provided by the gynecologist (Gynecological office Lončar, Karlovac, Croatia). The study protocol was approved by the Ethical Committee for the Research on People from the Josip Juraj Strossmayer University of Osijek, Faculty of Food Technology Osijek. Subjects were informed in detail about the study and written consent was obtained from all the participants.

Anthropometry Methods

Women recruited for the study were measured for body weight with a Tanita BC-601 (Tanita Corporation, Japan) scale (± 0.1 kg), body height without shoes with the position of the head in the Frankfurt plane (± 0.1 cm) with portable stadiometer Seca 123 (Seca, Germany) and waist and hip circumferences with measuring tape NCD Medical / Medical Prestige (Prestige Medical, USA). Their body mass index (BMI) was calculated with measured weight and height data. BMI was used to group all the women in one of the following categories: underweight ($\text{BMI} < 18.5 \text{ kg/m}^2$), normal (18.5 to 24.9 kg/m^2), overweight (25.0 to 29.9 kg/m^2) and obese (30.0 to $> 40.0 \text{ kg/m}^2$) (Lee and Nieman, 2010).

Diet Quality Assessment Method

The assessment of the dietary intake was performed with the 24-Hour Diet Recall, conducted with the "multi-pass protocol". This method was selected due to its convenience and because it showed the smallest error in terms of underestimating or overestimating nutritional intake (Scagliusi et al., 2008). Nutritional intake of macro and micronutrients was calculated by processing data from the 24-Hour Recall with a computer program "NutriPro" (Faculty of Food Technology Osijek, Croatia) that uses Croatian tables on the composition of foods and beverages (Kaić-Rak and Antičić, 1990). The results obtained were compared with the "Recommended Daily Allowance" (RDA) values (USDA, 2010).

Statistical Analysis

The chosen level of significance was $p=0.050$. Categorical variables were presented as absolute and relative frequencies while the numerical data were presented with the use of descriptive statistical methods, that is, mean and standard deviation. A Chi-square test was used to compare categorical variables within and between the groups. The differences between the two dependent groups were tested with the t-test for dependent measurements, that is, with using the t-test for independent measurements in the groups and the variables. An analysis of variance was performed with ANOVA. The Pearson correlation test was used for calculating the correlation of the numerical data.

Results and discussion

Mean BMI of the subjects was $27.4 \pm 8.5 \text{ kg/m}^2$ with mean waste hip ratio (WHR) of 0.9 ± 0.2 , which is not unusual for women with a PCOS diagnosis.

Responses to the "Survey on the Basic Data, Dietary Habits, and Physical Activity" showed that women with PCOS have a tendency towards higher GI foods (for example: 33.3% women with PCOS eat cakes/sweets every day, 33.3% women with PCOS eat bakery products with dinner, 16.6% women with PCOS never put additional sugar in their food/beverage), which corresponds to previous research on this topic (Altieri et al., 2013; Graff et al., 2013; Moran et al., 2013b; Douglas et al., 2006). When it comes to physical activity subjects were mostly sedentary.

The total daily energy intake of analyzed women was calculated as the percentage of the RDA intake based on the nutritional requirements for women of reproductive age. In this study, subjects had an

average daily energy intake of 82.3% of the RDA, i.e. 2333 kcal/day. Graff et al. (2013) and Moran et al. (2013b) found that women with PCOS have a higher calorie intake than women who do not suffer from

PCOS. However, women with PCOS in this study had higher total daily energy intake than Croatian healthy women of reproductive age in the study by Banjari (2012).

Table 1. Total energy intake per subject with percentage of RDA and distribution of energy intake by macronutrient

Subject	Total daily energy (kcal)	Energy % of RDA	Protein % in total energy	Fat % in total energy	CHO % in total energy
1	1702	72.9	25.6	35.0	40.8
2	1622	69.5	12.4	46.2	41.8
3	684	29.3	24.5	47.0	30.4
4	1278	54.8	16.5	31.7	52.7
5	4118	176.5	16.9	47.6	35.8
6	2714	116.3	14.0	39.4	47.2
7	2405	103.1	15.1	26.0	61.3
8	1455	62.4	10.0	44.9	46.8
9	1536	65.9	19.5	47.7	33.6
10	1022	43.8	21.1	33.6	45.4
11	2513	107.7	16.4	47.8	37.5
12	1978	84.8	20.0	34.1	48.4
Average	1919	82.3	17.7	40.1	43.5

RDA – Recommended Daily Allowance; CHO – carbohydrates

When analyzing the contribution of separate macronutrients to daily energy intake (Table 1), discrepancies from the recommendations (USDA, 2010) were found. The main discrepancies were high contribution of fats (40.1%) and increased intake of proteins (17.7%) (Fig. 1), both involved in abdominal obesity and pancreatic functioning, i.e. insulin secretion, which consequently can result in PCOS (Escott-Stump et al., 2012; Teede et al., 2007). It is interesting to note that the contribution of separate

macronutrients to the total daily energy intake differ significantly from the one reported by Banjari (2012) for Croatian women of reproductive age. Banjari (2012) found that for healthy women of reproductive age in Croatia contribution of macronutrients is as follows: 51.0% from carbohydrates, 12.6% from proteins, and 35.9% from fats. On the other hand, Altieri et al. (2013) did not find a difference in macronutrients and energy intake between healthy women and women with PCOS.



Fig. 1. Contribution of separate macronutrients to the total daily energy intake of women with PCOS as compared to the recommendations (US Department of Agriculture and US Department of Health and Human Services 2010)

Total fat intake showed a statistically significant positive correlation with higher levels of abdominal fat, as measured by waist circumference and waist to hips

ratio (Table 2). Abdominal fat can result in insulin resistance, which is instrumental in the etiology of PCOS (Escott-Stump et al., 2012; Teede et al., 2007).

Table 2. Correlation of Selected General and Socio-Economic Variables with Energy and Fat Intake Based on a 24-Hour Dietary Recall

Selected variable	kcal	Fat total	SF	MUFA	PUFA	Linoleic acid	Chol
Age	0.56	0.44	0.44	0.38	0.42	0.46	0.43
Household members	-0.26	-0.13	-0.23	-0.12	-0.23	-0.25	-0.45
Smoking	-0.47	-0.38	-0.43	-0.38	-0.34	-0.34	-0.29
BMI	0.44	0.50	0.36	0.39	0.28	0.28	-0.17
Waist (W)	0.55	0.62*	0.49	0.50	0.42	0.42	-0.12
Hips (H)	0.42	0.49	0.37	0.39	0.35	0.35	-0.08
W/H ratio	0.53	0.59*	0.47	0.47	0.39	0.38	-0.17

*statistical significance at p<0.05, the Pearson correlation test

SF–saturated fats; MUFA–monounsaturated fats; PUFA–polyunsaturated fats; Chol–cholesterol

In addition, a statistically significant correlation was found between age and intake of total carbohydrates and plant proteins (Table 3). Thus, according to these results, as women with PCOS get older they show an increased tendency towards higher consumption of carbohydrates and plant proteins. Moreover, distribution of age and BMI showed that BMI increased with age (six subjects aged 18-26 had mean BMI of 24.5±7.9 kg/m² and six subjects aged 29-39 had mean BMI of 30.4±8.7 kg/m²). This suggests

that, some age groups of women with PCOS in Croatia, when trying to lose weight in an attempt to control PCOS symptoms, should focus on reducing consumption of total carbohydrates and plant proteins. Moreover, if results from the “Survey on the Basic Data, Dietary Habits, and Physical Activity” are taken into account, these women should focus on reducing high GI foods (mainly simple carbohydrates) and increasing their physical activity (improving cellular metabolism of glucose).

Table 3. Correlation of Selected General and Socio-Economic Variables with Protein and Carbohydrate Intake Based on a 24-Hour Dietary Recall

Selected variable	Protein total	Plant protein	Animal protein	CHO total	CHO simple	CHO complex	CHO fiber
Age	0.50	0.61*	0.43	0.61*	0.49	0.51	0.55
Household members	-0.19	-0.35	-0.13	-0.41	-0.56	-0.33	-0.49
Smoking	-0.48	-0.42	-0.41	-0.49	-0.47	-0.44	-0.52
BMI	0.17	0.43	-0.00	0.37	0.28	0.29	0.30
Waist (W)	0.24	0.51	0.05	0.46	0.36	0.37	0.39
Hips (H)	0.16	0.40	0.01	0.33	0.26	0.24	0.29

*statistical significance at p<0.05, the Pearson correlation test, CHO– carbohydrates, BMI – body mass index

Study results indicated a potentially negative correlation between the number of household occupants and smoking with the intake of macronutrients. For women with higher number of household members negative correlations showed that more time spent in care for others (especially children and elderly) resulted in lower daily intake of energy (and all macronutrients; Tables 2 and 3). In addition, a negative correlation between smoking and daily intake of energy (and all macronutrients; Tables 2 and 3) showed that, as expected, smokers eat less. When asked, 50% of women participating in the study expressed their interest in involvement in a

prospective study with dietary intervention if it could change the course and symptoms of their PCOS. This shows the potential for educational intervention on women with PCOS about the PCOS symptoms and weight gain, especially around waist.

The main limitation of the study is the low number of participants. Thus, we believe that inclusion of a larger number of women with PCOS might have resulted in different statistical significance for some or all of the identified parameters that showed a potential influence on the overall diet quality and the symptoms of PCOS in Croatian women.

Conclusions

The discovered discrepancies with dietary recommendations when it comes to macronutrient intake, correlations between socio-economic circumstances and macronutrient intake and tendency towards high GI foods on this small sample indicate that there is a potential for a larger scale study of dietary patterns in Croatian women with PCOS.

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Conflict of interest statement

None declared.

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PHYSICOCHEMICAL PROPERTIES OF PRO-VITAMIN A CASSAVA-WHEAT COMPOSITE FLOUR BISCUIT

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Summary

In this research work, the physicochemical properties of biscuit made from wheat flour and pro-vitamin A cassava flour mixed in different ratios of 100:0, 90:10, 80:20, 70:30 and 60:40 (w/w) was investigated. The addition of pro-vitamin A flour to wheat flour influenced the functional properties such as water absorption, swelling and pasting properties. Peak, trough, breakdown and final viscosities of 100% wheat flour was generally lower than the mixes. However, the setback viscosity of the mixes were lower than that of wheat flour. This was attributed to possible complex formation between the starch components of the flours and the carotenoid. Protein (10.80-15.45%), fat (11.87-21.35%) and carbohydrate (60.08-70.99%) were the major components of the biscuits. The protein and fat contents of the biscuit decreased with increasing proportions of pro-vitamin A cassava flour. But, the carotenoid contents of the biscuits increased. Sensory results showed that biscuit prepared from wheat flour and pro-vitamin A cassava flour in ratio 90 to 10 had similar overall acceptability rating (6.50) and total rating score (31.8) to the control (Overall acceptability rating; 6.50 and total rating score; 32.5). The research has established that acceptable biscuits which could be potentially used to address protein, energy and vitamin A challenges in developing nations of the world.

Keywords: biscuit, cassava, physicochemical properties, pro-vitamin A, pasting

Introduction

Biscuit is a ready to eat, convenient and inexpensive food products (Kulkarni, 1997). In many parts of the world including Nigeria, biscuit is one of the confectioneries consumed mostly by children. It is produced essentially from wheat flour, fat, sugar and water, but other ingredients may be added to enhance desired sensory attributes (Oyeyinka et al., 2014). Wheat flour is the most widely used flour for making biscuit. However, flours from other tuber crops such as cassava have also found application in the biscuit industry. These non-conventional flours have been used either singly or in combination with wheat flour as composites in making bread (Eriksson, 2013; Eriksson et al., 2014) and biscuits (Akubor and Ukwuru, 2003; Oluwole and Karim, 2005; Akingbala et al., 2011; Oluwamukomi et al., 2011; Osundahunsi et al., 2012; Thomas, 2012; Obadina et al., 2014; Oyeyinka et al., 2014). The use of composite flours in biscuit production has several implications. It may result in the reduction of importation cost for wheat flours to developing nations, promote the use of lesser known or neglected food crops and may facilitate the delivery of important micronutrients such as vitamin A to the desired population.

Micronutrient deficiency such as Vitamin A deficiency is one of the major challenge facing developing nations. Vitamin A deficiency can lead to night blindness and other acute diseases. In many parts of the world, several intervention programmes aimed at reducing the associated menace caused by Vitamin A deficiency and other micronutrients deficiencies have been suggested. One of such interventions is the use of staples and snacks such as biscuits as a medium to reach the vulnerable groups, which in most cases are the pregnant women and infants.

Cassava (*Manihot esculenta crantz*) with improved nutritional value such as pro-vitamin A cassava is currently been used as an aid in reducing the prevalence of dietary Vitamin A deficiency due to its high content of β -carotene (Omodamiro et al., 2012). According to Aniedu and Omodamiro (2012), pro-vitamin A cassava have the potential of providing up to 25% of daily vitamin A requirements of children and women. These authors reported the use of 10% high quality cassava flour made from pro-vitamin A cassava root in the production of bread, chin-chin, cakes, strips and salad cream (Aniedu and Omodamiro, 2012). Whole cassava flour has also been successfully used in the production of biscuit

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with comparable sensory qualities to that of wheat biscuit (Akingbala et al., 2011). However, the nutritional quality in terms of protein content of the wheat biscuit was more than double those of the cassava flour biscuits (Akingbala et al., 2011). The replacement of wheat flour with flours from other food crops in the production of biscuit may influence the physicochemical properties of the biscuit. For example, Oluwamukomi et al. (2011) found that biscuit weight increased with increase in the levels of added cassava flour. The protein content of biscuit made from 90% wheat flour and 10% cassava flour was not significantly different from that produced from 100% wheat flour (Oluwamukomi et al., 2011). The incorporation of different ingredient may influence functional and nutritional properties of the resulting products. In this study, the physicochemical properties of biscuit produced from varying ratios of wheat and pro-vitamin A cassava flour was investigated.

Materials and methods

Materials

Pro-vitamin A cassava (TMS 01/1368 variety) was obtained from Oyo State Agricultural Development Farm, Ogbomoso, Nigeria, while wheat flour, sugar, salt, fat and baking powder were purchased from a local market in Ogbomoso, Nigeria.

Flour production, formulation of composite flour and biscuit production

Flour was produced from freshly harvested pro-vitamin A cassava roots according to the modified method of Onabolu et al. (1998). The resulting flour was sieved (sieve size: 200 μm) and mixed with wheat flour sieved with the same sieve size. The ratio of wheat flour and pro-vitamin A cassava flour was 100:0, 90:10, 80:20, 70:30 and 60:40 w/w). Other ingredients used in biscuit production were added as described previously (Akinwande et al., 2008).

Functional and pasting properties of composite flour

Water absorption capacity, swelling power and bulk density was done as described by (Oyeyinka et al., 2014). The pasting properties of wheat-pro-vitamin A composite flour was determined using a Rapid Visco Analyzer (Newport Scientific, Australia), according to the method described by Oyeyinka et al. (2016a).

Chemical composition of flour and biscuits

Moisture, fat and ash contents of flour and biscuit was determined using AOAC (2000) methods. Protein content was determined by Kjeldahl method ($6.25 \times \text{N}$) and total carbohydrate was calculated by difference. Total cyanide (mg/kg) was determined as reported by Oluwamukomi et al. (2011) while carotenoid analysis was carried out as described previously (Aniedu and Omodamiro, 2012).

Physical properties of biscuit

The diameter and thickness of biscuits were measured with a venire caliper. Spread ratio was calculated from the ratio of diameter to thickness as described by Gains (1991) method. The average of weight biscuit (5 pieces) was measured in (g).

Sensory evaluation of biscuit

Consumers acceptability of biscuits was conducted using semi-trained panelist (N=50) consisting of students in the Department of Food Science and Engineering, Ladoke Akintola University of Technology, Nigeria. Most panelists were between the ages: 18–30 and these were regular consumers of biscuits. A 7 point hedonic scale (1- extremely dislike; 7- extremely like) was used.

Statistical analysis

Experiments were conducted in triplicate. Data were analyzed using analysis of variance and means were compared using Fischer's Least Significant Difference Test ($p < 0.05$).

Results and discussion

Carotenoid and cyanide content of pro-vitamin A cassava-wheat composite flour

The carotenoid and cyanide contents of freshly harvested pro-vitamin A cassava tuber were 7.54 $\mu\text{g/g}$ and 24.99 mg/kg respectively (Table 1). Composite flour prepared from wheat flour and pro-vitamin A cassava flour had carotenoid content ranging between 4.09 and 6.84 $\mu\text{g/g}$, while the cyanide content varied between 0.08 and 0.34 mg/kg. Expectedly, as the amount of pro-vitamin A cassava flour increased in the mix, both carotenoid and cyanide contents increased. The cyanide and the carotenoid in the cassava variety used may explain the increase in these components in the composite flour mix.

Table 1. Carotenoid and cyanide content of pro-vitamin A cassava tuber and its composite flour

Wheat flour	Cassava flour	Carotenoid Content ($\mu\text{g/g}$)	Cyanide Content (mg/kg)
100	0	4.02 ± 0.01^e	0.02 ± 0.14^d
90	10	4.09 ± 0.01^d	0.08 ± 0.02^d
80	20	5.14 ± 0.12^c	0.12 ± 0.01^c
70	30	5.76 ± 1.20^c	0.13 ± 0.02^c
60	40	6.84 ± 1.20^b	0.34 ± 0.01^b
Raw cassava tuber		7.54 ± 0.01^a	24.99 ± 0.12^a

Mean \pm SD. Mean with different superscript letters along the same column are significantly different ($p < 0.05$).

Functional properties of pro-vitamin A cassava-wheat composite flour

Bulk density of the flours varied slightly ($0.72\text{-}0.76 \text{ g/cm}^3$) with the addition of pro-vitamin A cassava flour to wheat flour (Fig. 1). But the water absorption capacity decreased (approx. 25%) with the addition of up to 40% pro-vitamin A cassava flour. Differences in the size of starch granules in the respective flours used in the formulation may have accounted for the variation in water absorption capacity of the composite flours. Similar conclusion was reported by previous researchers (Oyeyinka et al., 2014). Further, the presence of different hydrophilic carbohydrates as previously reported (Maninder et al., 2007) and the reduction in proteins due to decrease in the amounts of wheat flour may also explain the variations in the water absorption capacity of the composite flours. The swelling power of the 100% wheat flour was significantly ($p < 0.05$) lower than those of the composite flours (Fig. 1). Cereal flours including wheat flour contain endogenous lipids which are known to restrict swelling of starch in foods. The restriction in swelling is associated with the formation of complexes between amylose in starch and the lipids (Putseys et al., 2010; Oyeyinka et al., 2016b). Lipids may also cover the surface of starch with film preventing hydration and swelling (Kim and Walker, 1992).

Pasting properties such as peak viscosity, pasting temperature and setback viscosity of the composite flours varied significantly (Table 2). The peak viscosity of the flours generally increased with increase in the pro-vitamin A cassava flour. Wheat flour (100%) showed the lowest peak viscosity, while that of composite flour i.e. 60% wheat and 40% cassava flour was the highest. Peak viscosity of flours is influenced by many factors including amylose content, starch granule size, presence of lipids and proteins. During cooking of flours, amylose in starch leaches out into the surrounding medium to form a viscous paste. The degree of interaction between the various components of the

flours may explain the variation in the paste peak viscosity. High starch content in cassava flour may have enhanced the increased peak viscosity of the composite flours. Pasting temperature provides an indication of the minimum temperature required to cook the flour and also represent the ease of cooking. The pasting temperature ($87.25 \text{ }^\circ\text{C}$) of wheat flour (100%) was higher than the composite flours, which showed approximately $74 \text{ }^\circ\text{C}$ (Table 2). High pasting temperature in the control wheat flour could be due to the presence of endogenous lipids, which may interact with starch in the flour to form amylose-inclusion complex. These complexes are known to be resistant to swelling and rupturing during cooking. Previous studies associated high pasting temperature with the presence of starch which is highly resistant to swelling and rupturing (Maninder et al., 2007). This explanation is in agreement with the swelling power result (Fig. 1), where 100% wheat flour showed restricted swelling when compared with the composite flours.

The setback viscosity of the flour decreased from 1255 to 890 RVU with increase in the levels of cassava pro-vitamin A. Setback viscosity is an indication of the retrogradation tendencies of cooked starch, which is dependent on the extent of re-association of leached amylose. The reduction in setback viscosity following the addition of pro-vitamin A cassava flour suggest low retrogradation tendencies of the composite flours. Nwokocha et al. (2009) reported lower setback viscosity for cassava starch compared to cocoyam starch.

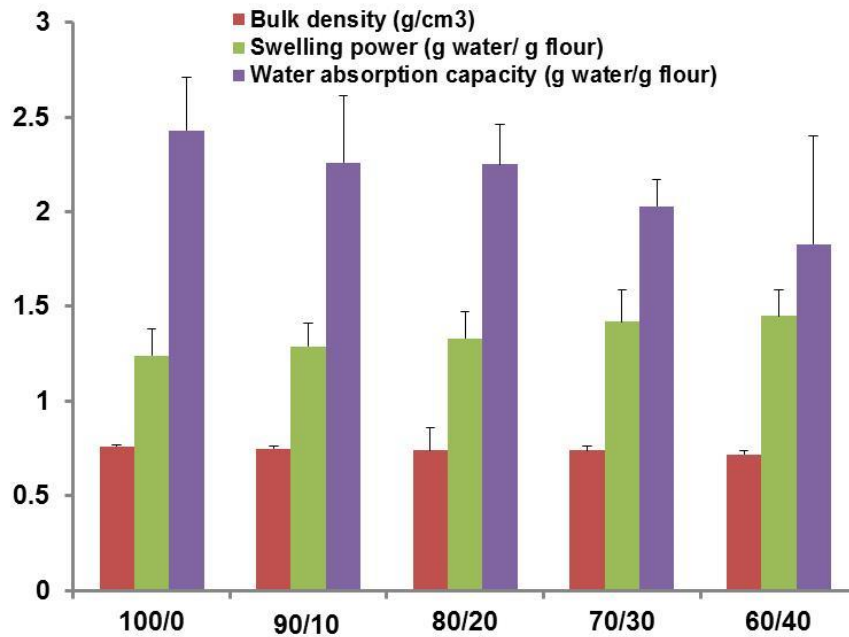


Fig. 1. Selected functional properties of wheat-pro-vitamin A cassava composite flour

Table 2. Pasting properties of wheat-pro-vitamin A cassava composite flour

Wheat flour	Cassava flour	PV (RVU)	TV (RVU)	BV (RVU)	FV (RVU)	SV (RVU)	Peak Time (min)	PT (°C)
100	0	2100 ^b	1234 ^c	799 ^b	2489 ^b	1255 ^a	6.1 ^a	87.2 ^a
90	10	2323 ^a	1437 ^b	866 ^{ab}	2572 ^a	1135 ^b	6.0 ^a	75.1 ^b
80	20	2344 ^a	1511 ^a	833 ^{ab}	2532 ^a	1021 ^b	5.9 ^a	73.4 ^c
70	30	2293 ^b	1494 ^{ab}	886 ^a	2429 ^b	935 ^c	5.8 ^b	73.4 ^c
60	40	2407 ^a	1521 ^a	888 ^a	2411 ^{bc}	890 ^{cd}	5.5 ^{bc}	73.5 ^c

Mean with different superscript letters along the same column are significantly different ($p < 0.05$).

PV: Peak viscosity, TV: Trough viscosity, BV: Breakdown viscosity, FV: Final viscosity, SV: Setback viscosity, PT: Pasting temperature

Physical properties of biscuit

The physical properties including weight, diameter, thickness, and spread ratio of biscuit made from wheat and pro-vitamin A composite flour are presented in Table 3. With increasing levels of pro-vitamin A cassava flour, the weight and spread ratio increased from 5.46 to 6.49 g and 7.44 to 8.27 respectively while diameter and thickness decreased from 4.91 to 4.63 cm

and 0.66 to 0.56 cm respectively. Aly and Seleem (2015) similarly reported increase in weight and spread ratio and a decrease in diameter and thickness of gluten-free biscuits made from cassava and extruded soy and pumpkin powder. The diameter of the biscuit in this study is within the range reported for biscuit made from wheat-cassava composite flour (Akingbala et al., 2011; Oluwamukomi et al., 2011; Aly and Seleem 2015).

Table 3. Physical property of wheat-pro-vitamin A cassava composite biscuit

Wheat flour	Cassava flour	Weight (g)	Diameter (cm)	Thickness (cm)	Spread Ratio
100	0	5.46 ± 0.01 ^b	4.91 ± 0.01 ^a	0.66 ± 0.01 ^a	7.44 ± 0.18 ^{bc}
90	10	5.67 ± 0.02 ^b	4.85 ± 0.01 ^a	0.65 ± 0.01 ^a	7.46 ± 0.18 ^{bc}
80	20	5.88 ± 0.01 ^b	4.81 ± 0.01 ^{ab}	0.64 ± 0.07 ^a	7.58 ± 0.06 ^b
70	30	6.18 ± 0.04 ^a	4.65 ± 0.01 ^c	0.59 ± 0.07 ^b	7.95 ± 0.12 ^{ab}
60	40	6.49 ± 0.01 ^a	4.63 ± 0.01 ^c	0.56 ± 0.01 ^b	8.27 ± 0.18 ^a

Mean ± SD. Mean with different superscript letters along the same column are significantly different ($p < 0.05$).

Proximate composition and carotenoid content of biscuit

Proteins, fats and carbohydrates were the major components of biscuit made from wheat-pro-vitamin A cassava composite flour (Table 4). The protein content of biscuit prepared from the control 100% wheat flour was the highest (approx. 16%), while that of biscuit prepared from composite flour (60% wheat and 40% pro-vitamin A flour) was the lowest (approx. 11%). Addition of pro-vitamin A cassava flour to wheat flour resulted in a reduction in protein and fat contents of the biscuits, while ash, fibre and carbohydrate contents increased. The observed composition of the formulated biscuit is similar to values reported in other studies on composite biscuits (Hooda and Jood, 2005; Oluwamukomi et al., 2011; Oyeyinka et al., 2014). Decrease in protein and fat contents of the biscuit could be attributed to the

reduction in the ratio of wheat flour in the biscuit formulation. Wheat flour is a richer source of protein and fat compared to cassava flour, which has the bulk of nutrient as carbohydrate. The carotenoid contents of the formulated biscuits increased with increasing amounts of pro-vitamin A cassava flour in the formulation from 246.80 to 352.40 $\mu\text{g}/100\text{ g}$ (Table 4). These values are within the carotenoid content range (219-428 $\mu\text{g}/100\text{ g}$) reported for biscuit prepared from sweet potato flour and mango mesocarp flour (Sengev et al., 2016) and bread with added mango mesocarp (Badifu et al., 2006). Sengev et al. (2016) found that biscuit formulated from 60% sweet potato flour and 40% mango mesocarp flour showed the highest carotenoid content of 428 $\mu\text{g}/100\text{ g}$. Both sweet potato and mango are relatively good source of beta carotene, which may explain the higher values reported by this authors compared to values in this study.

Table 4. Chemical composition of biscuit from wheat-pro-vitamin A cassava composite flour

Parameters	Samples				
	100:0	90:10	80:20	70:30	60:40
Moisture (%)	3.26 ± 0.01 ^{bc}	3.99 ± 0.01 ^a	3.70 ± 0.03 ^{ab}	3.51 ± 0.01 ^b	3.07 ± 0.10 ^c
Protein (%)	15.47 ± 1.07 ^a	13.00 ± 0.66 ^b	10.93 ± 0.74 ^c	10.80 ± 0.74 ^c	10.87 ± 0.28 ^c
Fat (%)	21.35 ± 1.21 ^a	20.02 ± 0.01 ^{ab}	17.54 ± 0.01 ^c	12.49 ± 0.01 ^d	11.87 ± 1.12 ^d
Ash (%)	0.61 ± 0.07 ^{cd}	0.75 ± 0.01 ^c	1.29 ± 0.01 ^b	1.39 ± 0.01 ^b	1.79 ± 0.02 ^a
Fibre (%)	0.23 ± 0.01 ^{cd}	0.28 ± 0.10 ^c	0.33 ± 0.01 ^b	0.28 ± 0.01 ^c	0.41 ± 0.17 ^a
CHO (%)	60.08 ± 0.53 ^d	60.96 ± 0.76 ^d	64.21 ± 0.98 ^c	68.53 ± 0.09 ^b	70.99 ± 1.47 ^a
Carotenoid ($\mu\text{g}/100\text{ g}$)	246.80 ± 0.04 ^d	303.60 ± 0.24 ^c	334.00 ± 0.42 ^b	331.60 ± 1.12 ^b	352.40 ± 1.83 ^a

Mean ± SD. Mean with different superscript letters along the same row are significantly different ($p < 0.05$).

Sensory properties of biscuit

Sensory properties of the biscuit were slightly affected by the added pro-vitamin A cassava flour (Fig. 2). In general, the ratings recorded for the sensory attributes of the biscuit decreased with increase in the level of pro-vitamin A cassava flour. Colour is an important sensory attribute of any food because of its influence on acceptability. The brown colour resulting from maillard reaction is always associated with baked goods. Biscuit made from wheat flour (100%), which served as the control had the highest rating for colour, aroma, texture and overall acceptability. The higher rating observed for the control sample may be due to the fact that panel members are familiar with wheat biscuit and this could have influenced their rating for the control sample. Obviously, the added pro-vitamin A cassava flour changed the sensory properties of the

biscuit as evident in the rating recorded by the panellist. The total rating score obtained for the control biscuit was 32.5 out of a maximum score of 35. Biscuit prepared from wheat flour and pro-vitamin A cassava flour in ratio 90 to 10 had similar overall acceptability rating (6.50) and total rating score (31.8) to the control (Overall acceptability rating; 6.50 and total rating score; 32.5).

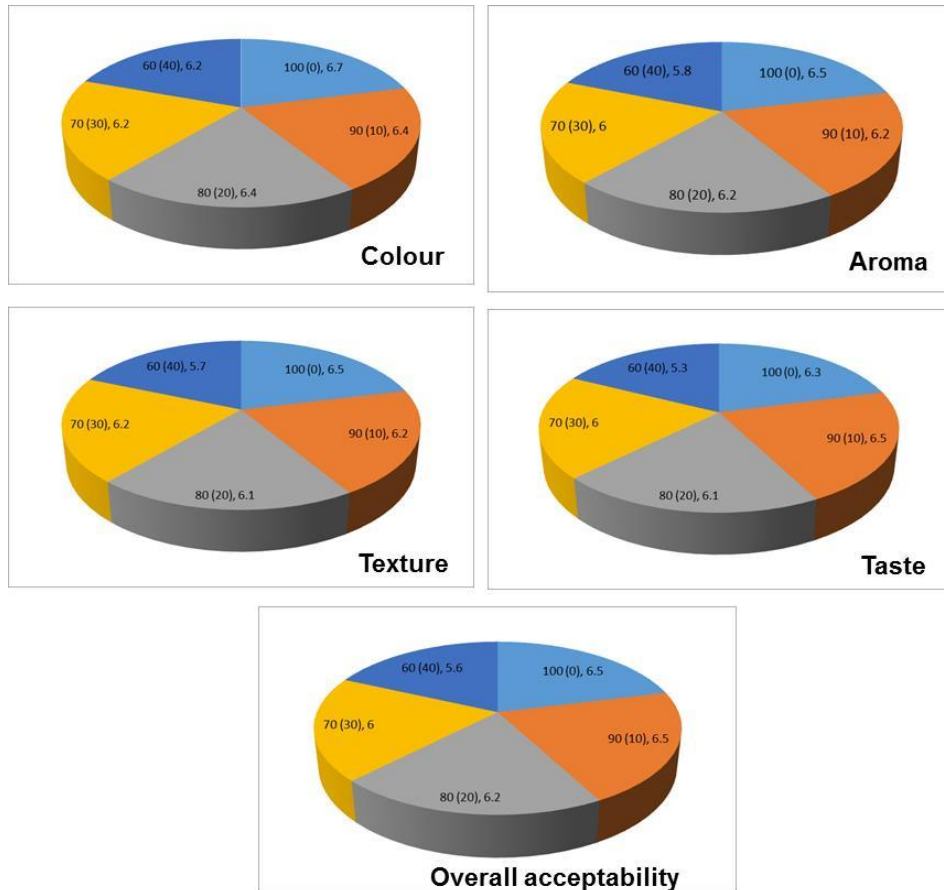


Fig. 2. Plot of mean sensory scores of biscuit from wheat-pro-vitamin A cassava composite Flour

Values before the bracket: Percentage of wheat flour
 Values in the bracket: Percentage of pro-vitamin A cassava
 Values after the bracket: Mean sensory scores

Conclusions

Biscuits with improved nutritional and sensory qualities can be prepared from a mixture of pro-vitamin A cassava flour and wheat flour at ratio 10 to 90 respectively. Functional properties of wheat flour was affected by addition of pro-vitamin A cassava flour. The formulated biscuits are good sources of proteins, energy and carotenoids, which can be potentially used to address protein-energy malnutrition and vitamin A deficiency among vulnerable groups especially in developing countries.

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BOLESTI OKA I TERAPIJSKI UČINAK MEDA

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Pregledni rad

Sažetak

Čovjek med koristi više od 8000 godina, kako u prehrani tako i u terapijske svrhe. U terapijske svrhe med se može koristiti od bolesti oka kao što su: konjunktivitisi, ulkusi rožnice, katarakta, glaukom, trahom i dijabetička retinopatija. Vitamini u medu imaju veću farmakološku vrijednost u odnosu na vitamine koji se dobiju sintetičkim putem. Biogene tvari iz meda pojačavaju djelovanje vitamina kao i ljekovito djelovanje meda (enzimi, fitohormoni, mikroelementi). Literaturni podaci ukazuju na brojne potencijale meda u liječenju bolesti oka. Primjena meda kod bolesti oka je raznolika, od sindroma suhog oka, u terapiji konjunktivitisa rezistentnih uzročnika kao i u terapiji katarakte gdje primjena meda usporava procese zamućenja leće. Radovi koji ukazuju na primjenu meda kod glaukoma, otvaraju novo poglavlje u pristupu liječenja ovoj bolesti oka. Promjena boje dužice nakon primjene meda u vidu kapi, širi indikacijsko polje u primjeni meda, kada je oko u pitanju. Na kraju se može zaključiti kako med kao pluripotentna kemijska tvar, kao hrana i kao lijek, se može primijeniti u terapiji različitih bolesti oka i kao takav može imati prednost u odnosu na sintetske kemijske tvari koje se koriste u liječenju bolesti oka.

Ključne riječi: med, bolesti oka, suho oko, katarakta, glaukom

Uvod

Prvi podaci o upotrebi meda, od strane čovjeka, potiču tisućama godina unazad. Prvi pisani tragovi o upotrebi meda nalaze se na stijinama u Valenciji (Španjolska) i datiraju od prije 8000 godina (Slika 1). Arheolozi su otkrili zdjele s medom u egipatskim grobnicama faraona, gdje je med sačuvao svoja svojstva i bio za upotrebu i nakon toliko godina. Još je Aristotel (350 godina p.K.) rekao: „Med je dobar spasitelj za problematične oči“. U narodu se med koristi u liječenju opekotina, otvorenih rana, posjekotina i raznih infekcija kože, a sve ove indikacije se zasnivaju na antimikrobnom djelovanju meda (Kochan, 2013). Airolla P., svjetski poznati nutricionista, ukazuje da med može djelovati protuupalno kod upala rožnice i u drugim stanjima (Airolla, 1978). Med se može upotrijebiti i u liječenju suhog oka, a otopina meda kada se koristi u tu svrhu ima bolje karakteristike nego tzv. umjetne suze. Otopina od 20 % meda u fiziološkoj otopini može se koristiti u tretmanu infekcija oka (Forester i Thompson, 1987). Američko društvo za apikoterapiju je ukazalo na znatno poboljšanje promjena kod ulkusa rožnice nakon upotrebe meda kad ni antibiotici ni antimikotici ni kortikosteroidi nisu mogli pomoći (Albietz i Lenton, 2006). U literaturi se navodi da se med može koristiti u sljedećim bolestima oka: katarakta, konjunktivitis, bol u oku, trahom, glaukom, degeneracija retine i dijabetička retinopatija.



Slika 1. Prvi pisani tragovi o medu na stijinama u Valenciji, Španjolska

Fig. 1. The first written traces of honey on the rocks in Valencia, Spain

Kemijski sastav meda

Med sadrži više od 350 za organizam važnih sastojaka. Na šećere otpada 75 % meda (fruktoza 38 %, glukoza 30 %, saharoza 1-2 % a ostatak su maltoza i ostali disaharidi), voda čini 18 % meda, organske kiseline 0,3 % (jabučna, vinska, limunska, oksalna, mliječna kiselina), enzimi (invertaza, diastaza, katalaza, fosfataza, dehidraza, oksidaza, peroksidaza), minerali 0,2 % (željezo, bakar, mangan, silicij, klor, kalcij, kalij, natrij, magnezij), vitamini B, C, A, K i E kao i fitokemikalije (flavonoidi i fenoli)

koji djeluju kao antioksidansi. Peludna zrnca u medu sadrže bjelančevine (Kochan, 2013).

Vitamini u medu imaju veću farmakološku aktivnost od onih koji se dobivaju sintetičkim putem, a biogene tvari iz meda pojačavaju djelovanje vitamina iz meda kao i samo njegovo ljekovito djelovanje (enzimi, fermenti, fitohormoni, mikroelementi) (da Silva i sur., 2016).

Cilj rada bio je ukazati na upotrebu meda, kao alternativnog terapijskog sredstva, u različitim bolestima oka kao što su: suho oko, infekcije oka, katarakta i glaukom. Upotreba meda kod želje za promjenom boje očiju od strane pacijenta, predstavlja još jedan u nizu učinaka meda na tkivo oka, odnosno dužicu.

Upotreba meda kod sindroma suhog oka

Studija koja je provedena u Brisbane-u, Australija, imala je za cilj ukazati na terapijski učinak meda kod pacijenata koji su koristili med kao otopinu u vidu tzv. umjetnih suza u odnosu na kontrolnu grupu pacijenata koji su tretirani konvencionalnim umjetnim suzama (Albietz i Lenton, 2006). Prvu grupu pacijenata tretiranih medom činili su pacijenti koji nisu mogli biti tretirani umjetnim suzama zbog nošenja kontaktnih leća ili zbog alergijske reakcije na benzalkonij hidroklorid (BAK), konzervans koji je u podlozi većine oftalmoloških preparata. Drugu grupu pacijenata u ovoj studiji činili su pacijenti koji su bili tretirani uobičajenim terapijskim postupkom umjetnim suzama i ova grupa pacijenata je imala češće infekcije oka u odnosu na prvu grupu. Tretman pacijenata s medom je trajao tri mjeseca i za taj period je primijećeno da je došlo do poboljšanja nekoliko parametara koji se tiču površine oka (kao npr. pojačano lučenje suza, smanjena hiperemija konjunktive, promjena denziteta goblet ćelija rožnice i dr.). Incidencija konjunktivalne infekcije kod sindroma suhog oka u pacijenata tretiranih medom je bila znatno manja u odnosu na grupu pacijenata koji su bili tretirani na uobičajeni način (Albietz i Lenton, 2006; Albeitz i Lenton, 2015).

Primjena meda u tretmanu sindroma suhog oka po principima tzv. *ayurveda* medicine (shushkakshipaka-sindrom suhog oka) preporučuje se kod pacijenata gdje drugi vidovi tretmana nisu mogući za provesti u cilju postizanja i subjektivnih i objektivnih simptoma poboljšanja ovog sindroma (Dhiman, 2011).

Upotreba meda kod upalnih bolesti oka

U grupu upalnih bolesti oka, a koja se mogu tretirati medom, prema dostupnoj literaturi, spadaju:

konjunktivitisi, blefaritisi i ulkusi rožnice. Antibakterijski učinak meda se zasniva na izostanku pojave rezistencije bakterija na ovo terapijsko sredstvo za razliku od terapije antibioticima (Levy i Marshall, 2004). Karakteristike koje čine med tako moćnim antibakterijskim čimbenikom se zasniva na sljedećem: nizak pH, visoka osmolarnost, niski stupanj hidrofilnosti, što omogućava prirodni put nastanka vodikovog peroksida. Med zaustavlja rast bakterija na staničnoj razini (Cernak i sur., 2012). Treba istaknuti da sve vrste meda nemaju iste karakteristike pa prema tome imaju i različite stupnjeve antibakterijskog djelovanja i mogu se svrstati u one koji djeluju kao inhibitorni čimbenici do onih koji imaju antimikrobnu efikasnost (Mulu i sur., 2004).

Stafilokok je najčešći uzročnik infekcija, ali i najteže ga je liječiti (primjer MRSA infekcije metilicilin rezistentnim tipom stafilokoka). Kad se ove bakterije razmnože u oku, one na površini stvaraju barijeru poznatu kao biofilm. Postojeći antibiotici teško probijaju ovu barijeru, dok za med biofilm nije prepreka jer 85 % bakterija nestaje poslije tretmana medom. Mehanizam terapijskog djelovanja meda se sastoji u sprečavanju vezivanja biofilma sa fibronektin-proteinom koji nastaje na površini humane stanice uslijed upale ili opekotine (Al-Waili, 2004).

Med ima pH od 3,2 do 4,5 što ga čini izuzetno kiselim. Mnoge bakterije ne podnose ovakav niski pH kao na primjer *E. coli*, *Salmonella sp.*, *Streptococcus sp.*, koje traže pH od 4,0 do 4,5 da bi se mogle razmnožavati. S druge strane, voda iz meda je slabo raspoloživa za bakterije jer 15 - 21 % vode iz meda je vezano za molekule šećera, pa jako malo vode ostaje slobodno za rast i razvoj bakterija. Ova slobodna voda, poznata kao „aktivna voda“ za med se kreće od 5,6 do 6,2 a za rast bakterije traže aktivnu vodu u rasponu od 9,4 do 9,9 (Kochan, 2013).

Med sadrži enzim glukoza-oksidadazu, čijom aktivnošću nastaje vodikov peroksid ukoliko se med rastvori u vodi i čijim oslobađanjem nastaje antiinfektivni i antiseptički učinak meda. Ukoliko se med koristi u nerazblaženom obliku, može doći do oštećenja tkiva (djelovanje kiseline na tkivo). Upravo iz gore navedenog, med razblažen sa vodom, predstavlja idealno sredstvo za liječenje infekcija oka (Kochan, 2013).

Ispitivanje terapijskog učinka meda u tretmanu vernalnog konjunktivitisa u studiji koja je provedena u Iranu (Salehi i sur., 2014) potvrdilo je terapijski učinak meda i potvrđeno je manje izraženo crvenilo očiju, smanjenje eozinofilije kao i smanjenje papila koje se javljaju kod ove bolesti u pacijenata koji su koristili med u terapiji. Ali je kao nuspojava

primijećeno kako je u grupi koja je koristila med došlo do blagog povećanja intraokularnog tlaka koje je bilo prolaznog karaktera.

Al-Waili (2004) u svom radu navodi da se upotreba meda razrijeđenog sa destiliranom vodom u omjeru 1:1 preporučuje kod različitih infekcija oka kao što su: blefaritisi, maibomianitisi, halacioni, hordeolumi, konjunktivitisi.

U studiji Černaka i autora (2012) upotreba meda se ispitala u profilaksi postoperativnog endoftalmitisa s ciljem potvrde antimikrobnog učinka meda kao i sredstva za poboljšanje zarastanja rane. Grupa pacijenata koji su tretirani predoperativno sa medom (N=49) i broj pacijenata tretiranih sa ofloksacinom (N=52) i nakon 7 dana primjene terapije nije bilo značajne razlike u antibakterijskom učinku među grupama.

Važno je napomenuti da med zajedno sa ostalim antibioticima ne mijenja njihovo djelovanje i ne dolazi do neželjenih interakcija između samih terapijskih sredstava ukoliko se istovremeno primjenjuju (Boateng i Diunase, 2015).

Upotreba meda kod katarakte

Istraživanje ruskih autora pacijenata sa kataraktom tretiranih medom, za period od 7 godina praćenja takvih pacijenata, utvrdilo je da u 55,9 % pacijenata nije došlo do pogoršanja vidne oštine. Čak je bilo registrirano i blago poboljšanje vida za jedan red na optotipu (Golychev, 1990). Vjeruje se da flavonoidi iz meda utječu na leću u oku i djeluju kao antioksidansi (Isaacs, 2013).

U nekim zemljama (Meksiko, Polinezija) otopina meda se koristi kao sredstvo za prevenciju nastanka katarakte, kao dio kulturnog nasljeđa (Traynor, 2015).

Med od eukaliptusa (manuka med) prednjači u karakteristikama u odnosu na ostale vrste meda kad je u pitanju primjena u mogućem tretmanu katarakte jer je sadržaj vitamina C, kalcija i natrija drugačiji u odnosu na ostale vrste meda. Med ne liječi kataraktu već usporava progresiju zamućenja leće pa se med u terapiji katarakte preporučuje kao moguće sredstvo u usporavanju procesa koji dovode do katarakte (Find Home Remedy, 2016; Singh, 2014; Beck, 2013; Albietz i Lenton, 2015).

Upotreba meda kod glaukoma

U literaturi se upotreba meda u terapiji glaukoma uglavnom zasniva na principima tzv. *ajurveda* medicine, gdje primjena prirodnih preparata ima prednost u odnosu na sintetičke lijekove. Stoga se, u cilju smanjenja vrijednosti intraokularnog tlaka i

poboljšanja protoka očne vodice, preporučuje primjena rastvora meda s kamforom, đumbirom i limunom, u vidu kapi kod pacijenata s glaukomom (Lee, 2016; Always Ayurveda, 2016).

Japanski autori su eksperimentalno postavljali saće sa medom zečevima u vidu adhezivne barijere kod operativnih filtracionih zahvata glaukoma. Saće s medom koje je postavljano u unutrašnjost filtracionog jastučeta koje se napravi prilikom operacione tehnike rješavanja glaukoma u zečeva, bilo je praćeno sa manjim vrijednostima intraokularnog tlaka kao i s manjim postotkom upalnih postoperativnih komplikacija u odnosu na drugu grupu zečeva koji su tretirani mitomicinom kod filtracionih operacija glaukoma (Okuda i sur., 2009).

Upotreba meda u cilju promjene boje očiju

Med je poznat kao prirodni izbjeljivač i upotrebljava se kod izbjeljivanja tamnih fleka po koži. Može se koristiti i za izbjeljivanje kose i kao lijek za alergiju na koži kapaka (Shenoy i sur., 2009).

Prema anketi koja je boja očiju najatraktivnija, dobiveni su sljedeći odgovori: na prvom mjestu zelene 20,3 %, a na zadnjem mjestu po stupnju atraktivnosti su bile oči smeđe boje 5,9 % (Wikia, 2016). Mogući učinak promjene boje očiju nastaje uslijed razlike pH oka i meda. Čovjekovo oko ima pH 7,5 a pH meda oko 3,9 pa razlika u pH vrijednostima dovodi do promjene aktivnosti melanocita što ima za posljedicu inhibiciju melanina. Međutim, treba imati na umu da ukoliko se med primjeni u nerazblaženom obliku, s ciljem dobivanja svjetlije boje dužice, med može izazvati neželjene učinke na oku u vidu opekotina (Wikia, 2016).

Zaključci

Prema podacima iz literature, med se koristi u gastroenterološkim, kardiovaskularnim, upalnim bolestima. Antioksidativni učinak tvari iz meda ima vrlo važnu ulogu u terapiji i indikacijama-od različitih vrsta bolesti do fiziološkog stanja kao što je starenje.

U oftalmološkoj praksi vrlo često se i danas srećemo s podacima pacijenata o tradicionalnim načinima liječenja, a u cilju dobivanja terapijskog učinka, bilo da su u pitanju upalni procesi na oku ili promjene u vidu zamućenja leće kao što je katarakta.

Primjena nekonvencionalnih preparata, kao što je na primjer upotreba meda, sa znanstvenog i istraživačkog aspekta, nastaje kao način izbjegavanja mogućih neželjenih učinaka dugotrajne upotrebe kemijskih tvari kao što su na primjer kortikosteroidi.

Primjena meda od strane pacijenata u liječenju bolesti oka ima prvenstveno financijski aspekt a i utjecaj tradicionalne medicine se ne može zanemariti.

Podaci iz literature, već dugi niz godina, skreću pažnju na značaj i primjenu meda kod različitih vrsta bolesti, ali i na značaj primjene meda u prevenciji bolesti i stanja čovjekovog organizma. U modernoj medicini treba naći mjesto za potvrdu starih tradicionalnih postupaka koje mogu biti potvrđene ili potpuno isključene u terapiji bolesti oka. Za znanstveno dokazani terapijski učinak potrebno je provesti dobro osmišljenu studiju s ciljem dobivanja vlastitih rezultata kad je u pitanju primjena meda kod bolesti oka, bilo da su to promjene na koži kapaka ili želja za promjenom boje očiju.

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DISEASES OF THE EYE AND THE THERAPEUTIC EFFECT OF HONEY

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Review paper

Summary

Honey has been used by man more than 8000 years, both as a food or for therapeutic purposes. For therapeutic purposes honey is used to treat eye diseases, such as: conjunctivitis, ulcers of the cornea, cataract, glaucoma, trachoma and diabetic retinopathy. Vitamins in honey have a greater value in relation to the pharmacological vitamins produced synthetically. Biogene components from honey increase the activity of honey's vitamin, and also enhance healing effects of honey (enzymes, microelements, fitohormoni). Literature data show the number of beneficial effects of honey in treatment of eye diseases. Application of honey in the treatment of eye diseases are diverse, from dry eye syndrome, the treatment of conjunctivitis cause with pathogen resistant and in the treatment of cataracts, where the application of honey slows down the processes blur of the lens. Studies that indicate the application of honey on glaucoma, open up a new chapter in the approach to the treatment of this disease of the eye. Change of the color of the iris after applying honey in the form of drops widens the field of application. In the end, honey as pluripotent chemical substance, as food and as medicine, can be applied in the treatment of eye diseases and as such can have an advantage in relation to other synthetical chemical substances that are used in the treatment of eye diseases.

Keywords: honey, diseases of the eye, dry eye, cataracts, glaucoma

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