

Effect of Annealing Treatment on Resistant Starch Content and Prebiotic Properties of High-Carbohydrate Foods: Meta-Analysis Study

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Abstract: Annealing treatment is a physical modification technique widely used to increase resistant starch levels in foodstuffs. Annealing technique (ANN) is a modification carried out using a large concentration of water (more than 40%) and given heat treatment below the starch gelatinization temperature. This study aims to analyze the types of carbohydrate foods that significantly affect the content of resistant starch and prebiotic properties by means of annealing treatment techniques. The PRISMA guidance method was used, which used 1038 researched and selected articles to produce 30 articles. The data is formulated based on the percentage value of the Hedges'd Effect Size (standardized mean difference/SMD) and the confidence interval (CI) value using the help of OpenMEE software. The meta-analysis revealed that the annealing technique significantly increased the resistant starch content and prebiotic properties in high-carbohydrate foods (SMD 15.289; 95% CI: 11.860 to 18.719; $p < 0.001$). This meta-analysis study concluded that the annealing treatment technique significantly improved based on a 95% confidence level to increase resistant starch content and prebiotic properties in high carbohydrate foods.

Keywords : annealing treatment; meta-analysis; prebiotic properties; resistant starch; starch modification

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1. Introduction

The starch fraction that digestive enzymes cannot digest is resistant starch which has resistant properties to stomach acid to reach the large intestine to be fermented by probiotic bacteria [1]. Resistant starch is a fraction that cannot be digested in the small intestine. It has physiological effects that are beneficial to health, such as colon cancer prevention, and has hypoglycemic and hypocholesterolemic effects [2]. Starch is categorized based on its digestibility by a starch-breaking enzyme, namely the amylase enzyme, to be converted into simpler parts (simple sugars) because starch is only composed of amylose and amylopectin [3].

Physical modifications generally have a principle of heating. Physical modification techniques include parboiling, extrusion, roasting, microwave irradiation, steam cooking, and hydrothermal [4]. Physical modifications are said to increase the levels of resistant starch [5]. Physical treatment that is still under-researched is the annealing method. Annealing technique

(ANN) is a modification carried out using a large concentration of water (more than 40%) and given heat treatment below the starch gelatinization temperature. Annealing using heat and water makes annealing a physical treatment for starch granules. During ANN, starch granules will have excess weight (>60% w/w) with intermediate moisture content (40% w/w) and are maintained at a temperature below gelatinization for a certain period [4,5,6].

Physical changes of starch with annealing have been reported, including (1) increased granule stability; (2) crystal perfection; (3) Starch chains can interact in granular crystals, and amorphous domains; (4) the formation of a helical double chain; (5) increase in gelatinization temperature; (6) narrowing of the gelatinization temperature range; (7) reduction of granular swelling; and (8) decreased amylose leaching [5,7,8]. However, each change depends on the starch source, crystallinity, amylose-lipid interaction, and susceptibility to acid. Enzyme hydrolysis has been shown to decrease due to annealing modification [1].

The annealing treatment for fixing starch has been extensively studied [9,10]. Generally, retrogradation occurs after the temperature is below gelatinization for a certain period, affecting starch digestibility [11]. However, many studies using annealing techniques in food have produced different prebiotic properties and resistant starch content. This meta-analysis study may contain updated information on improving prebiotic properties and resistant starch content in annealed foodstuffs. This study aims to analyze the types of carbohydrate foods that significantly affect the content of resistant starch and prebiotic properties by means of annealing treatment techniques.

2. Materials and Methods

2.1. Materials.

References in the meta-analysis study are articles from reputable international journals accredited by Scopus from online web servers publishers such as Taylor & Francis Online, Springer Link, Google Scholars, Wiley Online Library, and Science Direct. The tools used were Zotero [version 5.0.97 (2021), Mendeley software [version 1.19.8 (2020), Microsoft Excel [version 16.53 (2019)], and OpenMEE software [version 10.10 (2020)] used to analyze the data.

2.2. Library search strategy.

Literature analysis and selection were carried out by following the Preferred Reporting Items for Systematic Review and Meta-Analyses (PRISMA) rules, a series of analysis processes to select the desired library. The PRISMA analysis method can simplify the process of selecting research articles. The process of selecting articles is carried out in several stages using parameters: text completeness, title and duplication, abstract, and method. Searching and selecting references through the web server database Science Direct, Wiley Online Library, Taylor & Francis Online, Springer Link, and Google Scholars using keywords "physical modification of starch, annealing treatment, and prebiotic properties". Three keywords are combined using Boolean operators with expressions "and" by adding the choice of a year of publication (2011-2021) to narrow the search.

2.3. Study library selection.

Research literature was selected based on the screening of titles, and abstracts were analyzed to determine suitability with the selected inclusion and exclusion criteria. Inclusion criteria are a way of selecting reputable and internationally accredited articles. The selected articles are studies with data published within the last ten years (2013-2022). There is data on resistant starch content before the annealing technique is carried out (control data). After the annealing, the technique is carried out (experimental data), which is then limited to research using annealing techniques. Exclusion criteria by looking at the research results using starch processing methods, analysis of prebiotic properties, and additional treatment methods such as annealing treatment (autoclaving, linearization, and microwave cooling).

2.4. Data collection.

Research data from selected references from the web server of Scopus-indexed international journals were analyzed using Zotero software. Data from Zotero analysis was then extracted into a Microsoft Excel worksheet. Data were collected based on the type of food, the mean and standard deviation of control and experimental resistant starch content, the author's name, and the year of publication.

2.5. Statistical analysis.

Hedges'd effect (Standardized Mean Difference/SMD) with a 95% confidence interval (confidence interval value) was used in the data analysis process. OpenMEE software is used to process Effect Size data [12]. Data were collected from the selected references used for the meta-analysis: the number of trial repetitions, the average, the standard deviation, and the standard error. SMD analysis results, with corresponding 95% CI, were pooled using a random-effects model. Exploration of heterogeneity between studies was carried out using an index of I^2 ($I^2 > 50\%$ indicates good heterogeneity) [12]. Moderator variables for the sub-group analysis were prebiotic properties, diet, and site. The OpenMEE software is used to process and assist in the interpretation of meta-analysis data so that it can produce Forest Plot output.

3. Results and Discussion

3.1. Library selection.

Based on the results of library selection, a database of 1.038 libraries was obtained. Overall, the libraries were entered into the Zotero software to remove the duplicated libraries and get 402 libraries, followed by a selection based on the abstracts obtained from 97 libraries. A total of 28 references did not explain the evaluation of prebiotic properties, and quantitative data in resistant starch content before modification (control data) and after modification (experimental data) were eliminated from the meta-analysis study. Library selection is also based on literature published in reputable international databases. The selected literature is the result of primary data research published within the last ten years (2013-2022) and is limited to articles using annealing techniques. The 30 libraries used as relevant material in meta-analytic research can be seen in Figure 1.

3.2. Data analysis.

Analysis of data on resistant starch levels for each selected library obtained 14 data. You can see a summary of the data for each study in table 1. The whole data is then processed in the OpenMEE worksheet to determine the effect size, p-value, and heterogeneity value (I^2). The effect size value is defined as the value of Hedges'd analysis (Standardized Mean Difference/SMD) used to analyze the effect of the related treatment. Effect size values for each research parameter were further analyzed using OpenMEE software. The Effect Size value determines the result of the combined effect measurement with a 95% confidence interval (CI) and a significance level of 0.05.

The annealing treatment was carried out using a large water concentration (more than 40%). It was given heat treatment at a temperature below the starch gelatinization temperature, causing the pasta viscosity to increase and undergo a retrogradation process [11]. Retrogradation can increase the level of RS type 3. The hypothesis tested in a meta-analysis study was that the more significant the increase in resistant starch levels, the higher the effect of modified annealing (ANN) techniques on foodstuffs.

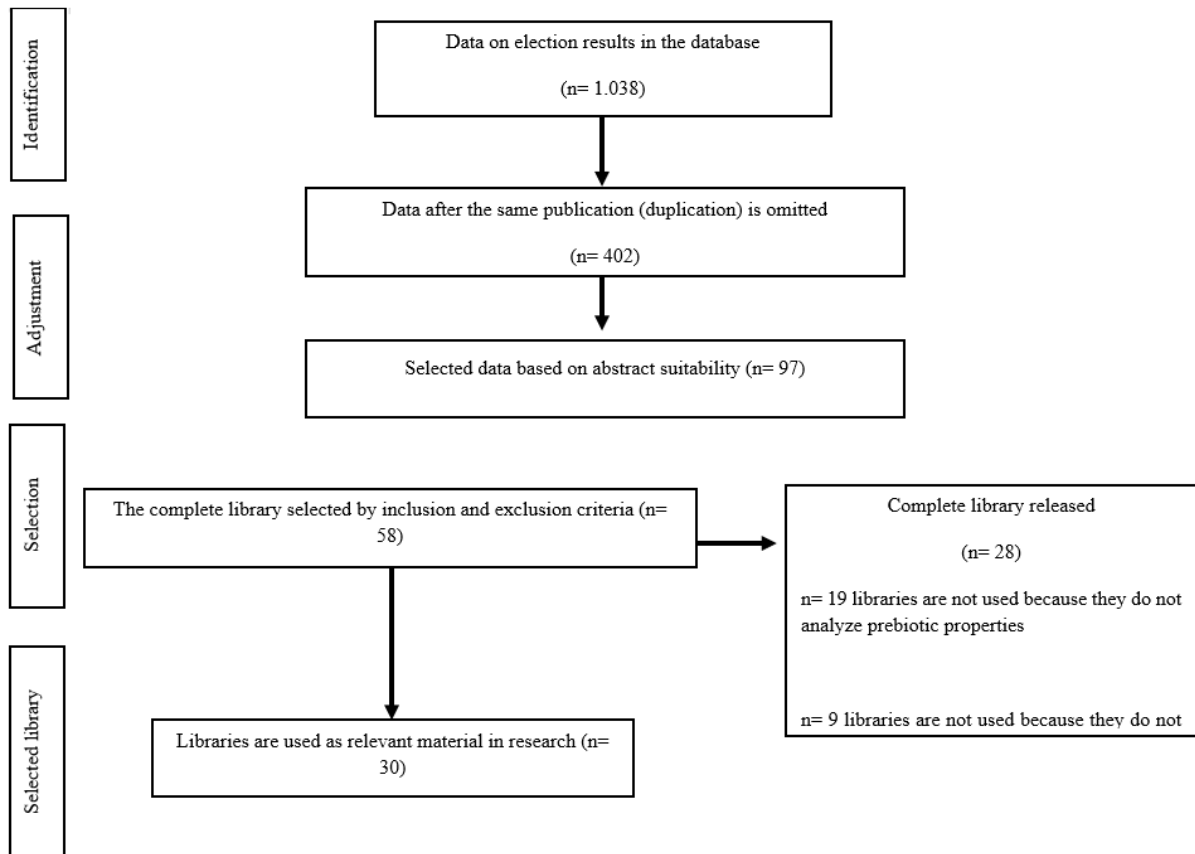


Figure 1. The process of selecting literature for further meta-analysis.

Table 1. Transformation data of resistant starch content in foodstuffs.

No.	Foodstuffs	(%) Control resistant starch	(%) Resistant starch after modification	(%) Changes in resistant starch	Literature
1	Rice	43.61	56.9	13.29	[13]
2	Bean seed	15.32	35.66	20.34	[1]
3	Wheat	71.72	94.03	22.31	[4]
4	Rice	17.32	48.7	31.38	[14]
5	Bean seed	7.45	10.2	2.75	[15]
6	Maize starch	7.41	7.85	0.44	[5]
7	Corn starch	4.34	26.81	22.47	[6]
8	Wheat	21.78	43.17	21.39	[9]

No.	Foodstuffs	(%) Control resistant starch	(%) Resistant starch after modification	(%) Changes in resistant starch	Literature
9	Wheat	10.41	68.96	58.55	[16]
10	Banana	12.04	42.78	30.74	[17]
11	Rice	5.38	8.39	3.01	[18]
12	Bean seed	18.3	30.77	12.47	[10]
13	Buckwheat	5.39	36.52	31.13	[19]
14	Bean seed	11.02	37.4	26.38	[20]
15	Maize starch	37.34	62.66	25.32	[21]
16	Banana	10.5	24	13.5	[22]
17	Wheat	2.67	14.67	12	[7]
18	Corn starch	3.71	14.02	10.31	[23]
19	Tubers	9.42	29.1	19.68	[8]
20	Pea starch	4.39	10.43	6.04	[24]
21	Corn starch	12.44	24.39	11.95	[25]
22	Cassava	7.82	42.36	34.54	[26]
23	Rice	22.17	44.94	22.77	[27]
24	Rice	7.68	23.31	15.63	[28]
25	Buckwheat	33.5	53.63	20.13	[29]
26	Rice	2.42	7.43	5.01	[30]
27	Ferox starch	18.44	86.02	67.58	[31]
28	Tubers	14.9	20.67	5.77	[32]
29	Rice	15.96	49.8	33.84	[3]
30	Wheat	13.71	82.04	68.33	[33]
		Average resistant starch content of control (n= 30) 15.61%			
		The average resistant starch content after modification (n= 30) 37.92%			
		The average increase in resistant starch content (n= 30) 22.30%			

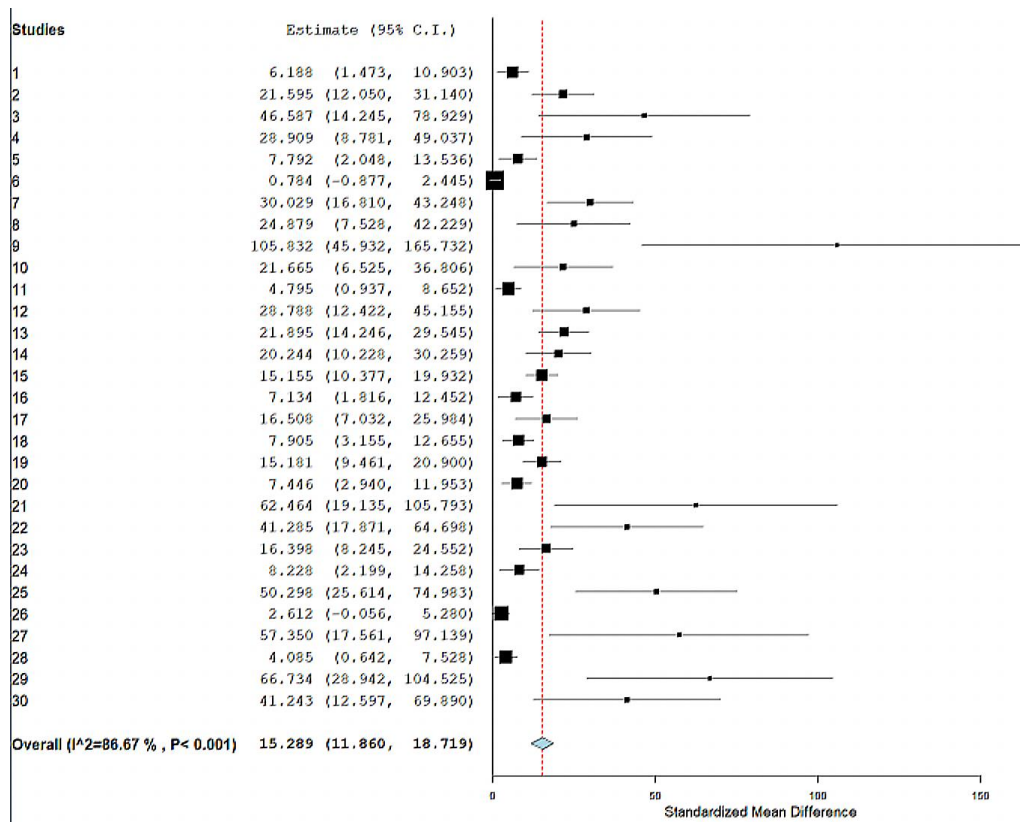


Figure 2. Forest plot of the results of the meta-analysis study of all data.

The annealing process significantly affects the increase in resistant starch content, as evidenced by the results of a forest plot meta-analysis using OpenMEE software (Figure 2). This is indicated by the combined SMD effect value of 15.289 with 95% CI (11.860 to 18.719) $p < 0.001$ and the heterogeneity value (I^2) of 86.67. The meta-analysis used Continuous Random-Effects Model analysis to see differences between one study and another. So, it requires a heterogeneity value to see the diversity between the analyzed studies. A meta-

analysis study can be good if it has a heterogeneity value close to 100% [34]. The higher the heterogeneity value between studies, the more heterogeneous and can represent the diversity of the data for each analysis.

3.3. The effect of differences in carbohydrate foods on increasing levels of resistant starch.

Table 1 presents 30 study data that reported increased resistant starch levels after the modified annealing technique. A total of 22.30% of study data reported an increase in the level of resistant starch. A total of 11 carbohydrate foods were used in the study, each of which gave different results of increased levels of resistant starch showed before or after the modified annealing technique.

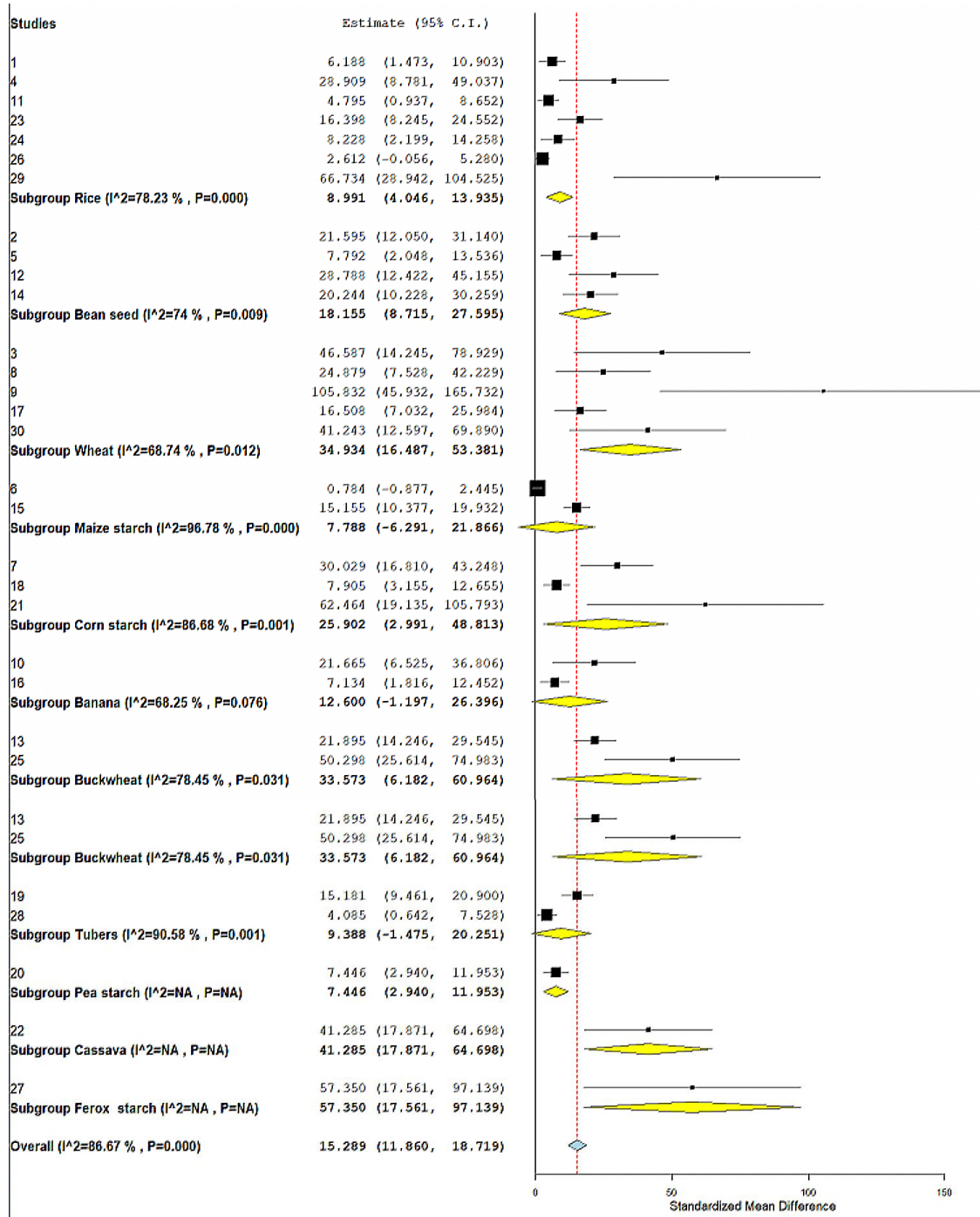
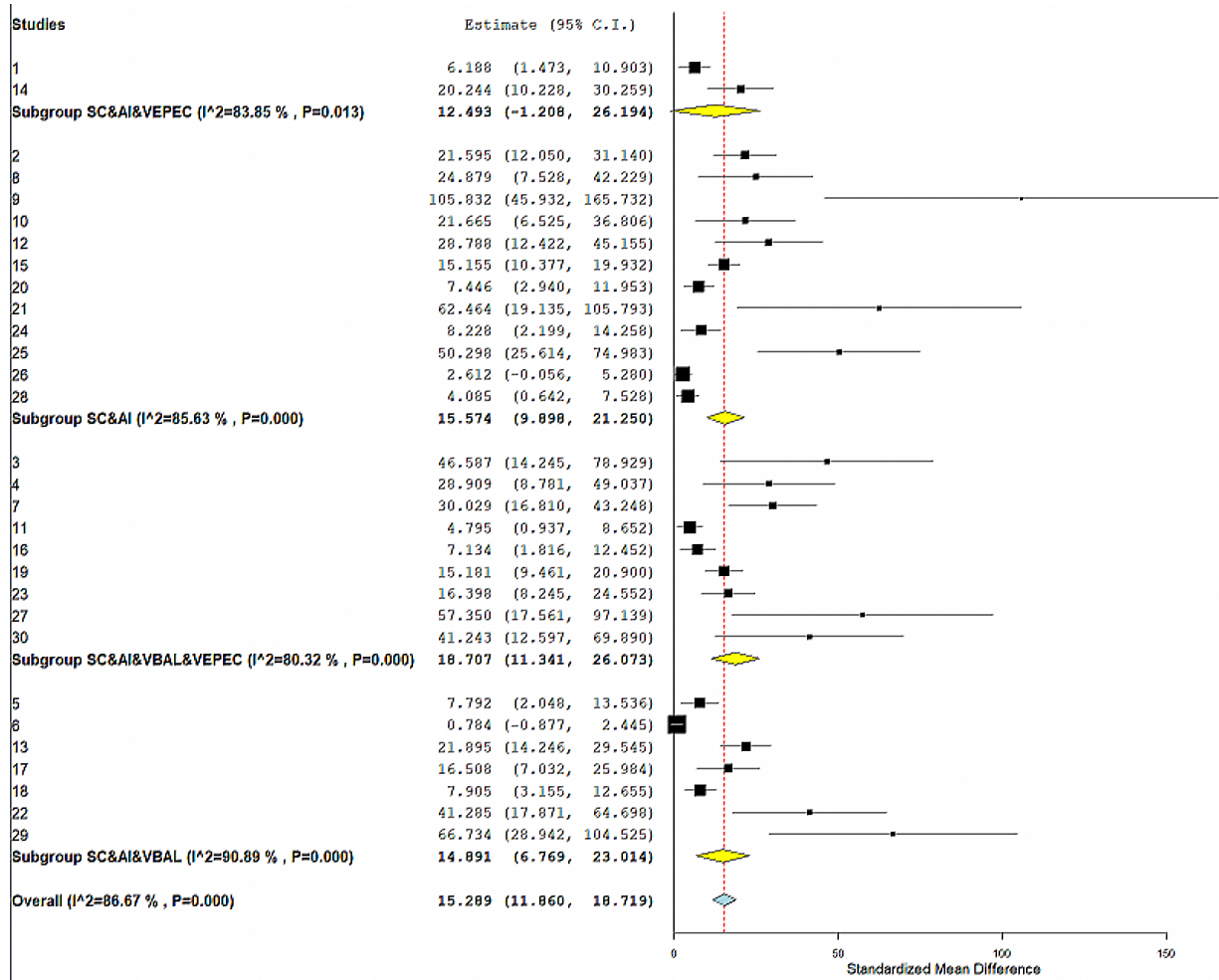


Figure 3. Forest plot meta-analysis of the effect of various types of carbohydrate foods on increasing levels of starch resistance.

Annealing increases the RS content of natural starch. Research on RS content of natural starch has been investigated exclusively from wheat (4.6%), peas (10.0%), and lentils (9.1%) significantly increased by 8.7% (wheat), 11.2% (peas), and 11.4% (lentils) [16]. This rate of change is more pronounced due to the greater degree of crystal perfection and the formation of short amylose chains [35]. Meanwhile, the increase in RDS levels in ANN may be due to forming a more porous structure [36]. The formation of a porous structure will allow greater access of hydrolytic enzymes to the granular interior. ANN modification of lentils has increased the RS content from 7.3% to 19.3% [15].

The difference in the increase in levels of resistant starch was caused by differences in the levels of amylose and amylopectin in foodstuffs [22]. The starch modification process affects starch digestibility, the composition of amylose and amylopectin ratios, the degree of starch retrogradation, starch gelatinization, fibre content and reducing sugar content [26]. The forest plot data from the analysis of different types of carbohydrate foods are presented in Figure 3. The forest plot results show that high-carbohydrate foods have a significant effect on increasing levels of resistant starch with the SMD effect value of 15.289 with 95% CI p<0.001 and a high heterogeneity value (I^2) of 86.67. The results of the forest plot for the carbohydrate food sub-group data stated that, in general, the overall annealing treatment for foodstuffs had a significant effect on increasing levels of high-carbohydrate food-resistant starch.



Information:

AI: Amylose Interaction SC: Starch Composition
 VEPEC: Viability EPEC VBAL: Viability BAL

Figure 4. Forest plot meta-analysis of studies of the effect of dietary carbohydrates on prebiotic properties.

3.4. Effect of prebiotic properties on carbohydrate food ingredients.

An in-depth analysis was carried out to determine the effect of carbohydrate food on the prebiotic properties. Each test of prebiotic properties is divided into the amylose interaction (AI) starch composition (SC), viability of *Enteropathogenic Escherichia coli* (VEPEC), viability of lactic acid bacteria (VBAL), in the literature are analyzed and obtained forest plot data can be seen in Figure 4.

Food has good prebiotic properties if it is metabolized selectively by probiotic bacteria such as *Lactobacillus plantarum*, *L. acidophilus*, and *Bifidobacterium* sp. but is not metabolized selectively by pathogenic bacteria such as EPEC [16,19,37]. Based on the forest plot results, the carbohydrate food type significantly affected prebiotic properties of food ingredients with an SMD effect value of 15.289 with a CI of 95%. (11.860 to 18.719) $p < 0.001$. The value of the effect size of food ingredients has a proportional influence on the prebiotic properties. The effect size value describes the average distribution of each meta-analytical study [38].

4. Conclusion

This meta-analysis study concluded that the annealing treatment significantly increased the levels of resistant starch and prebiotic properties of high carbohydrate foods with a 95% confidence level. The annealing technique for carbohydrate foods is a recommendation to increase the levels of resistant starch that can be done on a home or factory industrial scale. Various starchy foodstuffs have different physicochemical components, such as the ratio of amylose and amylopectin and the degree of polymerization, so further research is needed on a laboratory scale to increase the levels of resistant starch by using annealing treatment.

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Conflicts of Interest

In the preparation of the article the authors stated that there was no conflict of interest.

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